The Effect of Plasma TriglycERide Lowering Therapy on Organ FunctiOn in HypertRiglyceride Mia-induced Acute Pancreatitis: a multicenter, registry-based, observational study (PERFORM)

Protocol and Statistical Analysis Plan Amendment History

1. Original protocol, final protocol, summary of changes

The Effect of Plasma TriglycERide Lowering Therapy on Organ FunctiOn in HypertRiglyceride Mia-induced Acute Pancreatitis: a multicenter, registry-based, observational study (PERFORM)

Protocol Ver:1.0 dated Dec.2019

Primary Investigator :Professor. Weiqin Li

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SUMMARY INFORMATION TYPE	SUMMARY DETAILS
Acronym (Short Title)	PERFORM
Long Title	The Effect of Plasma Triglyceride Lowering Therapy on Organ Function in Hypertriglyceridemia-induced Acute Pancreatitis: a multicenter, registry-based, observational study
Version	1.0
Date	10th December 2019
Trial registration	ChiCTR2000039541
Study Design	A multi-centered, register-based, observational study
Type of Participants to be studied	Patients aged 18 to 70 years older who were diagnosed with hypertriglyceridemia-induced acute pancreatitis (See the study population in detail)
Setting	estimated 30-40 hospitals across China
Study Aim	This study aims to evaluate the TG-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.
Primary Outcomes Measure(s)	organ failure-free days (OFFDs) to 14 days after enrollment.
Secondary Outcome Measure(s)	Part I: Secondary outcomes during the index admission 1. New-onset organ failure; 2. New-onset multiple-organ failure (MOF); 3. New-onset persistent organ failure (POF); 4. New receipt of organ support; 5. Requirement of ICU admission; 6. ICU free days to day 14; 7. Hospital free days to day 14; Part II: Secondary outcomes within 60 days after enrollment 1. Mortality censored at 60 days after enrollment; 2. AP severity grade (Based on the Revised Atlanta Classification); 3. Incidence of infected pancreatic necrosis (IPN); 4. Incidence of septic shock; 5. Incidence of abdominal bleeding; 6. Incidence of gastrointestinal fistula.
Number of Participates to be studied	estimated 200 patients
Duration	estimated 18 months

Sponsor	Jinling Hospital of Nanjing University
Chief Investigators	Professor. Weiqin Li

1. Abrreviations

HREC	Hospital Human Research Ethics Committee
CDMC	Coordinating and Data Management Center
AP	Acute pancreatitis
HTG-AP	Hypertriglyceridemia-induced AP
TG	Triglyceride
ASFA	American Society for Apheresis
TPE	Therapeutic plasma exchange
RCTs	randomize controlled trials
OF	organ failure

2. Study Administrative information

2.1 Steering and management committee

The steering and management committee is responsible for the approval of the full protocol, database, and related methods. The members of the committee will also oversee the implementation of the study and play an advisory role.

Members of the steering committee are listed below

Prof. Weiqin Li

Prof. Zhihui Tong

Prof. Lu Ke

Prof. John Windsor

Dr. Jing Zhou

Dr. Longxiang Cao

2.2 Coordinating and data management center

Coordinating and data management centers (CDMC) will be organized before implementation of the current study in the CAPCTG. They are responsible for the day to day management of the trial, assistance for ethic application in each center, protocol and case report form design, online database

design and maintenance, protocol training for the participating centers, data entry and quality control, severe adverse event monitor and notification and data analysis. The CDMC plans to meet before enrollment, three months after initial enrollment and six months after initial enrollment to ensure qualified data entry.

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Dr. Longxiang Cao and all the research nurses and coordinators from the participating centers.

2.3 Writing and publication committee

The writing and publication committee is responsible for drafting the manuscript and submission of the manuscript to adequate journals. The Writing and publication committee will also decide on the authorship of this study. After the conclusion of this study, every participating centers are welcome to submit proposals for post-hoc analysis to the writing and publication committee, which is responsible for reviewing and rating all the proposals for further analysis.

Members are listed below

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2.4 Registration

The PERFORM study was registered at the Chinese Clinical Trials Registry (ChiCTR2000039541) before enrollment.

3. Background and rationale

Acute pancreatitis (AP) is a potentially life-threatening inflammatory disease with multiple etiologies, such as alcohol, gallstones and hypertriglyceridemia. HTG is the third most common cause of AP, accounting for 4-10% of cases globally, and the increasing prevalence of HTG-AP had been reported in recent studies[1-4]. In China, HTG had been the second leading cause of AP, and previous studies showed that HTG-AP patients had a higher risk of severe acute pancreatitis and multiple organ dysfunction syndrome (MODS) than other types of AP[2, 5-7].

Although the pathophysiology underlying HTG-AP remains controversial, it is widely accepted that free fatty acid (FFA) is one of the driving factors[8]. FFA, produced by the hydrolysis of triglyceride (TG), can initiate or worsen the disease by triggering inflammatory reactions, damaging the pancreatic cell, and promoting microvascular thrombosis within the pancreatic tissue[9]. Nawaz et al.[6] found that elevated serum TG levels in AP patients were independently and proportionally correlated with persistent organ failure (POF) regardless of etiology. In an observational study conducted by Lu et al. [10]timely reduction of serum TG during the early phase of HTG-AP was found to be associated with decreased incidence of POF

Over the past years, several attempts had been made to lower serum TG more efficiently during the acute phase of the disease, including medical treatment with insulin and/or heparin, blood purification, and genetic therapy in cases[11]. Medical treatment is convenient and safe and is considered the first-line choice for TG-lowering therapy[4]. Heparin stimulates the release of endothelial lipoprotein lipase into circulation, while insulin activates lipoprotein lipase, thereby increasing the clearance of chylomicrons from plasma[12]. However, the impact of medical therapy on clinical outcomes is uncertain, and an observational study is ongoing to figure it out[13]. Blood purification, especially plasmapheresis, is also widely used as a TG-lowering therapy. Plasmapheresis rapidly removes triglycerides from plasma and is considered one of the most efficient TG-lowering therapies [14]. Technically, it is a therapeutic procedure in which the blood of the patient is passed through a medical device that separates plasma from other components of blood. The plasma is removed and replaced by a replacement solution (e.g., albumin and/or plasma) or a combination of crystalloid/colloid solution[15]. Double filtration plasmapheresis (DFPP) is a semi-selective apheresis method based on a double filter system, which can remove macromolecules

selectively[16]. Both techniques are widely adopted, while plasmapheresis is thought to be more effective in removing FFA [17]. Other blood purification modalities were also reported effective in lowering plasma TG, including hemoperfusion and hemofiltration[18, 19]. A randomized control trial (RCT) reported that high-volume hemofiltration (HVHF) decreased TG levels more efficiently than medical therapy[18].

For the target of TG-lowering therapy, it is regarded that reducing the TG level to 5.65 mmol/L might be clinically sufficient[20]. Lu et al.[10]found that patients with earlier TG levels of < 5.65 mmol/L were less likely to develop POF. However, the optimal TG lowering target and choice of therapies in early HTG-AP are unclear due to the lack of high-quality studies. Given the paucity of evidence in the literature and the variation in the management of HTG-AP, we conducted this multicenter, observational study and built “The effect of plasma triglyceride-lowering therapy on the evolution of organ function in early hypertriglyceridemia-induced acute pancreatitis patients with worrisome features”(PERFORM) registry to evaluate the TG-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.

4. Study Design

4.1 Study aim

The PERFORM study aims to evaluate the association between different TG-lowering therapies and clinical outcomes in HTG-AP patients with worrisome features.

4.2 General study setting

The present clinical study will be performed in 30-40 different hospitals across China. It is a multi-centered, register-based, prospective, observational study.

5. Study population

5.1 Patient recruitment

We are going to collect approximately 200 patients presenting to the participating ICUs across China during a two-year period. Based on the volume of all the participating centers, the aim should

be able to be achieved within the study period.

5.2 Eligibility Criteria

5.21 Inclusion Criteria

1. Informed consent form obtained from the patient or next of kin;
2. Age between 18 to 70 years old;
3. Within 72 hours of onset,
4. With a primary diagnosis of AP based on Abdominal Pain Suggestive of AP, Serum Amylase at Least three Times the Upper Limit of Normal, and/or Characteristic Findings of AP on Computed Tomography or Less Commonly Magnetic Resonance Imaging (MRI) or Transabdominal Ultrasonography According to the Revised Atlanta Criteria[1].
5. When enrolled, TG>1000mg/dL (11.3mmol/L), accompanied by the clinical worrisome features of any one or more of the following[2]:
   1) Signs of hypocalcaemia (calcium levels less than 2.1mmol/L)
   2) Lactic acidosis (Lactate levels more than 2mmol/L and PH＜7.35)
   3) Signs of worsening systemic inflammation (two or more):
      a) Temperature >38.5°C or <35.0°C
      b) Heart rate of >90 /min
      c) Respiratory rate of >20 breaths/min or PaCO2 of <32 mmHg
      d) WBC count of >12,000 cells/mL, <4000 cells/mL, or >10 percent immature (band) forms
   4) Signs of worsening organ dysfunction or multi-organ failure as defined by Modified Marshall scoring system for organ dysfunction ；

5.22 Exclusion Criteria

1. Failure to obtain informed consent;
2. Pregnant or lactating women; or have a pregnancy plan within a month after the study (including male subjects);
3. Researchers' family members who are directly involved in the study;
4. Patients are expected to die within 48 hours after enrollment, defined as patients with norepinephrine usage at a dose of 25 mg/min or more under full-fluid resuscitation, with a systolic blood pressure <90mm Hg and serum pH values <7.0.

The judgment will be made by the treating physician.

5.Additional exclusion criteria for this analysis: Patients undergo any invasive blood purification therapy other than plasmapheresis.

6. Patient management

We recommend all all patients receive standard treatment that follows the "Acute Pancreatitis Treatment Guidelines" issued by the American College of Gastroenterology (ACG) in 2013 and the standard treatment plan for acute pancreatitis provided in the "Evidence-Based Guidelines for the Treatment of Acute Pancreatitis" issued by the International Association of Pancreatology (IAP) and the American Pancreatic Association (APA)[23, 24]. Since the PERFORM study is an observational one, all treatment decisions will be at the discretion of the treating physicians, including the use of different TG-lowering therapies.

7. Outcome measures

7.1 Primary outcome

The primary outcome is organ failure-free days (OFFDs) to 14days after enrollment. Only the final period of OF-free days is included, and patients who have OF at day 14 or died before day14 are assigned to zero OF-free days.

7.2 Secondary outcome

Secondary outcome measures

Part I: Secondary outcomes during the index admission

1. New-onset organ failure;
2. New-onset multiple-organ failure (MOF);
3. New-onset persistent organ failure (POF);
4. New receipt of organ support;
5. Requirement of ICU admission;
6. ICU free days to day 14;
7. Hospital free days to day 14;

Part II: Secondary outcomes within 60 days after enrollment

1. Mortality censored at 60 days after enrollment;
2. AP severity grade (Based on the Revised Atlanta Classification);
3. Incidence of infected pancreatic necrosis (IPN);
4. Incidence of septic shock;
5. Incidence of abdominal bleeding;
6. Incidence of gastrointestinal fistula.

Definition of seconday outcomes:1)An individual SOFA score of 2 or more for the respiration, cardiovascular, or renal system is defined as the presence of organ failure. 2)New-onset organ failure is defined as organ failure that is not present at any time in the 24 h after enrollment. Multiple organ failure is defined as two or more organ failures present at the same time. 3)Persistent organ failure is defined as organ failure that persists for more than 48 h. 4)ICU free days to day 14 after enrollment is defined as the number of days alive and not admitted to an ICU after the patient’ s latest discharge from the ICU before day 14. If the patient is admitted to an ICU on day 14 or dies prior to day 14, ICU-free days will be 0. Hospital-free days to day 14 after enrollment is defined as the number of days alive and not admitted to the hospital after the patient’ s final discharge from the hospital before day 14.

8. Ethics and dissemination

This study has been approved by the ethics committee of the Jinling Hospital (No. 2020NZKY-016-01). Ethic approval of each participating center is required before initiation of enrollment.

8.1 Consent and confidentiality

Informed consent is required for each participant of this study, either signed by the patients himself or next of kin. All the data stored in the electronical database are de-identified to guarantee patients' privacy.

8.2 Dissemination policy

All the primary investigators and the sponsor will have full access to the data after conclusion of the study. Anyone who want to do a post-hoc analysis need to submit a formal writing proposal to the expert panel. Only approved author can have access to the database.

9. Data collection

A web-based electronic database (access through the website of the CAPCTG, https://capctg.medbit.cn/) is used for data collection and storage. All data are de-identified and input by the primary investigator or nominated investigators (less than two for each participating center) approved by the primary nvestigator, and a double check will be done by the research coordinator. Training for data entry is performed by the provider of the electrical database (Unimed Scientific, Inc, Wuxi, China) and the coordinating and data management center of the CAPCTG. Data including demographic characteristics, baseline characteristics, daily laboratory test, daily TG-lowering treatment, daily SOFA score, and follow-up characteristics. Demographic characteristics include age and sex. Baseline characteristics include body mass index (BMI), SOFA score on admission, Acute Physiology and Chronic Health Evaluation II (APACHE II) score on admission, the systemic inflammatory response syndrome (SIRS) on admission. Daily laboratory tests include serum total cholesterol (TC), triglyceride (TG), high-density lipoprotein cholesterol (HDL-c), low-density lipoprotein cholesterol (LDL-c), apolipoprotein A1 (Apo A1), apolipoprotein B (Apo B), apolipoprotein E (Apo E), lipoprotein a [LP(a)], free fatty acids (FFA), C-reactive protein (CRP), and procalcitonin (PCT). Daily TG-lowering treatment includes blood purification treatment (e.g., plasma exchange, hemoperfusion, and hemofiltration) and medical treatment (e.g., insulin and heparin). Follow-up characteristics include ICU days, hospital days, in-hospital cost, revision of the Atlanta classification on admission, CT severity index (CTSI) score (Based on the last image before discharge or death), mortality, and incidence of major complications on day 60. According to the schedule shown in Table 1, the investigators are required to collect data during the index admission and on day 60 after enrollment. And a follow-up on day 60 will be implemented through telephone.

TABLE 1 | Schedule of enrollment, assessment and follow up.

	Study period			
	Enrollment	**Observational period**		**Discharge**
Time point	<72 h	Day0[a]	Day1-Day14[b]	Day28 and Day60
Enrollment:				
Eligibility screen	X			
Informed consent	X			
Laboratory test	X			
Imaging(CT scan etc.)	X			
Assessment:				
Organ failure		X	⟷	
Laboratory test		X	⟷	
Major treatment		X	⟷	
Adverse effects		X	⟷	
Follow up:				
Vital status				X
Major complication				X
ICU days and hospital days				X
Cost				X

[a]*Day0 is defined as the day from enrollment to 8 am the next day.*
[b]*DayX is defined as the day from 8 am day X after enrollment to 8 am the next day.*
⟷*: Assessments during this period need to be repeated on a daily basis.*

10. Statistical analysis

10.1 Data collection methods:

A web-based electrical database will be used for data collection and storage. All data will be input by the primary investigator or nominated investigator(less than two for each participating center) approved by the primary investigator. Training for data entry will be performed by the supplier of the electrical database and the sponsor of the PERFORM study, and the coordinating center of CAPCTG.

10.2 Statistical methods

Continuous normally distributed data were reported as means with SDs. Skewed continuous data were reported as medians andby counts and percentages. The intergroup difference will be compared by Student’ s t-test or Wilcoxon rank-sum test for continuous variable depending on their normality and chi-square test for categorical data. To evaluate the association between TG decline and OF free days, the study patients will be dichotomized depending on whether the TG level reaches 5.65 mmol/L on Day 3 (the day of enrollment is labeled Day 1, the next day labeled Day 2, and the following day Day 3). For the primary outcome comparison, Wilcoxon rank-sum test will be employed. However, since OF could be evaluated with a time-to-event analysis censored at 14 days to account for the mortality as a competing event, Fine and Gray competing risk regression is used to assess the group difference as a supportive analysis. For the association between different TG lowering therapy and OF free days, we considered the possibility that baseline characteristics, which were expected to be prognostic for OF,differ according to the choice of TG lowering therapies (i.e., blood purification treatment and medical treatment). A propensity score matching will be further used to compensate for the intergroup unbalance. For secondary outcomes, a multivariate analysis generalized linear model (GLM) model will be performed to identify its association with TG decline and TG-lowering therapy with proper link and distribution function. The variable included in the model will be age, sex, TG level at enrollment, and other baseline variables that have significant differences between groups.

11. References

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Protocol Ver:2.0 dated June.2020

Primary Investigator：Professor. Weiqin Li

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Trial registration	ChiCTR2000039541
Study Design	Multicenter, registry-based, observational study
Type of Participants to be Studied	Patients aged 18 to 70 years older who were diagnosed with hypertriglyceridemia-induced acute pancreatitis (See the study population in detail)
Setting	Multicenter across China
Study Aim	This study aimed to describe the practice variation in triglyceride-lowering for HTG-AP patients with worrisome clinical features. It also aimed to evaluate the triglyceride-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.
Primary Outcomes Measure(s)	Organ failure-free days (OFFDs) to 14 days after enrollment
Key Secondary Outcome Measure(s)	Part I: Secondary outcomes during the index admission 8. a composite of death from any cause by day 28 and the presence of at least one organ failure at day 7 9. ICU free days to day 14 10. hospital free days to day 14, 11. new onset organ failure 12. requirement of ICU admission and development of infected pancreatic necrosis (IPN). Part II: Secondary outcomes within 60 days after enrollment

	1. Mortality censored at 60 days after enrollment; 2. hospital free days to day28 and day60 of enrollment
Number of Participates to be studied	PERFORM is designed to be a long-running registry. The phase-I goal is to recruit 300 patients.
Duration	Long-running
Sponsor	Jinling Hospital of Nanjing University
Chief Investigators	Professor. Weiqin Li

2. Abrreviations

HREC	Hospital Human Research Ethics Committee
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AP	Acute pancreatitis
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TPE	Therapeutic plasma exchange
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2. Study Administrative information

2.1 Study timelines

June 2019	Protocol manuscript
December 2019	Protocol finalized
	Initial expressions of interest sought from sites
	Commence study organization
June 2020	Hospital Human Research Ethics Committee (HREC) submissions
	Study material V.1.0 finalized
	HREC approvals obtained
December 2020	Patient recruitment commences (in plan)

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2.5 Registration

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2.6 Funding

This study was funded by Key Research and Development Program Foundation of Jiangsu Province of China (No. BE 2016749) and the National Science Foundation of China (No. 81900592).

3. Background and rationale

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Although the pathophysiology underlying HTG-AP remains controversial, it is widely accepted that free fatty acid (FFA) is one of the driving factors[8]. FFA, produced by the hydrolysis of triglyceride (TG), can initiate or worsen the disease by triggering inflammatory reactions, damaging

the pancreatic cell, and promoting microvascular thrombosis within the pancreatic tissue[9]. Nawaz et al.[6] found that elevated serum TG levels in AP patients were independently and proportionally correlated with persistent organ failure (POF) regardless of etiology. In an observational study conducted by Lu et al. [10]timely reduction of serum TG during the early phase of HTG-AP was found to be associated with decreased incidence of POF

Over the past years, several attempts had been made to lower serum TG more efficiently during the acute phase of the disease, including medical treatment with insulin and/or heparin, blood purification, and genetic therapy in cases[11]. Medical treatment is convenient and safe and is considered the first-line choice for TG-lowering therapy[4]. Heparin stimulates the release of endothelial lipoprotein lipase into circulation, while insulin activates lipoprotein lipase, thereby increasing the clearance of chylomicrons from plasma[12]. However, the impact of medical therapy on clinical outcomes is uncertain, and an observational study is ongoing to figure it out[13]. Blood purification, especially plasmapheresis, is also widely used as a TG-lowering therapy. Plasmapheresis rapidly removes triglycerides from plasma and is considered one of the most efficient TG-lowering therapies [14]. Technically, it is a therapeutic procedure in which the blood of the patient is passed through a medical device that separates plasma from other components of blood. The plasma is removed and replaced by a replacement solution (e.g., albumin and/or plasma) or a combination of crystalloid/colloid solution[15]. Double filtration plasmapheresis (DFPP) is a semi-selective apheresis method based on a double filter system, which can remove macromolecules selectively[16]. Both techniques are widely adopted, while plasmapheresis is thought to be more effective in removing FFA [17]. Other blood purification modalities were also reported effective in lowering plasma TG, including hemoperfusion and hemofiltration[18, 19]. A randomized control trial (RCT) reported that high-volume hemofiltration (HVHF) decreased TG levels more efficiently than medical therapy[18].

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the evolution of organ function in early hypertriglyceridemia-induced acute pancreatitis patients with worrisome features"(PERFORM) registry to evaluate the TG-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.

4. Study Design

4.1 Study aim

This study aimed to 1) describe the practice variation in triglyceride-lowering for HTG-AP patients with worrisome clinical features; 2)evaluate the triglyceride-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.

4.2 General study setting

The present clinical study will be performed in multiple sites across China . It is a multicenter, registry-based, prospective, observational cohort study.

5. Study population

5.1 Patient recruitment and sample size considerations

Based on the feasibility and patient flow of the potential participating sites, a sample size of 300 patients was expected, with an average of 15 patients per month within 2 years. Considering an estimate20% rate of incomplete data or losses of follow-up, our expected sample size (240 patients) would provide 87% to detect a 2- days (SD: 5) or 82.5% for 1.5-days (SD: 4) improvement of organ failure free days between patients achieve target TG and those not. The PERFORM study is aimed to be a long-running registry, so the collection of patients will be continuous after achieving the phase-I goal(300 patients).

5.2 Eligibility Criteria

5.21 Inclusion Criteria

6. Informed consent form obtained from the patient or next of kin;
7. Age between 18 to 70 years old;
8. Within 72 hours of onset,
9. With a primary diagnosis of AP based on Abdominal Pain Suggestive of AP, Serum Amylase

at Least three Times the Upper Limit of Normal, and/or Characteristic Findings of AP on Computed Tomography or Less Commonly Magnetic Resonance Imaging (MRI) or Transabdominal Ultrasonography According to the Revised Atlanta Criteria[21].

10. When enrolled, TG>1000mg/dL (11.3mmol/L), accompanied by the clinical worrisome features of any one or more of the following[22]:

    1) Signs of hypocalcaemia (calcium levels less than 2.1mmol/L)

    2) Lactic acidosis (Lactate levels more than 2mmol/L and PH＜7.35)

    3) Signs of worsening systemic inflammation (two or more):

        a) Temperature >38.5°C or <35.0°C

        b) Heart rate of >90 /min

        c) Respiratory rate of >20 breaths/min or PaCO2 of <32 mmHg

        d) WBC count of >12,000 cells/mL, <4000 cells/mL, or >10 percent immature (band) forms

    4) Signs of worsening organ dysfunction or multi-organ failure as defined by Modified Marshall scoring system for organ dysfunction ；

5.22 Exclusion Criteria

1. Failure to obtain informed consent;
2. Pregnant or lactating women; or have a pregnancy plan within a month after the study (including male subjects);
3. Researchers' family members who are directly involved in the study;
4. Patients are expected to die within 48 hours after enrollment, defined as patients with norepinephrine usage at a dose of 25 mg/min or more under full-fluid resuscitation, with a systolic blood pressure <90mm Hg and serum pH values <7.0. The judgment will be made by the treating physician.
5. *Additional exclusion criteria for this analysis:* Patients undergo any invasive blood purification therapy other than plasmapheresis.

6. Patient management

We recommend all patients receive standard treatment that follows the “Acute Pancreatitis Treatment Guidelines” issued by the American College of Gastroenterology (ACG) in 2013 and the

standard treatment plan for acute pancreatitis provided in the “Evidence-Based Guidelines for the Treatment of Acute Pancreatitis” issued by the International Association of Pancreatology (IAP) and the American Pancreatic Association (APA)[23, 24]. Since the PERFORM study is an observational one, all treatment decisions will be at the discretion of the treating physicians, including the use of different TG-lowering therapies.

7. Outcome measures

7.1 Primary outcome

The primary outcome is organ failure-free days (OFFDs) to 14days after enrollment. Only the final period of OFFDs is included, and patients who have OF at day 14 or died before day14 are assigned to zero OFFDs.

7.2 Secondary outcome

Secondary outcome measures

Part I: Secondary outcomes during the index admission

1. a composite of death from any cause by day 28 and the presence of at least one organ failure at day 7
2. ICU free days to day 14;
3. Hospital free days to day 14;
4. Requirement of ICU admission;
5. new onset organ failure to day 14;
6. $SOFA_{RANK}$ to day 14;
7. $\Delta SOFA_{max}$ to day 14;
8. ΔSOFA14 to day14

Part II: Secondary outcomes within 60 days after enrollment

1. Mortality censored at 60 days after enrollment;
2. Hospital free days to day28 and day60 of enrollment;
3. Development of infected pancreatic necrosis (IPN);

Definition of seconday outcomes:： New-onset organ failure is defined as organ failure that is not present at any time in the first 24 h after enrollment. $SOFA_{RANK}$ is a ranking parameter based on the cumulative daily delta SOFA score from day 1 to day 14. For each patient, it is calculated as a sum of the daily delta SOFA score (defined as the daily total SOFA score minus the baseline SOFA score) over the first 14 study days. Discharge is counted (from the day of discharge forward) as a score of 0 minus baseline score, and death is counted (from the day of death forward) as a maximum score of 24 minus baseline score. $\Delta SOFA_{max}$ is defined as the maximum SOFA score within 14 days minus the baseline SOFA score, and $\Delta SOFA_{14}$ as the SOFA score at day14 minus the baseline. The definition of other secondary outcomes can be found in the published protocol

8. Ethics and dissemination

This study has been approved by the ethics committee of the Jinling Hospital (No. 2020NZKY-016-01). Ethic approval of each participating center is required before initiation of enrollment.

8.1 Consent and confidentiality

Informed consent is required for each participant of this study, either signed by the patients himself or next of kin. All the data stored in the electronical database are de-identified to guarantee patients' privacy.

8.2 Dissemination policy

All the primary investigators and the sponsor will have full access to the data after conclusion of the study. Anyone who want to do a post-hoc analysis need to submit a formal writing proposal to the expert panel. Only approved author can have access to the database.

9. Data collection

A web-based electronic database (access through the website of the CAPCTG, https://capctg.medbit.cn/) is used for data collection and storage. All data are de-identified and input by the primary investigator or nominated investigators (less than two for each participating center) approved by the primary nvestigator, and a double check will be done by the research coordinator.

Training for data entry is performed by the provider of the electrical database (Unimed Scientific, Inc, Wuxi, China) and the coordinating and data management center of the CAPCTG. Data including demographic characteristics, baseline characteristics, daily laboratory test, daily TG-lowering treatment, daily SOFA score, and follow-up characteristics. Demographic characteristics include age and sex. Baseline characteristics include body mass index (BMI), SOFA score on admission, Acute Physiology and Chronic Health Evaluation II (APACHE II) score on admission, the systemic inflammatory response syndrome (SIRS) on admission. Daily laboratory tests include serum total cholesterol (TC), triglyceride (TG), high-density lipoprotein cholesterol (HDL-c), low-density lipoprotein cholesterol (LDL-c), apolipoprotein A1 (Apo A1), apolipoprotein B (Apo B), apolipoprotein E (Apo E), lipoprotein a [LP(a)], free fatty acids (FFA), C-reactive protein (CRP), and procalcitonin (PCT). Daily TG-lowering treatment includes blood purification treatment (e.g., plasma exchange, hemoperfusion, and hemofiltration) and medical treatment (e.g., insulin and heparin). Follow-up characteristics include ICU days, hospital days, in-hospital cost, revision of the Atlanta classification on admission, CT severity index (CTSI) score (Based on the last image before discharge or death), mortality, and incidence of major complications on day 60. According to the schedule shown in Table 1, the investigators are required to collect data during the index admission and on day 60 after enrollment. And a follow-up on day 60 will be implemented through telephone.

Table1 Schedule of enrollment, assessment and follow up.

	Study period			
	Enrollment	Observational period		Discharge
Time point	<72h	Day1	Day2-Day14	Day60
Enrollment:				
Eligibility screen	X			
Informed consent	X			
Laboratory test	X			
Imaging (CT scan etc.)	X			
Assessment:				
Organ failure		X	← →	
Laboratory test		X	← →	
Major treatment		X	← →	
Adverse effects		X	← →	
Follow up:				
Vital status				X
Major complication				X
ICU days& hospital days				X
Cost				X

10. Statistical analysis

10.1 Data collection methods:

A web-based electrical database will be used for data collection and storage. All data will be input by the primary investigator or nominated investigator(less than two for each participating center) approved by the primary investigator. Training for data entry will be performed by the supplier of the electrical database and the sponsor of the PERFORM study, and the coordinating center of CAPCTG.

10.2 Statistical methods

Continuous normally distributed data were reported as means with SDs. Skewed continuous data were reported as medians and by counts and percentages. The intergroup difference will be compared by Student's t-test or Wilcoxon rank-sum test for continuous variable depending on their normality and chi-square test for categorical data. To evaluate the association between TG decline

and OFFDs, the study patients will be dichotomized depending on whether the TG level reaches 5.65 mmol/L on Day 3 (the day of enrollment is labeled Day 1, the next day labeled Day 2, and the following day Day 3). For the primary outcome comparison, Wilcoxon rank-sum test will be employed. However, since OF could be evaluated with a time-to-event analysis censored at 14 days to account for the mortality as a competing event, Fine and Gray competing risk regression is used to assess the group difference as a supportive analysis. For the association between different TG lowering therapy and OFFDs, we considered the possibility that baseline characteristics, which were expected to be prognostic for OF, differ according to the choice of TG lowering therapies (i.e., blood purification treatment and medical treatment). A propensity score matching will be further used to compensate for the intergroup unbalance. For secondary outcomes, a multivariate analysis generalized linear model (GLM) model will be performed to identify its association with TG decline and TG-lowering therapy with proper link and distribution function. The variable included in the model will be age, sex, TG level at enrollment, and other baseline variables that have significant differences between groups.

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Summary of protocol changes

List of Changes

Protocol Amendment (version 1 to version 2)

Page/ Line No.	Original Text	New Text	Reason
Page 2,Table,**Setting**	estimated 30-40 hospitals across China	Multicenter, registry-based, observational study	Expected sites to be enrolled will not be limited.
Page 2, Table,**Study Aim**	This study aims to evaluate the TG-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.	This study aimed to describe the practice variation in triglyceride-lowering for HTG-AP patients with worrisome clinical features. It also aimed to evaluate the triglyceride-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.	Add Study Aim details.
Page2,Table, **Secondary Outcome Measure(s)**	Part I: Secondary outcomes during the index admission 1. New-onset organ failure; 2. New-onset multiple-organ failure (MOF); 3. New-onset persistent organ failure (POF); 4. New receipt of organ support;	Part I: Secondary outcomes during the index admission 13. a composite of death from any cause by day 28 and the presence of at least one organ failure at day 7 14. ICU free days to day 14 15. Hospital free days to day 14, 16. new onset organ failure 17. requirement of ICU admission and development of infected pancreatic necrosis (IPN). Part II: Secondary outcomes within 60 days after enrollment	Only key secondary outcome measures are listed.

	5. Requirement of ICU admission; 6. ICU free days to day 14; 7. Hospital free days to day 14; Part II: Secondary outcomes within 60 days after enrollment 1. Mortality censored at 60 days after enrollment; 2. AP severity grade (Based on the Revised Atlanta Classification); 3. Incidence of infected pancreatic necrosis (IPN); 4. Incidence of septic shock; 5. Incidence of abdominal bleeding; 6. Incidence of gastrointestinal fistula.	1. Mortality censored at 60 days after enrollment; 2. hospital free days to day28 and day60 of enrollment	
Page 2,Table, **Number of Participates to be studied**	estimated 200 patients	PERFORM is designed to be a long-running registry. The phase-I goal is to recruit 300 patients.	The PERFORM registry will be long-running.
Page 2,Table, **Duration**	estimated 18 months	Long-running	Same as above.

Page 4,line 13	Study Administrative information without **timeline**	Add 2.1 timeline June 2019 Protocol manuscript December 2019 Protocol finalized Initial expressions of interest sought from sites Commence study organization June 2020 Hospital Human Research Ethics Committee (HREC) submissions Study material V.1.0 finalized HREC approvals obtained December 2020 Patient recruitment commences (in plan)	Describe in more detail about timeline.
Page 5,line 28	Study Administrative information without **funding**	Add **2.6 Funding** This study was funded by Key Research and Development Program Foundation of Jiangsu Province of China (No. BE 2016749) and the National Science Foundation of China (No. 81900592).	Describe funding of the study.
Page7,line 19	The PERFORM study aims to evaluate the association between different TG-lowering therapies and clinical outcomes in HTG-AP patients with worrisome features.	This study aimed to 1) describe the practice variation in triglyceride-lowering for HTG-AP patients with worrisome clinical features; 2)evaluate the triglyceride-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.	Add Study Aim detals.

Page7,line 22	The present clinical study will be performed in 30-40 different hospitals across China. It is a multi-centered, register-based, prospective, observational study.	The present clinical study will be performed in multiple sites across China . It is a multicenter, registry-based, prospective, observational cohort study.	The PERFORM registry will be long-running.
Page 7,line 27	We are going to collect approximately 200 patients presenting to the participating ICUs across China during a two-year period. Based on the volume of all the participating centers, the aim should be able to be achieved within the study period.	Based on the feasibility and patient flow of the potential participating sites, a sample size of 300 patients was expected, with an average of 15 patients per month within 2 years. Considering an estimate20% rate of incomplete data or losses of follow-up, our expected sample size (240 patients) would provide 87% to detect a 2- days (SD: 5) or 82.5% for 1.5-days (SD: 4) improvement of organ failure free days between patients achieve target TG and those not. The PERFORM study is aimed to be a long-running registry, so the collection of patients will be continuous after achieving the phase-I goal(300 patients).	Add sample size estimation.
Page 9,line 22	Part I: Secondary outcomes during the index admission 1. New-onset organ failure; 2. New-onset multiple-organ failure (MOF); 3. New-onset persistent organ failure (POF); 4. New receipt of organ support;	Part I: Secondary outcomes during the index admission 1. a composite of death from any cause by day 28 and the presence of at least one organ failure at day 7 2. ICU free days to day 14; 3. Hospital free days to day 14; 4. Requirement of ICU admission; 5. new onset organ failure to day 14; 6. SOFARANK to day 14; 7. ΔSOFAmax to day 14; 8. ΔSOFA14 to day14	Changes in secondary outcomes including the addition of a composite outcome, and some other organ failure measures.

	5. Requirement of ICU admission; 6. ICU free days to day 14; 7. Hospital free days to day 14; Part II: Secondary outcomes within 60 days after enrollment 1. Mortality censored at 60 days after enrollment; 2. AP severity grade (Based on the Revised Atlanta Classification); 3. Incidence of infected pancreatic necrosis (IPN); 4. Incidence of septic shock; 5. Incidence of abdominal bleeding; 6. Incidence of gastrointestinal fistula.	Part II: Secondary outcomes within 60 days after enrollment 1. Mortality censored at 60 days after enrollment; 2. Hospital free days to day28 and day60 of enrollment; 3. Development of infected pancreatic necrosis (IPN);	
Page 10，line 11	Definition of seconday outcomes:1)An individual SOFA score of 2 or more for the respiration, cardiovascular, or renal system is defined as the presence of organ failure. 2)New-onset organ failure is	Definition of seconday outcomes:New-onset organ failure is defined as organ failure that is not present at any time in the first 24 h after enrollment. SOFARANK is a ranking parameter based on the cumulative daily delta SOFA score from day 1 to day 14. For each patient, it is calculated as a sum of the daily delta SOFA score (defined as the daily total SOFA score minus the baseline SOFA score) over the first 14 study days. Discharge is counted (from the day of discharge forward) as a score of 0 minus baseline score, and death is counted (from the day	Reword the definition and description of the secondary outcoems.

	defined as organ failure that is not present at any time in the 24 h after enrollment. Multiple organ failure is defined as two or more organ failures present at the same time. 3)Persistent organ failure is defined as organ failure that persists for more than 48 h. 4)ICU free days to day 14 after enrollment is defined as the number of days alive and not admitted to an ICU after the patient's latest discharge from the ICU before day 14. If the patient is admitted to an ICU on day 14 or dies prior to day 14, ICU-free days will be 0. Hospital-free days to day 14 after enrollment is defined as the number of days alive and not admitted to the hospital after the patient's final discharge from the hospital before day 14.	of death forward) as a maximum score of 24 minus baseline score. ΔSOFAmax is defined as the maximum SOFA score within 14 days minus the baseline SOFA score, and ΔSOFA14 as the SOFA score at day14 minus the baseline. The definition of other secondary outcomes can be found in the published protocol	

The Effect of Plasma TriglycERide Lowering Therapy on Organ FunctiOn in HypertRiglycerideMia-induced Acute Pancreatitis: a multicenter, registry-based, observational study (PERFORM)

Statistical Analysis Plan

Ver.1.0 dated Nov. 2020

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1. INTRODUCTION

Acute pancreatitis (AP) is a potentially life-threatening inflammatory disease with multiple etiologies, such as alcohol, gallstones and hypertriglyceridemia. HTG is the third most common cause of AP, accounting for 4-10% of cases globally, and the increasing prevalence of HTG-AP had been reported in recent studies[1-4]. In China, HTG had been the second leading cause of AP, and previous studies showed that HTG-AP patients had a higher risk of severe acute pancreatitis and multiple organ dysfunction syndrome (MODS) than other types of AP[2, 5-7].

Although the pathophysiology underlying HTG-AP remains controversial, it is widely accepted that free fatty acid (FFA) is one of the driving factors[8]. FFA, produced by the hydrolysis of triglyceride (TG), can initiate or worsen the disease by triggering inflammatory reactions, damaging the pancreatic cell, and promoting microvascular thrombosis within the pancreatic tissue[9]. Nawaz et al.[6] found that elevated serum TG levels in AP patients were independently and proportionally correlated with persistent organ failure (POF) regardless of etiology. In an observational study conducted by Lu et al. [10]timely reduction of serum TG during the early phase of HTG-AP was found to be associated with decreased incidence of POF

Over the past years, several attempts had been made to lower serum TG more efficiently during the acute phase of the disease, including medical treatment with insulin and/or heparin, blood purification, and genetic therapy in cases[11]. Medical treatment is convenient and safe and is considered the first-line choice for TG-lowering therapy[4]. Heparin stimulates the release of endothelial lipoprotein lipase into circulation, while insulin activates lipoprotein lipase, thereby increasing the clearance of chylomicrons from plasma[12]. However, the impact of medical therapy on clinical outcomes is uncertain, and an observational study is ongoing to figure it out[13]. Blood purification, especially plasmapheresis, is also widely used as a TG-lowering therapy. Plasmapheresis rapidly removes triglycerides from plasma and is considered one of the most efficient TG-lowering therapies [14]. Technically, it is a therapeutic procedure in which the blood of the patient is passed through a medical device that separates plasma

from other components of blood. The plasma is removed and replaced by a replacement solution (e.g., albumin and/or plasma) or a combination of crystalloid/colloid solution[15]. Double filtration plasmapheresis (DFPP) is a semi-selective apheresis method based on a double filter system, which can remove macromolecules selectively[16]. Both techniques are widely adopted, while plasmapheresis is thought to be more effective in removing FFA [17]. Other blood purification modalities were also reported effective in lowering plasma TG, including hemoperfusion and hemofiltration[18, 19]. A randomized control trial (RCT) reported that high-volume hemofiltration (HVHF) decreased TG levels more efficiently than medical therapy[18].

For the target of TG-lowering therapy, it is regarded that reducing the TG level to 5.65 mmol/L might be clinically sufficient[20]. Lu et al.[10]found that patients with earlier TG levels of < 5.65 mmol/L were less likely to develop POF. However, the optimal TG lowering target and choice of therapies in early HTG-AP are unclear due to the lack of high-quality studies. Given the paucity of evidence in the literature and the variation in the management of HTG-AP, we conducted this multicenter, observational study and built “The effect of plasma triglyceride-lowering therapy on the evolution of organ function in early hypertriglyceridemia-induced acute pancreatitis patients with worrisome features”(PERFORM) registry to evaluate the TG-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.

2. STUDY DESIGN AND OBJECTIVE

This study is an analysis of data collected from the PERFORM study (ChiCTR2000039541, https://www.chictr.org.cn/index.aspx). This study aimed to 1) describe the practice variation in triglyceride-lowering for HTG-AP patients with worrisome clinical features; 2)evaluate the triglyceride-lowering effects of different therapies and their impact on clinical outcomes in HTG-AP patients with worrisome features.

3. ELIGIBILITY CRITERIA

3.1 Patient recruitment

Based on the feasibility and patient flow of the potential participating sites, a sample size of 300 patients was expected, with an average of 15 patients per month within 2 years. Considering an estimate20% rate of incomplete data or losses of follow-up, our expected sample size (240 patients) would provide 87% to detect a 2-days (SD: 5) or 82.5% for 1.5-days (SD: 4) improvement of organ failure free days between patients achieve target TG and those not. The PERFORM study is aimed to be a long-running registry, so the collection of patients will be continuous after achieving the phase-I goal(300 patients).

3.*2 Inclusion criteria for the PERFORM study*

11. Informed consent form obtained from the patient or next of kin;
12. Age between 18 to 70 years old;
13. Within 72 hours of onset,
14. With a primary diagnosis of AP based on Abdominal Pain Suggestive of AP, Serum Amylase at Least three Times the Upper Limit of Normal, and/or Characteristic Findings of AP on Computed Tomography or Less Commonly Magnetic Resonance Imaging (MRI) or Transabdominal Ultrasonography According to the Revised Atlanta Criteria[21].
15. When enrolled, TG>1000mg/dL (11.3mmol/L), accompanied by the clinical worrisome features of any one or more of the following[22]:

    1) Signs of hypocalcaemia (calcium levels less than 2.1mmol/L)

    2) Lactic acidosis (Lactate levels more than 2mmol/L and PH＜7.35)

    3) Signs of worsening systemic inflammation (two or more):

    a) Temperature >38.5°C or <35.0°C

    b) Heart rate of >90 /min

    c) Respiratory rate of >20 breaths/min or PaCO2 of <32 mmHg

    d) WBC count of >12,000 cells/mL, <4000 cells/mL, or >10 percent immature (band) forms

    4) Signs of worsening organ dysfunction or multi-organ failure as defined by Modified Marshall scoring system for organ dysfunction ；

3.3 Exclusion criteria for the PERFORM study

1. Failure to obtain informed consent;

2. Pregnant or lactating women; or have a pregnancy plan within a month after the study (including male subjects);

3. Researchers' family members who are directly involved in the study;

4. Patients are expected to die within 48 hours after enrollment, defined as patients with norepinephrine usage at a dose of 25 mg/min or more under full-fluid resuscitation, with a systolic blood pressure <90mm Hg and serum pH values <7.0. The judgment will be made by the treating physician.

3.4 Additional exclusion criteria for this analysis

1. Patients undergo any invasive blood purification therapy other than plasmapheresis.

4. PATIENT MANAGEMENT

We recommend all all patients receive standard treatment that follows the "Acute Pancreatitis Treatment Guidelines" issued by the American College of Gastroenterology (ACG) in 2013 and the standard treatment plan for acute pancreatitis provided in the "Evidence-Based Guidelines for the Treatment of Acute Pancreatitis" issued by the International Association of Pancreatology (IAP) and the American Pancreatic Association (APA)[23, 24]. Since the PERFORM study is an observational one, all treatment decisions will be at the discretion of the treating physicians, including the use of different TG-lowering therapies.

5. OUTCOME MEASURES

5.1 The primary outcome

The primary outcome was organ failure free days(OFFD) to study day14. The diagnosis of organ failure was made when an individual sequential organ failure assessment(SOFA) score of two or more for the respiration, cardiovascular, or renal system. In patients with transient organ failure resolution, only the final periods of OFFD were counted. Patients who had organ failure on day14 or died before day14

were assigned zero OFFD.

5.2 Secondary outcomes

Secondary outcomes included a composite endpoint of death from any cause by day28 and the presence of at least one organ failure at day7, SOFArank, ΔSOFAmax, and ΔSOFA14 to day14, ICU free days to day14, hospital free days to day14, day28, and day60, new onset organ failure (defined as organ failure that is no present at any time in the first 24h of enrolment) to day14, requirement of blood purification and ICU admission during the index admission, mortality and incidence of infected pancreatic necrosis(IPN) by day60. SOFArank was calculated as a sum of the daily delta SOFA score (defined as the daily total SOFA score minus the baseline SOFA score) over the first 14 days of enrolment. Discharge was counted (from the day of discharge forward) as a score of 0 and death as 24. ΔSOFAmax was defined as the maximum SOFA score within 14 days minus the baseline, and ΔSOFA14 as the SOFA score at day14 minus the baseline.

6. ETHICS AND DISSEMINATION

This PERFORM study has been approved by the ethics committee of the Jinling Hospital. Ethic approval of each participating center will also be obtained before the initiation of enrollment.

6.1 Consent and confidentiality

Informed consent will be required for each participant of this study, either signed by the patients or next of kin. All the data stored in the electronical database will be de-identified to guarantee the patients' privacy.

6.2 Dissemination policy

All the investigators will have full access to the data after the conclusion of the study. Anyone who wants to do a post-hoc analysis needs to submit a formal writing proposal to the expert panel. Only the approved authors can have access to the database.

7. DATA COLLECTION AND FOLLOW UP

In the original PERFORM study, a web-based electrical database (access through the website of the CAPCTG, https://capctg.medbit.cn/) will be used for data collection and storage. All data will be input by the primary investigator or nominated investigators (less than two for each participating center) approved by the primary investigator, and a double check will be done by the research coordinator from the coordinating and data management center of the CAPCTG. Training for data entry will be performed by the provider of the electrical database and the coordinating and data management center of the CAPCTG. According to the data collection schedule, the investigator will collect data during the index admission and on day 60 after enrollment.

8. STATISTICAL PLAN

8.1 General Considerations

8.1.1 Reporting guidelines

We will follow the STrengthening the Reporting of OBservational studies in Epidemiology guideline for explanation and elaboration of the study results.

8.1.2 Participant disposition and Flow chart

A flowchart will be drawn up showing the number of patients screened, enrolled, and followed up in each exposure group.

The number screened and not enrolled, and the reasons for non-enrollment will be reported, as well as the number and reasons of patients who are lost for follow up, or who are withdrawn from the study.

8.1.3 Data Summaries

Continuous normally distributed data will be reported as means with SDs. Skewed continuous data will be reported as medians and interquartile ranges (IQRs). Categorical data will be summarized by counts and percentages.

Mean values for some continuous outcomes (e.g., triglyceride, SOFA scores) by group will be plotted. Kaplan-Meier plots will be produced to display the time-to-event data.

8.1.4 Planned Covariates

Covariate analysis will be performed, in particular, for the primary outcome. The prespecified covariates in this study will be:

- Age
- Sex
- Body Mass Index (BMI)
- baseline Acute Physiology and Chronic Health Evaluation II (APACHE II) score
- baseline triglyceride

8.1.5 Missing Data

For baseline covariates and efficacy outcomes, missing data less than 5% will be assumed to be missing at random (MAR) and thus will not be imputed in the primary analysis. However, if greater than 5% of data that should be available for analysis are missing, multiple imputation will be undertaken in the analysis.

8.2. Statistical Analysis

Continuous data were reported as means and SDs or medians and interquartile ranges (IQR) as appropriate, depending on their normality. The normality of data was determined by Shapiro-Wilk tests. Categorical data were summarised by counts and percentages. The intergroup differences were compared by Student’s t-test or Wilcoxon rank-sum test for continuous variables depending on their normality and Fisher's exact test for categorical data.

Two statistical approaches were applied to assess the association between triglyceride-lowering effects(Triglyceride decline) and clinical outcomes. First, all the study individuals with triglyceride levels on study day3 were dichotomised depending on whether the Triglyceride level was ≤5.65 (target-reaching). Comparisons were made between those who reached the target or not. For the primary outcome comparison, Wilcoxon rank-sum test was employed, and median difference (95% CI [confidence intervals]) was calculated by the Hodges–Lehmann estimation. For the sensitivity analysis of the primary outcome, the adjusted generalised linear model (GLM) was performed to assess its association with target-reaching. Furthermore, the Fine and Gray

competing risk model (competing factor=death) was adopted to assess the association between target-reaching and organ failure resolution.

Second, the decline rate of triglyceride for the first three days of enrolment (decline slope) was obtained from the linear regression with time as a dependent and TG as an independent variable, and the regression coefficient was the slope of the Triglyceride decline. The second approach involves patients with triglyceride levels measured on either day2 or day3. The adjusted GLMs were performed to assess the association between decline slope and OFFD. The confounders were the same as the primary analysis.

For the comparison of secondary outcomes, the adjusted generalised linear model (GLM) was performed to identify its association with target-reaching. The variables included in the model were the same as the adjusted analysis for the primary outcome.

We then assess the impact of blood purification on clinical outcomes. Propensity score matching (PSM) analysis was used to control potential confounders. Patients who received blood purification were matched 1:1 with patients who received conventional treatment using their propensity score. The propensity score was based on the same confounding variables for the adjusted GLM. Genetic Matching with a calliper width of 0.2 was used in the PSM. Comparisons of differences between groups in the PSM cohort were performed using Wilcoxon signed-rank test and McNemar's test for matched data. To evaluate the robustness of our findings, we also performed the inverse probability of treatment weighting (IPTW) analysis with the same variables as PSM. Comparisons of differences between groups in the IPTW cohort were performed using Wilcoxon rank-sum test and Fisher's exact test.

We performed data conversions (including log, reciprocal, and square root transformations) for the continuous outcomes with skewed distribution in the GLM models. Statistical tests were two-sided, and p values <0.05 were deemed significant unless otherwise stated. All statistical analyses were done in the SPSS 26.0 software and the R 4.2.1 software.

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