Thymosin alpha 1 in the prevention of pancreatic infection following acute necrotizing pancreatitis (TRACE trial)

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Abstract

Abstract Background: IPN and its related septic complications are the major causes of death in patients with ANP. Therefore, prevention of pancreatic infection is of great clinical value in the treatment of ANP. This study aimed to evaluate the role of Thymosin Alpha 1 in treating ANP. Methods: Eligible patients were randomised to the treatment group(Thymosin alpha 1 group) or the control group. Occurrence of IPN will serve as the primary outcome measure of the TRACE trial. Occurrence of new-onset organ failure and persistent organ failure; CRP, HLA-DR, WBC day0, day 7 and day14; other related pancreatic complications and prognosis of the patients were also calculated. Discussion: The TRACE trial is designed to test the effect of a new therapy focusing on the immune system in preventing secondary infection following ANP, which is a potentially lethal complication causing substantial morbidity and mortality. We also aimed to prospectively investigate the effect of immunomodulatory treatment with thymosin alpha 1 in patients with different severity of diseases with predefined subgroup analysis. The results of the TRACE trial would potentially provide a novel therapeutic option in the early management of ANP and identify the patient population who may benefit most from immunomodulation. The trial was registered on the June 16, 2015 in the CT.gov register (NCT02473406)

Background

Infected pancreatic necrosis (IPN) and its related septic complications are the major causes of death in patients with acute necrotizing pancreatitis(ANP)[1].When compared with patients with sterile necrosis, patients with IPN suffered substantial increase in mortality ranging from 14% to 69% due to sepsis and its related multiple organ failure, despite advances in critical care, surgical interventions and antibiotics[2]. Therefore, prevention of pancreatic infection is of great clinical value in the treatment of ANP. In past years, numerous attempts had been made to prevent or delay the development of IPN including antibiotic prophylaxis, selective gut decontamination and probiotics, but none of them has been clinically proved with high-quality evidence[3-5].

Immunosuppression and disorders characterized by decreased HLA-DR expression and unbalanced CD3/CD4+/CD8+ T cells of peripheral blood mononuclear cell (PBMC) are reported to be associated with the development of IPN[6, 7]. Our previous study also found that early enteral could moderate the excessive immune response during the early stage of severe acute pancreatitis without leading to subsequent immunosuppression, and ultimately improve the outcome of the patients[8].Thus immunomodulatory treatment could potentially intervene in the evolution of secondary infection of pancreatic necrosis, resulting in better outcome. Unfortunately, study regarding the immune status in patients with any type of acute pancreatitis (AP) is rare, let alone appropriate treatment aiming to balance patients' immune function.

Thymosin alpha 1 has been shown to have immunomodulatory properties and is reported to be beneficial in patients with sepsis[9, 10], majorly through the involvement of distinct Toll-like receptors (TLRs) acting on different dendritic cells (DC) subsets and involving the MyD88-dependent signaling pathway. However,

the effect of thymosin alpha 1 in patients with AP is rarely reported in the literature. The only randomized clinical study conducted by our group years before proved that the use of Thymosin alpha 1 is associated with improved cellular immunity and reduced infection rate in a group of ANP patients[11]. However, no further study was published yet and the clinical significance of this study is limited due to small sample size and single-center nature. Therefore, a multi-center, randomized study with large sample size and proper design is warranted to evaluate the role of Thymosin Alpha 1 in treating ANP.

Methods

Study Design

The present study is an investigator-initiated, randomized, multi-centered, double-blinded, placebo-controlled comparison study. We aimed to evaluate the efficacy of Thymosin Alpha 1 in prevention of IPN and its impact on immune function and other clinical outcomes in patients with ANP.

Study population

The present study is a clinical interventional study being performed in 12 hospitals across China. All adult patients admitted to the participating centers with AP will be assessed for eligibility after their hospital admission. The inclusion and exclusion criteria are as follows:

Inclusion criteria

1. Symptoms and signs of acute pancreatitis 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[12]; 2. Within a week from the onset of abdominal pain; 3.Age between 18 to 70 years old; 4. Acute Physiology and Chronic Health Evaluation(APACHEII) score ≥8 and Balthazar CT score ≥5 (presence of pancreatic necrosis)[13].

Exclusion criteria

1. Pregnant pancreatitis; 2. Receiving intervention or early surgery due to abdominal compartment syndrome or other reasons before admission.4. Patients with a known history of severe cardiovascular, respiratory, renal, hepatic, hematologic, or immunologic disease defined as (1) greater than New York Heart Association class II heart failure, (2) active myocardial ischemia or (3) cardiovascular intervention within previous 60 days, (4) history of cirrhosis or (5) chronic kidney disease with creatinine clearance< 40 mL/min, or (6) chronic obstructive pulmonary disease with requirement for home oxygen; 5.Patients with preexisting immune disorders such as AIDS.

Randomization and blinding methods

The sequence generation was performed by the Randomization Allocation Tool v. 1.1.4 (RAT, Ratjin, Nanjing). After the initial enrollment, participants will be randomly assigned in a 1:1 ratio to either the treatment group or the control group in a double-blinded manner according to internet-based computer-generated random numbers in block sizes of 4. The random assignment will be conducted by a third party independent of the study, and the assignment records will not be disclosed until the end of the study. After the acquisition of written informed consent and the completion of baseline measurements, the allocation will be sequentially delivered to the clinical investigator who is not involved in outcome assessment and is responsible for patient care after entering basic information into the RAT. Participants, data collectors, and investigators assessing outcome data will be blinded to the treatment allocation to minimize potential sources of bias.

Study protocol and Interventional arms

After confirming that a participant meets the inclusion criteria and does not meet any of the exclusion criteria, the study protocol flow of participants is outlined in Figure1. The investigational drug (as allocated by the online tool) will be delivered to the clinical investigator who is responsible for treatment of the recruit and baseline measurement would be completed on the same day. The recruited patients will start to receive either thymosin Alpha 1 or placebo (normal saline) subcutaneously from the day after the allocation day as described above. The drug regimen will last for 14 consecutive days as shown in Figure2. If the patient die or is discharged before complete the designed regimen, the rest of the investigational drug will be waived and recycled by the sponsor. The enrolled patients will be followed up until hospital death or discharge.

General treatment regimen

All patients received initial standard treatment including fluid resuscitation, early enteral nutrition, routine medical treatment, mechanical ventilation if needed and continuous renal replacement therapy (CRRT) if needed in the light of recent published guidelines. All participating centers are able to offer appropriate intensive care in case the patients require organ support or continuous monitor. The necrotic collection will be intervened when infection was suspected or confirmed, but the intervention should be optimally delayed for 4 weeks when the patient could tolerate the symptoms.

When pancreatic infection occurs, either surgical or endoscopic step-up approach on the basis of the location of the necrotic collection and technical availability in each participating center will be applied. Initial drainage is optimally performed after the necrotic collection is well walled-off. Principally, either percutaneous catheter drainage or endoscopic transluminal placement of double pig-tail stents are the primary choices of treatment.

Primary outcome measure

All patients will be followed until hospital death or discharge. Occurrence of IPN will serve as the primary outcome measure of the TRACE trial. The diagnosis of IPN will be based on the international guidelines when one or more of the following were present: gas bubbles within (peri) pancreatic necrosis on computed tomography; a positive culture of (peri) pancreatic necrosis obtained by image-guided fine-needle aspiration; a positive culture of (peri) pancreatic necrosis obtained during the first drainage and/or necrosectomy[12].

Secondary outcome measures

1.Occurrence of new-onset organ failure and persistent organ failure; 2. C-reactive protein(CRP), HLA-DR expression on monocytes and other laboratory results like white blood cell(WBC) count and lymphocyte count on day0, day 7 and day14 of the observation;3.Occurence of bleeding requiring intervention;4.Occurence of gastrointestinal fistula;5.In-hospital mortality; 6.Pancreatic fistula; 7.Need of percutaneous or endoscopic transluminal drainage; 8. Need of minimally-invasive necrosectomy; 9.Need of open surgery; 10.Length of intensive care unit(ICU) stay; 11. Length of hospital stay; 12. In-hospital cost.

Sample size estimation and statistical analysis

The prevalence of pancreatic infection was reported to be around 25% in ANP episodes. To demonstrate a 40% reduction in the prevalence of pancreatic infection considering the results of our pilot study [11] with 80% power at a two-sided alpha level of 0.05, we projected an estimated sample size of 500 participants using the PASS software (PASS, NCSS software, Kaysville, USA). Considering possible 4% withdrawal, we planned to randomize 520 patients in total. The statistical analysis for outcome measures will be based on the intention-to-treat (ITT) population. Missing data will be handled by multiple imputation to evaluate the robustness of the primary endpoint analyses[14]. Normality of continuous variables were examined using skewness and kurtosis. Continuous variables were compared using the Mann-Whitney method or Student's T test depending on the distribution of the data[15]. Categorical data were expressed as number and percentage. Between-group difference will be compared using the chi-squared test. Statistical tests will be two-sided, and p values < 0.05 will be accepted as significant for the endpoints. Given that some very sick patients can die very early without a chance of infection during the disease courses (although very rare), which may cofound the analysis, those who die during the first week after onset of abdominal pain will be considered as missing data for the analysis of the primary outcome measure, namely, the occurrence of pancreatic infection. They will be included for analyses of other outcome measures. No interim unblinding and analysis is planned for the TRACE trial.

Pre-defined subgroup analyses

Considering that patients with organ failure are much more likely to develop infection as reported in the literature, namely, these patients are at higher risk of IPN, a subgroup analysis will be done for the occurrence of the primary endpoint (infection of pancreatic necrosis) in patients with or without organ failure. Other outcome endpoints will also be additionally assessed in the two subgroups to comprehensively evaluate the effect of thymosin alpha 1 in patients with different severity of AP. Moreover, subgroup analyses for the primary outcome will be performed for patients with different age (dichotomized at 60 years), gender, different etiologies (biliary and others) and extent of pancreatic necrosis (>50 vs.50 and below).

Data collection and management

A web-based electrical database (based on the website of chinese acute pancreatic trial group) 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 and double check will be done by the research coordinator. Training for data entry will be performed by the supplier of the electrical database and the sponsor of the TRACE trial. The final data will be assessed by the first author and independent statistician. An interim analysis is not planned.

Recruiting success

The trial was registered on the June 16, 2015 in the CT.gov register (NCT02473406 https://www.clinicaltrials.gov/ct2/show/NCT02473406?term=NCT02473406&rank=1)The first patient was randomized on the 22nd of March 2017. So far, 197 patients have been randomized and inclusion process keeps to the schedule.

Discussion

The TRACE trial is designed to test the effect of a new therapy focusing on the immune system in preventing secondary infection following ANP, which is a potentially lethal complication causing substantial morbidity and mortality. We also aimed to prospectively investigate the effect of immunomodulatory treatment with thymosin alpha 1 in patients with different severity of diseases with predefined subgroup analysis. The results of the TRACE trial would potentially provide a novel therapeutic option in the early management of ANP and identify the patient population who may benefit most from immunomodulation.

Considering the difficulty and complexity in treating IPN prevention the development of this life-threatening complication is of great clinical interests and studies on this topic are abundant in the

literature. The use of prophylactic antibiotics is a long-term controversial issue repeatedly mentioned in different versions of guidelines. In recent years, based on latest clinical evidence, it is no longer supported by mainstream guidelines[16, 17], although controversy remains[18]. Other efforts were made on preventing translocation of bacteria toward the necrotic collection like selective decontamination[4] and enterally administration of probiotics, but both failed in reducing the risk of infectious complication[5]. Enteral feeding is the only proved treatment for protecting the gut-mucosal barrier and randomized clinical trials(RCTs) have strongly suggested that early enteral nutrition could significantly decrease the risk for interventions for necrosis, namely, the risk of secondary infection of the necrotic collection[19]. However, as enteral feeding initiated with 24-72hours after admission has almost become the standard of treatment, other treatment which could further reducing the risk of infection is required and immunomodulation has the potential to be future clinical practice considering the evidence showing that immunosuppression happens early and strongly in more severe patients[7, 20].

Immunomodulation is of significant clinical value in critically ill settings and treatment of sepsis[9]. While, acute pancreatitis, which has a lot in common with sepsis like overwhelmed inflammation and infection related complications, might be another good target for immunomodulatory therapy. In the past years, a few attempts have been made in this field using drugs like lexipafant and octreotide, but the evidence can not hitherto support solid clinical benefits in terms of major clinical outcomes [21]. In general, both lexipafant and octreotide were aimed to control cytokines, which are thought to be the pivotal part in early inflammatory response of AP, rather than preventing secondary infection. However, like what we learn in sepsis, immunosuppression quickly following the initial inflammatory cascades should be the target of treatment during the course of ANP, especially in those with organ failure[7, 22]. Our group published our pilot study several years ago in which we found that the administration of thymosin α1 could improve compromised monocyte HLA-DR expression and reduce infection rate in a small group of patients (n=24) with severe acute pancreatitis. The result of this study is encouraging which drive us to implement this large multi-center RCT to obtain more reliable clinical evidence[11].

The TRACE trial was sponsored by the Jinling Hospital, which is the national transferal center for acute pancreatitis (AP) admitting more than 600 cases of AP annually. The trial was aimed to be performed in 12 centers across China recruiting 520 patients to meet the estimated sample size. Due to the limitation of the budget and technical availability, we can not perform comprehensive immune test in multiple time points and alternatively we choose monocyte HLA-DR, which is a representative parameter of the immune system majorly reflecting the antigen presentation capacity to assess the immunomodulatory effect of thymosin alpha 1 and also be widely used in previous studies regarding immune function in different diseases like sepsis[9, 23].

In conclusion, the TRACE trial majorly aimed to assess the efficacy of thymosin α1 administered early during the disease course of ANP on the rate of necrosis infection and other major clinical outcomes, thus offering a novel therapeutic option in treatment of ANP patients.

Abbreviation List

TRACE Thymosin alpha 1 in the prevention of pancreatic infection following acute necrotizing pancreatitis

IPN Infected pancreatic necrosis

ANP acute necrotizing pancreatitis

PBMC peripheral blood mononuclear cell

AP acute pancreatitis

TLRs Toll-like receptors

DC different dendritic cell

MRI magnetic resonance imaging

APACHEII Acute Physiology and Chronic Health Evaluation

RAT Randomization Allocation Tool

CRRT continuous renal replacement therapy

CRP C-reactive protein

ICU intensive care unit

ITT intention-to-treat

RCT randomized clinical trial

Declarations

Ethics approval and consent to participate:

This study was approved by the ethic committee of Jinling Hospital. Even central ethical approval has been confirmed, we will not begin recruiting at other centres in the trial until local ethical approval has been obtained. Besides, the consents for this study were obtained from each patient or his next of kin.

Consent for publication

Not Applicable.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

Study concept and design: L K, Z T

Acquisition of data; analysis and interpretation of data: J Z, G L, B Y, Z Z, W M

Drafting of the manuscript; J Z, L K

Critical revision of the manuscript for important intellectual content: Z Z, W L

Obtained funding; administrative, technical, or material support: Z T, W L

Study supervision: W L, J L

* All authors have read and approved the final version of the manuscript and ensure this is the case.

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Figures

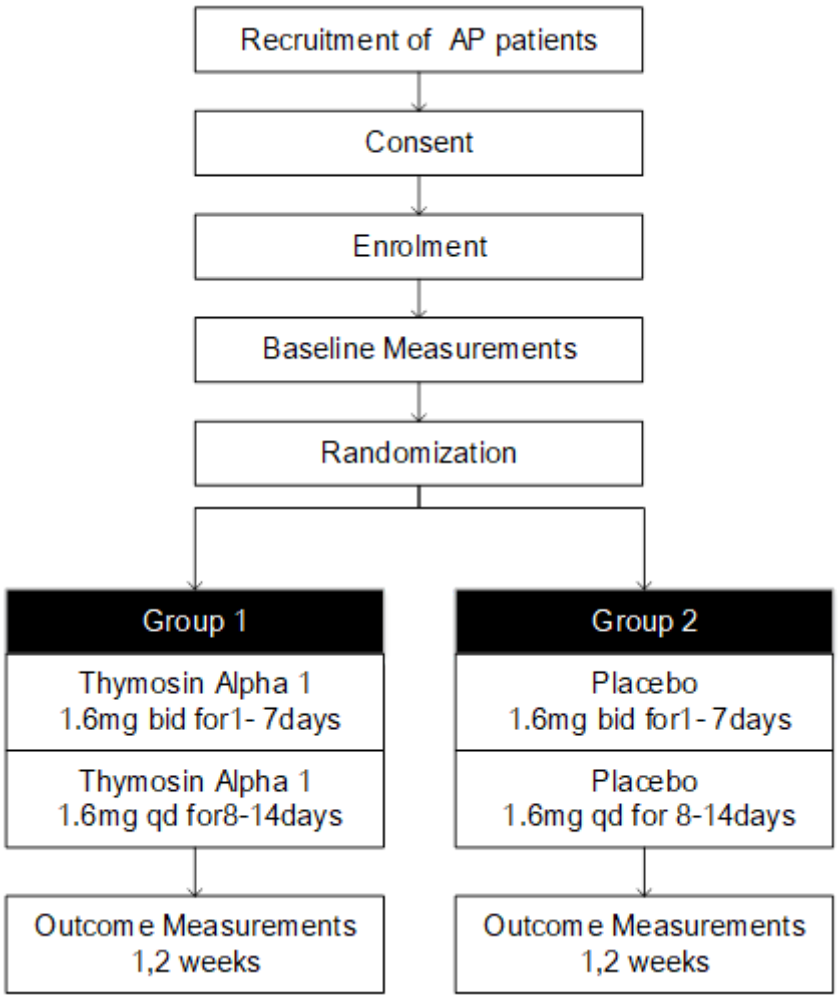

Figure 1

Flow of participants through the trial

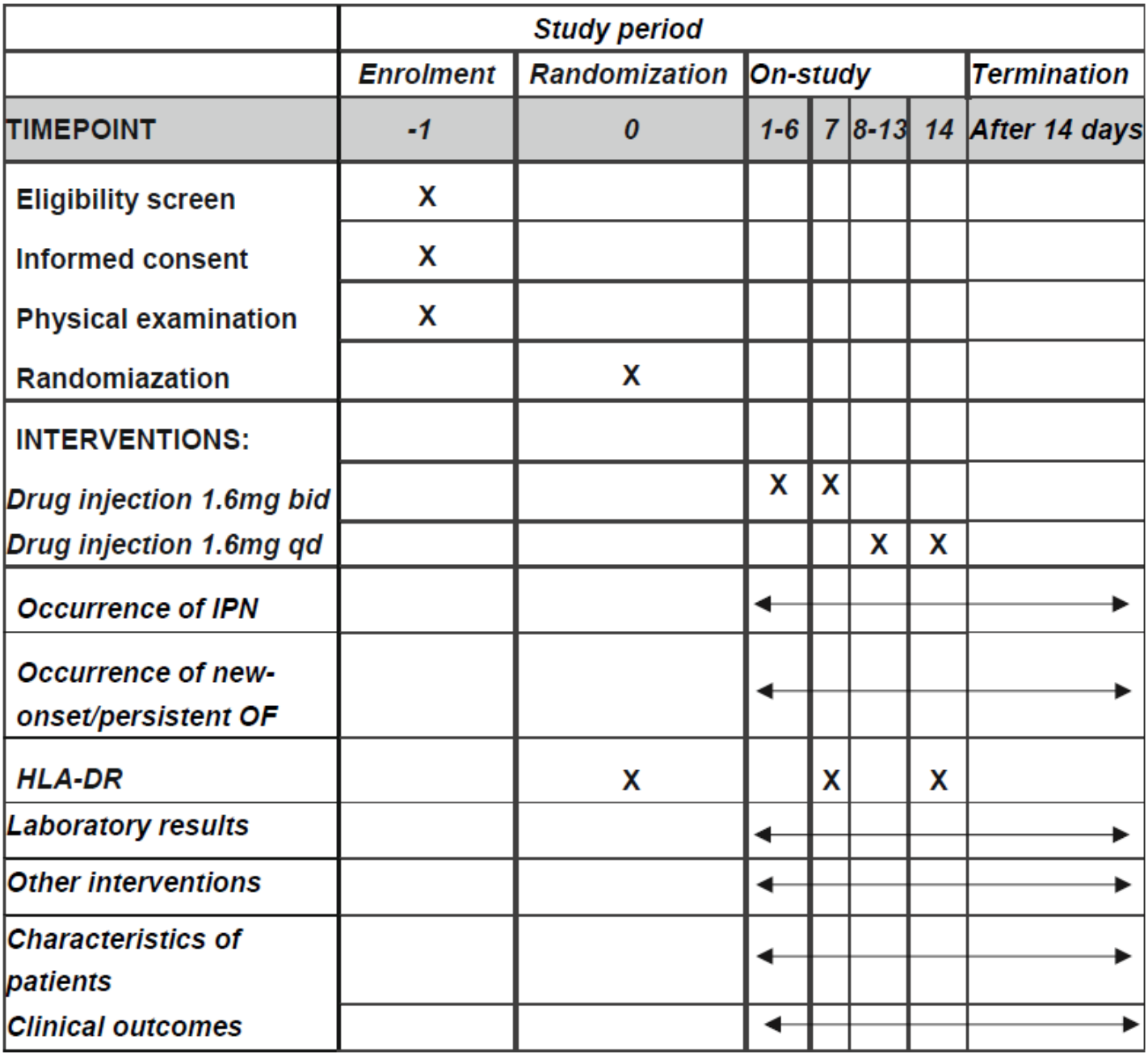

Figure 2

Trace figure showing times points for enrolment, interventions and assessments. IPN infected pancreatic necrosis, OF organ failure.

Supplementary Files

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