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TITLE:

A Clinical Trial Assessing the Safety, Efficacy, and Delivery of Olive-Oil-Based Three-Chamber Bags for Parenteral Nutrition

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KEYWORDS:

Parenteral nutrition, soybean oil, triple-chamber bags, compounded bags, olive oil, efficacy, lipids, oxidative stress

SUMMARY:

Here, we present a protocol for a study comparing the efficacy, safety, and delivery of olive-oil-based 3CB and soybean-oil-based CoB formulations in adults requiring parenteral nutrition. The results revealed that olive-oil-based 3CBs is non-inferior and well tolerated compared to soybean formulations.

ABSTRACT:

Limited evidence exists to precisely estimate efficacy and safety differences between parenteral nutrition (PN) prepared using olive-oil-based three-chamber bags (3CBs) and soybean-oil-based compounded bags (CoBs) in hospitalized adult patients. We designed a multicenter, randomized, prospective, open-label, noninferiority protocol to compare the efficacy, safety, and distribution of olive-oil-based 3CBs and soybean-oil-based CoB formulations in adult Chinese patients requiring PN during surgical intervention. Subjects were randomized to receive either one of the study treatments using an interactive voice or web-based recognition system in accordance with the randomization code. Randomization was further stratified based on the study site and surgical category. Both treatment groups received similar amounts of calories and protein. In addition, the two study treatments contained a similar composition of the amino-acid component. The only difference between the two PN formulations was the lipid constitution. The duration of administration of study treatments was a minimum of 5 days up to a maximum of 14 days after the surgical procedure. The primary efficacy endpoint was serum prealbumin levels on day 5 of the study. Noninferiority was proved if the anti-log of the lower bound of the 95% confidence interval (CI) of the treatment difference was at least 0.80. Other efficacy

measures included treatment preparation time; duration to achieve tolerability of oral nutrition; associated infectious complications; length of hospitalization; and laboratory assessment of markers of nutrition, inflammation, metabolism, and oxidative stress. A total of 458 patients were enrolled in the study. The results showed that olive-oil-based 3CBs were non-inferior to soybean-based CoBs, besides being well tolerated. The infection rate was found to be significantly lower in the olive-oil-based 3CB group. Thus, this study may be used as a reference for future research on lipid emulsion and 3CBs.

INTRODUCTION:

Parenteral nutrition is an essential component of overall therapy for a wide spectrum of indications, such as major gastrointestinal surgery, transient enteral intolerance, severe burns, coma; or for use in critically ill patients. Improvements in intravenous (IV) nutritional formulations and knowledge advancement regarding the implementation of therapy allow the safe and clinically efficacious administration of IV nutrition. These characteristics are particularly important in a metabolically stressed patient[1].

Parenteral nutrition is commonly administered to patients by mixing nutrients that are compounded in the hospital pharmacy. Compounding total parenteral nutrition solutions from individual components is a multi-step, time-intensive process associated with a greater risk of human error. Recently, triple-chamber bag (3CB) systems have been developed in which individual components are separated by nonpermanent breakable seals. The contents of a 3CB include a glucose solution, an amino-acid solution, a lipid emulsion, with or without electrolytes. Prior to administration, the seal separating the various components of the 3CB is broken, enabling the components of the chambers to be admixed. The advantages offered by the 3CB includes increased physio-chemical shelf life of components, reduction the extent of contamination during preparation, and cutting down on the steps required in the preparation of a PN product[2].

Lipid emulsion is an important ingredient in a PN formula; it can produce different clinical effects, depending upon the constituent fatty acids. Soybean-oil-based lipid emulsions primarily consist of long-chain linoleic acid (ω-6 polyunsaturated fatty acid [ω-6 PUFA]), which is mainly proinflammatory. Experimental data suggest that ω-6 PUFA-rich lipid emulsions may amplify the inflammatory response during stress and traumatic conditions, as well as increasing the infection rate[3]. On the other hand, olive-oil-based lipid emulsions, which consist of long-chain oleic acid (ω-9 monounsaturated fatty acids, [ω-9 MUFAs]), have a neutral response on the immune system[3,4]. Substituting soybean-oil-based ω-6 PUFAs with olive-oil-based ω-9 MUFAs can make the PN safe and further widen its clinical application[5,6]. However, there are limited clinical data in this connection.

Therefore, the present study aims to evaluate the rate of infections in two different lipid emulsions that varied in the content of linoleic acid, in addition to having the primary objective of assessing the safety and efficacy of 3CBs compared to CoBs for delivering PN. The assessment was carried out in adult hospitalized patients scheduled to undergo surgery for whom enteral nutrition was either not possible, inadequate, or inadvisable.

PROTOCOL:

For this prospective, randomized, multicenter, active-controlled, parallel-group investigational trial, the Ethics Committees of Shanghai Sixth People's Hospital approved the study protocol.

1. Patient recruitment and enrollment

1.1. Recruit patients as per the inclusion criteria specified in the protocol. Seek informed consent from patients and perform screening assessments not more than 3 days prior to the scheduled surgery.

1.2. Randomize enrolled subjects to one of the two study treatments (olive-oil-based 3CBsor soybean-oil-based CoBs), based on the randomization schedule.

1.2.1. Randomize subjects undergoing surgery who were not scheduled for preoperative PN after a surgical procedure (**Figure 1**). Randomize subjects undergoing surgery and who were scheduled for preoperative PN prior to the surgery (**Figure 2**). Randomize subjects who were not scheduled to undergo surgery if they meet the enrollment criteria (**Figure 3**).

1.2.2. Do not blind the data management personnel, biostatistician, and/or the personnel at the central laboratory for the assignment of study treatment.

1.3. Unblind the designated pharmacist for the preparation of the study treatment. Enter the blinded data into the database. Reveal the treatment group assignments after the database lock.

NOTE: For this study, the study treatment assignment was delegated to the site pharmacist.

1.4. Perform the allocation of patient numbers using an interactive voice-recognition system/interactive web-based recognition system (IVRS/IWRS) according to the randomization code contained in the randomization list.

1.4.1. Stratify randomization by study site and surgical category (no surgery, medium-complexity, and high-complexity, i.e. each type of surgery is categorized based on the complexity and the length of surgery) within each study site. In addition, specify the block size in the randomization code algorithm.

2. Study population

2.1. Inclusion criteria

2.1.1. Enroll subjects into the study if all the following criteria are met:

2.1.1.1. Ensure that the subject is male or female aged 18 to 80 years.

2.1.1.2. Ensure that the subject is inpatient but hospitalized for fewer than 14 days prior to enrollment in the study.

2.1.1.3. Ensure that the subject requires PN because enteral nutrition was not feasible, inadequate, or inadvisable

2.1.1.4. Ensure that the subject is capable of completing at least 5 days of study treatment and having a functional visible peripheral vein for IV delivery of PN.

2.2. Exclusion criteria

2.2.1. Exclude subjects from the study if any of the following criteria are met:

2.2.1.1. Exclude if the patient has a life expectancy of fewer than 6 days from the start of study treatment, as determined by the investigator.

2.2.1.2. Exclude if the patient has established hypersensitivity to the individual constituents of any of the study treatments.

2.2.1.3. Exclude if the patient has used forbidden medications (e.g., glucocorticosteroids or antitumor chemotherapeutic agents) within 30 days prior to enrollment in the study.

2.2.1.4. Exclude patients with a confirmed clinically relevant serious condition that prevents inclusion in the study (e.g., congestive heart failure with New York Heart Association class IV) or severe renal insufficiency without adequate compensation (e.g., hemofiltration, hemodialysis, or peritoneal dialysis), etc. or known chronic active hepatitis, alanine aminotransferase (ALT) >4x upper limit of normal (ULN), aspartate aminotransferase (AST) >4x ULN; total serum bilirubin >2x ULN; history of human immunodeficiency virus infection

2.2.1.5. Exclude patients with confirmed inborn abnormalities related to the metabolism of amino acids (e.g., phenylketonuria, maple syrup urine disease, homocystinuria, or tyrosinemia, etc.), severe dyslipidemia with triglyceride level >2x ULN or >4.52 mM (>400 mg/dL).

2.2.1.6. Exclude patients with severe hyperglycemia; serum glucose levels >20 mM (>360 mg/dL); and with clinically relevant abnormalities of plasma electrolytes, such as sodium (<130 mM or >150 mM), potassium (<3.0 mM or >5.5 mM), magnesium (<0.70 mM or >1.10 mM), calcium (<2.0 mM or >3.0 mM), or phosphorus (<0.96 mM or >1.62 mM).

2.2.1.7. Exclude pregnant and lactating patients.

2.2.1.8. Exclude patients previously enrolled in the current study, or who participated in a study of any investigational drug or device concurrently or within 30 days before enrollment in this

study.

2.2.1.9. Exclude for any reason, as per the investigator's opinion, that renders the subject unsuitable for the trial.

2.3. Removal of patients from therapy or assessment

2.3.1. Discontinuation of study treatment/early release from study

2.3.1.1. Exclude patients who do not continue to meet the inclusion and exclusion criteria after surgery (assessed either in the recovery room or just after transfer to the surgical ward).

2.3.1.2. Exclude and consider the patient discontinued if any enrolled patient who, continued the study after surgery, and for whom study treatment was prematurely terminated (i.e., completion of <5 full days of post-surgery study treatment).

2.3.2. Withdrawal from study

2.3.2.1. Withdraw the patient if the patient did not continue to fulfill the inclusion or exclusion criteria.

2.3.2.2. Withdraw the patient if the patient experienced an adverse event (AE) or developed an inter-current illness, condition, or procedural complication that would interfere with continued participation

2.3.2.3. Withdraw the patient if the patient voluntarily withdrew consent/authorization for the study

2.3.2.4. Withdraw the patient if the patient was found to be in violation of the protocol and for whom the physician deemed it in their best medical interests to terminate involvement in the study

3. Method and clinical parameters

3.1. Efficacy Assessments

3.1.1. Assess the primary efficacy outcome measure of the serum prealbumin level on day 5.

NOTE: Secondary efficacy parameters included:

1. Time for preparation of the study treatment (**Table 1**)
2. Duration to achieve tolerability of oral nutrition
3. Infectious complication
4. Length of hospitalization
5. Nutrition markers: Albumin and fatty acids (including linoleic, oleic acid, arachidonic acid,

and eicosapentaenoic acid)
6. Infection and inflammatory markers: Including procalcitonin, C-reactive protein, interleukin-6, and intercellular adhesion molecule-1
7. Oxidative stress markers: Including malondialdehyde and F2-isoprostane
8. Metabolism markers: Including urine markers of metabolism (urinary urea nitrogen, urinary 3-methylhistidine); hormonal markers of metabolism (thyroid panel, cortisol, growth hormone, insulin-like growth factor-1 [IGF-1], and testosterone)

3.1.2. Draw venous blood via a peripheral vein in a contralateral appendage from the peripheral IV study treatment administration at the time points indicated in the schedule of assessment (**Table 1**).

3.1.3. Transfer the samples to a central laboratory for analysis of efficacy parameters. Use EDTA anticoagulant tubes for the hematology test. Use common serum tubes which are free of additives for the serum biochemical and serological tests.

3.1.4. Collect 2-3 mL of blood per sample. If the test was performed 4 hours after collection, store the blood in the 4 °C refrigerator.

3.1.5. Collect urine over a 6-hour period, according to the schedule of assessment (**Table 1**). Record the collected volume of the urine for the applicable treatment day and store in the 4 °C refrigerator. Take an aliquot from the urine collection and transfer it to a central laboratory for analysis of efficacy parameters.

3.2. Safety assessments

NOTE: The safety assessments in this study included:
1. Adverse events and SAEs
2. Vital sign assessment and physical examination findings, including body weight and injection-site rating by the investigator
3. Clinical laboratory assessments: Hematology, serum chemistry, urinalysis, and other required laboratory assessments by the investigator

4. Study treatments

4.1. Dosage Forms and Administration

NOTE: For this study, the treatments were intended to provide 25 kilocalories per kilogram per day (kcal/kg/day) using a PN admixture that contained 910 kcal/1.5 L, with a dextrose-to-lipid ratio of 62:38 and 5.4 grams (g) nitrogen/1.5 L. The study treatment was delivered over 12 to 22 hours.

4.1.1. For Treatment A (test treatment) which comprises olive-oil-based 3CBs, use dextrose (D-glucose) solution (final mixed concentration 80 g/L) with calcium (final mixed concentration 2

mmol/L); a middle chamber that contains a solution of 15 amino acids (final mixed concentration 22 g/L), with electrolytes including sodium (final mixed concentration 21 mmol/L),potassium (final mixed concentration 16 mmol/L), magnesium(final mixed concentration 2.2 mmol/L), and phosphate (final mixed concentration 8.5 mmol/L); and a smaller outer chamber that contains a lipid emulsion comprising 80 % olive oil and 20 % soybean oil (final mixed concentration 20 g/L).

4.1.2. For Treatment B (control treatment), use a compounded ternary PN admixture that has the same volume, energy, nitrogen and dextrose-to-lipid ratio as Treatment A. It comprises soybean-oil based CoBs, compound amino acid, and intralipid compounded as a 1.5-L admixture in the pharmacy.

4.1.3. Give each research site a table containing the exact composition of different weights to ensure the consistency of prescriptions. Electrolytes, vitamins, minerals, and trace elements were allowed to be prescribed for addition to either treatment group according to the clinical requirement of different patients.

4.1.4. Administer the treatments through a peripheral IV catheter via a control pump. If the infusion is not possible or inadvisable via peripheral IV due to the integrity of the peripheral vein, infuse the study treatment via a peripherally inserted central catheter or a central IV line. Increase the flow rate gradually during the first hour.

4.1.5. Provide a daily volume of 46.6 mL/h and limit the administer duration between 12-22 h. Gradually increase the flow rate during the first hour (50-100 mL/h at the first hour). Adjust the administration rate considering the dose being administered, the daily volume intake, and the duration of the infusion. Do not increase the rate of intake over 150 mL/h.

4.1.6. Administer the treatments for a minimum of 5 days up to 14 days. On days 1 through 5, administer only the study treatment (i.e., PN). On day 6 and until the end of the study period, allow the addition of liquid oral nutrition to the study treatment in order to meet the calculated daily nutrition requirements.

4.1.7. Stop the study treatment once the subject receives at least 80% of the calculated daily nutrition requirements by the administered liquid oral nutrition or the completion of day-14 study treatment, whichever comes first.

4.1.8. Terminate treatment for patients who met the discontinuation or withdrawal criteria. Upon completion or termination of treatment, perform the end-of-treatment procedures for subjects.

4.2. Contraindicated medications and therapies

4.2.1. Do not administer additional lipids or amino acids during the clinical trial (from screening through completion of the study treatment period, inclusive), to the study subjects.

Do not allow the use of glucocorticosteroids within 30 days prior to enrollment.

4.2.2. Administer glucocorticosteroids during the clinical trial (from enrollment through completion of the study treatment period, inclusive) only if medically necessary. Similarly, do not allow the use of antitumor chemotherapeutic agents, whether for treatment of cancer or for other diseases, within 30 days prior to enrollment.

4.2.3. Administer antitumor chemotherapeutic agents during the clinical trial (from enrollment through completion of the study treatment period, inclusive) only if medically necessary. Record the administered concomitant medications on the case report forms (CRFs).

4.2.4. Allow the use of dextrose-containing IV solutions for maintenance of fluid status from randomization until the study treatment is discontinued.

4.3. Acceptable medications and therapies

4.3.1. Dilute the medications that need to be delivered via the IV route in saline-based solutions; however, limit the volume to 50 mL per medication if a medication is diluted in a dextrose-containing IV solution.

4.3.2. Record these medications on the CRFs, including details about the type of IV solution used as the diluent. Administer dextrose solutions for the treatment of hypoglycemia, and record details on the CRFs.

4.3.3. Do not restrict the use of other concomitant therapies (e.g., drugs, blood and plasma transfusion products, albumin, and other treatments) if these treatments are commercially available. Maintain a detailed record (including dose and duration) of medical treatments, including vitamins, electrolytes, and trace elements.

5. **Statistical methods**

NOTE: The assumption for the sample size calculation in this noninferiority trial was made such that the true ratio was 1, the coefficient of variance was 0.5, and the noninferiority margin was 20 %. A sample size of 98 patients per study treatment came out of all the assumptions, providing 90% power to claim noninferiority between groups for the primary efficacy endpoint (i.e., prealbumin levels on day 5).

5.1. To achieve a total of 400 subjects, required for primary efficacy assessment, randomize approximately 500 subjects. Determine the sample size based on the assumption that up to 20% of the randomized subjects would drop out of the study before the day-5 analysis.

5.2. As the intention-to-treat (ITT) population is the primary analysis population for demographic and other baseline characteristics, draw statistical inferences for the 2 treatment groups.

5.3. Use analysis of variance (ANOVA) to compare continuous data, and use the chi-square test/Fisher's Exact test to compare categorical data. In the case of significant imbalances for certain variables, review the results using covariate adjustments and subgroup analyses.

5.4. Use the analysis of covariance (ANCOVA) model to analyze the log-transformed primary efficacy endpoint. For the stated model, treatment and study site were the main effects and baseline serum prealbumin was the covariate.

5.5. Test the interaction between the 2 main effects and remove from the model if it was statistically insignificant. Determine the least squared geometric mean ratio of the treatment difference for olive-oil-based 3CBs to soybean-oil-based CoBs, as well as the 2-sided 95% CI.

NOTE: The primary comparison hypothesis was that olive-oil-based 3CBs were non-inferior to soybean-oil-based CoBs for increasing or maintaining serum prealbumin levels. Noninferiority was ascertained if the anti-log of the lower limit of the 95 % CI of treatment difference was at least 0.80.

5.6. Use the Kruskal-Wallis test to analyze differences between treatment groups to determine the preparation time of the study treatment (days 1 to 5).

5.7. Use the Kaplan-Meier method to summarize the time required to achieve tolerability of oral nutrition, and the parameter was compared using a log-rank test. Fatty acids, such as linoleic and oleic acids; infection and inflammatory markers; markers of oxidative stress; markers of nutrition; and markers of metabolism were analyzed in the same fashion as the primary endpoint—using the ANCOVA model with the treatment and study site as the main effects. The least squared geometric mean ratio of the treatment differences for olive-oil-based 3CBs to soybean-oil-based CoBs, as well as the 2-sided 95% CI, was determined.

5.8. For the safety variables, make statistical comparisons between the two treatment groups. Use the ANOVA method to compare continuous data; use the chi-square test/Fisher's exact test to compare categorical data. Use the Cochran-Mantel-Haenszel test with modified ridit scores to compare the relationship and severity of AEs.

5.9. Complete statistical analyses using statistical analysis software. A p-value <0.05 was considered to be statistically significant.

REPRESENTATIVE RESULT:

Patient Disposition

Out of the 480 patients who gave their consent, a total of 458 patients were enrolled and randomized in the study. The ITT population included all randomized patients, of whom 226 constituted the test group and 232 the control group. The safety population included a total of 453 patients, of whom 222 belonged to the test group and 231 to the control group. The modified intention-to-treat (mITT) population comprised a total of 443 patients, of whom 219

were in the test group and 224 in the control group (**Figure 4**).

A total of 373 patients comprised the per-protocol (PP) population, of whom 183 were in the test group and 190 in the control group. A comparable percentage of patients discontinued from the study in both the groups. Additionally, for both the groups, the two main reasons for discontinuation were AEs and the withdrawal of consent by the patient (**Figure 4**).

Demographic and Baseline Clinical Characteristics

The demographic and baseline clinical characteristics of patients in the two treatment groups (ITT population) were comparable. Sixty-one percent of the total patients were male. The majority of the patients were identified as Chinese Han (95%) and had a mean age of 56 years. A total of 62% of patients underwent high-complexity surgery of a mean duration of 3 hours (**Table 2**).

Endpoint's Result

In relation to the primary endpoint of the study, olive-oil-based 3CBs were found to be non-inferior to soybean-oil-based CoBs in increasing or maintaining the levels of serum prealbumin at day 5 in both the mITT population (p=0.0002) and the PP population (p=0.0006).

A similar trend was also observed when the subgroup analyses for age, gender, no surgery, surgery of medium complexity, and surgery of high complexity was performed for the two groups (**Figure 5**).

Increased levels of prealbumin and albumin were observed for the test group, while the control group showed decreased levels of both proteins. Serum prealbumin and albumin levels on day 5 of the study were found to be significantly higher in the test group compared to the control group. No statistically significant difference was observed in serum IGF-I levels at day 5 between both the groups. However, at day 14, serum IGF-I levels were found to be significantly higher in the test group compared to the control group. No statistically significant differences were observed during the between-group analysis for 6 h urinary urea nitrogen and 6 h urinary excretion of 3-methylhistidine.

Lipid Endpoints

A significant increase in serum oleic acid levels were observed in both the groups; however, the observed increase in the olive-oil-based 3CB group was greater. There were no statistically significant differences observed at any time point between the treatment groups for serum levels of linoleic acid, arachidonic acid, and EPA.

Inflammation, Oxidation, and Infections

A small but statistically significant difference was observed on day 5 in serum levels of interleukin (IL)-6 between the two treatment groups. There was a decrease in the level of IL-6 in both the groups.

No significant differences were observed in serum levels of cortisol, procalcitonin, C-reactive

protein, or ICAM-1 between the two treatment groups. Additionally, for the serum levels of malondialdehyde or F2-isoprostane, no significant differences were observed on day 5 or day 14 of the study between the two treatment groups.

The overall incidence rate of infections was found to be low in the study. Patients in the control group had a significantly higher infection rate compared to the test group (**Table 3**). The most common infections observed in the study were lung infections, followed by incision/wound infections. No bloodstream infections were reported in the study[7].

Preparation Time

The preparation time for study treatment was found to be significantly lower for the test group compared to the control group on all assessment days (**Figure 6**).

FIGURE LEGENDS:

Table 1: Schedule of assessment—Pre-study treatment period through study treatment period (day 1 through day 14) and end of treatment

Table 2: Patient demographics and baseline characteristics (Intention-to-treat [ITT] population)

Table 3: Treatment-emergent adverse event infections in safety population

Figure 1: Representative schematic for subjects undergoing surgery without preoperative parenteral nutrition (PN). (Treatment A (test treatment) is olive oil-based 3CBs; Treatment B (control treatment): Soybean-oil based CoBs)

Figure 2: Representative schematic for subjects undergoing surgery with preoperative parenteral nutrition (PN) (Treatment A (test treatment) is olive oil-based 3CBs; Treatment B (control treatment): Soybean-oil based CoBs)

Figure 3: Representative schematic for subjects not undergoing surgery. (Treatment A (test treatment) is olive oil-based 3CBs; Treatment B (control treatment): Soybean-oil based CoBs)

Figure 4: Flowchart for patient disposition in study. (Treatment A (test treatment) is olive oil-based 3CBs; Treatment B (control treatment): Soybean-oil based CoBs)

Figure 5: Representation of efficacy analyses of olive oil in the modified intention-to-treat (mITT) population and prespecified patient subgroups. (LSGM ratio is the antilog of (log (GM) ± 1.96SE); $p<0.05$ Abbreviations: CI confidence interval, LSGM least square geometric means, mITT modified intention-to-treat, PN parenteral nutrition, SD standard deviation, SE standard error)

Figure 6: Preparation time of olive oil-based 3CBs (Clinomel N4) and Soybean-oil based CoBs

(PN admixture) (Day 1 through day 5). (*p<0.05 by Kruskal-Wallis was considered statistically significant. Error bars indicate standard deviations.)

DISCUSSION:

The randomized clinical trial protocol is a multi-purpose document. It not only provides guidance for the conduct of trial to the investigators, but it also makes ethics committees and institutional review boards aware of appropriate measures adopted to protect participants' safety and interests. A proper design is crucial for the success of a clinical trial. It is often noted that the design of a trial is connected with its successes/failures[8].

Additionally, selection of the intervention and control groups is a key step in protocol design. Compounded PN is the current standard for the administration of PN to patients who cannot obtain adequate nutrition intake from diet or enteral nutrition. A placebo would not represent ethical treatment for patients requiring IV nutrition for the prevention or treatment of malnutrition. Therefore, active treatment (compounded PN admixture) was used as a control instead of a placebo in this study, to maintain the standard of care that subjects required. Another reason for which the compounded PN admixture was selected is the stated research objective. This made it possible to deliver both similar amounts of calories (primarily from dextrose and lipids) and protein to both groups. Furthermore, the two study treatments contained a similar amino-acid component. The only intentional difference in the PN formula was lipid source.

The double-blind design is preferable for a clinical trial. For this study, blinding was not impractical because, as per the basic clinical practice, the clinician or nurse made sure that the integrity of the combined components in the PN formulation was maintained throughout the length of the infusion. Although it was an open-label study, the bias in the reporting of treatment effects was minimized by implementing blinding for the data management personnel, biostatisticians, and researchers at the central laboratory.

Inherent in all clinical trials is the issue of con-founder of relationship. Randomization is necessary to equally distribute both known and unknown con-founders to the study and control groups; this can reduce bias[9]. In this study, an IVRS/IWRS system was used by the sponsor to enroll/randomize patients to different treatments arms. It generated a unique enrollment/randomization number for each patient. This configurable-and-customizable system was accessible via telephone or the Web from anywhere in the world. It empowered the sponsor to proactively manage key aspects of their clinical trials, including enrollment/randomization, dosing/drug dispensation, clinical supplies, and unbinding, etc. In addition, it has eliminated the risk of bias at sites by automated randomization, dispensation, and unbinding. Furthermore, the system also has the potential to decrease the work burden of research staff[10]. Stratified randomization includes construction of strata based on various variables, such as age groups, race, or practice and randomizing within these developed strata. In this study, randomization was stratified by study site to maintain approximately equal proportions of patients randomized to each study treatment at each study site. Randomization within each study site was further stratified by the surgical category. This approach reduced the

impact of operational differences between study sites on statistical comparisons. Stratified randomization ensures that randomization is achieved in a balanced manner for the important predefined confounder variables. It further helps in allowing researchers to analyze various subgroups[11].

Block randomization divides randomized participants within different subgroups, termed as 'blocks,' in order to ensure equal distribution of participants to each group. The limitation of the block randomization approach is the predictable distribution of participants, leading to a selection bias in unmasked study groups. Selection bias in the block randomization approach can be reduced by ensuring random block sizes and by blinding the investigator with respect to the size of the block[12].The block size was specified in the randomization code algorithm in this study.

The eligibility criteria of a clinical trial protocol should ensure that the enrolled participants in the trial are alike to the maximum extent and that the results obtained from them are applicable to the general populations as well[9]. In this study, we selected surgical patients incapable of receiving the required nutrition via the enteral or oral route, representing a population that may benefit from PN therapy when enteral nutrition is not feasible, insufficient, or challenging. We also enrolled non-surgery patients, aiming to make the results applicable to the nonsurgical population as well. The purpose of the exclusion criteria was to reduce noise and ensure the safety of the trial. To fulfill these, we excluded severely ill and dying patients, patients with contraindication to PN or allergic to PN components, and patients with a medical history that would interfere with metabolism.

The primary study endpoint must correlate directly to the investigational study product, should be clinically accordant, and assessable conveniently in a clinical trial. Endpoints are usually a biomarker or a patient-specific structural or functional endpoint[13]. In this study, the prealbumin level was selected as the primary endpoint, following a discussion involving various experts. Serum prealbumin level, one of the commonly used nutritional endpoints, serves as a composite pointer for the amino acid supply, protein-synthesizing extent, inflammation, and catabolism. However, several confounding variables affect this endpoint, including synthesis and degradation status and inflammation level. Thus, for the secondary endpoints, albumin and IGF-1 levels were selected to assess anabolism; excretion of nitrogen and 3-methylhistidine helped in assessing catabolism. Oxidation, lipid profile, glucose levels, insulin, and electrolytes levels helped in assessing the metabolic status; clinical outcomes are assessed based on infections, hospital stay, morbidities, mortality, and preparation time. In addition to establishing the efficacy of 3CBs and lipids in supporting the claim, the trial should also prove the safety profile, which includes the assessment of lab parameters, vital signs, and injection-site reactions.

More and more clinical trials are being undertaken in order to evaluate whether the new treatment is as efficacious as standard treatment. The new treatment possesses various advantages such as a good safety profile, ease of administration, and is economical, making it fruitful to establish noninferiority with regard to the efficacy parameter. The noninferiority trial

offers the advantage of statistical significance to be only 1 tailed, as there is no assumption that the analysis addresses whether the treatment is better. The predefining of a noninferiority margin for the primary outcome measure is of prime importance while designing a noninferiority trial[14]. In this study, the noninferiority margin was defined as −20%. The noninferiority in the trial was justified if the anti-log of the lower limit of the treatment difference 95% CI was at least 0.80.

In the majority of hospitalized patients, the ability to absorb nutrients via the gastrointestinal route will return to normal levels within 1 to 2 weeks of the medical/surgical event that caused a disruption in feeding. It is, therefore, practical to compare olive-oil-based 3CB to a CoB (soybean-oil-based) for a course of therapy that is long enough to evaluate a signal of the comparative efficacy and safety, yet does not delay the re-administration of oral or enteral feeding. It is always a critical decision to specify the duration of follow-up, to come to a meaningful result. The relatively short duration of follow-up, i.e. a maximum of 14 days, may be considered as the main limitation of this study. Despite the short duration of follow-up, it is possible that with a longer duration of PN, additional differences may have been noted between treatment groups.

To sum up, several aspects of the trial design are appropriately selected to ensure the value of the study is duly considered for this standard protocol involving lipid emulsion and 3CBs. This study may be used as a reference for future research.

DISCLOSURES

The authors have nothing to disclose.

ACKNOWLEDGMENT

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REFERENCES

1. Jeejeebhoy, K.N. Parenteral nutrition in the intensive care unit. *Nutrition Reviews.* **70** (11), 623-630 (2012).
2. Simmer, K., Rakshasbhuvankar, A., Deshpande, G. Standardised parenteral nutrition. *Nutrients.* **5** (4), 1058-70 (2013).
3. Vanek, V.W. et al. A.S.P.E.N. position paper: Clinical role for alternative intravenous fat emulsions. *Nutrition in Clinical Practice*. **27** (2), 150-192 (2012).
4. Singer, P. et al. ESPEN guidelines on parenteral nutrition: Intensive care. *Clinical Nutrition*. **28** (4), 387-400 (2009).
5. Calder, P.C., Jensen, G.L., Koletzko, B.V., Singer, P., Wanten, G.J. Lipid emulsions in parenteral nutrition of intensive care patients: Current thinking and future directions. *Intensive Care Medicine.* **36** (5), 735-749 (2010).
6. Umpierrez, G.E. et al. A double-blind, randomized clinical trial comparing soybean oil-based versus olive oil-based lipid emulsions in adult medical-surgical intensive care unit patients

requiring parenteral nutrition. *Critical Care Medicine*. **40** (6), 1792-1798 (2012).
7. Jia, Z.Y. et al. Safety and efficacy of an olive oil-based triple-chamber bag for parenteral nutrition: A prospective, randomized, multi-center clinical trial in China. *Nutrition Journal.* **14**, 119 (2015).
8. Tetzlaff, J.M., Moher, D., Chan, A.W. Developing a guideline for clinical trial protocol content: Delphi consensus survey. *Trials.* **24** (13), 176 (2012).
9. Glasser, S.P., Howard, G. Clinical trial design issues: At least 10 things you should look for in clinical trials. *Journal of Clinical Pharmacology*. **46** (10), 1106-1115 (2006).
10. Ruikar, V. Interactive voice/Web response system in clinical research. *Perspectives in Clinical Research.* **7** (1), 15-20 (2016).
11. Kulkarni, P.M., Meadows, E.S., Ahuja, S., Muram, D., Plouffe, L. Rationale for a non-inferiority clinical trial design focused on subpopulations. *Current Medical Research and Opinion*. **20** (10), 1641-1647 (2004).
12. Efird, J. Blocked randomization with randomly selected block sizes. *International Journal of Environmental Research and Public Health.* **8** (1), 15-20 (2011).
13. Teitelbaum, D.H. et al. Proceedings From FDA/A.S.P.E.N. Public Workshop: Clinical Trial Design for Intravenous Fat Emulsion Products, October 29, 2013. *Journal of Parenteral and Enteral Nutrition*. **39** (7), 768-786 (2015).
14. Pocock, S.J., Clayton, T.C., Stone, G.W. Challenging issues in clinical trial design: Part 4 of a 4-Part series on statistics for clinical trials. *Journal of the American College of Cardiology*. **66** (25), 2886-2898 (2015).

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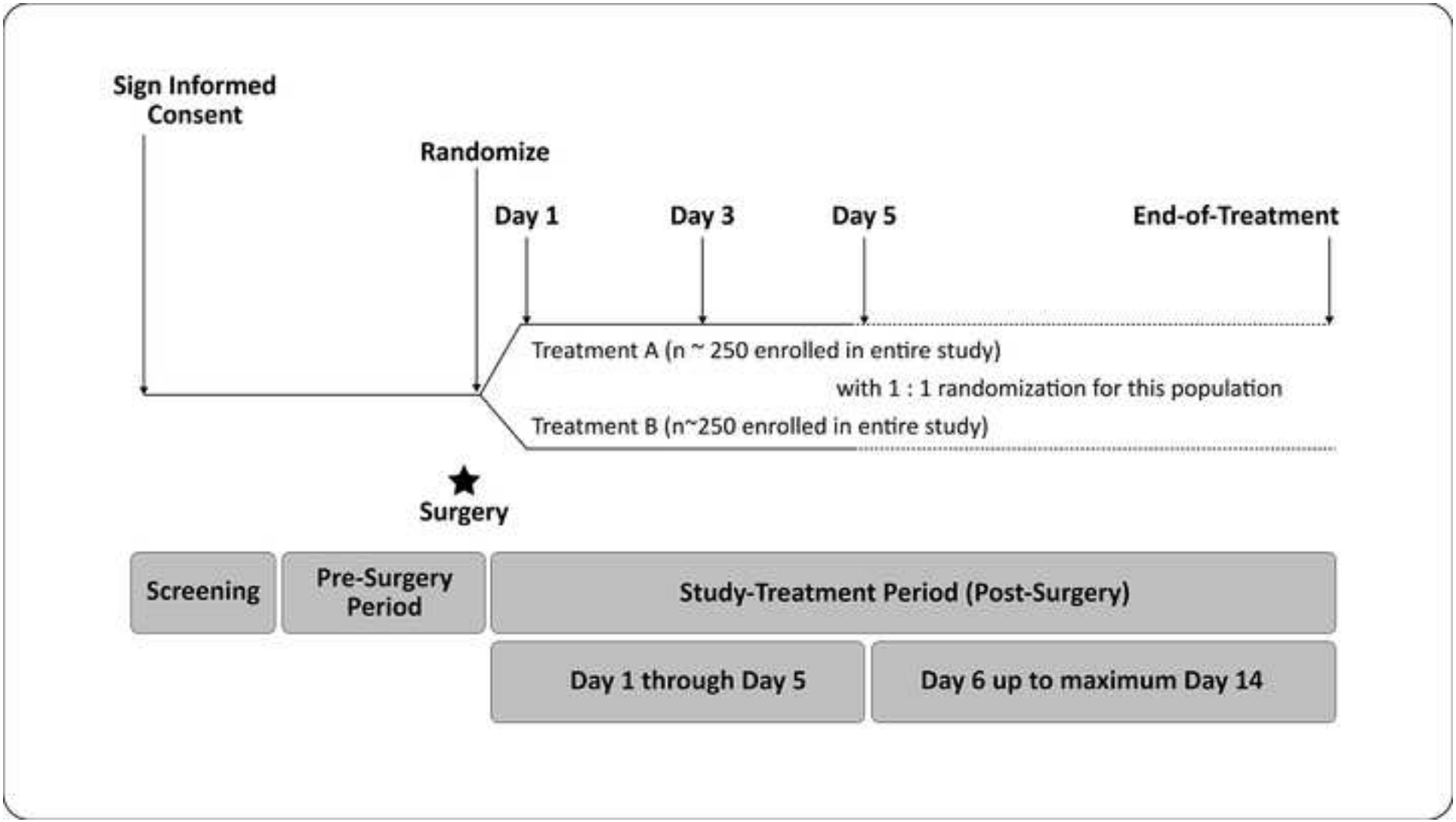

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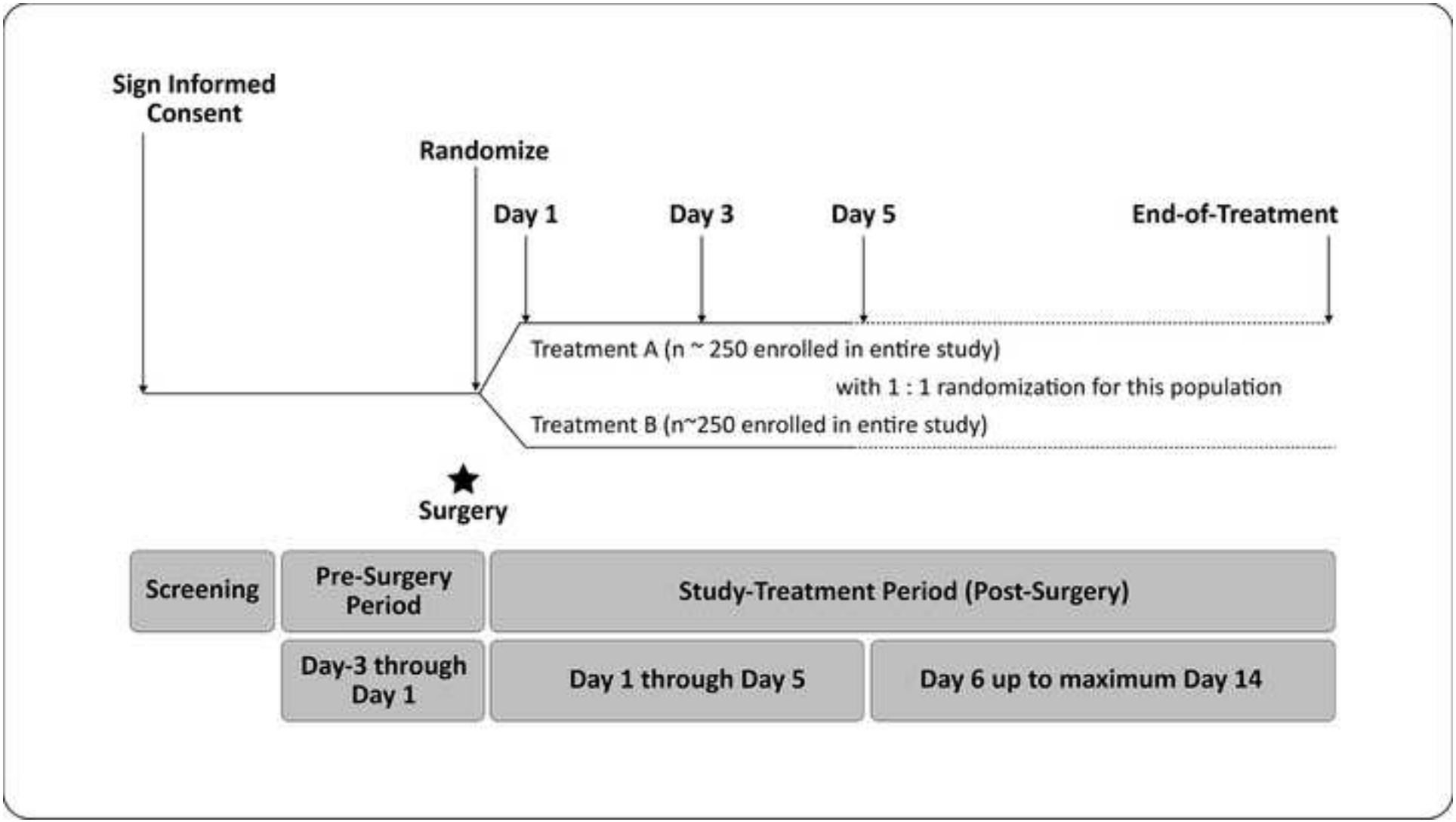

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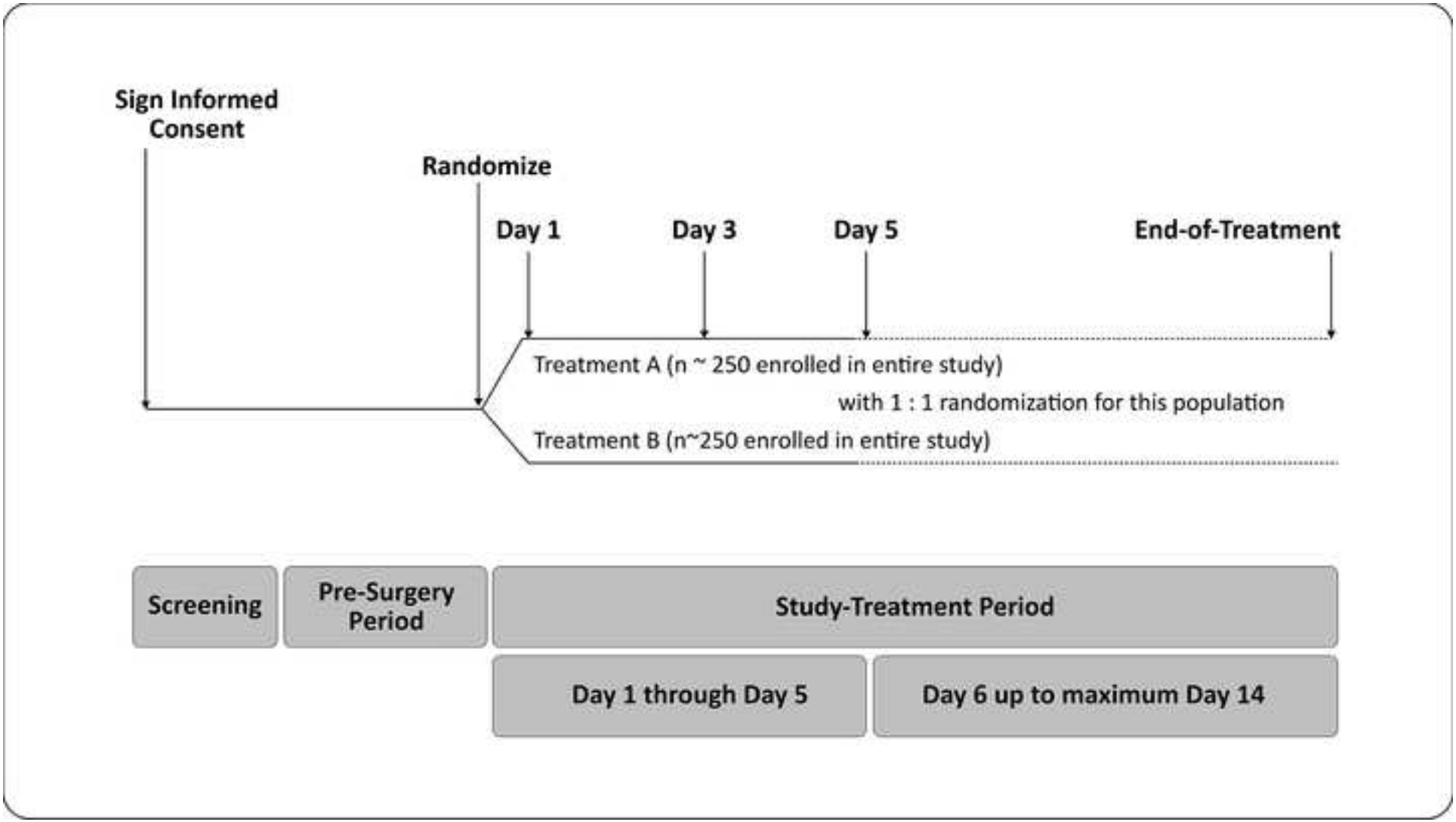

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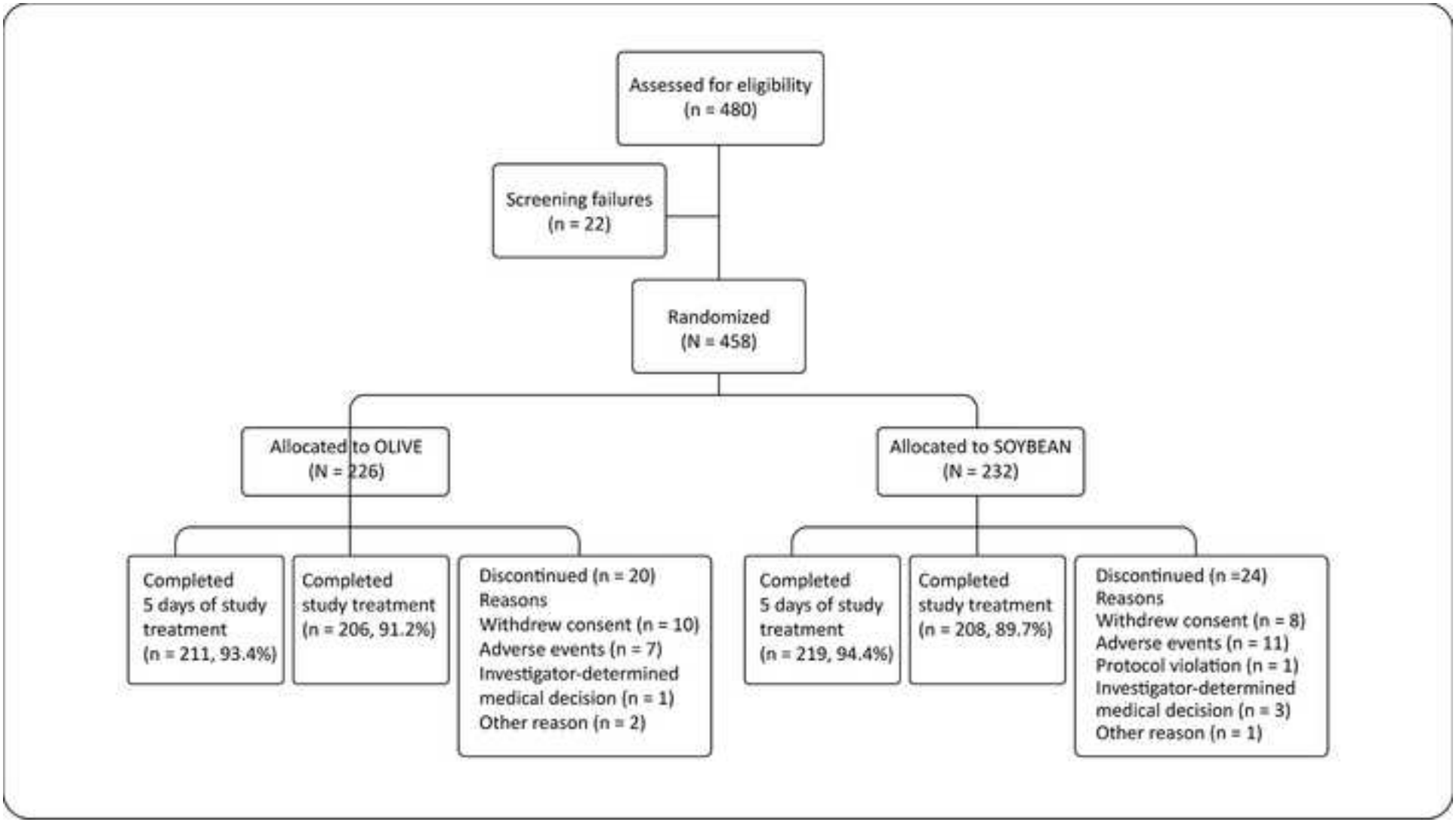

Figure 5

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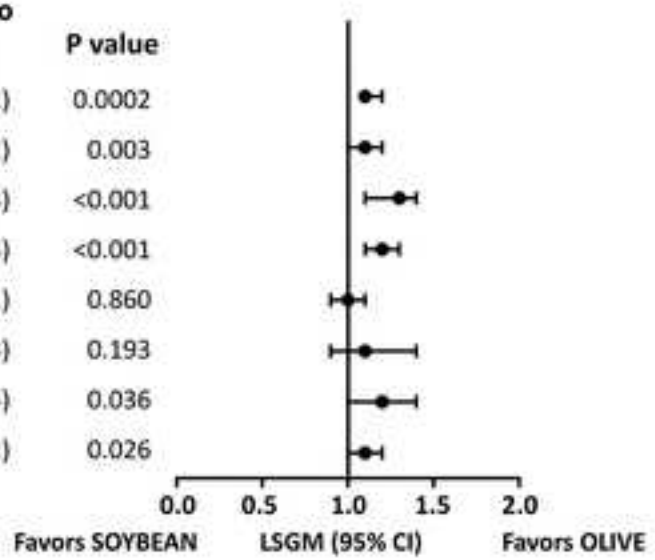
Population	Prealbumin Day 5 (mean ± SD) OLIVE	SOYBEAN	LSGM Ratio (95% CI)	P value
mITT population	15.7 ± 5.1	14.0 ± 5.1	1.1 (1.1, 1.2)	0.0002
Age <65 years	16.0 ± 5.1	14.5 ± 5.1	1.1 (1.0, 1.2)	0.003
Age >65 years	14.9 ± 5.2	12.1 ± 4.6	1.3 (1.1, 1.4)	<0.001
Male	16.2 ± 5.0	13.4 ± 4.9	1.2 (1.1, 1.3)	<0.001
Female	14.9 ± 5.2	14.9 ± 5.2	1.0 (0.9, 1.1)	0.860
No surgery	17.1 ± 6.1	14.2 ± 5.7	1.1 (0.9, 1.4)	0.193
Medium complexity sugery	15.3 ± 5.6	13.3 ± 5.2	1.2 (1.0, 1.4)	0.036
High complexity surgery	15.4 ± 4.7	14.0 ± 4.6	1.1 (1.0, 1.2)	0.026



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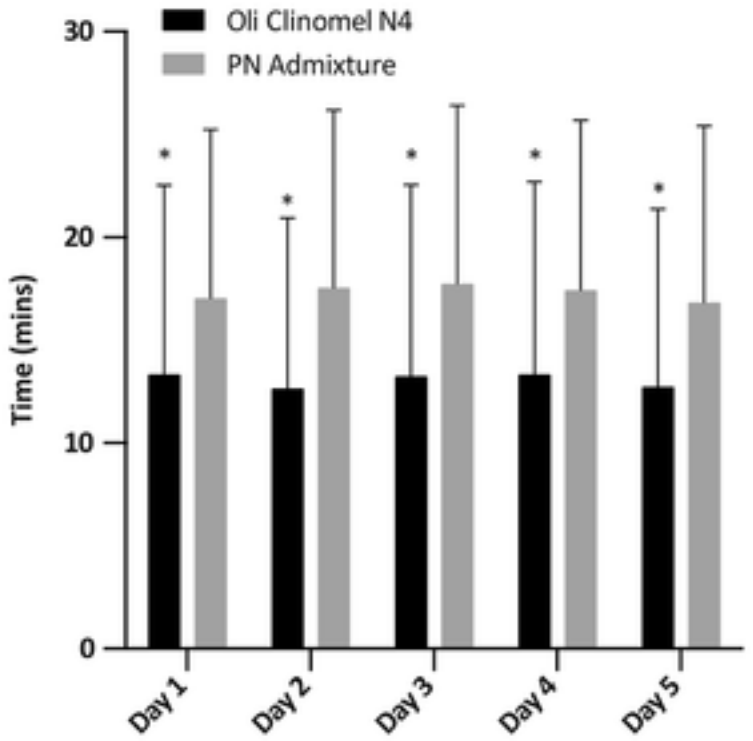

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Study period	Screening	Pre-surgery [a]	Post-surg	
			Baseline	
Visit	**1**	**2-4**	**5**	**6**
Time	Screening	Days -3 to -1	Hour 0	Day 1
Procedure/assessment		Before surgery [b]		
Informed consent/ authorization [c]	X			
Inclusion/exclusion Criteria	X	X		
Medical history	X			
Demographics	X			
Height	X			
Body weight [d]	X		X	X
Vital signs [e]	X	X	X	X
Physical examination: Complete	X [f]		X [f]	
Physical Examination: Focused				X [g]
Randomization [h]	X [h]		X [h]	
Study treatment [i]		X		X
Oral/enteral nutrition				
Calculation of daily nutrition		X	X	
Calculation of proportion of daily nutrition requirement [j]				
Documentation of peripheral IV		X		X
Clinical efficacy assessments [k]	X	X	X	X
Local clinical laboratory (safety) assessments [l]	X		X	
Venous blood for central laboratory (efficacy) assessments	X [m]		X [m]	
Urine for central laboratory (efficacy) assessments			X [p]	
Pre-study medications	X			
Concomitant medications		X	X	
Adverse events [q]	X	X		X

a: Pre-surgery study treatment period refers to the time period (up to 3 days) when the subje

b: If a subject underwent surgery and started study treatment preoperatively, the study proce

c: Informed consent/authorization must be obtained prior to performing any protocol-specific

d: Body weight must be measured and recorded at screening and at EOT. Body weight, if meas

e: Vital signs: Include body temperature, respiration rate, pulse rate, and systolic and diastolic

f: Complete physical examination assessing the major body systems.
g: Focused physical examination.
h: The inclusion/exclusion criteria were confirmed prior to randomization. For a patient under
i: Administered using an infusion pump set to the daily volume per prescribing orders. The pr
j: Calculation of the proportion of daily nutrition requirement that was administered by liquic
k: Clinical efficacy assessments include: Time for study treatment preparation (day 1 through
l: Insulin: Hematology: Red blood cells (RBC), hemoglobin, hematocrit, white blood cells (WBC
m: Venous blood samples for efficacy assessment were collected during screening and at hour
n: Venous blood samples were collected within 3 to 4 hours from the initiation of study treatm
p: Venous blood samples were collected at least 1 hour after the last study treatment. These s
q: Serious adverse events (SAEs) were collected from the time of signing the informed consent
r: Any day from day 6 through day 13 was the final treatment day if the subject received at le
o: Day-14 procedures were performed for subjects who still required at least 20% of their daily

ery study treatment period				Study treatment				
				Day 1 through day 14				
7	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**
Day 2	Day 3	Day 4	Day 5	Day 6	Day 7 [r]	Day 8 [r]	Day 9 [r]	Day 10 [r]
Study treatment only				Study treatment plus liquid or				
X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X
X [g]	X [g]	X [g]	X [g]	X [g]	X [g]	X [g]	X [g]	X [g]
X	X	X	X	X	X	X	X	X
				X	X	X	X	X
X	X	X	X	X	X	X	X	X
				X	X	X	X	X
X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X
	X				X			
	X [n]		X [m, o]					
			X p					
X	X	X	X	X	X	X	X	X
X	X	X	X	X	X	X	X	X

ct received study treatment prior to surgery. If the subject did not undergo surgery, or if the sul
dures during the pre-surgery period were performed and the initiation of the first study treatm
: assessments.
sured for routine standard of care, should be recorded for each treatment day and, in particular
: blood pressure. Body temperature was measured as an axilla temperature and was obtained ir

going surgery without peroperative parenteral nutrition (PN), inclusion/exclusion criteria confir
escribed and administered (actual) volumes and duration of each study treatment were record
l oral or enteral nutrition was performed daily from day 6 through the remainder of the study p
day 5, documented in pharmacy record); surgical incision assessment (day 1, day 5, and end of s
C) with differential, platelet count, and prothrombin time. Serum chemistry: glucose, blood urea
0 (just prior to initiation of study treatment on day 1) and within 3 to 4 hours from initiation of
ment on day 3, for measurement of hormones. These samples were processed and sent to the c
samples were processed and sent to the central laboratory. Samples to assess serum prealbumi
t/authorization to EOT. AEs were collected from the initiation of study treatment to EOT. AEs oc
ast 80% of their daily energy requirement from liquid oral or enteral nutrition. At the completio
y energy requirements from the study treatment on day 14.

period				End of treatment
16	17	18	19	EOT [r]
Day 11 [r]	Day 12 [r]	Day 13 [r]	Day 14 [s]	Varies [r]
al/enteral nutrition				
X	X	X	X	X
X	X	X	X	X
X [g]	X [g]	X [g]	X [g]	X [g]
X	X	X	X	
X	X	X	X	
X	X	X	X	
X	X	X	X	
X	X	X	X	
X	X	X	X	X
X			X	
			X [m]	X [o]
			X [p]	X [p]
X	X	X	X	X
X	X	X	X	X

bject underwent surgery without preoperative study treatment, this time period did not apply and t
ient after surgery was hour 0.

, at baseline (hour 0) and on day 5.
n the supine or sitting position; respiration rate, pulse rate, and systolic and diastolic blood pressure

mation and randomization were performed after surgery.
ed.
period.
study [EOT]); intravenous (IV) site assessment; length of hospitalization, mechanical ventilation (if ap
a nitrogen (BUN), creatinine, bicarbonate (total carbon dioxide), sodium, potassium, chloride, phosp
the study treatment on day 5 and day 14 (if applicable).
entral laboratory.
n were drawn for all subjects at EOT. For all other efficacy laboratory measurements, venous blood s
curring after the initiation of study treatment were considered treatment-emergent.
on of the final treatment, EOT procedures were performed.

:he initiation of study treatment was hour 0.

: were obtained after the subject had been in the supine position for a minimum of 5 minutes. (

pplicable), and ICU (if applicable); date of first bowel movement after surgery.
horus, triglycerides, total cholesterol, conjugated bilirubin, total bilirubin, alanine aminotransfe

samples were drawn only from subjects who received their final study treatment on day 9 and l

Clinically relevant changes in vital sign measurements consistent with worsening clinical status v

erase (ALT), aspartate aminotransferase (AST), alkaline phosphatase, and gamma-glutamyl trans

later. A urine sample collection was obtained prior to initiation of study treatment, on day 5 an

*n*ere recorded as adverse events (AEs).

sferase (GGT). These parameters were measured in the local clinical laboratory. The investigato

d on day 14 (if applicable), or EOT. On day 5 and day 14 (if applicable), the collection began at le

r was expected to review the laboratory data in real-time, by signing and dating laboratory prin

east 1 hour from initiation of study treatment and continued for 6 hours. For subjects who recei

touts (or other media). Clinically relevant changes in safety laboratory values consistent with w

ived their final study treatment on day 9 through day 13, the EOT urine collection began at leas

orsening clinical status were recorded as AEs.

t 1 hour from the end of last study treatment and continued for 6 hours. The volume of the uri

ne collection was recorded, and then an aliquot from the urine collection was taken. These sam

ıples were processed and sent to the central laboratory.

Table 2 Click here to access/download;Table;Tab2.xlsx

Variable	OLIVE 3CBs (n=226)	SOYBEAN CoBs (n=232)	p-value
Sex, n (%)			0.482
Male	134 (59.3)	145 (62.5)	
Female	92 (40.7)	87 (37.5)	
Race, n (%）			0.673
Chinese Han	216 (95.6)	220 (94.8)	
Chinese (Other minority)	8 (3.5)	11 (4.7)	
Other	2 (0.9)	1 (0.4)	
Underwent surgery, n (%)	195 (86.3)	202 (87.1）	0.805
Complexity of surgery, n (%)			0.859
Medium complexity	49 (21.7)	48 (20.7)	
High complexity	140 (61.9)	143 (61.6)	
Missing	37 (16.4)	41 (17.7)	
Age, years, mean ± SD	55.8 ± 13.1	56.3 ± 11.7	0.656
BMI, kg/m^2, mean ± SD	21.7 ± 3.9a	21.8 ± 3.9 b	0.667
Duration of surgery, (hours) mean ± SD	2.9 ± 1.3	3.0 ± 1.4	0.645

BMI: Body mass index;kg/m2: kilogram per square meter;SD: Standard deviation;p value <0.05 was cc

a= (n = 217) ;b= (n = 226); * Information on the complexity of the surgery was missing

›nsidered statistically significant

Table 3

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Infection	OLIVE 3CBs (n=222) (%)	SOYBEAN CoBs (n=231) (%)
Total infections	8	26*
Total patient infected	8 (3.60)	24*(10.4）
Lung infections	2 (0.09)	13 (5.62)
Incision/wound site infections	5 (2.25)	3 (1.29)
Urinary tract infections	1 (0.04)	2 (0.086)
Abdominal/gastrointestinal infections	0 (0)	2 (0.086)
Scrotal infections	0 (0)	1 (0.043)
Non-specified infections (Systemic infection, site not identified)	0 (0)	5 (2.16)
*P <0.01		

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Editorial comments:

Changes to be made by the Author(s):

Note: We have checked all the following items and made the alterations according to these comments. For comment 4,9 and 13, Responses have been given respectively.

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9. Figures 1-3: In the Figure legend please briefly explain what Treatment A and Treatment B are. Is the surgery day considered as day 0? Please label it if so.
   As per the methodology Day 0 is not the surgery
10. Figure 4: The vertical line connecting the box of “Completed study treatment (n = 208, 89.7%)” is missing. Please add it. It will also be helpful if you can label Treatment A and Treatment B. Should the number of those assessed for eligibility add up to 500?
11. Figure 5: Please explain P value in the figure legend.
12. Figure 6: Please revise the figure legend to be clear. At least explain what are Oli Clinomel N4 and PN Admixture. Please use SI abbreviations for time: min instead of mins
13. Please obtain explicit copyright permission to reuse any figures from a previous publication. Explicit permission can be expressed in the form of a letter from the editor or a link to the editorial policy that allows re-prints. Please upload this information as a .doc or .docx file to your Editorial Manager account. The Figure must be cited appropriately in the Figure Legend, i.e. “This figure has been modified from [citation].”
   Not applicable, as no figures are copied
14. Table 2: Please explain what the variable “Missing” refers to.
15. Table 3: Please specify what “*” means.
16. Please upload each table separately as an xls/xlsx file.
17. Please place the superscripted numbered reference before the punctuation.
18. Please revise the Introduction to include all of the following:

a) A clear statement of the overall goal of this method

b) The rationale behind the development and/or use of this technique
c) The advantages over alternative techniques with applicable references to previous studies
d) A description of the context of the technique in the wider body of literature
e) Information to help readers to determine whether the method is appropriate for their application
19. The Protocol should contain only action items that direct the reader to do something. Please move the discussion about the protocol to the Discussion.
20. Please ensure that all text in the protocol section is written in the imperative tense as if telling someone how to do the technique (e.g., “Do this,” “Ensure that,” etc.). Avoid usage of phrases such as “could be,” “should be,” and “would be” throughout the Protocol. Any text that cannot be written in the imperative tense may be added as a “Note.” However, notes should be concise and used sparingly. Please include all safety procedures and use of hoods, etc.
21. The Protocol should be made up almost entirely of discrete steps without large paragraphs of text between sections. Please simplify the Protocol so that individual steps contain only 2-3 actions per step and a maximum of 4 sentences per step.
22. Please remove all commercial language from your manuscript and use generic terms instead. All commercial products should be sufficiently referenced in the Table of Materials and Reagents.
For example: SAS version 9.2 (SAS Institute Inc, Cary, North Carolina USA), etc.
23. Please adjust the numbering of the Protocol to follow the JoVE Instructions for Authors. For example, 1 should be followed by 1.1 and then 1.1.1 and 1.1.2 if necessary. Please refrain from using bullets or dashes. See substeps in 4.1.
24. 2.4: Should it be “prior to a surgical procedure” instead of “after a surgical procedure”?
25. Lines 239-244: Please break up this paragraph into numbered substeps.
26. Lines 274-278: Please explain how these are actually done.
27. Line 281: Please spell out EOT and explain how this procedure is performed.
28. Please revise Steps 4.2, 4.3, and 5 of the protocol to include several action steps. Any text that cannot be written in the imperative tense may be added as a “Note.”
29. Please note that we can film only steps 3.1 and 4.1. If you wish to have a video of the protocol, then we need an actual protocol that can be filmed. Patient exclusion/inclusion criteria as well as recruitment/enrollment and statistical methods are not appropriate to be filmed.
30. Please highlight 2.75 pages or less of the Protocol (including headings and spacing) that identifies the essential steps of the protocol for the video, i.e., the steps that should be visualized to tell the most cohesive story of the Protocol. Remember that non-highlighted Protocol steps will remain in the manuscript, and therefore will still be available to the reader.
31. Please ensure that the highlighted steps form a cohesive narrative with a logical flow from one highlighted step to the next. Please highlight complete sentences (not parts of sentences). Please ensure that the highlighted part of the step includes at least one action that is written in imperative tense.
32. Please revise to explain the Representative Results in the context of the technique

you have described, e.g., how do these results show the technique, suggestions about how to analyze the outcome, etc. The paragraph text should refer to all of the figures.

33. please shorten the Discussion to explicitly cover the following in detail in 3-6 paragraphs with citations:

a) Critical steps within the protocol

b) Any modifications and troubleshooting of the technique

c) Any limitations of the technique

d) The significance with respect to existing methods

e) Any future applications of the technique

34. Please revise the table of the essential supplies, reagents, and equipment. The table should include the name, company, and catalog number of all relevant materials in separate columns.

35. Please ensure that the references appear as the following: [Lastname, F.I., LastName, F.I., LastName, F.I. Article Title. Source. Volume (Issue), FirstPage – LastPage (YEAR).] For more than 6 authors, list only the first author then et al.

36. Please do not abbreviate journal titles.