Informed Consent

Information for Subjects

Dear Ms./Mr:

We are going to conduct a study on effects of late evening snack on nutritional status and metabolic pattern of patients with primary hepatocellular carcinoma after hepatectomy, and you may be eligible for enrollment in the study, therefore, we invite you to participate in the study, which is sponsored by Drum Tower Hospital Affiliated to the School of Medicine of Nanjing University, and the principal investigators of the study in our center are Bian Xiaojie, Associate Chief Pharmacist and Du Yao, Pharmacist-in-Charge.

This informed consent form will tell you about the purpose of the study, the procedure, the benefits to you, the risks, inconveniences or discomforts you will have to bear, and the main aspects of the study, as well as the other treatment options available to you and your right to withdraw from the study at any time. Please read it carefully and make your decision about whether or not to participate in the study. This informed consent form may contain words or information that you do not understand, so be sure to ask your study pharmacist and you will get answers until you are satisfied. Before making a decision, you may take the unsigned informed consent form home to think about or talk to your family, friends, or anyone you choose. If you decide to participate in this study, please sign and date this informed consent form. Your signature does not deprive you of any legal rights, and the original signed informed consent form will be kept with the researcher and another copy will be kept by you.

1. Research background

Primary liver cancer (hereinafter referred to as hepatocellular carcinoma) is one of the common malignant tumors of the digestive system worldwide, and hepatectomy is considered to be the most effective treatment for early-stage liver cancer. Meanwhile, the liver is the target organ for most nutrient metabolism, and the high metabolic status and organ invasion of cancer often expose liver cancer patients to malnutrition, with an incidence rate as high as 74.36% according to the survey. In addition, hepatectomy will cause liver cell damage to a certain extent, resulting in the metabolic balance of sugar, protein, fat and hormone in the patient's body being disrupted, making the body's catabolic metabolism stronger than the anabolic metabolism, and making it more likely to suffer from the decrease of albumin synthesis level and the significant decrease of nutrient intake after the surgery, which will then cause the decline of patients' ability of resisting infections, the increase of the risk of complications and the decrease of the quality of life, as well as a series of bad prognosis. Therefore, in recent years, the mode of Late-evening snack (LES) has been recommended by

various guidelines and consensus of the European Society of Clinical Nutrition and Metabolism (ESPEN), the International Society for Hepatic Encephalopathy and Nitrogen Metabolism, etc. LES refers to moving part of the food to be eaten before bedtime under the condition that the total amount of daily food remains unchanged, and to shorten the interval between meals as much as possible, which can be effective for the relief of accelerated hunger and associated protein hydrolysis. In most studies, various combinations of LES (total calories 200-275kcal, protein 11.5g-18g, complex carbohydrates 25-55g) have been shown to be meaningful in improving liver function indices and nutritional indices. Therefore, the aim of this study was to investigate the optimal management of patients' eating time and frequency through bedtime meal addition and dietary guidance, with a view to improving the accelerated catabolic and nutritional status of patients and reducing complications.

2. Research Purpose

The aim of this study was to use LES to optimize the management of subjects' eating time and frequency with a view to reducing complications and improving clinical outcomes, and to collect blood samples remaining after the subjects' routine review tests in a one-step study to investigate the effects of LES on subjects' metabolic patterns, providing a theoretical basis for the implementation of effective nutritional strategies in clinical practice to promote the subjects' recovery.

3. Conditions to be fulfilled for participation in studies/research institutes

Inclusion Criteria

1) Age 18-75 years;

2) Meet the diagnostic criteria of China's "Guidelines for Diagnosis and Treatment of Primary Liver Cancer (2022 Edition)", clinically diagnosed with primary liver cancer, hospitalized with radical hepatectomy as the main surgical treatment, no indication of metastasis of the tumor to extra-hepatic organs in preoperative tests and examinations, no absolute contraindications to surgery, complete resection of the liver tumor in the operation, and hepatocellular carcinoma confirmed by postoperative pathological diagnosis； 3. Child-Pugh grades A and B;

4) Preoperative Eastern Cooperative Oncology Group Physical Status Score (ECOG-PS) of 0 to 2;

5) The patient is conscious, has normal verbal communication, and is able to cooperate with the relevant examinations;

6) Fully informed about the study and voluntarily signed an informed consent form.

Exclusion Criteria

1) Failure to meet selection criteria;

2) Nutritional assessment as cachexia;

3) Presence of contraindications to enteral nutrition (EN) or EN intolerance, such as acute gastrointestinal bleeding, intestinal obstruction. (≥grade 3, National Cancer Institute-Common Terminology Criteria for Adverse Events [NCINCI-CTCAE v 5.0]);

4) Simultaneous combination of malignant tumors in other parts of the body;

5) Combined hepatic encephalopathy or definite infection on admission;

6) Known refractory metabolic diseases (e.g., poorly controlled diabetes mellitus or fasting glucose ≥10 mmol/L, hyperthyroidism, hypothyroidism, metabolic acidosis);

7) Decreased renal function (defined as serum creatinine Cr level ≥176.8 μmol/L);

8) Intravenous or oral nutritional supplements, such as proteins, amino acids, etc., applied within one month prior to admission to the hospital;

9) Patients with severe stress or severe complications such as respiratory failure with severe cardiac, hepatic, renal and other insufficiencies；

10) Persons with mental and neurological disorders who are unable to cooperate with a physician;

11) Alzheimer's disease, cerebral atrophy, acute stage or sequelae of cerebrovascular disease, cognitive impairment;

12) Previously poor adherence to medication and nutritional counseling;

13) Critically ill and difficult to assess;

14) On the liver transplant waiting list or under consideration for liver transplantation, as such patients may discontinue follow-up before the end of the study;

15) Less than 12 months since last localized treatment (TACE or HAIC or ablative therapy);

16) Other circumstances that the researcher considers inappropriate for participation in the study.

Specifics of withdrawal from the study/trial:

1) The subject does not wish to continue in the clinical trial and, in accordance with the Declaration of Helsinki and the informed consent form, the subject has the right to withdraw at any stage of the trial, and his/her subsequent treatment and follow-up will not be affected as a result;

2) Disease progression/treatment failure or worsening of the disease (unless the investigator

believes that there is still a potential benefit to be gained by continuing treatment);

3) the occurrence of a serious adverse event that does not improve with optimal medical therapy and is not tolerated and, in the judgment of the investigator, requires termination of treatment;

4) a general deterioration in health or, in the judgment of the Investigator, termination of the study in the best interest of the subject;

5) Use of other nutritional supplements by the subject that are not taken at the prescribed times, or other medications (other than those permitted by the protocol) that, in the judgment of the investigator, would have a greater impact on the efficacy of the treatment;

6) Postoperative pathology suggestive of non-primary or metastatic hepatocellular carcinoma;

7) Patients who did not take more than 7 additional meals before bedtime during the follow-up period.

4. Number and duration of research

This study is planned to run for 2 years and 106 subjects will be recruited.

5. Is it mandatory to participate and complete this study?

Your participation in this study is completely voluntary and if you decide to participate, you will be asked to sign an informed consent form and will be given a copy of this informed consent form. If you take part in this study, you can still ask to leave at any time, and your standard treatment will not be affected if you leave.

6. Research/Research process

This study is a prospective cohort study using the grouping method of patients' wishes. You will learn about the 2 dietary patterns, bedtime meal and regular diet, and then choose to enter the different groups according to your wishes. Patients who volunteer for the bedtime meal diet will be in the experimental group and those who volunteer for the regular diet will be in the control group. Both groups will have dietary counseling and discharge follow-up, with or without the bedtime meal pattern being the difference.

The experimental group treatment protocol is:

patients were given additional meal (total calories 200-275kcal, protein 11.5g-18g, complex carbohydrates 25-55g) 1h before bedtime. the LES regimen was: ① whole protein type enteral nutrition; ② whey protein powder + bread or cookie. The addition of meals was carried out 1h before bedtime. The dietary regimen of the experimental group will be deducted from the corresponding late-evening snacks intake.

(1) Preoperative: ① Patients without nutritional risk: independent intake according to the issued diet program card. ② Patients with nutritional risk: daily oral whole-protein enteral nutrition on the basis of general diet after admission.

(2) Postoperative: start drinking small amount of water on the 1st postoperative day, over to liquid diet after ventilation, and may over to autonomous semi-liquid diet + oral whole protein enteral nutrition during the day + LES program on the 2nd postoperative day. As the patient's voluntary intake meets 60% of the body's needs, the daytime oral enteral nutrition support can be gradually stopped and the LES program can be continued.

(3) Discharge: daily intake needs to be on time according to the LES regimen.

Subsequent night snacks until disease recurrence or gastrointestinal intolerance or the occurrence of other conditions specified in the program that require termination of treatment, whichever occurs first.

The control group treatment protocol is:

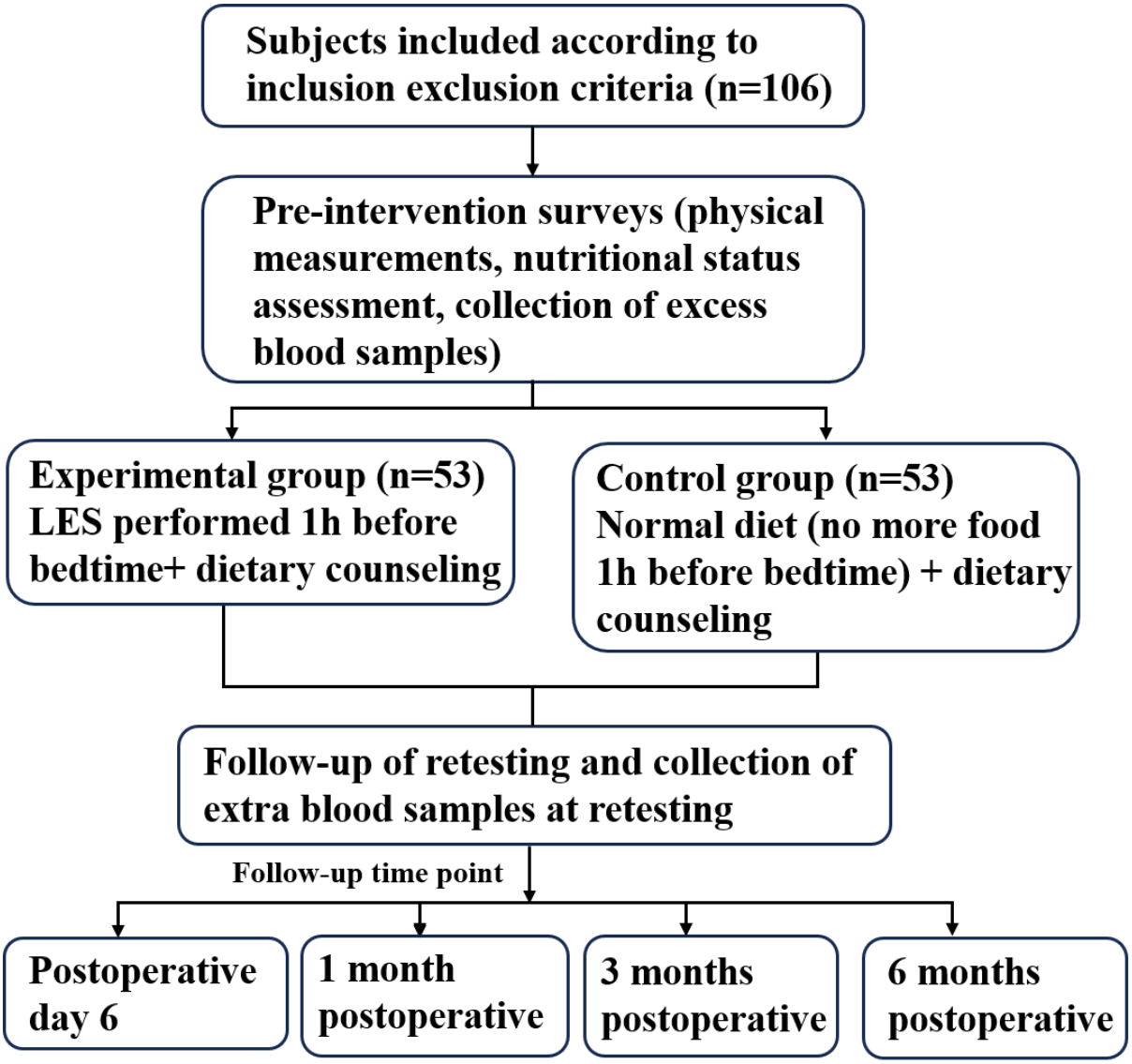
no more food 1h before bedtime.

(1) Preoperative: same as the experimental group.

(2) Postoperative: start drinking small amount of water on the 1st postoperative day, over to liquid diet after ventilation, and can over to autonomous semi-liquid diet + oral whole protein type enteral nutrition during the day on the 2nd postoperative day. As the patient's voluntary intake meets 60% of the body's needs, enteral nutrition support can be gradually stopped.

(3) Discharge: Doctors judge whether the patients need to continue nutritional support according to their dietary recovery, and if they do, their nutritional supplementation should be completed before 1h before bedtime.

The process of the entire study is the following flowchart, and the study period amounted to 6 months:



7. Participation in the study and what you need to do with your cooperation

- provides accurate past medical history and current condition information.
- Tell the pharmacist in charge of the study that you need to tell your doctor about any health problems you develop during the study, whether you think they are related to your treatment or not.
- Tell the pharmacist in charge of the study about any new drugs, medicines, vitamins, nutritional supplements, or herbs you are taking during the study.
- Please be sure to take your bedtime meal correctly if you are in the experimental group, or if you are in the control group if you need to take a nutritional preparation. Both groups will be visited as required and will be fasting at the time of the visit so that you can review your blood draws.
- During the study period, refer to the issued dietary guidance card intake, to ensure that the body needs daily calories.

- Follow the guidance of the researcher and research pharmacist.
- Feel free to ask if there is anything unclear at any time.

8. Alternative treatment options if you do not participate in the study

You may choose not to participate in the study, which will have no adverse effect on your access to conventional treatment. At present, the conventional treatment for your health condition is: eat small meals. The subjects eating less and more meals can reduce the postoperative abdominal distension and make it easier to consume the daily body requirement.

9. Possible side effects, risks, and discomfort associated with study participation

For the experimental group:

Your study pharmacist will monitor side effects such as gastrointestinal discomfort caused by extra meals before bed. If you experience any side effects or discomfort during the study, it is important that you report them immediately to your study pharmacist or doctor. The research pharmacist may give you other medications to control side effects. If you or your study pharmacist believe that you cannot tolerate these side effects, late-evening snacks will be stopped completely, and you may withdraw from the study.

For the control group:

Your study pharmacist will monitor for side effects, such as gastrointestinal discomfort, that may occur when you need to supplement whole-protein nutritional preparations if your self-intake is insufficient. If you experience any side effects or discomfort during the study, it is important that you report them immediately to your study pharmacist or doctor. The research pharmacist may give you other medications to control side effects. If you or your study pharmacist determines that you cannot tolerate these side effects, the supplement will be discontinued and you may withdraw from the study.

About nutrition preparations: Whole-protein enteral nutrition preparations may cause gastrointestinal symptoms such as bloating and diarrhea after taking. In order to prevent the occurrence of the above symptoms, the subject should warm the nutritional preparation in warm water and take it orally. If the above symptoms cannot be alleviated, stop taking it to observe whether it is caused by the eating pattern of late-evening snacks. If the symptoms persist, please inform the study pharmacist in time or seek medical attention nearby.

About Blood collection: Blood samples will be collected for this study. The blood sample comes from the excess blood sample drawn during your review and will not add additional burden to you. Temporary discomfort and/or bruising may occur during blood drawing.

10. Possible benefits of participating in a study

Participating in this study, you may be able to improve your nutritional status, such as improved albumin, prealbumin, alanine aminotransferase and other nutritional indicators and liver function indicators, slow weight loss, reduce lipid oxidation, etc., but we cannot guarantee this. This research could also help determine which treatments are safer and more effective in treating other patients with conditions similar to yours.

11. New information during the study

During the course of the research project, new information about late-evening snacks or nutritional supplements may emerge. If new information comes to light, your study pharmacist will inform you in a timely manner and discuss with you whether you wish to continue participating in the study. If you decide to discontinue participation in the study, your study pharmacist will arrange follow-up treatment for you. If you decide to continue to participate in the study, you may be asked to sign a new informed consent form. Or if your study pharmacist believes it is in your best interest to withdraw from the study, he/she will explain why and arrange for follow-up treatment.

12. Your rights

Participation in the study is entirely voluntary. You can withdraw your informed consent at any time without giving a reason. Whether you make a decision to participate or not to participate, it will not lead to bias against you or affect your medical care. If you do not participate in the study or drop out of the study, there are many alternative treatments that you do not have to choose to participate in the study in order to treat your disease. If you need to withdraw from the study, for your safety and objective evaluation of the effects of the drug, please cooperate with the study pharmacist to complete the relevant evaluation and laboratory examination after the study.

If you have any questions during the study, you can always consult your research pharmacist.

13. Costs of participation in this study/Study and treatment in the event of a research-related injury

Participating in this study, we will provide you with appropriate nutrition support plan formulation and individualized diet guidance free of charge. The cost of nutritional preparations and review costs need to be paid by you. The specific amount of costs is determined by your illness and cannot be estimated. The review items and nutritional preparations included in this study are all required for the original treatment. For patients in the experimental group, we will only guide your eating pattern and eating frequency; For the

control group, we only instruct you on the use of nutritional preparations and your daily diet. No matter which group you choose, there will be no additional cost to you.

14. Privacy confidentiality

Any information and data obtained about you personally during the course of the study will be kept strictly confidential. Your blood sample will be identified by a study number/number rather than your name, and information that identifies you will not be disclosed to members outside the study group unless you have given your permission. All research members are requested to keep your identity confidential. Your files will be kept in a locked cabinet and accessible only to researchers. In order to ensure that the research is carried out in accordance with the regulations, the government administration or the members of the ethics committee may consult the relevant information about your participation in the research unit as required, but they will ensure that your information will not be disclosed to other parties. Although the results of the research may be published, your identity will not be revealed in these publications. The data of this study will be stored in Drum Tower Hospital Affiliated to the School of Medicine of Nanjing University.

By signing this written informed consent, you consent to the study pharmacist's collection and processing of your personal information for the study (" Study Data "), including your birth date, gender, race, and physical and mental health status, which means that your study data will remain available until your informed consent is withdrawn. If you withdraw your informed consent, your personal data will no longer be used by the research pharmacist, but personal data that was shared prior to the withdrawal of informed consent can still be used.

Research pharmacists will use the study data to conduct clinical studies. You have the right to request the personal data held by the research pharmacist, and you also have the right to request the correction of inaccuracies in your personal data; You have the right to withdraw informed consent at any time, if you have the above request, please contact the research pharmacist.

15. Treatment after the study

After the study, your doctor and pharmacist will discuss your future treatment options with you.

16. Contact information

◆If a study-related injury occurs, or if you have any questions about the study, please contact:

Name of clinical pharmacist: __Yao Du __ address：__ Drum Tower Hospital Affiliated to the School of Medicine of Nanjing University__ Contact number：__86-18351887978___

◆ If you have any questions related to subjects' rights and interests, please contact the Medical Ethics Committee of Drum Tower Hospital Affiliated to Medical School of Nanjing University, telephone: 025-68182923.

Informed consent signing page

Subject Informed Consent Statement:

• I have read this informed consent form and obtained the background, purpose, steps, risks and benefits of this study. I have enough time and opportunity to ask questions about this clinical study and have received satisfactory answers.

• I understand that participation in this study is voluntary.

• I consent to the use and sharing of my medical information as described in the informed consent form.

• I know that I can withdraw from the study at any time without suffering loss of interest or other adverse consequences.

• I am willing to cooperate with the researchers to do the relevant examination or treatment.

• I understand that the identity and privacy of individuals participating in this study will be strictly confidential.

• I was also told who to contact when I had questions or wanted further information.

• I will obtain a signed and dated copy of this informed consent.

Subject's signature (printed)：_____ Contact number：_____

Subject's signature (handwritten): _____ Date: _____

Guardian signature [if applicable] (in print, please indicate the direct relationship with the subject) : _____ Contact number: _____

Guardian's signature (handwritten) :_____ Date: _____

The researcher performing informed consent states:

I or my research team have fully explained and explained to the subject the background, purpose, procedure, risks and benefits of this clinical study, and given him/her enough time to read the informed consent, discuss with others, and answer his/her questions about the study; I have given the subject contact information in case of problems; I have informed the subject (or guardian) that he/she may withdraw from the study at any time during the study period without any reason.

Investigator signature (printed)：_____ Contact number：_____

Investigator signature (handwritten): _____ Date: _____