Cover page:

Official title of the study:

Efficacy and safety of Astragalus for symptomatic alleviation in patients with high-grade lymphoma treated with chemotherapy: a double-blind randomized placebo-controlled trial

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Introduction

Lymphoma is a malignant disease of the lymph nodes. There are different types of lymphoma: when some of the lymphomas are indolent and require only follow-up, in cases of aggressive lymphoma or indolent lymphoma with a high disease burden, treatment is necessary, which usually includes chemotherapy with or without biological drugs. These drugs often cause significant damage to the quality of life, various symptoms including exhaustion, neuropathic pain, nausea/vomiting, cognitive decline, as well as a drop in blood counts with serious life-threatening infections that cause a delay in treatment initiation that may affect prognosis.[1] Conventional interventions for symptomatic relief and improvement in quality-of-life in patients with lymphoma have only a partial effect and various side effects.[1]

Astragalus (scientific name: Astragalus membranaceus, common name: Huang Qi) is a perennial flowering plant common in China with a long history of use in Traditional Chinese Medicine (TCM).[2] It is given in various forms in combination with other herbs, medicinal soups and as tablets, capsules, solutions, ointments for local use, or as subcutaneous or intravenous injection.[2] In patients with oncological diseases, a systematic review (4 trials on a total of 342 patients) showed an improvement in nausea and vomiting as well as in the immune system in patients with colon cancer who received chemotherapy.[3] A meta-analysis of 27 studies on 1,843 patients with colon cancer also demonstrated a reduction in chemotherapy-induced nausea in patients who received a combination of astragalus with chemotherapy compared to chemotherapy alone.[4] In another meta-analysis that included 45 trials on 3,236 patients with liver cancer, an improvement in the 12-month survival was reported with products containing astragalus.[5] In a randomized study of 136 patients with lung cancer, three treatment cycles with astragalus by injection in combination with chemotherapy (vinorelbine and cisplatin) resulted in a significant improvement in quality-of-life compared to the control group that received chemotherapy alone.[6] Similar results were found in 23 patients with metastatic malignancy receiving astragalus by injection in combination with chemotherapy.[7] In addition, a study on 90 patients with advanced cancer showed an improvement in fatigue in patients who received astragalus.[8] Also, a pilot study on 60 patients with breast cancer showed that intravenous administration of astragalus improved blood counts in patients who received it in combination with chemotherapy compared to chemotherapy alone.[9] In patients with hematological malignancies, a study conducted on 498 patients with acute myeloid leukemia showed that the use of astragalus or plants containing astragalus was associated with improved survival compared to patients who did not receive it.[10] In the context of lymphoma, a pharmacological study in vivo on animals with lymphoma showed that a combination of an astragalus-containing formula with adriamycin-type chemotherapy improved the effect of

adriamycin for the treatment of B-cell lymphoma.[11] Another study in patients who had completed conventional chemo-immunotherapy with R-CHOP for diffuse large B-cell lymphoma showed that the administration of a formula containing astragalus improved immune function and blood counts.[12] Finally, a randomized placebo-controlled study showed that giving a formula containing astragalus to patients under R-CHOP chemo-immunotherapy as first line for high-risk diffuse large B cell lymphoma significantly reduced the occurrence of side effects and improved response to chemotherapy compared to the control group.[13]

Side effects attributed to astragalus include fatigue, weakness, headaches and hypotension,[14] but in the cited studies and meta-analyses, no increase in the rate of side effects was found in the groups of patients who received this plant compared to the control groups, and no safety problems were described in the use of astragalus in combination with chemotherapy.

In light of the multitude of symptoms that affect the quality-of-life and adherence to treatment in patients with lymphoma receiving chemotherapy, and in light of the encouraging findings in the literature about symptomatic improvement as a result of combining astragalus during chemotherapy in various malignancies, we are interested in examining the efficacy and safety of combining astragalus during the treatment of patients with lymphoma with a high degree of malignancy on symptomatic relief and adherence to chemotherapy, in a randomized, double-blind, placebo-controlled study.

Methods

Study design: This is a double-blind randomized-controlled trial examining the efficacy and safety of Astragalus in patients receiving chemotherapy as a first-line treatment for high-grade lymphoma. The study protocol was reviewed and approved by the Institutional Review Board (IRB) in accordance with the Helsinki Declaration (BNZ-0036-24). Trial methods and results will be reported according to the Consolidated Standards of Reporting Trials (CONSORT) 2010 guidelines.

Study population: Inclusion criteria include age over 18, diagnosis of high-grade lymphoma, need to start treatment for lymphoma that includes chemotherapy as first line, possibility to answer questionnaires once a month and of signing an informed consent form. Patients will be excluded if they are taking in the week before the randomization a drug with the potential for an interaction of major clinical significance with Astragalus according to a professional database, if they received chemotherapy in the past, if they have a significant disturbance in liver function tests (AST and/or ALT above 5 times the upper limit of normal) or a significant renal dysfunction (GFR of less than 30 ml/min according to CKD EPI equation), if they have an active autoimmune

disease, in the case of pregnancy or breastfeeding, if they actively participate in another interventional study or if they have a psychiatric disorder with impaired cognition.

Randomization and blinding: After a screening period of up to 10 days, patients who fit the study according to inclusion and exclusion criteria and agree to participate, will undergo a 1:1 randomization according to computer software between the intervention and the placebo group. The random placement and distribution of the capsules will be performed by an independent coordinator to enable blinding of the process for both researchers and patients. The Astragalus and placebo capsules will be kept by the coordinator who will issue them according to a number that will appear both on the boxes and on the secret list of the random assignment. In the event of an unusual event involving safety and requiring disclosure of the patient group, the investigators may request the principal investigator to perform unblinding.

Intervention: Patients in the intervention group will receive capsules of Astragalus (scientific name: Astragalus membranaceus, common name: Huang Qi) at an initial dose of 2 grams (4 capsules) per day with the option to increase the dose to up to 4 grams (8 capsules) per day according to evaluation of the study investigators and patient tolerance. The dosage was determined by a focus group of 5 herbalists and according to data from the literature on oncology patients. Placebo capsules that look similar to Astragalus capsules will be given to patients from the control group, between 4 and 8 capsules per day according to the researcher's assessment and tolerance. The duration of the treatment will be up to one month from the end of the ongoing chemotherapy treatment (usually up to six months).

Follow-up: During the research treatment, the patients will undergo normal follow-up as usual for patients with high-grade lymphoma receiving chemotherapy, including a blood count at least once a week, a physical examination and imaging tests (CT, PET CT). In addition, patients will be required to answer questionnaires including MYCAW and EORTC QLQ-C30 once a month and EQ-5D once every 3 months for the duration of the intervention. Information will also be collected from the patients and their medical files including comorbidities, concomitant medications, nutritional supplements, use of complementary medicine, results of laboratory and imaging tests, adherence to the chemotherapy treatment, use of drugs for supportive care, transfusions and G-CSF. Of course, adverse events and serious adverse events safety reports will be disclosed. In addition, compliance with the research product will be checked according to the dispensing and counting of the capsules at each research visit (once a month). Finally, potential drug interactions of drugs and nutritional supplements that the patient receives with Astragalus will be checked by a clinical pharmacist. If there is a safety problem with the drug combination, the investigators will consider switching to another drug, delaying treatment with the study drug, or removing the patient from the study. After the

end of the study period, survival monitoring will continue for two years which will include an assessment of response to lymphoma treatment, lymphoma relapse or death. These findings will be reported in the manual CRF (Word document).

Visit	Screening (0-10 days)	Randomization	1, 2, 4, 5 months	3, 6 months	EOT	9, 12, 15, 18, 21, 24 months
Inclusion exclusion	X	X				
History	X					
CM	X		X	X	X	
Dietary supplements	X		X	X	X	
Chemo	X		X	X	X	
Interactions	X		X	X		
G-CSF data			X	X	X	
Transfusions data			X	X	X	
CBC data			X	X	X	
EORTC-QLQ-C30	X		X	X	X	
MYCAW	X		X	X	X	
EQ-5D	X			X	X	
Astragalus vs placebo		2 grams (4 capsules) daily	Increase to 3-4 grams daily as of investigator decision	2-4 grams daily		
Compliance to IP			X	X		
AE/SAE			X	X	X	X
Response to lymphoma treatment				X	X	X
Survival				X	X	X

Legend: AE: Adverse events; CBC: Complete blood count; Chemo: chemotherapy/biological therapies; CM: concomitant medications; EOT: End of treatment; IP: Investigation product; SAE: Severe adverse events

Criteria for exclusion from the trial: Patients may be excluded from the trial in the case of a major side effect of Astragalus treatment according to the researcher's assessment. Patients whose treatment with the investigational product is discontinued but who have not withdrawn their participation in the study will continue to be monitored as required by protocol.

Outcomes: The primary outcome is an improvement in the quality-of-life of patients with high-grade lymphoma receiving chemotherapy under the influence of the astragalus plant, according to the EORTC-QLQ-C30 questionnaire score.

Secondary outcomes include:

-General symptomatic improvement according to the MYCAW questionnaire

-Improvement in fatigue according to the EORTC-QLQ-C30 questionnaire fatigue score

-Improvement in nausea and vomiting according to the EORTC-QLQ-C30 questionnaire nausea and vomiting score

-Cognitive improvement according to the EORTC-QLQ-C30 questionnaire cognitive functioning score

-Decrease in the incidence of severe neutropenia (Absolute Neutrophil Count<0.5 x 10^9/L) during the course of treatment

-Decrease in the use of blood transfusions during the course of treatment

-Decreased use of G-CSF during the course of treatment

-Decrease in the incidence of infections during the course of treatment

-Decrease in the rate of hospitalizations during the course of treatment

-Improvement in adherence to chemotherapy treatment: according to relative dose intensity (RDI) calculation

-Improvement in lymphoma prognosis including ORR, CR, PFS, OS

-Safety of Astragalus during high-grade lymphoma chemotherapy according to reports of adverse events, serious adverse events and interactions with astragalus

-Economic evaluation according to EQ-5D values and calculation of QALY

Minimum sample size: In a randomized-controlled study that compared two different doses of Astragalus in 23 patients with metastatic gastrointestinal malignancies, it was found that patients who received treatment with Astragalus at each dose had an improvement in quality-of-life according to the EORTC QLQ-C30 questionnaire with a score decrease of 50.56±9.11 to 43.68±5.28. Therefore, and according to a two-way hypothesis with acceptable measures of alpha 0.05 and a power of 80%, a minimum sample size of 34 patients was calculated according to G*Power version 3.1 software. To perform additional analyzes as described above, it is necessary to add a 50% confidence interval, therefore at least 50 patients are required to undergo random assignment (that is, 25 in the intervention group and another 25 in the control group).

Data analysis: The data analysis will be performed using the IBM SPSS Statistics software. Demographic and clinical data will be analyzed at baseline to measure balance

between study groups. Quantitative variables will be described using mean and standard deviation or median with range (minimum and maximum) according to their distribution. Qualitative variables will be described using distribution, frequency and percentages. To compare variables that are normally distributed between the study groups, t-test for independent samples will be used. To compare variables that are not normally distributed between the study groups, Mann-Whitney test will be used. To compare qualitative variables between the research groups, Chi-square and Fisher's exact tests will be used. Survival analyzes will be performed using the Kaplan-Meier product survival limit estimator with log-rank comparison between groups, and Cox regression will be performed for multivariate adjustment of potential confounders. The hazard ratio (HR) with confidence intervals (CI) will be calculated for each independent variable and controlled for all other independent variables in the regression. All comparisons will be two-sided with a significance level set at $p<0.05$.

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