

## The GLOBE Trial: Efficacy and Safety of L-Glutamine Plus Hydroxyurea Versus Hydroxyurea Alone in Sickle Cell Anemia – A Double-Blind, Randomized Study

Shakibazad N. et al.: L-Glutamine Plus HU in Sickle Cell Anemia

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### ABSTRACT

**Background:** Hydroxyurea (HU) reduces complications of sickle cell anemia (SCA), but the response is variable. L-glutamine, an antioxidant that improves redox balance, addresses a distinct pathophysiological pathway and may provide additional clinical benefit when added to HU. We evaluated HU plus L-glutamine versus HU alone in pediatric/adolescent SCA.

**Methods:** In a 6-month, double-blind, placebo-controlled trial, 53 patients with HbSS or HbS/ $\beta^0$ -thalassemia were randomized to HU + L-glutamine (n=27) or HU + placebo (n=26) while continuing HU (~20 mg/kg/day). The primary endpoint was vaso-occlusive crisis (VOC) frequency; secondary endpoints included acute chest syndrome (ACS), hospitalizations, and hematologic parameters. Analyses were intention-to-treat with baseline-adjusted models for key outcomes.

**Results:** Over 6 months, the HU + L-glutamine group experienced significantly fewer VOCs ( $1.00 \pm 0.73$  vs.  $1.65 \pm 0.80$ ;  $p = 0.003$ ) and ACS episodes ( $0.19$  vs.  $0.77$ ;  $p = 0.006$ ). Hospitalizations declined by 40% ( $p = 0.04$ ). Hemoglobin rose more in the combination arm ( $+0.78$  vs.  $+0.32$  g/dL;  $p = 0.028$ ), with larger reductions in reticulocytes ( $p = 0.04$ ) and greater HbF increase ( $+6.2\%$  vs.  $+1.6\%$ ;  $p < 0.001$ ). Adherence exceeded 80% in both arms, and no serious adverse events occurred.

**Conclusions:** Adding L-glutamine to HU significantly reduced VOCs, ACS, and hospitalizations, while improving hemoglobin and hemolysis markers, without added toxicity. The combination's efficacy likely reflects synergistic effects on oxidative stress and sickle cell pathophysiology. This well-tolerated combination may improve SCA control, but larger confirmatory trials are needed.

**Keywords:** Sickle Cell Anemia, Hydroxyurea, L-Glutamine, Vaso-Occlusive Crisis, Pediatric Hematology

### Introduction

Sickle cell anemia (SCA) affects approximately 300,000 newborns annually, mainly in sub-Saharan Africa but also in the Middle East and South Asia. In southern Iran, the carrier rate is 1.43%, and the prevalence is 0.1% [1,2,3]. A  $\beta$ -globin mutation (Glu→Val) produces hemoglobin S, leading to sickling, vaso-occlusion, hemolysis, and

complications such as vaso-occlusive crisis (VOCs), acute chest syndrome (ACS), stroke, and organ damage. The disease causes substantial morbidity, premature mortality, and a high socioeconomic burden [4,5]. Hydroxyurea (HU) remains the standard therapy, increasing fetal hemoglobin (HbF) and reducing crises [6,7]. New agents (L-glutamine, crizanlizumab, voxelotor) have expanded the treatment options for SCD. L-glutamine improves redox balance and, in a pivotal trial including HU-treated patients, reduced VOCs, hospitalizations, and ACS, but high cost limits uptake [8,9,10,11,12,13]. Given their complementary mechanisms (HU increases HbF; L-glutamine reduces oxidative stress), combination therapy is considered safe and recommended for patients with suboptimal HU response. Inadequate response to HU alone often cannot be addressed by dose escalation due to toxicity [11,14,15]. In children and adolescents with SCA on stable HU, we compared HU + L-glutamine versus HU alone to quantify incremental benefit, hypothesizing fewer VOC/ACS events and improved hematologic outcomes.

## **Materials and Methods**

### ***Patients and Study Design***

This randomized, double-masked, placebo-controlled trial compared HU plus L-glutamine with HU plus placebo in SCA patients (HbSS or HbS- $\beta^0$ ) over 6 months. All participants received standard-dose HU and either L-glutamine or a placebo. The study was approved by the Ethics Committee (IR.BPUMS.REC.1400.107), registered (IRCT20210715051904N1, IRCT.IR), and conducted per CONSORT guidelines (Fig. 1). Written informed consent (and assent  $\geq 7$  years) was obtained. Medications and travel support were provided; patients in the placebo arm were later offered L-glutamine.

Eligible patients were  $\geq 5$  years, confirmed HbSS/HbS- $\beta^0$ , with  $\geq 2$  severe VOCs in the prior year, and stable HU doses (about 20 mg /kg/day) for  $\geq 3$  months. Key exclusions included HbSC or HbS- $\beta^+$  thalassemia, central organ insufficiency, recent transfusion, investigational therapy, or prior L-glutamine.

### ***Randomization and Blinding***

Patients were randomized 1:1 to HU plus L-glutamine or HU plus placebo via a secure, web-based permuted-block system (fixed block size 6) with the sequence generated by an independent statistician. Allocation concealment was ensured using opaque, sealed envelopes and centralized dispensing. L-glutamine and placebo sachets were indistinguishable in appearance, taste, and packaging, preserving blinding of participants, treating clinicians, investigators (including outcome assessors and analysts), and event adjudicators; only the dispensing pharmacist—who had no role in patient care—assembled sequentially numbered medication kits and held the code until database lock and trial completion.

### ***Sample Size***

Based on the sample size estimation reported by Niihara et al.[8], we estimated that 114 participants (57 per arm) were needed to achieve 80% power to detect a difference in VOC rate (two-sided  $\alpha=0.05$ , and power  $(1-\beta) = 0.80$ ), assuming an approximately 25–30% reduction in VOC with add-on L-glutamine and variability consistent with the prior trial. Due to feasibility constraints at our center, we targeted 60 enrollments and ultimately randomized 53 patients, yielding about 70% post hoc power; therefore, the study should be considered as exploratory/pragmatic, with limited power for secondary endpoints, although the primary outcome remained statistically significant with wider confidence intervals.

### ***Intervention***

L-glutamine (Endari<sup>®</sup>) was administered orally at a dose of 0.3 g/kg twice daily (maximum 30 g per day), provided in 5 g, 10 g, or 15 g powder sachets to be mixed with food or beverages. Placebo sachets contained weight-matched inert maltodextrin powder. HU dosing was maintained at the pre-study stable dose, averaging 20 mg/kg/day, with adjustments for tolerance. Adherence was reinforced through monthly visits, weekly calls, and sachet counts. Both groups received folic acid, preventive penicillin, and occasional transfusions or analgesics.

### ***Outcome Measures***

The primary endpoint was the rate of VOCs during 6 months of treatment. A VOC was defined as an acute sickle-related pain episode requiring medical evaluation and parenteral analgesia; ACS, sequestration, stroke, or priapism were also counted as VOC-equivalent events [14]. ACS was characterized by a new pulmonary infiltrate, fever, and/or respiratory symptoms [15].

Secondary endpoints included the proportion of patients with  $\geq 1$  VOC, time to first VOC, number of hospitalizations for VOC/ACS, emergency department (ED) visits for sickle pain, and changes in hemoglobin, hematocrit, reticulocyte count, and HbF from baseline to 6 months. Increases in HbF and mean corpuscular volume (MCV), along with decreases in reticulocytes and absolute neutrophil count (ANC), indicated the HU effect and

adherence. Safety endpoints included the incidence of adverse events, serious adverse events, and deaths, with investigator-attributed causality.

### **Data Collection and Monitoring**

Baseline evaluation included medical history/physical examination and laboratory testing (CBC with differential, reticulocyte count, hemoglobin electrophoresis for %HbA/%HbF/%HbS, liver/renal function, lactate dehydrogenase (LDH), and infection screening; pregnancy testing when indicated), followed by monthly visits for 6 months to document interval events, perform targeted examinations, assess adherence (interview and pill/sachet counts), and repeat CBC/reticulocytes/chemistry, with electrophoresis repeated at study end. All assays were performed in the hospital central laboratory under routine Quality control (HPLC Bio-Rad VARIANT II for hemoglobin fractions; Sysmex XN-series fluorescence flow cytometry for reticulocytes; Roche Cobas c501 for LDH/biochemistry), with inter-assay coefficients of variation <3% for HbF and <5% for reticulocytes/chemistry and personnel blinded to allocation. Suspected VOC/ACS events were independently adjudicated by two blinded pediatric hematologist–oncologists using predefined criteria and de-identified clinical/imaging data (third adjudicator prespecified but not required). Discrepancies were resolved by consensus, with a third hematologist available if needed, though not used. Chest X-rays were interpreted by radiologists blinded to treatment status, while the DSMC reviewed interim safety and endpoint data for any patterns.

### **Adherence Assessment**

Adherence to study medications was monitored via pill counts and self-reports, with rates exceeding 80% in both treatment arms, indicating good compliance. Pharmacokinetic or biomarker-based assessments were not conducted. Safety was overseen by an independent Data and Safety Monitoring Committee, with no concerns raised after an interim analysis of 25 patients.

### **Statistical Analysis**

Analyses followed intention-to-treat. Continuous variables are reported as mean±SD or median (IQR) and categorical variables as n (%). Normality was assessed using the Shapiro–Wilk test and Q–Q plots. Between-group comparisons used t-test or Mann–Whitney U for continuous data and chi-square or Fisher’s exact tests for categorical data; within-group changes used paired t-tests or Wilcoxon signed-rank tests. In contrast, comparisons between groups were conducted with two-sample t-tests or Mann–Whitney tests, as applicable. ANCOVA models were used to enhance precision and adjust for baseline values in key outcomes, including Hb, HbF%, reticulocytes, and VOC counts, with adjustments for the baseline annualized VOC rate. The time-to-first-VOC was analyzed using Kaplan–Meier curves, with log-rank testing and a Cox model to estimate hazard ratios; proportional hazards were assessed using Schoenfeld residuals. A two-sided p-value of less than 0.05 was deemed significant; no multiplicity adjustment was implemented due to the exploratory design. Missing data constituted less than 5% and were addressed through complete-case analysis, following confirmation of missing-at-random. All analyses were conducted using SPSS version 26, with figures produced in GraphPad Prism 9 and SPSS.

## **Results**

### **Baseline Characteristics**

Between June 2019 and January 2021, 60 patients were screened for eligibility. Seven were excluded (5 did not meet the inclusion criteria, and two declined to participate). Fifty-three patients were randomized to HU + L-glutamine (n = 27) or HU + placebo (n = 26). All randomized participants completed the 6-month outcome assessments and were included in the final analysis (Fig. 1). The CONSORT flow diagram in Figure 1 shows that the main reasons for participant exclusion were as follows: in the L-glutamine + HU group, 3 participants were lost to follow-up, 2 withdrew consent, and 1 had a protocol violation; in the Placebo + HU group, 2 withdrew consent, and 1 had a protocol violation. Despite these exclusions, all randomized participants were included in the intention-to-treat analysis.

The mean age of participants was  $13.5 \pm 7.4$  years, and nearly half (49%) were younger than 12 years. The genotype distribution was balanced between groups (HbSS 45%, HbS/β<sup>0</sup>-thalassemia 55%), with no HbSC or other variants. During the trial, all patients received a nearly constant dose of HU (20 mg/kg/day), with an average dose of 19.8 mg/kg (SD 2.5). Both groups had evidence of mild-to-moderate baseline anemia and hemolysis, typical for SCD. Clinical and laboratory characteristics are detailed in Table 1. The baseline characteristics were essentially the same for all treatment groups. For example, hemoglobin levels were about 9.5 g/dL, fetal hemoglobin levels were about 15% (a little higher in the L-glutamine group), reticulocyte counts were around  $260 \times 10^3/\mu\text{L}$ , and lactate dehydrogenase levels were around 580 U/L. The dosages of HU and folate were also similar. The HU + L-glutamine group showed a slightly higher pre-enrollment VOC burden (mean 2.6 vs.

2.0/year; median 2 for both;  $p=0.10$ ). Baseline ACS history and organ-damage markers were similar (ACS uncommon; median 0), which means that randomization worked even if there were some minor differences.

### **Primary Outcome: Pain Crisis Frequency**

The 6-month trial indicated that patients receiving HU + L-glutamine had significantly fewer VOCs than those receiving only HU ( $1.00 \pm 0.73$  vs.  $1.65 \pm 0.80$  crises per patient; 95% CI: 0.23–1.07;  $p=0.003$ ), reflecting a 40% reduction. The HU + L-glutamine group demonstrated a decrease of 0.65 crises per patient relative to the HU-only group (95% CI: 0.23 to 1.07 fewer). This corresponds to a similar 40% relative reduction from a baseline of 1.65 crises per patient (Fig. 2). More patients were crisis-free with L-glutamine (7/27, 26%) than with control (2/26, 8%). Both groups improved from baseline, but the reduction was greater with L-glutamine ( $p<0.001$  vs  $p=0.08$ ). ANCOVA adjusted for baseline VOC rate showed a significant benefit for HU + L-glutamine ( $p<0.001$ ). Crisis-free survival was more prolonged (120 vs 80 days), and time-to-first VOC favoured combination therapy ( $HR\approx 0.58$ ), but these time-to-event differences were not statistically significant ( $p\approx 0.10$ – $0.11$ ), likely due to limited power; overall, VOC reduction and more crisis-free patients support an additive effect.

### **Secondary Outcome: Acute Chest Syndrome (ACS)**

ACS was less frequent with HU + L-glutamine (5/27 [19%] vs 14/26 [54%];  $RR\approx 0.35$ ;  $p=0.055$ ), with fewer episodes per patient (0.19 vs 0.77;  $p=0.006$ ). Although both incidence proportions and event rates indicate fewer ACS episodes in the L-glutamine + HU group, the small number of events means these findings should be interpreted with caution and are considered hypothesis-generating due to the limited statistical power of this exploratory study.

### **Hospitalizations and Hematologic Parameters**

The HU + L-glutamine group had fewer sickle cell-related hospitalizations than controls (44% vs. 69%, mean 0.46 vs. 1.0 admissions,  $p = 0.04$ ), with shorter total hospital days (55 vs. 88). Combination therapy led to a modest but significant hemoglobin increase ( $+0.46$  g/dL; 95% CI:  $+0.05$ – $0.87$ ;  $p=0.028$ ;  $+0.78$  vs.  $+0.32$  g/dL), with a higher proportion of patients achieving  $Hb \geq 10$  g/dL (59% vs. 42%). Reticulocyte counts declined more markedly ( $-19$  vs.  $-8 \times 10^3/\mu L$ ,  $p = 0.04$ ), accompanied by lower LDH trends, indicating reduced hemolysis (Fig. 4A–B). HbF rose significantly with HU + L-glutamine ( $+6.2\%$  vs.  $+1.6\%$ ,  $p < 0.001$ ), reaching 22.8% vs. 15.4% at 6 months, with an absolute increase of  $+4.6\%$  (95% CI:  $+2.0$ – $7.2\%$ ) indicating a statistically robust effect (Fig. 5). Although L-glutamine does not directly induce HbF, its combination with HU likely enhanced erythropoiesis and redox stability. HU adherence exceeded 80% in both groups, minimizing compliance bias. Non-admission ED visits were lower with HU + L-glutamine (17 vs 33), consistent with fewer VOCs and reduced acute healthcare utilization.

### **Safety and Adherence**

Adherence to HU and L-glutamine exceeded 80%, confirmed by pill counts and follow-up, with stable MCV/ANC and expected HU-related changes (increased HbF, reduced reticulocytes) supporting comparable HU exposure and objective adherence. L-glutamine was well tolerated, with no serious adverse events or discontinuations; mild, transient gastrointestinal symptoms occurred in  $\sim 15\%$ , and laboratory parameters remained stable, supporting its safety and feasibility as an adjunct to HU.

## **Discussion**

### **Principal Findings and Interpretation**

In the GLOBE study, adding oral L-glutamine to HU improved pediatric/adolescent SCA outcomes over 6 months, reducing VOCs, ACS, and hospitalizations and modestly improving hemoglobin and hemolysis markers (reticulocytes, LDH). Although L-glutamine does not directly induce HbF production, the greater HbF rise observed with combination therapy likely reflects indirect effects (e.g., improved redox environment and erythropoiesis). The observed increase in HbF in the HU + L-glutamine group is intriguing but unexpected, as L-glutamine is not known to directly induce HbF production. Although baseline adjustments were made to account for potential confounders, alternative explanations may also account for these findings. For instance, improved adherence to HU therapy due to enhanced patient support or a potential regression to the mean effect could have contributed to the HbF increase. Further studies are needed to clarify the biological mechanisms underlying this effect [16,17].

Overall, L-glutamine may potentiate HU's benefits by mitigating oxidative stress and improving HU responsiveness and adherence, thereby sustaining red cell integrity and clinical stability.

Our results corroborate and extend Niihara et al.'s Phase 3 trial [8], which showed reduced VOCs with L-glutamine. Whereas Niihara et al. [8] reported a 25% lower median annual pain-crisis rate, we observed a 40% relative

reduction in VOC over 6 months in HU-treated patients, supporting an additive benefit. The primary endpoint remains robust: baseline-adjusted analyses continue to show significantly lower VOC rates with HU + L-glutamine, despite baseline imbalance. Unlike Niihara et al. [8], we observed improvements in hemoglobin and HbF, potentially reflecting our younger, uniformly HU-treated cohort and intensive adherence monitoring. These findings are consistent with Phase 2 data from Ebeid et al. [18], which showed fewer acute SCD complications with L-glutamine.

Our trial reaffirmed the safety of the combination, showing no increase in adverse events or laboratory toxicity with L-glutamine, consistent with recent studies [18,19]. Gastrointestinal symptoms were mild with no discontinuations, and comparable neutrophil/cytopenia profiles suggest L-glutamine does not exacerbate HU myelosuppression. High adherence likely reflected good tolerability and easy administration, supporting the practicality of combination therapy; adherence is reinforced by education, monitoring, access, family support, and strong provider relationships [9,20,21].

### ***Mechanistic Implications***

The findings indicate an oxidative-stress mechanism: L-glutamine, as a NAD precursor, enhances redox defences (e.g., glutathione), reduces hemolysis and endothelial adhesion, and may improve vascular function and nitric oxide bioavailability. This is consistent with improved hemoglobin and reticulocyte levels, as well as fewer crises, thereby reinforcing the need for multimodal therapy beyond HU-driven hemoglobin F induction [12,16,17,18,19,22,23].

### ***Study Limitations***

Due to feasibility constraints, we targeted 60 participants and randomized 53 (~70% post hoc power). This single-center study with 6-month follow-up should therefore be viewed as a pragmatic, exploratory trial with wider confidence intervals and limited power for some secondary endpoints (e.g., ACS). Nonetheless, the primary outcome remained statistically significant, and prespecified baseline-adjusted analyses, together with consistent, clinically meaningful improvements in VOCs, hospitalizations, and hematologic indices, support the robustness and clinical relevance of the findings and warrant confirmation in larger multicenter studies.

### **Conclusions**

This randomized trial shows that adding L-glutamine to HU significantly reduces VOCs and ACS, while improving hemoglobin and hematologic indices without added toxicity. The combination's efficacy likely reflects synergistic effects on redox balance and sickle cell pathophysiology, with excellent adherence and tolerability.

### **Conflict of Interest and Funding Statement**

Authors declare no financial or commercial conflicts. None has received honoraria, consultancy fees, or personal compensation from entities interested in the submitted work. Bushehr University of Medical Sciences (Grant No. 1755) sponsored the trial and covered the costs of pharmaceutical purchases; there was no funding involvement in study design, data collection, analysis, interpretation, or paper writing.

### **Ethics Approval and Consent to Participate**

This study followed the Helsinki Declaration. The Bushehr University of Medical Sciences Ethics Committee approved the study (IR.BPUMS.REC.1400.107) and registered it at IRCT (IRCT20210715051904N1, irct.ir). It followed CONSORT criteria. Written informed consent was obtained from parents/guardians, with assent from children  $\geq 7$  years. Before review and publishing, study participant photos were de-identified. Participants (where applicable) and their parents or legal guardians provided consent for the use of images.

### **Data Availability**

De-identified participant data and the analysis code used to generate figures and tables will be made available upon reasonable request to the corresponding author (nshakibazad@gmail.com), subject to institutional and ethical approvals and data-use agreements. Data will be shared for non-commercial academic purposes within 30 days of a qualified request.

### **Author Contributions**

Medical Practices: N.S., M.MZ., M.R.; Concept: N.S., M.R.; Design: N.S., M.R.; Data Collection or Processing: M.MZ., N.S., M.M., B.A., M.R.; Analysis or Interpretation: M.M., B.A.; Literature Search: M.M., N.S.; Writing: N.S., M.M., B.A., M.MZ., M.R.

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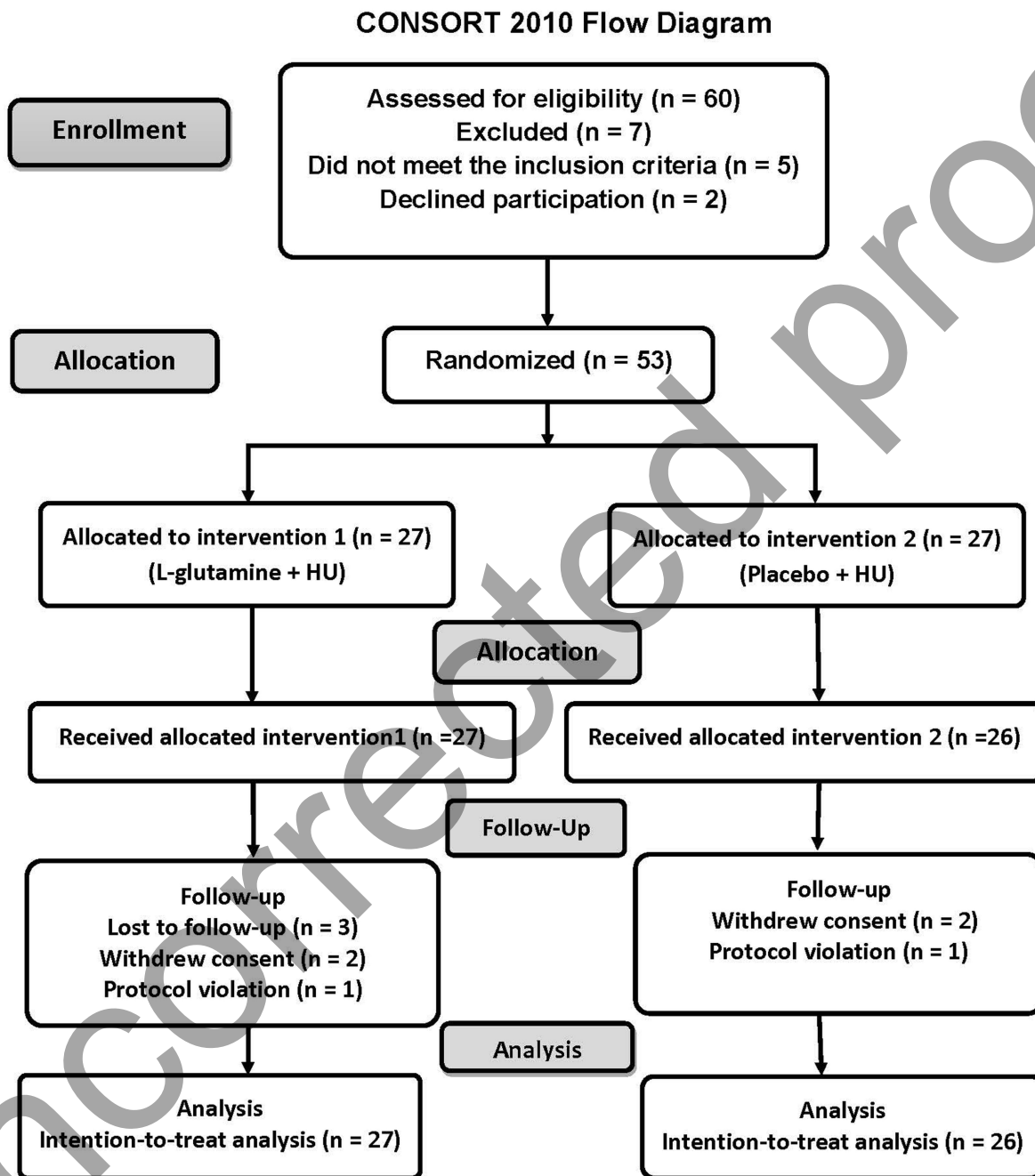
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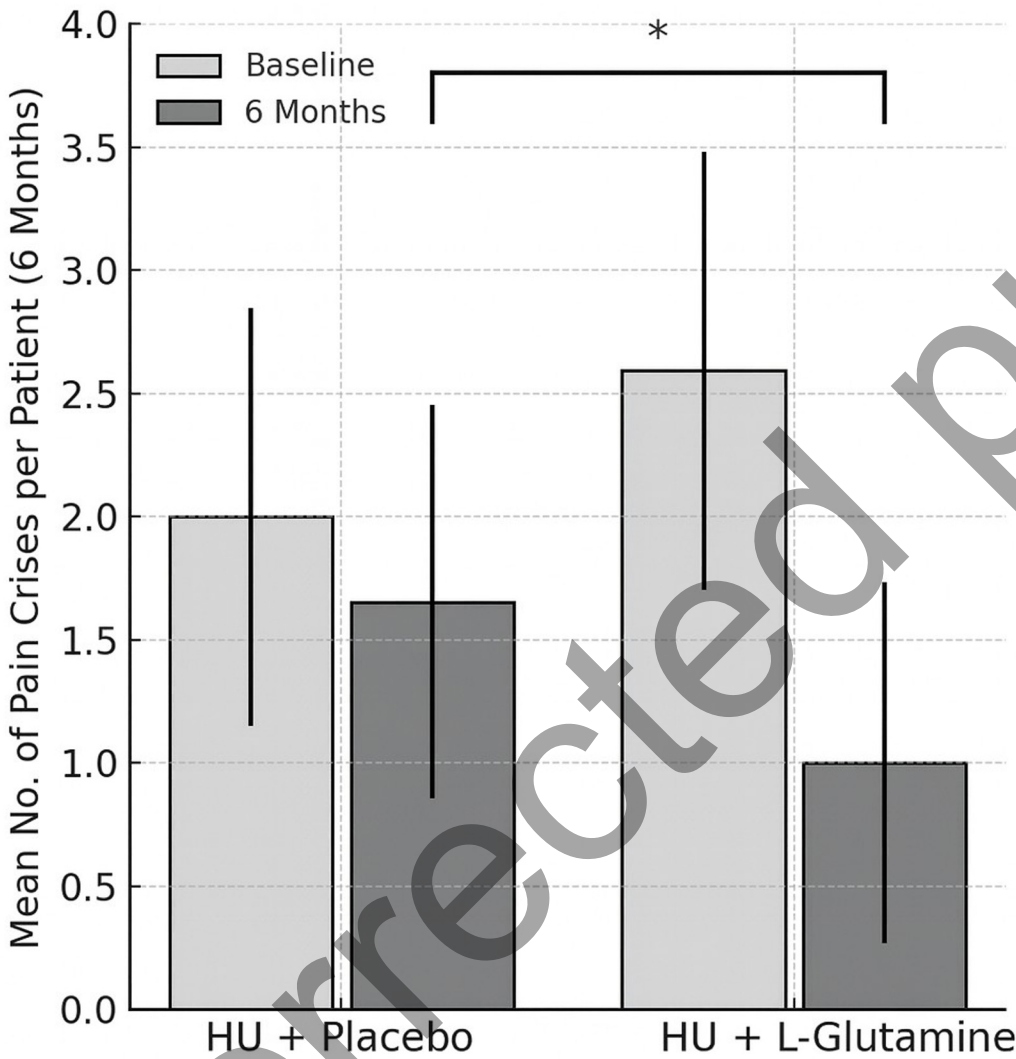
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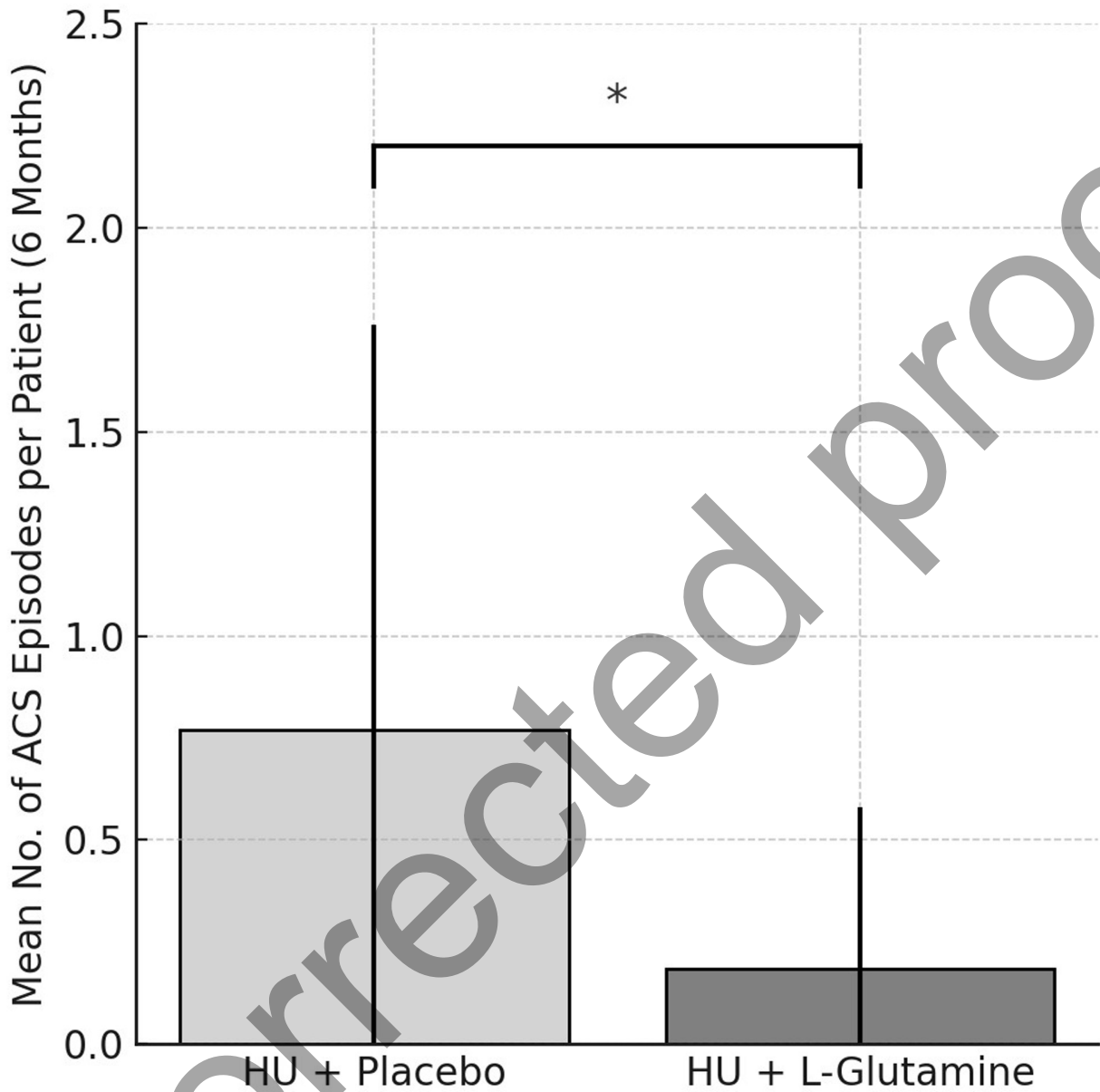


**Figure 1.** CONSORT flow diagram of the trial. A total of 60 patients were screened for eligibility. Seven were excluded (5 did not meet the inclusion criteria, 2 declined participation), leaving 53 participants who were randomized 1:1. In the L-glutamine + HU group, 6 participants did not complete follow-up: 3 were lost to follow-

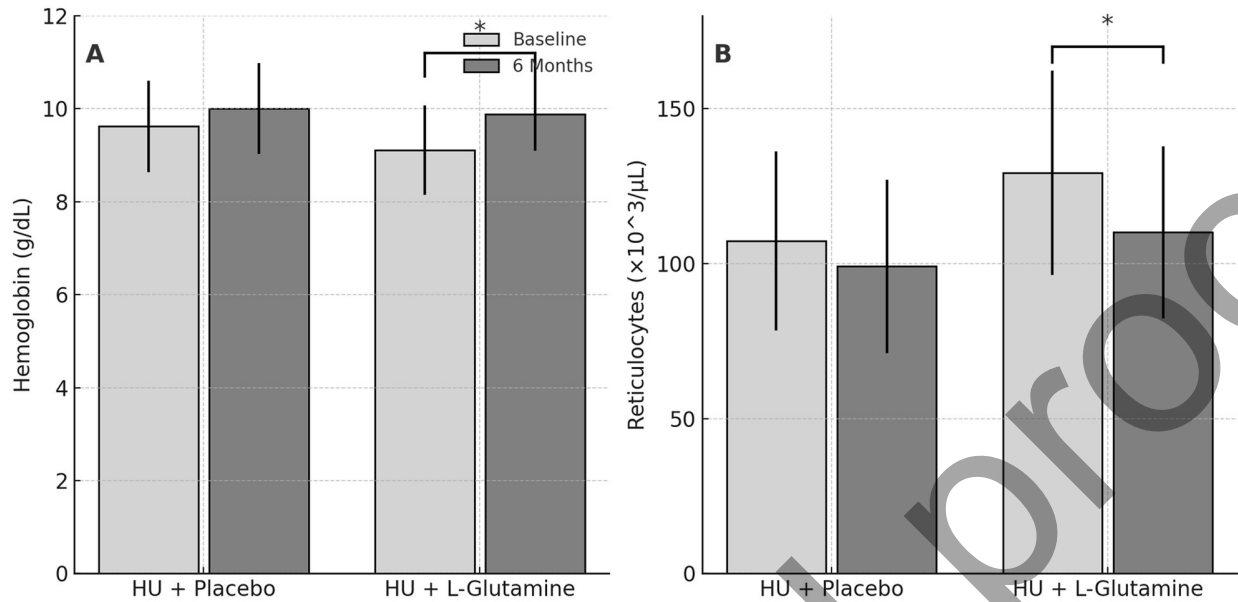
up, 2 withdrew consent, and 1 had a protocol violation. In the Placebo + HU group, 3 participants did not complete follow-up: 2 withdrew consent, and 1 had a protocol violation. Despite these exclusions, all randomized participants were included in the final intention-to-treat analysis. (ITT= intention-to-treat, HU= Hydroxyurea).



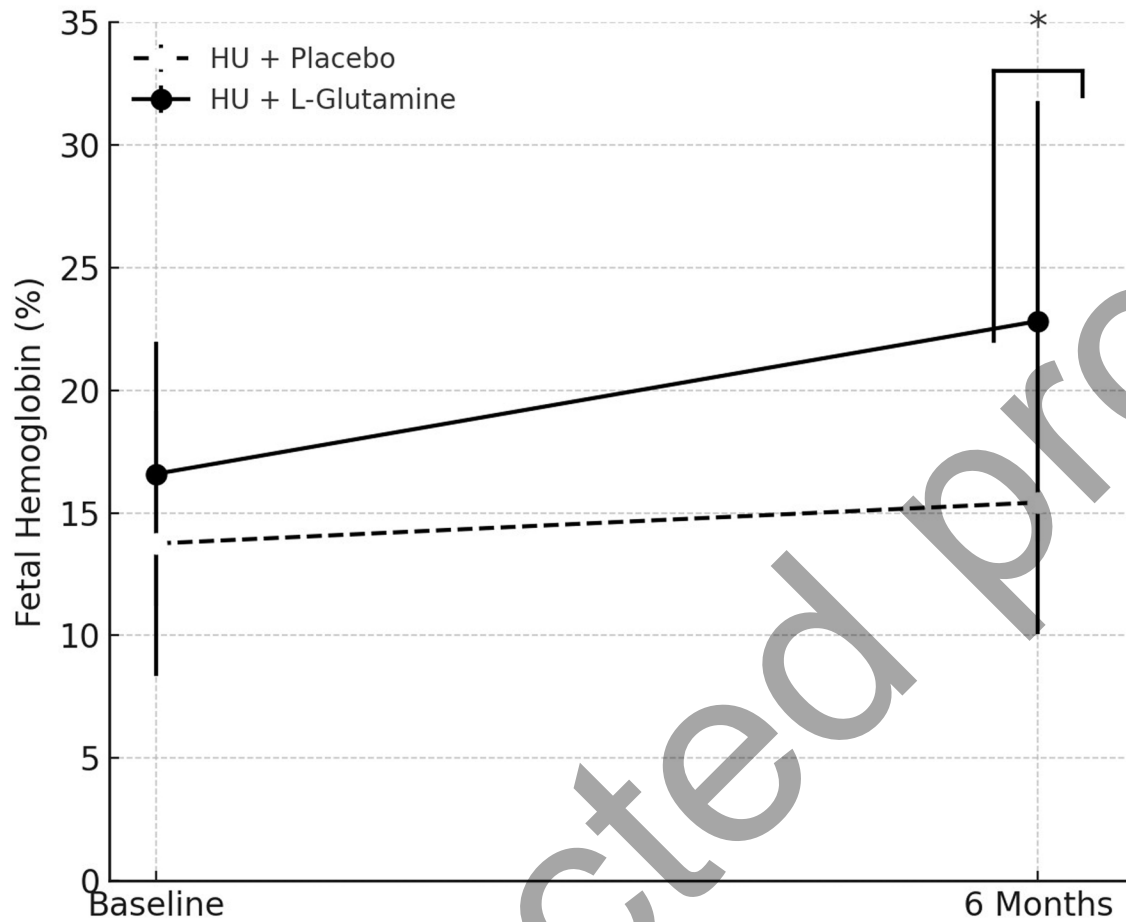
**Figure 2.** Mean number of vaso-occlusive crises per patient at baseline and after 6 months of treatment in each arm. The HU + L-glutamine group's VOC rate declined from 2.6 (baseline) to 1.0 at 6 months, versus a change from 2.0 to 1.65 in the HU + placebo group ( $p = 0.003$  between groups).



**Figure 3.** Mean number of acute chest syndrome (ACS) events per patient over 6 months in each group. HU + L-glutamine arm averaged 0.19 ACS episodes vs 0.77 in HU + placebo ( $p=0.006$ ). We note that the incidence proportions were 19% vs 54%. The ACS analysis had limited statistical power due to the small number of events, which should be considered when interpreting the results.



**Figure 4A-B. (Hemoglobin and Reticulocytes).** (4A) Hemoglobin: mean hemoglobin levels at baseline and 6 months for both groups. The HU + L-glutamine group showed a significant increase in Hb (from ~9.4 to 10.2 g/dL on average), whereas the HU-only group showed a more minor increase (from ~9.5 to 9.8 g/dL); error bars indicate SD. (4B) Reticulocyte count: mean absolute reticulocyte counts (ARC) at baseline and 6 months. The HU + L-glutamine group had a larger decrease in reticulocyte count, indicating reduced hemolysis, compared to minimal change in the HU-only group; error bars indicate SD.



**Figure 5. Fetal Hemoglobin (HbF).** HbF levels at baseline and after 6 months in both groups. The HU + L-glutamine group increased from 16.6%  $\pm$  SD to 22.8% ( $p < 0.001$ ), whereas the HU + placebo group went from 13.8% to 15.4% (no significant change). Error bars show SD.

**Table 1. Baseline Characteristics of the Study Population**

Variable	HU + Placebo (n = 26)	HU + L-glutamine (n = 27)	p-value
Demographics	Demographics	Demographics	Demographics
Age, years (mean $\pm$ SD)	13.6 $\pm$ 7.2	13.5 $\pm$ 7.4	0.84
Male sex, n (%)	12 (46%)	13 (48%)	0.88
Female sex, n (%)	14 (54%)	14 (52%)	0.88
HbSS genotype, n (%)	12 (46%)	12 (44%)	0.89
HbS/ $\beta^0$ -thalassemia genotype, n (%)	14 (54%)	15 (56%)	0.90
Hemoglobin, g/dL (mean $\pm$ SD)	9.62 $\pm$ 0.98	9.11 $\pm$ 0.97	0.06
HbF, % (mean $\pm$ SD)	13.75 $\pm$ 5.41	16.59 $\pm$ 5.38	0.06
Absolute reticulocyte count (ARC), $\times 10^3/\mu\text{L}$ (mean $\pm$ SD)	107.3 $\pm$ 28.9	129.3 $\pm$ 32.9	0.012*
MCV, fL (mean $\pm$ SD)	95.1 $\pm$ 3.5	95.0 $\pm$ 3.2	0.81
ANC, $\times 10^9/\text{L}$ (mean $\pm$ SD)	5.48 $\pm$ 1.30	5.20 $\pm$ 1.25	0.67

VOCs in past 6 months, median [IQR]	2 [1–3]	3 [2–4]	0.020*
ACS history, n (%)	13 (50%)	14 (52%)	0.49
HU dose (mg/kg/day, mean $\pm$ SD)	19.8 $\pm$ 2.5	19.8 $\pm$ 2.5	—

Values are presented as mean  $\pm$  SD, median [IQR], or n (%). P-values compare groups at baseline using Student's t-test or Mann–Whitney U test for continuous variables and  $\chi^2$  or Fisher's exact test for categorical variables, as appropriate. \*p < 0.05.

Abbreviations: HU, hydroxyurea; HbF, fetal hemoglobin; ARC, absolute reticulocyte count; MCV, mean corpuscular volume; ANC, absolute neutrophil count; VOC, vaso-occlusive crisis; ACS, acute chest syndrome.