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# BMJ Open

## Multicentric, double-blind, randomised controlled trial of hyperbaric-oxygen therapy (HBOT) versus sham for treating Vaso-Occlusive Crisis (VOC) in sickle cell disease (SCD) after 8 years old (HBOT-SCD).

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**Multicentric, double-blind, randomised controlled trial of hyperbaric-oxygen therapy (HBOT) versus sham for treating Vaso-Occlusive Crisis (VOC) in sickle cell disease (SCD) after 8 years old (HBOT-SCD).**

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## ABSTRACT

**Introduction:** Sickle cell disease (SCD) is one of the most common genetic diseases in the world. Vaso-occlusive crisis (VOC) is the most frequent complication of SCD, leading to bone pain or thoracic pain. Hyperbaric oxygen therapy (HBOT) is a safe and well-established method of increasing tissue oxygen delivery immediately by up to 10–20 fold. In context of VOC, HBOT has the potential to limit sickling. A previous pilot study of 9 patients showed the safety and potential benefits of HBOT on VOC induced pain. Our study aims to assess the clinical safety and effectiveness of HBOT for treating VOC, its biological mechanisms of actions as well as its cost-effectiveness.

**Methods and analysis:** This is a double-blinded, randomised, multicentric, controlled trial. Patients diagnosed with a major form of SCD, aged 8 years or older, presenting at one of the participating centres' Emergency Departments (EDs) with a VOC requiring level 3 analgesia, without exclusion criteria will be eligible. All patients will receive the usual care for VOCs. After informed, patients will be randomised into the HBOT intervention group (2.0 Atmosphere Absolute [ATA], 95 min, FIO<sub>2</sub> = 1) or the sham group (1.3 ATA, 95 min, FIO<sub>2</sub> = 0.21), with 1 to 3 sessions. Difference in the VAS pain score before and after HBOT and other outcomes will be compared between the intervention and sham groups. Our primary outcome will be pain at 4 hours after first HBOT session and our secondary outcomes will be morphine usage, length of stay, biological parameters and cost.

**Expected results and their impact:** HBOT is expected to be safe and reduce: pain experienced, duration of the crisis, number of transfusions required, the number of morphine doses, length of stay and frequency of ACSs and VOCs.

**Ethics and dissemination:** Ethical approval CER Geneva 2019-01707

**Trial registration number** NCT04978116

**KEYWORDS :** Hyperbaric Oxygen Therapy, Sickle-cell disease, vaso-occlusive crisis

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STRENGTHS AND LIMITATIONS OF THIS STUDY

- Innovative study in a very fragile population.
- RCT with Hyperbaric Oxygen Therapy and validated sham control
- Accessibility of hyperbaric chambers in countries with high Sickle-cell disease prevalence would be straightforward, and could be a good alternative to current Vaso-occlusvie crisis treatments.
- Primary outcome (pain) is influenced by treatment used (morphine and its derivatives)

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## INTRODUCTION

Sickle cell disease (SCD) is the most frequent autosomal recessive disease, resulting in the production of sickle haemoglobin (HbS). Reducing life expectancy in sufferers, its major forms of SCD regroup SS, SC, S $\beta_0$  and S $\beta_+$  - thalassaemias. Vaso-occlusive crisis (VOC) is the most common complication of SCD with an annual incidence of approximately 60% in adult sufferers. VOC usually causes bone, chest and/or abdominal pain and is the leading cause of death in SCD patients (1). Acute Chest Syndrome (ACS) is a specific clinical presentation of VOC defined by a triad of symptoms (chest pain, fever and radiological infiltration) and caused by pulmonary capillary occlusion. It remains of real concern in adults, being a major cause of hospitalisation and death.

### Physiopathology of Sickle-cell disease

Hypoxia, acidosis, infection, dehydration or exposure to cold can trigger VOC/ACS by facilitating falciformation, due to a HbS polymerisation, leading to acute vaso-occlusion, haemolysis (2), inflammation, and ultimately, to vascular damage and tissue hypoxia with multi-organ injury (3, 4). Other cellular mechanisms involved in CVO/ACS are: adherence of sickle cells to the vascular endothelium, involving intercellular adhesion molecules such as ICAM-1, VCAM-1, PAF and CD 36 (5-8); erythrocyte-leukocyte-platelet endothelium interactions (9) with leukocytes (10) and platelets activation (11); an increase in the number of circulating leukocytes (12) and formation of reactive oxygen forms (Reactive Oxygen Species -ROS-) which cause a major endothelial dysfunction (13). Intravascular haemolysis and cell adhesion promote both vasculopathy and vaso-occlusion (14-16), as well as promoting inflammation (17). Mitochondrial dysfunction is an additional element of this perpetual cycle, promoting platelet activation, and the release of inflammation activators (18).

### Usual Care

The management of VOC is based on effective analgesic treatment (often morphine/morphine derivatives), abundant hydration, local warming, oxygen therapy (in case of pulmonary manifestations in particular chest pain and/or hypoxaemia SpO<sub>2</sub><94%), systematic folate administration, antibiotic therapy in the case of infection, and, when the crisis is not controlled by these measures, erythrapheresis/transfusions can be used. Additional indications for transfusions/erythrapheresis are severe ACS (19), ischaemic stroke, recurrent severe infection and side effects that limit the use of morphine. Unfortunately, when using blood products, alloimmunisation remains an extremely frequent consequence which excludes the possibility of further transfusions or exchange transfusions as a therapeutic option in certain patients.

### Hyperbaric Oxygen Therapy

Hyperbaric oxygen therapy (HBOT) is a long-established technique with several recognised indications, such as carbon monoxide poisoning, or anaerobic soft tissue infection. HBOT consists of intermittent periods of breathing 100% oxygen at a pressure above that of sea level. The increase in pressure produced in the compression chamber significantly increases the partial pressure of the inspired gases in the blood and peripheral tissues (20), reducing

ischaemia, and as a consequence reducing / reversing sickling. HBOT allows the enhanced oxygenation of tissues whose blood circulation is deficient, in that oxygen no longer needs the support of erythrocytes to circulate and to be diffused.

**Rationale for the study**

In the early stages of VOC, the falciformation process is reversible if conditions, especially oxygenation, improve. HBOT is effective in increasing tissue oxygenation by increasing dissolved O2 in the blood and limiting local hypoxaemia, a promoter of falciformation. This is often sufficient to break the vicious cycle of VOC and ACS. HBOT demonstrated an improvement in microcirculatory vascular flow, a decrease in leukocyte adhesion and an increase in tissue ATP in a rodent liver model (21). HBOT may influence the NO synthetase and NO secretion, particularly in the central nervous system, leading to increase regional brain blood flow by vasodilation (22). It also down-regulates cell adhesion proteins such as ICAM-1, VCAM-1, PAF and CD36 (16, 23-25). Thus, HBOT could counterbalance the effects of VOC which induces an overexpression of adhesion phenomena and leads to NO deficiency (26). Several cases of complications related to SCD treated with HBOT have already been documented since 1966 in clinical cases or retrospective series with favourable issue reported for number of patients (26-35). In hyperbaric medicine guidelines, SCD is a recognised indication of HBOT, with a low level of evidence (type C recommendations) (36). However, the North American guidelines did not include SCD as an indication (37). Our group published a pilot non-comparative study on 9 SCD patients, hospitalised for severe and resistant VOC, treated with HBOT (2.5 ATA, 90min, FIO2=1). We observed a significant decrease in mean Visual Analogic Scale (VAS) score after one HBOT session compared to the VAS evaluated before the session. For 2 patients, HBOT session was interrupted because of minor ear barotrauma (otalgia) during compression, but no problems relating to tolerance were reported in the 7 other patients (38).

This study aims to investigate the effect of HBOT in SCD-VOC in terms of efficacy on clinical outcomes (such as pain level, analgesic consumption, duration of hospitalisation) and its safety.

**METHODS AND ANALYSIS**

**Setting**

The study will be conducted in three or more academic hospitals in Switzerland and France: Geneva University Hospital (Switzerland), and other university hospitals in France. Recruitment is already in progress on the Geneva site.

**Study Design**

This is a superiority, double blinded, multicentric randomised controlled (RCT), comparing HBOT (intervention) superiority to sham in the treatment of VOC in SCD (Fig. 1).

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All patients with VOC presenting at the ED will receive the current standard treatment according to the adult or paediatric protocols: hydration, normobaric oxygen therapy to maintain  $SpO_2 \geq 94\%$ , analgesics (preferably intravenous morphine or derivatives via PCA pump [Patient Controlled Analgesia]), and antibiotic therapy (when needed). The first VAS will be evaluated by the patient on arrival and if all the inclusion criteria are met without exclusion criteria, a hyperbaric physician will verify that there are no contraindications to HBOT. The screening will ideally take place in the ED aiming at patient inclusion in the study within the first 4 hours of arrival where possible, (or up to 24 hours or possibly more in specific cases).

After informed consent, the patient will be transferred to the Hyperbaric Medicine Department (HD). There, the randomization between the intervention arm (2.0 ATA, 95 min session, FIO<sub>2</sub> 100%) and the sham arm (1.3 ATA, 95 min session, FIO<sub>2</sub> 21%) will be carried out. A HBOT session can be interrupted at any time in case of complications, and this would be documented. All patients will receive established standard of care, including PCA (if already in progress) during the HBOT sessions as per protocol. Solely the HD team will know in which study arm the patient has been randomised. Both the patient and the internal medicine clinician in charge will be blinded to group allocation.

For Geneva centre only: If adult participants specifically consent to the additional Biobank study, venous blood samples will be taken immediately before and after the first HBOT session. A third blood sample will be taken at 24 hrs post start time of the first HBOT session or up to 36 hours if H24 falls in the middle of the night. If the patient is discharged from hospital before H24, the third blood sample will be taken just before discharge.

A second VAS score will be evaluated four hours after the end of the initial HBOT session by non-HD staff in the ED/ward area. As HD staff will be aware of the allocated arm, they will not assess any of the patient outcomes. A second (and third) HBOT session will systematically be scheduled within 24 (up to 36) hours after the first session. Patients initially enrolled in the sham arm may receive a second (+/- 3rd) sham session and patients enrolled in the intervention arm will again receive HBOT. The second/third session will be cancelled in both arms of the study if a patient's condition deteriorates meaning that they need  $> 2l/min O_2$  to maintain their  $SpO_2 > 92\%$ . Although the protocol will be interrupted for these patients, they will be integrated into the analysis in "intention to treat". The VAS score evaluation will be undertaken immediately before and after the second and third sessions and again at four hours after the sessions. This second (and third) session will not be undertaken if the patient's VAS score  $\leq 2$  without morphine treatment. If necessary, transfusion therapy may be given. If a transfusion or exchange transfusion is given after the first (or the second) HBOT/sham session, the second (or respective third) session will be cancelled. The VAS scores considered for analysis will be those collected before transfusion/exchange. In certain strictly defined circumstances (only for patients needed more than 2 l/min of oxygen), VOC's will be eligible for HBOT, without randomization in this protocol.

### Sample size

The sample size calculation is based on our primary outcome, i.e., the identification of an effect of the intervention versus sham on the difference in VAS score between H0 and H6. The minimum difference in VAS score between H0 and H6 considered as clinically relevant is 1.3 points (on a scale of 0 to 10), particularly for the treatment of



VOC in ER (39, 40). In the sham group, the decreased VAS score between H0 and H6 is expected to be less marked than in the intervention group (difference more than 1.3). The expected standard deviation (SD) of the VAS score difference between H0 and H6 is based on a preliminary study of 9 patients. In this study, the standard deviation of the VAS score after HBOT was 2 and the SD of the difference between H0 and H6 was 1, which corresponds to a strong correlation (0.88) between the VAS score at H0 and H6. Assuming, conservatively, that the correlation between H0 and H6 is 0.50, the standard deviation of the VAS score difference between H0 and H6 is 2 in the HBOT group. Assuming that the standard deviation of the difference is the same in the sham group and a bilateral risk alpha of 0.05, the sample size to obtain a 90% power is 50 patients per group (100 patients in total).

**Study phase**

This is a phase 3 study. An RCT remains the only design adapted to answer the question of effectiveness of HBOT on VOC. This study follows the pilot study published by our group in 2012. As SCD is a rare disease, the considered sample size is sufficient to meet the phase 3 criteria.

**Patient population**

Any patient, 8 years of age or over with a VOC, with or without ACS, admitted to the Emergency Department (ED) and requiring level 3 analgesia (according to WHO definition) may be screened and included in this study unless they have exclusion criteria.

## Inclusion and exclusion criteria

Inclusion and exclusion criteria are listed in detail in box 1.

### BOX 1 : ELIGIBILITY CRITERIA

#### Inclusion criteria:

- Patients aged 8 or over
- Diagnosed with a major SCD disorder (SS, SC, Hb O Arab, S $\beta_0$  and S $\beta_+$ -thalassemias);
- A presentation of a VOC (with or without ACS) unresponsive to level 2 analgesics (WHO classification) which fulfils the criteria necessary for consultation at an ED;
- The ability to carry out the Valsalva maneuver;
- The ability to give informed consent and sign a written informed consent form (consent and signature of legal guardian 8 authorized).

#### Exclusion criteria:

- Pregnancy;
- Indication for mechanical ventilation (non-invasive ventilation/oro-tracheal intubation) ;
- Contraindication for HBOT established by a physician responsible for hyperbaric medicine;
- An anomaly in the results of prior transcranial Doppler (TCD) ultrasound (> 200 cm/sec) or a previous history of stroke (but TCD will not be performed for the study);
- Patients requiring more than 2 l/min of normobaric oxygen in order to maintain an SpO<sub>2</sub>  $\geq$  92% (because not safe to include these patients in placebo group).

## Recruitment

To optimise recruitment into this protocol, this study has been discussed with and presented to the Swiss Sickle Cell Association. Patient awareness of this protocol will be raised by the haematologist in charge of the out-patient follow-up of sickle cell patients. The protocol includes some ED team as investigators to optimise inclusion directly from the ER. Treatment by HBOT outside of the protocol for VOC will not be possible unless contraindications exist for inclusion in the sham arm.

## Randomisation

Randomisation will be carried out by the HBOT team. It will be stratified by site but also by the hourly dose of morphine received from arrival in the ED until the time of randomisation (high versus low, defined by the average hourly dose since admission converted into Morphine IV equivalent,  $\geq$  5mg/h IV or <5mg/h IV respectively in adult patients). Randomisation will be carried out between the intervention strategy (HBOT session at 2 ATA; 95

min; FIO2=1) and the sham (HBOT session at 1.3 ATA; 95 min; FIO2=0.21). Patients will be treated using the same strategy for the other 2 potential sessions.

**Intervention and sham**

Intervention Group

The pressure increase is achieved by introducing compressed air into the hyperbaric chamber. Changes in ambient pressure will also modify the volume that gas occupies, including in the body (eg. ear cavities, sinuses). In the hyperbaric chamber patients, in either a sitting or lying position, will breathe through an oxygen mask and can be offered the same care and monitoring, where necessary, as in a ward setting. A member of the HD team will monitor the patient throughout the entirety of each session. In a preliminary consultation treatment process is specified and manoeuvres to equalize pressure across tympanic membranes explained. During the HBOT session, an airlock allows entry into or exit from the main chamber without any pressure change. A second airlock allows the passage of any small pieces of equipment required in the event of an emergency. Conventional treatment (standard of care) will already have been initiated regardless of patients' group. A specific hyperbaric approved device allows morphine to be administered via a PCA pump whilst in the hyperbaric chamber. Vital signs will be measured before and after the HBOT to monitor for desaturation. In this protocol, the patient breathes FiO2 of 1 (pure oxygen) when allocated to the intervention group. When breathing 100% oxygen, there is no accumulation of nitrogen in the tissues which excludes the risk of decompression sickness. Both compression and decompression phases will take 10 min each (1m per minute). Compression will be performed on air and decompression on oxygen. Each session will therefore last 95 minutes (10 min of compression to 2.0 ATA, followed by 3 cycles of 20 minutes of oxygen at pressure and a 5 minutes on air, followed by 10 minutes of decompression).

Sham group

To best simulate HBOT for the sham arm, our group has previously validated a procedure that we will be employed in this study (41) . Using the method described below, the study identified a pressure of 1.3 ATA, FIO2=0.21 as being the most realistic sham to create the illusion of HBOT in healthy volunteers. It has the advantage of being extremely low risk (it is the equivalent of a pressure at 3 meters of depth in water) and has an FIO2 equivalent of 0.27, which has little effect on oxygen (42). With this approach, even if there is a slight positive effect from the minimal pressurisation in the sham group it should not affect overall results. The sham consists of using the same chamber as in the intervention arm, but with pressure limited to 1.3 ATA and the FIO2 at 0.21 (air administered via respiratory devices). As in the intervention arm the session will have a total duration of 95 mins (compression period, of 5 min, followed by 85 min at 1.3 ATA, then a decompression period of 5 min). The sham group (control group), will avoid any potential adverse effects of pressurisation.

**Blinding (masking)**

Randomisation will be double-blinded: neither the patient nor physician caring for the patient will be informed of the strategy. Only the HD team, in charge of the session, will be aware of the treatment arm which has been attributed.

A questionnaire will be given to the patient to evaluate their experience. To avoid influencing patients, the HD team will be excluded from involvement in the questionnaire and more importantly from the evaluation of the VAS which instead, will be carried out by the initial department (ED or hospital ward). The statistician will equally be blinded to the intervention/sham group allocation.

### Emergency Unblinding

If side effects are experienced, then the sessions (HBOT/sham) will be interrupted. An unblinding procedure may be proposed in the event of a serious adverse event (SAE) for example as barotrauma or seizure. This procedure will be initiated by the HD team, with the agreement of the coordinating investigator. Unblinding will allow the clinician in charge to investigate and treat the cause of the SAE. An identical unblinding procedure will also be implemented for the second and/or third session.

### Criteria for discontinuing or modifying interventions.

Adverse events may result in the interruption of HBOT/sham sessions. These however have been shown to be extremely infrequent (43, 44). In 99.3% of all HBO treatments, no adverse events were recorded. The most frequently occurring event was ear/sinus barotrauma (0.37%), followed by claustrophobia (0.16%) and symptomatic hypoglycemia (0.08%). More serious events, such as seizure (0.02%) or lung toxicity (0.01%), occurred very rarely. Only 1 instance of confirmed pulmonary barotrauma was identified out of 1.5 million treatments (44).

Middle ear/sinus barotrauma is the most frequent side effect. Patients may report difficulty with ear equalization, with a feeling of pressure, ear pain, or discomfort during compression (the initial phase of HBOT). These adverse events are limited by carrying out an ear examination before the HBOT session and by the systematic accompaniment of patients in the chamber during their first compression. In the event of a failure to equalize ears, a myringotomy (tympanic perforation) will be proposed. A separate consent from the patient will be sought before performing this. In the case of refusal, they will no longer continue to benefit from the chamber but will be analysed in the patient's randomised arm.

Panic attacks and other manifestations of anxiety or claustrophobia related to confinement in the chamber are prevented by the presence of the healthcare personnel who accompany patients during their first HBOT compression and who remain nearby afterwards.

The recognised presentation of CNS oxygen toxicity during clinical hyperbaric oxygen treatment is an oxygen toxicity seizure (Paul Bert Effect). Prodromal symptoms, (twitching, staring gaze, auditory hallucinations, visual changes, nausea, vertigo, anxiety, and irritability), have been reported, although they appear in <50% of cases of

oxygen toxicity. Tonic-clonic seizure activity rapidly follows these signs. This reversible condition can be resolved, with no residual neurological damage, by decreasing the inspired partial pressure of oxygen (PO<sub>2</sub>), resulting in a reduced cerebral PO<sub>2</sub> and if necessary, with the use of benzodiazepines. Combination of benzodiazepines with morphine should be considered with much caution in sickle cell patients, due to the increased risk of respiratory depression. Due to the particular sensitivity of sickle cell patients, the study's HBOT protocol limits pressure to 2.0 ATA with pure oxygen, to avoid potential convulsion. In children, transcranial dopplers are carried out regularly and abnormalities (>200cm/sec) are an exclusion criterion to limit the possible risk of seizures.

Other potential side effects include progressive myopia, headache, nausea, numbness, or pulmonary dyspnea. These usually recede after treatment ceases. In case of a serious adverse reaction during the first or the second session, additional sessions will be foregone. If the patient does not wish to undergo the second or third session, it will be cancelled. Since the main outcome is evaluated after the first session, any patients leaving the protocol will not be replaced.

**Standard of care given to all patients in both randomized groups**

The usual protocols for VOC / ACS should be implemented as soon as possible after the patient's arrival at the hospital.

All patients should benefit from a treatment that will combine:

- Normobaric oxygen therapy administered by nasal cannula to maintain a SpO<sub>2</sub>≥94%.
- Intravenous hydration: NaCl 1500 ml/24H (adapted in the case of paediatric inclusion).
- Analgesics: paracetamol and IV morphine (+/- PCA) according to defined adult or paediatric guidelines (A PCA-protocol will be established for unified management. This will involve boluses of 0.025 to 0.05mg/kg without background infusion initially).
- Antibiotic therapy for ACS, documented infectious syndrome or suspected secondary infection (Co-Amoxycillin + macrolide or Levofloxacin in case of allergy).

Transfusion therapy (transfusion or exchange transfusion) may be indicated according to the usual recommendations.

If transfusion therapy (simple or exchange transfusion) is administered because of VOC or ACS, this will be considered as a treatment failure and the protocol will be interrupted. If the indication for transfusion therapy is not related to VOC and/or ACS, it should be administered where possible, after the end of the 3<sup>rd</sup> HBOT session and after the last assessment with the VAS score i.e., 4 hours after the last session. If the indication is urgent, it may be administered without delay. Exchange transfusion may be indicated for the treatment of acute priapism (lasting more than 3 hours with little or no effect from intracavernous etilnephrine injections), ischemic stroke, and recurrent severe infection. It could also be indicated prior to surgery.

## Data safety monitoring board

An interim safety analysis will be performed to ensure that no serious adverse event occurs more significantly in the intervention group in comparison to the sham group. A comparative analysis will be carried out by Fisher's test, but the absolute number of events and the type of event will be qualitatively analyzed by the Data Safety Monitoring Board (DSMB) after removal of the blinding. The DSMB will benefit from regular interim (after 6, 12, then 50 patients) reports on safety outcomes and will have the option of requesting the discontinuation of the study for safety reasons.

## OBJECTIVES

### Primary Objective

To measure the effectiveness of HBOT, as measured by a decrease in the pain level 6 hours after initiation of HBOT in the treatment of SCD-VOC.

### Secondary Objectives

To analyse safety and other clinical, biological and cost outcomes of this HBOT procedure on SCD patients to observe the effects of HBOT on: relevant biological markers (CRP, LDH) during a sickle-cell crisis, duration of VOC, transfusion therapy indications, morphine doses and length of stay (LOS). In addition, we wish to analyse long term impact of HBOT by observing the frequency of VOC recurrence during the following year as well as analysing cost implications of the implementation of HBOT.

An ancillary study will consist of an analysis of the effect of HBOT on the evolution of biological markers of interest in the SCD-crisis in both the intervention and sham groups. As described earlier, this will be achieved by taking blood samples from adult patients included in the Geneva centre only, who consent for this additional study ("Biobank study").

## OUTCOMES

### Main Outcome

The difference in the global VAS pain score evaluated immediately before (H0) and 4hrs after (H6) the HBOT/sham session.

### Secondary main outcome (composite outcome)

The number of patients with a VAS pain score > 4 and/or a morphine dosage > 1 mg/h IV after the HBOT session.



**Other outcomes (Comparison to be made between the intervention and sham groups)**

Differences in VAS pain scores between H0 and H24, change in hourly doses of morphine; accumulated dose of parenteral opioid equivalent from baseline until VOC resolution or readiness for discharge; time until discontinuation of IV opioids; length of hospital stay; number of patients experiencing relief from pain at both H6 and H24 (with a reduction in VAS pain score of > 30% compared with H0); change in pain intensity from baseline to every 4 hours during awake time, until VOC resolution; pain location; patient’s “global impression of change” (using a 7-point rating scale with the options “very much improved,” “much improved,” “minimally improved,” “no change,” “minimally worse,” “much worse,” and “very much worse.”) (45); frequency of patients with VOC terminated at each visit (VOC is terminated when VAS<2, in the absence of painkillers of level III) and time until end of VOC (until 48 hours of follow-up); absence of parenteral opioid use; indications for and implementation of transfusion therapies during hospitalisation; complications (notably Acute Chest Syndrome, priapism, stroke or other) and values of appropriate biomarkers associated with a sickle cell crisis (LDH, CRP); readiness for discharge as judged by the patient or physician; evaluation of patient satisfaction via a questionnaire; recurrences of VOC during the following year; further hospitalisations during the following year; treatment costs, death during hospitalisation or after discharge.

**STATISTICAL METHODS**

**Analysis of the primary outcome:**

The primary outcome is the change in global VAS between before (H0) and after (H6) the first HBOT session. The mean change in VAS will be reported in each arm with the two-sided 95% confidence interval. The null hypothesis that the mean change in VAS is the same in both arms will be tested using linear regression model adjusted for the centre and the hourly dose of morphine prior to inclusion (high versus low, defined by the average hourly dose since admission converted into Morphine IV equivalent, respectively > 2mg/h IV or ≤ 2mg/h IV) and the baseline VAS. The risk of type I error will be 0.05 two-sided. The intervention’s effect will be assessed by the adjusted mean difference in change of VAS between study arms and will be reported with the two-sided 95% confidence interval. Other adjustments will be considered, for example depending on the homo or heterozygous type, treatment with hydroxycarbamide or a complication with an Acute Chest Syndrome.

**Analysis of the secondary outcomes:**

The proportion of patients with a VAS pain score >4 and/or a morphine dosage>1 mg/h IV (or equivalent in mcg/kg for the paediatric protocol) after the HBO session will be reported with the Clopper-Pearson exact 95% confidence interval in each study arm. The null hypothesis that the risk to have this outcome is the same in both study arms will be tested using binomial generalized linear regression models adjusted for the centre and the hourly dose of morphine prior to inclusion. The risk of type I error will be 0.05 two-sided. The adjusted difference of risk (from

a binomial regression model) and the adjusted risk ratio (from a log-binomial model) will be reported with the two-sided 95% confidence interval.

### Analysis of the other outcomes

The difference in VAS pain scores between H0 and H24, the difference in hourly doses of morphine between before and after HBOT and the difference in biomarkers (LDH, CRP) between before and after HBOT will be analysed using similar statistical methods than for the primary outcome.

The length of hospital stay will be described by median and quartiles and compared between study arms with non-parametric test since the distribution of the length of stay is expected to deviate considerably from the normal. The van Elteren test (a stratified version of the Mann-Whitney's test) will be used with stratification on centres and hourly dose of morphine prior to inclusion.

The frequency of patients with a reduction in VAS pain score of 30% or more from H6 to H24 and the frequency of patients with VAS<2 (in absence of painkillers of level III) at each visit will be analysed using similar statistical methods than for the secondary outcome.

Recurrences of VOC and further hospitalisations during the following year will be described. Survival analyses (Kaplan-Meier survival estimates and log-rank test stratified on centres and hourly dose of morphine prior to inclusion) will be conducted to compare the risk of the first recurrence and the first hospitalisations between intervention and sham. A Cox regression model will be used to assess the hazard ratio adjusted for the centre and the hourly dose of morphine prior to inclusion. If needed, survival models with competing risk will be used.

The patient's "global impression of change" will be described and compared between study arms using van Elteren's test.

The significance level will be two-sided  $\alpha = 0.05$  for all analyses. Analyses will be conducted with R (R Development Core Team (2008). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.).

Any deviation from the original statistical plan will be described and justified in the final trial report.

### Analysis population and missing data

**Dataset to be analysed:** The analysis will be carried out in intention to treat. A sensitivity analysis will be performed in per-protocol analysis (excluding patients who did not have the HBO/sham session). No method of managing missing data is envisaged. The main outcome should be available in all situations.

**Interim Analysis:** No interim effectiveness analysis will be performed. Only, an interim safety analysis is envisaged.



**PATIENT AND PUBLIC INVOLVEMENT**

The protocol was presented to, discussed with, and approved by the patients' association "Suisse-Drépano". Information will be disseminated to all its members, and an information session will be proposed by the principal investigator.

**ETHICS AND DISSEMINATION**

HBOT-SCD will be carried out in accordance with the research plan and the Declaration of Helsinki, the Swiss Law and Swiss regulatory authority's requirements as applicable. The application has been approved by the lead committee, that is, the Ethic Committee of Geneva (CER 2019-01707).

The results of the studies will be disseminated by several media, including publications in peer-reviewed international medical journals, and presentations at national and/or international conferences. The results can also be incorporated into international recommendations.

Data will be shared according to FAIR principles.

**DISCUSSION AND IMPLICATIONS**

The results of the HBOT-Sickle cell trial could have an important impact on the effective and rapid management of VOCs in SCD. These crises can be extremely painful and often lead to long hospitalisations with potentially serious consequences and a significant use of morphine and blood transfusions. A positive study would have a major impact on patients with sickle cell disease, improving the quality of life for patients not only in Europe but also worldwide. It is an opportunity to test the effectiveness of the HBOT in the VOC-SCD with a well-designed RCT and a validated sham.

The use of morphine and the need for iterative transfusions to manage the patients during VOCs and ACSs are associated with potential health consequences such as opioid dependence, and transfusion risks as well as the use of scarce resources. Expected benefits of HBOT are the reduction of: level and duration of pain experienced, duration of the crisis, length of stay, number of transfusions required, number of morphine doses administered, and reduction in the frequency of ACS and VOCs. Indeed, breaking of the vicious cycle of the crisis early should decrease the frequency/severity of further VOCs.

This RCT will increase the level of evidence concerning the efficacy of HBOT in SCD, and if the results are positive, could broaden its indications. If positive, HBOT could be proposed as an alternative to conventional treatments, creating a place for it amongst current therapeutic options. This could be of particular value in low-income countries, where the disease is frequent, and use of transfusions remains problematic.

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The medico-economic interest will of course have to be analysed, but the possible reduction of LOS and also the use of blood transfusions will potentially lead to major savings when compared with the cost of this technique. This study will use a hyperbaric chamber that is already marketed, licensed, and used in other diseases.

In conclusion, we are anticipating that this study will clarify the strength of evidence, either supporting or refuting indication for the use of HBOT in the treatment of VOC-SCD.

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**Contributors:** JS has the primary responsibility for the final content. Study concept and design : JS, JaS, TM, PL, CC and RP. Draft of manuscript and statistical analysis: JS, JaS, TM, PL, CC, KS, RP and SB. Revision of manuscript: all authors read and approved the manuscript for final publication.

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**Competing interests :** none

**Patient and public involvement:** Protocol presented to, discussed with and approved by the patients' association "Suisse-Drépano".

**Patient consent for publication** Not applicable.

**Provenance and peer review** Not commissioned, externally peer reviewed.

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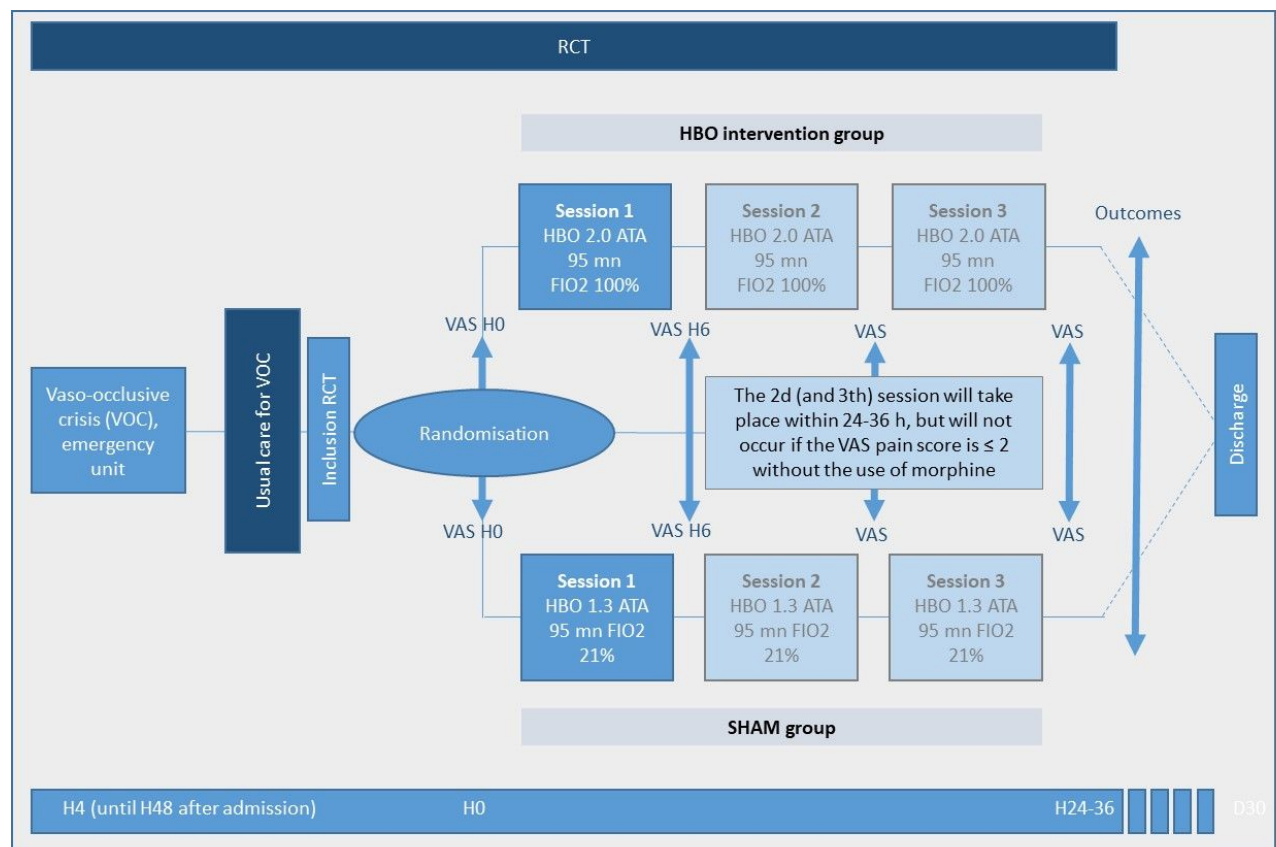


Fig 1: Schematic diagram of the Randomised Controlled Trial (RCT)

Table 1: Outcomes	
Outcomes parameters and measurement	
Main outcome	
Difference in the global visual analogue scale (VAS 0 to 10)	Difference between the global VAS score-H0 (immediately before 1 <sup>st</sup> HBOT session) and the global VAS score-H6 (4h after 1 <sup>st</sup> HBOT session)
Secondary main outcome	
Composite outcome: Number of patients with a VAS > 4 and/or a morphine dose within 4 hours after the HBOT session > 4 mg/4h IV morphine equivalent in addition to the usual ambulatory dose.	This evaluation will be made 4 hours after each HBOT session. The dose will be calculated based on PCA morphine usage data. All additional forms of morphine administered will be included in this calculation. An IV equivalent of this morphine will be calculated according to the recommendations of the Pain Network.
Other outcomes	
Length of stay	In the different acute wards (in days), starting from admission to the ED until discharge or transfer to a rehabilitation facility
VAS & Categorical Pain Score (CPS) ranges from 0 to 3 points in 7 body sites (the 4 limbs, ribs and sternum, head, and spine and pelvis).	Difference between the global CPS score-H0 (immediately before 1 <sup>st</sup> HBOT session) and the global CPS score-H6 (4h after 1 <sup>st</sup> HBOT session). Same measurements for 2nd and 3rd sessions.
Number of patients relieved at H6 and H24	Defined by a reduction of VAS score >30% with doses of level 3 painkillers inferior or equal to those of standard treatment (45)
Frequency of patients with VOC terminated at each visit	VOC is terminated when VAS<2, in the absence of level III painkillers
Duration of VOC since inclusion (in hours)	A VOC is considered terminated when at least 3 of the following 4 criteria are met: absence of fever for 8 hours; absence of pain progression and requirement of intravenous infusion of opioid analgesics for the last 8 hours; pain-free mobilisation; or absence of spontaneous pain with a CPS of 1 or less. The success rate will be defined as the percentage of VOC terminated without recourse to transfusion and or the occurrence of complications (46)
Indications and use of transfusions or transfusion exchanges	
The occurrence of complications during hospitalisation	

Vital signs	Heart rate, blood pressure and percutaneous oxygen saturation will be measured before and after each HBOT session
Relevant biological markers of sickle cell crisis	CRP, LDH, haemoglobin and reticulocytes levels, leukocytes, when carried out at inclusion and again at H24
Patient satisfaction	By questionnaire
The Patient Global Impression of Change scale (PGIC)	Assessed as recommended by IMMPACT for use in chronic pain clinical trials as a core outcome measure of global improvement with treatment (45)
Evaluation of VOC recurrences requiring hospitalisation	A new episode of VOC will be defined by the recurrence of painful spell after a free interval > 24 hours pain-free or with usual pain level. Number of patients with VOC at 3, 6 and 12 months.
A medico-economic analysis	On the basis of the actual expenditure in the intervention and control groups
Ancillary study "Biobank"	<p>Blood samples to measure these biomarkers will be taken immediately before and after the first HBOT session, with a third sample being taken at 24 to 36 hours after the beginning of the HBOT session or just before discharge if before 24 hours. Consequences of oxidative stress on lipids: Dosage of plasma Malondialdehyde (MDA), measurement of lipid peroxidation, dosage of Thiols Barbituric Acid Reactive Species (TBARS)</p> <p>Oxidative stress on proteins (Oxyblot): measurement of advanced glycation products (AGE), nitrosation products and protein glutathione formation</p> <p>Serum markers of inflammation: dosage of pro and anti-inflammatory cytokines (IL2-IL6, IL10, IL1-<math>\beta</math>, TNF<math>\alpha</math>, IL12)</p> <p>Adhesion proteins: ICAM-1, VCAM-1, glyocalix</p> <p>Tissue ischaemia: ischaemia modified albumin (IMA)</p>
Safety outcomes	Any side effects will be systematically documented both during and or following each HBOT session by the HD or ward team. Details will include date, precise time, duration and a detailed account of the event as well as the action and effect of the action taken. Probability of the HBOT session being accountable for the side effect and severity will be rated according

	to the usual grading. Any Serious Adverse Event must be reported to the Principal Investigator within 24h.
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# BMJ Open

## Protocol for a multicentric, double-blind, randomised controlled trial of hyperbaric-oxygen therapy (HBOT) versus sham for treating Vaso-Occlusive Crisis (VOC) in sickle cell disease (SCD) in patients aged 8 years or older (HBOT-SCD study).

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<b>Primary Subject Heading</b>:	Haematology (incl blood transfusion)
Secondary Subject Heading:	Genetics and genomics, Medical management
Keywords:	Emergency Service, Hospital, Anaemia < HAEMATOLOGY, INTERNAL MEDICINE

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Manuscripts

**Protocol for a multicentric, double-blind, randomised controlled trial of hyperbaric-oxygen therapy (HBOT) versus sham for treating Vaso-Occlusive Crisis (VOC) in sickle cell disease (SCD) in patients aged 8 years or older (HBOT-SCD study).**

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## ABSTRACT

**Introduction:** Sickle cell disease (SCD) is one of the most common genetic diseases in the world, annually affecting approximately 310,000 births and causing >100,000 deaths. Vaso-occlusive crisis (VOC) is the most frequent complication of SCD, leading to bone pain, thoracic pain (Acute Chest Syndrome [ACS]) and/or abdominal spasms. It is the main cause of mortality in patients with SCD, reducing life expectancy. Hyperbaric oxygen therapy (HBOT) is a safe and well-established method of increasing tissue oxygen delivery immediately by up to 10–20 fold. In context of VOC, HBOT has the potential to limit sickling. A previous pilot study of nine patients showed the safety and potential benefits of HBOT on VOC induced pain. Our study aims to assess the clinical safety and effectiveness of HBOT for treating VOC, its biological mechanisms of actions as well as its cost-effectiveness.

**Methods and analysis:** This is a multicentric, triple-blinded, randomised controlled trial. Patients aged 8 years or above with a diagnosed major form of SCD, presenting at one of the participating centres' Emergency Departments (EDs) with a VOC requiring level 3 analgesia (according to WHO definition) will be eligible. Exclusion criteria are: pregnancy, mechanical ventilation, previous history of stroke or prior transcranial doppler ultrasound anomaly, contra-indication to HBOT, and the need for above 2L/min of oxygen. All patients will receive the usual care for VOCs, including hydration, analgesics, normobaric oxygen therapy and when medically indicated, antibiotic therapy and/or transfusions. Within 24 hours of their arrival in the ED (or longer in specific cases), and after obtention of informed consent, patients will be randomised into the HBOT intervention group (2.0 Atmosphere Absolute [ATA], 90 min, FIO<sub>2</sub> = 1) or the sham group (1.3 ATA, 90 min, FIO<sub>2</sub> = 0.21). After their first HBOT session, patients will return to their acute-care ward. Patients in both arms will undergo a second and third session within 24 – 36 hours of the first, unless their pain-Visual Analogic Scale (VAS) is ≤ 2 without use of level 3 analgesics. The difference in the pain VAS before and after HBOT and other outcomes will be compared between the intervention and sham groups. Our composite primary outcome will be: 1) The change in global pain-VAS 6 hours after initiation of HBOT. 2) The number of patients with a VAS pain score > 4 and/or a morphine dosage > 1 mg/h IV after the HBOT/sham session. Other outcomes to be reported are: morphine usage, length of stay, biological parameters, satisfaction, complications and cost.

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**Ethics and dissemination:** *Ethical approval* CER Geneva 2019-01707 (last submission v5.1, 06.15.2023). The results of the studies will be disseminated by several media, including publications in peer-reviewed international medical journals, and presentations at national and/or international conferences.

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**Trial registration number** NCT04978116 (clinicaltrial.gov)

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**Strengths and limitations of this study**

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**Strengths**

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- Direct comparison of hyperbaric oxygen therapy to a control in Sickle cell disease vaso-occlusive crisis in a randomised trial.
  - Use of a validated sham treatment.
  - Triple blinding to avoid any bias.
  - Multi-disciplinary involvement in the study.

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**Limitations**

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- Logistically complex study to operationalize.

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Glossary

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ACS: Acute Chest Syndrome

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ATA: Atmosphere Absolute

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ED: Emergency department

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GVHD: graft-versus-host

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HBOT: Hyperbaric oxygen therapy

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HD: Hyperbaric Department

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PCA: Patient Controlled Analgesia

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RCT: randomised controlled trial

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SD: standard deviation

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VAS: Visual Analogic Scale

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VOC: Vaso-occlusive crisis

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SCD: Sickle cell disease

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## INTRODUCTION

Sickle cell disease (SCD) is the most frequent autosomal recessive disease, resulting in the production of sickle haemoglobin (HbS). Severe forms of SCD include haemoglobin SS resulting from the inheritance of HbS, S $\beta_0$ -thalassaemias due to co-inheritance of HbS with the  $\beta_0$ -thalassaemia mutation or other forms of co-inheritance of HbS with other  $\beta$ -globin gene mutation such as hemoglobin C (SC form) or  $\beta_+$ -thalassaemias (S $\beta_+$ ). Vaso-occlusive crisis (VOC) is the most common complication of SCD with an annual incidence of approximately 60% in adult sufferers. VOC usually causes bone, chest and/or abdominal pain and is a leading cause of death in SCD patients<sup>1</sup>. Acute Chest Syndrome (ACS) is a specific clinical presentation of VOC that is caused by pulmonary capillary occlusion and is defined by a triad of symptoms (chest pain, fever and radiological infiltration)<sup>2</sup>. It remains a major cause of hospitalisation and SCD-related mortality. Due to accompanying acute complications (such as VOC and ACS), with or without infections and chronic complications (such as stroke, pulmonary hypertension, chronic lung disease or nephropathy), SCD is associated with a reduced life expectancy and a median age of death of 43 years (IQR 31.5-55 years)<sup>3</sup>.

### Physiopathology of Sickle-cell disease

Hypoxia, acidosis, infection, dehydration or exposure to cold can trigger VOC/ACS by facilitating falciformation, due to HbS polymerisation. This leads to acute vaso-occlusion and haemolysis-mediated endothelial dysfunction<sup>2,4</sup>, inflammation, and ultimately, to vascular damage and tissue hypoxia with multi-organ injury<sup>5,6</sup>. Other cellular mechanisms involved in VOC/ACS are: adherence of sickle cells to the vascular endothelium, involving intercellular adhesion molecules such as ICAM-1, VCAM-1, PAF and CD 36<sup>7-10</sup>; erythrocyte-leukocyte-platelet endothelium interactions<sup>11</sup> with leukocytes<sup>12</sup> and platelet activation<sup>13</sup>; an increase in the number of circulating leukocytes<sup>14</sup> and formation of reactive oxygen forms (Reactive Oxygen Species -ROS-) which cause major endothelial dysfunction<sup>15</sup>. Intravascular haemolysis and cell adhesion promote both vasculopathy and vaso-occlusion<sup>16-18</sup>, as well as inflammation<sup>19</sup>. Mitochondrial dysfunction is an additional element of this vicious cycle, promoting platelet activation, and the release of inflammation activators<sup>20</sup>.

### Usual Care

Home-based management of a VOC is usually based on WHO classification level one and two analgesics (1: nonsteroidal anti-inflammatory drugs or acetaminophen, 2: codeine, tramadol). When VOC management is impossible with these analgesics, WHO classified level three analgesics are introduced (3: morphine or its derivatives e.g, fentanyl, oxycodone, buprenorphine or hydromorphone). Other VOC management is symptom appropriate and includes adapted hydration, local warming, oxygen therapy, folate administration, and antibiotic therapy. When these management strategies prove insufficient, or, in the presence of severe ACS<sup>21,22</sup>, ischaemic stroke, recurrent severe infection, or side effects that limit the use of morphine, erythropheresis and transfusions can be used. Alloimmunisation remains an extremely frequent consequence of the use of erythrocyte transfusion which, consequently, excludes the possibility of further transfusions or exchange transfusions as a therapeutic option in certain patients. Some patients become impossible to transfuse and HBOT is then an ultimate treatment



offered in case of CVO. The recent availability of L-glutamine, crizanlizumab and voxelotor provides an alternative or additional alternative to hydroxyurea, which remains the first choice for disease-modifying therapy<sup>3</sup>.

Overall survival rates remain high for individuals with SCD who have undergone allogeneic hematopoietic stem cell transplantation using matched sibling donors. Newer approaches to graft-versus-host (GVHD) prophylaxis and the incorporation of post-transplant cyclophosphamide have improved engraftment rates, reduced GVHD and have allowed for alternative donors for individuals in the absence of a HLA-matched sibling. Despite progress in this field, the increase in world SCD prevalence means management of VOCs remains a real and persistent problem, requiring the identification of alternative treatments outside of opioid and transfusion therapy.

**Hyperbaric Oxygen Therapy**

Hyperbaric oxygen therapy (HBOT) is a long-established technique with several recognised indications, including carbon monoxide poisoning, and anaerobic soft tissue infection<sup>23</sup>. HBOT consists of intermittent periods of breathing 100% oxygen at a pressure above that of sea level. The increase in pressure produced in the compression chamber significantly increases the partial pressure of the inspired gases in the blood and peripheral tissues<sup>24</sup>, reducing ischaemia, and as a consequence reducing / reversing sickling. HBOT allows the enhanced oxygenation of tissues where blood circulation is deficient, meaning oxygen no longer needs the support of erythrocytes to circulate and to be diffused.

In practice, the pressure increase is achieved by introducing compressed air into the hyperbaric chamber. Changes in ambient pressure modify the volume that gas occupies, including in the body (e.g., ear cavities, sinuses). In the hyperbaric chamber, when at the targeted pressure (outside of compression, decompression phases and during air breaks), patients breathe 100% oxygen through a specifically adapted mask or hood (if mask cannot be adjusted to patient morphology). A member of the HD team monitors patients at all times during hyperbaric oxygen therapy sessions. In a preliminary consultation prior to the first session, the treatment process and manoeuvres necessary to equalize pressure across tympanic membranes are explained. At the Geneva hospital, a multiplace hyperbaric chamber (HAUX, Germany) will be used. During sessions, the airlock system allows entry into or exit from the main chambers without any pressure change. A second airlock allows the passage of any small pieces of equipment required in the event of an emergency. When breathing 100% oxygen, there is no accumulation of nitrogen in the tissues thus excluding the risk of decompression sickness.

To best simulate HBOT in the sham arm, our group has previously validated a procedure that will be employed in this study<sup>25</sup>. Using the method described later, the study identified a pressure of 1.3 ATA, FIO2=0.21 as being most realistic in creating the illusion of HBOT in healthy volunteers. Its' advantages are of being extremely low risk (equivalent of a pressure at 3 meters of depth in water) and having an FIO2 equivalent of 0.27<sup>25</sup>. With this approach, even if there is a slight positive effect from the minimal pressurisation, equivalent to breathing air with a FIO2 of 0.27 in the sham group, it is considered as not affecting overall results. When breathing air at 1.3 ATA, there is no accumulation of nitrogen in the tissues thus excluding the risk of decompression sickness.



Adverse events of HBOT are extremely infrequent <sup>26 27</sup>. In 99.3% of all HBOT sessions, no adverse events were recorded. The most frequently occurring event was ear/sinus barotrauma (0.37%), followed by claustrophobia (0.16%) and symptomatic hypoglycaemia (0.08%). More serious events, such as seizure (0.02%) or lung toxicity (0.01%), occurred very rarely. Only one instance of confirmed pulmonary barotrauma was identified out of 1.5 million treatments <sup>27</sup>. To minimise the risk of middle ear/sinus barotrauma caused by difficulty with ear equalization, resulting in a feeling of pressure, ear pain, or discomfort, an ear examination will be carried out before the HBOT session. Patients will systematically be accompanied and “educated” by HD staff during their first compression (the initial phase of HBOT). The recognised presentation of CNS oxygen toxicity during clinical hyperbaric oxygen treatment is an oxygen toxicity seizure (Paul Bert Effect). Prodromal symptoms, (twitching, staring gaze, auditory hallucinations, visual changes, nausea, vertigo, anxiety, and irritability), have been reported, although they appear in <50% of cases of oxygen toxicity. Tonic-clonic seizure activity rapidly follows these signs. Other potential side effects include progressive myopia, headache, nausea, numbness, or pulmonary dyspnoea.

### Rationale for the study

In the early stages of VOC, the falciformation process is reversible if conditions, especially oxygenation, improve. HBOT is effective in increasing tissue oxygenation by increasing dissolved O<sub>2</sub> in the blood and limiting local hypoxaemia, a promoter of falciformation. This is often sufficient to break the vicious cycle of VOC and ACS. HBOT demonstrated an improvement in microcirculatory vascular flow, a decrease in leukocyte adhesion and an increase in tissue ATP in a rodent liver model <sup>28</sup>. HBOT may influence the NO synthetase and NO secretion, particularly in the central nervous system, leading to increase regional brain blood flow by vasodilation <sup>29</sup>. It also down-regulates cell adhesion proteins such as ICAM-1, VCAM-1, PAF and CD36 <sup>18 30-32</sup>. Thus, HBOT could counterbalance the effects of VOC which induces an overexpression of adhesion phenomena and leads to NO deficiency <sup>33</sup>. Several cases of complications related to SCD treated with HBOT have already been documented since 1966 in clinical cases or retrospective series with favourable outcomes reported for a number of patients <sup>33-42</sup>. In hyperbaric medicine guidelines, SCD is a recognised indication for HBOT <sup>19</sup>, with a low level of evidence (type C recommendations) <sup>23</sup>. The North American guidelines, however, did not include SCD as an indication <sup>43</sup>. Our group published a pilot non-comparative study on nine SCD patients, hospitalised for severe and resistant VOC, treated with HBOT (2.5 Atmosphere Absolute [ATA], 90min, FIO<sub>2</sub>=1). We observed a significant decrease in mean Visual Analogic Scale (VAS) score after one HBOT session compared to the VAS evaluated before the session. For two patients, the HBOT session was interrupted due to minor ear barotrauma (otalgia) during compression, but no problems relating to tolerance were reported in the seven other patients <sup>44</sup>.

This study aims to investigate the effect of HBOT in SCD-VOC in terms of efficacy on clinical outcomes (such as pain severity, analgesic consumption, duration of hospitalisation) and its safety.



An additional Biobank will be collected from patients recruited at the Geneva University Hospitals only. For adult participants who consent to this sub-study, venous blood samples will be taken immediately before and after the first HBOT session. A third blood sample will be taken at 24-36 hrs post start time of the first HBOT session (depending on time 24-hour point falls). If the patient is discharged from hospital before H24, the third blood sample will be taken just before discharge.

A VAS pain score will be evaluated four hours after the end of the initial HBOT session by blinded staff in the ED/ward. As HD staff know the allocated arm, they will not assess any of the patient outcomes to avoid bias. A second and third HBOT session will systematically be scheduled within 24 to 36 hours after the first session. Subsequent sessions will be consistent with the randomisation arm (sham or treatment). VAS pain scores will be evaluated immediately prior to and after the second and third sessions and again at four hours post session. These sessions will be cancelled if the patient's VAS score  $\leq 2$  without morphine treatment. In this case, we can consider that the VOC is finished. The second/third session will be cancelled in both arms of the study if a patient's condition deteriorates meaning that they need  $> 2\text{ l/min O}_2$  to maintain their  $\text{SpO}_2 > 92\%$ . Although the protocol will be interrupted for these patients, they will be integrated into the analysis in "intention to treat".

If a transfusion or exchange transfusion is administered after the first or second session, the specific effect of the HBOT treatment cannot be evaluated and thus, subsequent sessions will be cancelled. The VAS scores considered for analysis will be those collected before the transfusion/exchange. In certain strictly defined circumstances and only for patients needing more than  $2\text{ l/min}$  of oxygen, patients will not be recruited in the RCT, but could receive HBOT off protocol, as per usual practice in the institution. After inclusion, patients will be followed up for one year (by telephone or medical file).

### Sample size

We estimated the sample size calculation based on our primary, patient-focused, outcome: the difference in reduction in VAS score between H0 and H6 between study arms. The minimum difference in VAS score between H0 and H6 considered as clinically relevant is 1.3 points (on a scale of 0 to 10), particularly for the treatment of VOC in ER<sup>45 46</sup>. In the sham group, the decreased VAS score between H0 and H6 is expected to be less marked than in the intervention group (difference more than 1.3). The expected standard deviation (SD) of the VAS score difference between H0 and H6 is based on a preliminary study of 9 patients. In this study, the standard deviation of the VAS score after HBOT was 2.0 and the SD of the difference between H0 and H6 was 1, which corresponds to a strong correlation (0.88) between the VAS score at H0 and H6. With a more conservative assumption that the correlation between H0 and H6 is 0.50, the SD of the VAS score difference between H0 and H6 is 2 in the HBOT group. Assuming that the standard deviation of the difference is the same in the sham group and a bilateral risk alpha of 0.05, the sample size to obtain a 90% power is 50 patients per study arm (100 patients in total).

Study phase

BOX 1 : ELIGIBILITY CRITERIA

**Inclusion criteria:**

- Patients aged 8 or older
- Diagnosed with a major SCD disorder (SS, SC, Sβ<sub>0</sub> and Sβ<sub>+</sub>-thalassemias, other);
- A presentation of a VOC (with or without ACS) unresponsive to level 2 analgesics (WHO classification) which fulfils the criteria necessary for consultation at an ED.
- The ability to carry out the Valsalva manoeuver
- The ability to give informed consent and sign a written informed consent form (consent and signature of legal guardian is required for minor patients or those under guardianship).

**Exclusion criteria:**

- Pregnancy.
- Indication for mechanical ventilation (non-invasive ventilation/oro-tracheal intubation) ;
- Contraindication for HBOT established by a physician responsible for hyperbaric medicine;
- An anomaly in the results of prior transcranial Doppler (TCD) ultrasound (> 200 cm/sec) or a previous history of stroke (but TCD will not be performed for the study);
- Patients requiring more than 2 l/min (28%) of normobaric oxygen to maintain an SpO<sub>2</sub> ≥ 92% (the placebo group would receive equivalent of only 27% O<sub>2</sub>).
- Transfusion/ exchange transfusion just prior to the first session.

This is a phase 3 study. An RCT remains the only appropriate design to answer the question of effectiveness of HBOT on VOC. This study follows the pilot study published by our group in 2012. As SCD is a rare disease, the considered sample size is sufficient to meet the phase 3 criteria.

Patient population

Any patient, 8 years of age or older with a VOC, admitted to the Emergency Department (ED) and requiring level 3 analgesia (according to WHO definition) may be screened and included in this study unless they have exclusion criteria.

Inclusion and exclusion criteria

Inclusion and exclusion criteria are listed in detail in box 1.

The contraindication for HBOT established by a physician responsible for hyperbaric medicine as per ECHM/UHMS guidelines, e.g. untreated pneumothorax, eustachian tube dysfunction, claustrophobia or other usual contraindications <sup>47</sup>.

Due to the particular sensitivity of sickle cell patients, the study's HBOT protocol limits pressure to 2.0 ATA with 100% oxygen, in order to decrease the risk of seizure. In children, transcranial dopplers are carried out regularly and abnormalities (>200cm/sec) are an exclusion criterion to limit the possible risk of seizures.

### Recruitment

To optimise recruitment, this study has been presented to and discussed with the Swiss Sickle Cell Association. Patient awareness of this protocol will be raised by the haematologist responsible for out-patient follow-up of sickle cell patients. The study has been presented to the ED team on several occasions and some of the ED team have been included as investigators to optimise inclusion directly from the ER. Alerts to beeps and emails have been set up on the electronic files of sickle cell patients to notify the research and HD teams of the arrival of a patient with VOC to the ED. Treatment by HBOT outside of the protocol for VOC will not be possible unless contraindications exist for inclusion in the sham arm.

Potential patients will be identified on their arrival by the triage nurse or treating physician who will inform the hyperbaric physician of their arrival. The hyperbaric physician and/or research nurse will then verify the patient's inclusion and exclusion criteria. The hyperbaric physician is responsible for giving information concerning the study and obtaining the patient's /legal representative's informed consent. The patient will then be transferred to the HD, next to the ED. There, the HD team will carry out the randomisation and be responsible for giving the treatment/sham according to randomised arm. Data will be collected by research personnel from their respective participating centres. The data will be entered into the eCRF (REDCap software) with confidentiality storage.

### Randomisation

Randomisation will be carried out by the HBOT team using REDCap. It will be stratified by site but also by the average hourly dose of morphine received since arrival in the ED until the time of randomisation (high versus low, high = Morphine IV equivalent,  $\geq 5\text{mg/h IV}$ , low =  $<5\text{mg/h IV}$  in adult patients). Randomisation will be carried out between the intervention strategy (HBOT session at 2 ATA; 95 min; FIO<sub>2</sub>=1) and the sham (HBOT session at 1.3 ATA; 95 min; FIO<sub>2</sub>=0.21). Patients will be treated using the same strategy for all sessions. Randomisation will be triple blinded: the patient, research team and treating physician will not be aware of the randomisation arm, only the HD team.

To preserve the blinding, the randomisation blocks are completely random and the randomisation list created by a researcher completely external to the protocol. The size of randomised group is 1: 1 (same numbers of subjects in the two groups).

A questionnaire will be given to the patient to evaluate their experience and impressions. To avoid influencing patients, the HD team will be excluded from involvement in the questionnaire and more importantly from the evaluation of the VAS. The statistician will equally be blinded to the intervention/sham group allocation.

**Intervention and sham**

Conventional treatment (standard of care) will already have been initiated regardless of patients' group. A specific hyperbaric compatible device allows morphine to be administered via a PCA pump whilst the hyperbaric chamber is pressurised. Vital signs will be measured before and after the HBOT to identify the improvement or worsening of a patient's condition.

Intervention Group

During pressurisation, patients in the intervention group will breathe an FiO2 of 1 (pure oxygen) via a specifically adapted mask. Both compression and decompression phases will take 10 min each (1m per minute). Compression will be performed in air and decompression in oxygen. Each session will last 90 minutes (10 min of compression to 2.0 ATA, followed by 3 cycles of 20 minutes of oxygen at pressure with 5 minutes in air between cycles, followed by 10 minutes of decompression) [Figure 2]. Surveillance by a member of the HD team is assured throughout the entirety of each session.

Sham group

A previously validated sham <sup>25</sup>, will be used, using the same chamber as in the intervention arm, but with the pressure limited to 1.3 ATA and an FIO2 at 0.21 (via the same model of mask as the intervention arm). As described, additional measures will be taken to simulate the intervention session such as faster compression with ventilation during the fictitious compression time, heating at compression and cooling at decompression. The sham session will be identical in duration to the intervention group (total duration of 90 mins with 5 min compression period followed by 80 min at 1.3 ATA with air, then a decompression period of 5 min) [Figure 2]. The sham group (control group) will avoid only mild potential adverse effects of pressurisation such as minimal ear barotrauma.

**Unblinding emergency procedure**

If side effects are experienced, the sessions (HBOT/sham) will be interrupted. An unblinding procedure may be proposed in the event of a serious adverse event (SAE) for example as barotrauma or seizure. This procedure will be initiated by the HD team, with the agreement of the coordinating investigator. Unblinding will allow the treating clinician to investigate and treat the cause of the SAE. An identical unblinding procedure will also be implemented for the second and/or third session.



### Standard of care given to all patients in both randomised groups

The usual protocols for VOC / ACS should be implemented as soon as possible after the patient's arrival at the hospital.

All patients should benefit from a treatment that will include:

- Normobaric oxygen therapy administered by nasal cannula to maintain a  $SpO_2 \geq 94\%$ .
- Intravenous hydration: NaCl 1500 ml/24H (adapted in the case of paediatric inclusion).
- Analgesics: paracetamol and IV morphine (+/- PCA) according to defined adult or paediatric guidelines. (A PCA-protocol will be established for unified management. This will involve, for example, morphine boluses of 0.025 to 0.05mg/kg without background infusion initially in adult patients, with hydromorphone or fentanyl ften being used for paediatric patients).
- Antibiotic therapy for ACS, documented infectious syndrome or suspected infection (Co-Amoxycillin + macrolide or Levofloxacin in case of allergy).

Transfusion therapy (transfusion or exchange transfusion) may be indicated according to the usual recommendations.

### Criteria for discontinuing or modifying interventions.

Adverse events may result in the interruption of HBOT/sham sessions. In the event of a failure to equalize ears, a myringotomy (tympanic perforation) will be proposed. A separate consent from the patient will be sought before performing this. In the case of refusal, patients will no longer continue to benefit from the chamber but will be analysed in their randomised arm. Panic attacks, other manifestations of anxiety or claustrophobia related to confinement in the chamber are prevented by the presence of a healthcare personnel who accompany patients during their first HBOT compression and who remain nearby afterwards.

Seizures due to the oxygen toxicity, can be resolved, with no residual neurological damage, by decreasing the inspired partial pressure of oxygen ( $PO_2$ ), resulting in a reduced cerebral  $PO_2$ . If necessary, benzodiazepines are used. However, combining benzodiazepines with morphine should be considered with much caution due to the increased risk of respiratory depression (both drugs depress the brainstem areas responsible for regulating breathing in any hypoxic patients).

Other adverse events usually recede after treatment ceases. In the case of a serious adverse reaction during the first or the second session, additional sessions will be foregone. If the patient does not wish to undergo the second or third session, it will be cancelled. Since the main outcome is evaluated after the first session, any patients leaving the protocol will not be replaced.

If transfusion therapy (simple or exchange transfusion) is administered because of the VOC or ACS, this will be considered as a treatment failure and the protocol will be interrupted. If the indication for transfusion therapy is not related to VOC and/or ACS, it should be administered, where possible, after the end of the 3<sup>rd</sup> HBOT session,

after the last assessment with the VAS score i.e., 4 hours after the last session. If the indication is urgent, it may be administered without delay. Exchange transfusion may be indicated for the treatment of acute priapism (lasting more than 3 hours with little or no effect from intracavernous etilnephrine injections), ischemic stroke, and recurrent severe infection. It could also be indicated prior to surgery.

**Data safety monitoring board**

The appointed Data Safety Monitoring Board (DSMB) will be unblinded and will undertake an interim safety analysis to ensure that no serious adverse events occur significantly more frequently in the intervention group than in the sham group. A comparative analysis will be carried out by Fisher's test. The DSMB will benefit from regular reports on safety outcomes (after 6, 12, then 50 patients) and if necessary, will have the option of requesting the discontinuation of the study for safety reasons. Furthermore, an internal independent audit from the HUG also validated the continuation of the study to the extent that it would be funded, by proposing substantial improvements.

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## OBJECTIVES

### Primary Objective

To measure the effectiveness of HBOT in the treatment of SCD-VOC, as measured by a decrease in the pain level 6 hours after the initiation of HBOT.

### Secondary Objectives

To analyse safety and other clinical, biological and cost outcomes of this HBOT procedure on SCD patients and to observe the effects of HBOT on relevant biological markers (CRP, LDH) during a sickle-cell crisis, duration of VOC, transfusion therapy indications, morphine doses, length of stay (LOS) and the occurrence of adverse events. In addition, we wish to analyse the long-term impact of HBOT by observing the frequency of VOC recurrence during the following year as well as analysing cost implications of the implementation of HBOT.

An ancillary study will analyse the effect of HBOT on the evolution of biological markers of interest during a VOC in both the intervention and sham groups. As described earlier, blood samples will be taken from adult patients included at the Geneva centre only, who consent to this additional study ("Biobank study").

**OUTCOMES**

All outcomes are detailed in the Table 1.

**First Primary Outcome**

The difference in the global VAS pain score evaluated immediately before (H0) and 4hrs after (H6) the HBOT/sham session <sup>48</sup>.

**Second primary outcome (composite outcome)**

The number of patients with a VAS pain score > 4 and/or a morphine dosage > 1 mg/h (or 0.02 mg/kg/h) IV after the HBOT/sham session.

**Secondary outcomes (Comparison to be made between the intervention and sham groups)**

Differences in VAS pain scores between H0 and H24, difference between H0, H6 and H24 of Categorical Pain Score (CPS) ranges from 0 to 3 points in 7 body sites, change in hourly doses of morphine; accumulated dose of parenteral opioid equivalent from baseline until VOC resolution or until discharge; time until discontinuation of IV opioids; length of hospital stay; number of patients experiencing relief from pain at both H6 and H24 (with a reduction in VAS pain score of > 30% compared with H0); change in pain intensity from baseline to every 4 hours during awake time, until VOC resolution; pain location; patient's "global impression of change" (using a 7-point rating scale with the options "very much improved," "much improved," "minimally improved," "no change," "minimally worse," "much worse," and "very much worse.") <sup>49</sup>; frequency of patients with VOC terminated at each visit (VOC is terminated when VAS<2, in the absence of painkillers of level 3) and time until end of VOC (until 48 hours of follow-up); absence of parenteral opioid use; indications for and implementation of transfusion therapies during hospitalisation; complications (notably ACS, priapism, stroke or other) and values of appropriate biomarkers associated with a sickle cell crisis (LDH, CRP); readiness for discharge as judged by the patient or physician; evaluation of patient satisfaction via a questionnaire; recurrences of VOC during the following year; further hospitalisations during the following year; treatment costs; death during hospitalisation or after discharge; occurrence of adverse events of HBOT/sham.

The different outcomes will be identified in the patient's electronic charting system by the research team and entered into the eCRF. The specific outcomes (scores, biology, investigations) will be prescribed by the HD physician/research team and performed then documented by the clinical team in charge of the patient.

Table 1: Outcomes parameters and measurement

First Primary outcome	
Difference in the global visual analogue scale (VAS 0 to 10)	Difference between the global VAS score - H0 (immediately before 1 <sup>st</sup> HBOT session) and the global VAS score - H6 (4h after 1 <sup>st</sup> HBOT session). Pain network: <a href="#">Réseau douleur à Genève aux HUG - HUG</a> .
Second primary outcome	
Composite outcome: Number of patients with a VAS > 4 and/or a morphine dose within 4 hours after the HBOT session > 1 mg/h IV morphine equivalent in addition to the usual outpatient dose.	This evaluation will be made 4 hours after each HBOT session. The dose will be calculated based on PCA morphine usage data. All additional forms of morphine administered will be included in this calculation. An IV equivalent of this morphine will be calculated according to the recommendations of the Pain Network.
Secondary outcomes	
Length of stay	In the different acute wards (in days), starting from admission to the ED until discharge or transfer to a rehabilitation facility
Categorical Pain Score (CPS) ranges from 0 to 3 points in 7 body sites (the 4 limbs, ribs and sternum, head, and spine and pelvis).	Difference between the global CPS score - H0 (immediately before 1 <sup>st</sup> HBOT session) and the global CPS score - H6 (4h after 1 <sup>st</sup> HBOT session). Same measurements for 2nd and 3rd sessions.
Number of patients relieved at H6 and H24	Defined by a reduction of VAS score >30% with doses of level 3 painkillers inferior or equal to compared with H0 <sup>49</sup>
Frequency of patients with VOC terminated at each visit	VOC is terminated when VAS<2, in the absence of level III painkillers
Duration of VOC since inclusion (in hours)	A VOC is considered terminated when at least 3 of the following 4 criteria are met: absence of fever for 8 hours; absence of pain progression and requirement of intravenous infusion of opioid analgesics for the last 8 hours; pain-free mobilisation; or absence of spontaneous pain with a CPS of 1 or less. The success rate will be defined as the percentage of VOC terminated without recourse to transfusion and or the occurrence of complications <sup>50</sup>
Indications and use of transfusions or transfusion exchanges	
The occurrence of complications during hospitalisation	

Vital signs	Heart rate, blood pressure and percutaneous oxygen saturation will be measured before and after each HBOT session
Relevant biological markers of sickle cell crisis	CRP, LDH, haemoglobin and reticulocytes levels, leukocytes, when carried out at inclusion and again at H24
Patient satisfaction and impression questionnaire	Questions about : <ul style="list-style-type: none"> <li>- Satisfaction of reception, HBO team (communication, availability, privacy), local, security, comfort, information.</li> <li>- Sensations (temperature, noise, pain).</li> <li>- General impression of the management, the treatment and the feeling of having undergone “compression” or not.</li> </ul>
The Patient Global Impression of Change scale (PGIC)	Assessed as recommended by IMMPACT for use in chronic pain clinical trials as a core outcome measure of global improvement with treatment <sup>49</sup>
Evaluation of VOC recurrences requiring hospitalisation	A new episode of VOC will be defined by the recurrence of painful spell after a free interval > 24 hours pain-free or with usual pain level.
A medico-economic analysis	Based on the actual expenditure in the intervention and control groups
Ancillary study “Biobank”	Blood samples to measure these biomarkers will be taken immediately before and after the first HBOT session, with a third sample being taken at 24 to 36 hours after the beginning of the HBOT session or just before discharge if less than 24 hours. Consequences of oxidative stress on lipids: Dosage of plasma Malondialdehyde (MDA), measurement of lipid peroxidation, dosage of Thiols Barbituric Acid Reactive Species (TBARS) Oxidative stress on proteins (Oxyblot): measurement of advanced glycation products (AGE), nitrosation products and protein glutathione formation Serum markers of inflammation: dosage of pro and anti-inflammatory cytokines (IL2-IL6, IL10, IL1-β, TNFα, IL12) Adhesion proteins: ICAM-1, VCAM-1, glycocalix Tissue ischaemia: ischaemia modified albumin (IMA)
Safety outcomes	Any side effects will be systematically documented both during and/or following each HBOT session by the HD or ward team. Details will include date, precise time, duration and a detailed account of the event as well as the action and effect of the action taken. The probability of the HBOT session being accountable for the side effect and severity will be rated according to the usual grading: Mild (tolerable), Moderate (interferes with daily activity) or Severe (daily activities impossible). Any Serious Adverse Event must be reported to the Principal Investigator within 24h.

## STATISTICAL METHODS

### Analysis of the primary outcome:

The primary outcome is the change in global VAS between before (H0) and after (H6) the first HBOT session. The mean change in VAS will be reported in each arm with the two-sided 95% confidence interval. The null hypothesis that the mean change in VAS is the same in both arms will be tested using linear regression model adjusted for the centre and for the hourly dose of morphine prior to inclusion (high versus low, defined by the average hourly dose since admission converted into morphine IV equivalent, respectively  $> 5\text{mg/h IV}$  or  $\leq 5\text{mg/h IV}$ ). The risk of type I error will be 0.05 two-sided. The intervention's effect will be assessed by the adjusted mean difference in change of VAS between study arms and will be reported with the two-sided 95% confidence interval. Other adjustments will be considered, for example depending on the homo or heterozygous type, treatment with hydroxycarbamide or a complication with an Acute Chest Syndrome.

### Analysis of the second primary outcome (composite):

The proportion of patients with a VAS pain score  $>4$  and/or a morphine dosage  $>1\text{ mg/h IV}$  (or equivalent in  $\text{mcg/kg}$  for the paediatric protocol) after the HBO session will be reported with the Clopper-Pearson exact 95% confidence interval in each study arm. The null hypothesis that the risk to have this outcome is the same in both study arms will be tested using binomial generalized linear regression models adjusted for the centre and the hourly dose of morphine prior to inclusion. The risk of type I error will be 0.05 two-sided. The adjusted difference of risk (from a binomial regression model) and the adjusted risk ratio (from a log-binomial model) will be reported with the two-sided 95% confidence interval.

### Analysis of the secondary outcomes

The difference in VAS pain scores between H0 and H24, the difference in hourly doses of morphine between before and after HBOT and the difference in biomarkers (LDH, CRP) between before and after HBOT will be reported in each arm using the same two-sided 95% confidence interval used for the primary outcome.

The length of hospital stay will be described by median and quartiles and compared between study arms with non-parametric test since the distribution of the length of stay is expected to deviate considerably from the normal. The van Elteren test (a stratified version of the Mann-Whitney's test) will be used with stratification on centres and hourly dose of morphine prior to inclusion.

The frequency of patients with a reduction in VAS pain score of 30% or more from H6 to H24 and the frequency of patients with  $\text{VAS} < 2$  (in absence of painkillers of level III) at each visit will be reported with the Clopper-Pearson exact 95% confidence interval in each study arm as for the second primary outcome.

Recurrences of VOC and further hospitalisations during the following year will be described. Survival analyses (Kaplan-Meier survival estimates and log-rank test stratified on centres and hourly dose of morphine prior to

inclusion) will be conducted to compare the risk of the first recurrence and the first hospitalisations between intervention and sham. A Cox regression model will be used to assess the hazard ratio adjusted for the centre and the hourly dose of morphine prior to inclusion. If needed, survival models with competing risk will be used.

A comparative description can be provided of other outcomes (accumulated dose of parenteral opioid, absence of opioid use, time to discontinuation of IV opioids, pain location, transfusions, complications, patient's satisfaction, readiness for discharge, costs).

The patient's "global impression of change" will be described and compared between study arms using van Elteren's test.

The significance level will be two-sided  $\alpha = 0.05$  for all analyses. Analyses will be conducted with R (R Development Core Team (2008). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.).

Any deviation from the original statistical plan will be described and justified in the final trial report.

#### Analysis population and missing data

**Dataset to be analysed:** The analysis will be carried out in intention to treat. A sensitivity analysis will be performed in per-protocol analysis (excluding patients who did not have the HBO/sham session). No method of managing missing data is envisaged. The main outcome should be available in all situations.

**Interim Analysis:** No interim effectiveness analysis will be performed. Only, an interim safety analysis is envisaged.

#### PATIENT AND PUBLIC INVOLVEMENT

The protocol was presented to, discussed with, and approved by the patients' association "Suisse-Drépano" (Swiss Association of Sickle Cell Patients). Information will be disseminated to all its members, and an information session will be proposed by the Principal Investigator.

#### ETHICS AND DISSEMINATION

HBOT-SCD study will be carried out in accordance with the research plan and the Declaration of Helsinki, the Swiss Law and Swiss regulatory authority's requirements as applicable. The application has been approved by the lead committee, that is, the Ethic Committee of Geneva (CER 2019-01707). All protocol modification will be approved by the Ethic Committee. Specific insurance has been taken out to cover possible complications of the protocol for patients.

The results of the studies will be disseminated by several media, including publications in peer-reviewed international medical journals, and presentations at national and/or international conferences. The results can also be incorporated into international recommendations. The authorship will follow the GCP rules.

Data will be shared according to FAIR principles.

## DISCUSSION AND IMPLICATIONS

A feasibility and safety phase was initiated at the Geneva university Hospitals in 2022 (first inclusion the 30.09.2022). Fifteen patients were included with DSMB analyses after the inclusion of 6 and 12 patients recommending the continuation of the study in its current state.

The results of the HBOT-Sickle cell disease study could have an important impact on the effective and rapid management of VOCs in SCD. Often extremely painful and leading to long hospitalisations, these crises have potentially serious consequences and require significant use of both morphine and blood products. A positive study would have a major impact on patients with sickle cell disease, improving the quality of life for patients not only in Europe but also worldwide. This is an opportunity to test the effectiveness of the HBOT in VOC-SCD management with a well-designed RCT and a validated sham.

The use of morphine and the need for iterative transfusions to manage the patients during VOCs and ACSs are associated with potentially serious health consequences such as opioid dependence, and transfusion risks as well as the use of scarce resources. Expected benefits of HBOT are the reduction of the: level and duration of pain severity, crisis duration, hospital length of stay, number of transfusions required, cumulative quantity of opioids, and frequency of ACS and VOCs. Indeed, breaking of the vicious cycle of the crisis early should decrease the frequency/severity of further VOCs and their consequences.

This RCT will increase the level of evidence concerning the efficacy of HBOT in SCD-VOC, and if the results are positive, could broaden its indications. If positive, HBOT could be proposed as an alternative to conventional treatments, creating a place for it amongst current therapeutic options. This could be of particular value in low-income countries, where the disease is frequent, and use of transfusions remains problematic. The introduction of mobile and easy installed hyperbaric chambers could then be considered in these countries.

The medico-economic interest will of course have to be analysed, but the possible reduction of LOS and the use of blood transfusions, will potentially lead to major savings when compared with the cost of this technique. This study will use a hyperbaric chamber that is already marketed, licensed, and used in other pathologies.

In conclusion, we are anticipating that this study will clarify the strength of evidence, either supporting or refuting indication for the use of HBOT in the treatment of VOC-SCD.



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**Contributors:** JS has the primary responsibility for the final content. Study concept and design: JS, JaS, TM, PL, CC and RP. Draft of manuscript and statistical analysis: JS, JaS, TM, PL, CC, KS, RP and SB. Revision of manuscript: all authors, notably HBOT-SCD study group, read and approved the manuscript for final publication. JS is the PI coordinator and the PI of Geneva Center; GC and PC are respectively PI for Lyon and Toulouse. JS is the guarantor; he accepts full responsibility for the finished work and/or the conduct of the study, had access to the data, and controlled the decision to publish.

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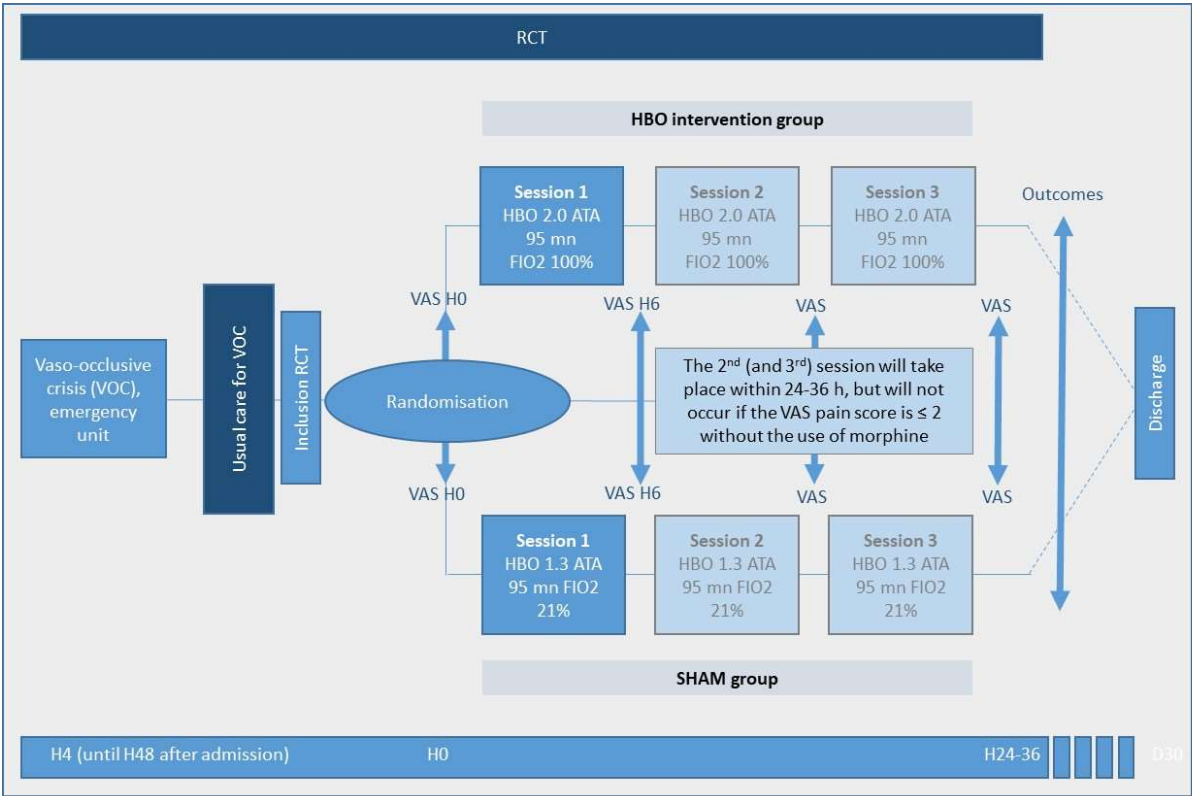
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Figure 1: Schematic diagram of the Randomised Controlled Trial (RCT)

Figure 2: Representation of procedures for intervention (A) and sham (B).

Intervention (A): 10 min of compression to 2.0 ATA, followed by 3 cycles of 20 minutes of oxygen at pressure with 5 minutes in air between cycles, followed by 10 minutes of decompression (total of 90 mn)

Sham (B): 5 min compression period followed by 80 min at 1.3 ATA with air, then a decompression period of 5 min (total of 90 mn)



VAS: visual analogue scale; VOC: Vaso-occlusive crisis

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