Cerebral venous thrombosis in adult patients with acute lymphoblastic leukemia or lymphoblastic lymphoma during induction chemotherapy with l-asparaginase: The GRAALL experience

Marie-Anne Couturier, Françoise Huguet, Patrice Chevallier, Felipe Suarez, Xavier Thomas, Martine Escoffre-Barbe, Victoria Cacheux, Jean-Michel Pignon, Caroline Bonmati, Laurence Sanhes, et al.

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MA Couturier* (1); F Huguet (2); P Chevallier (3); F Suarez (4); X Thomas (5); M Escoffre-Barbe (6); V Cacheux (7); JM Pignon (8); C Bonmati (9); L Sanhes (10); P Bories (11); E Daguindau (12); V Dorvaux (13); O Reman (14); J Frayfer (15); C Orvain (16); V Lhéritier (5); N Ifrah (16); H Dombret (17); M Hunault-Berger (18); A Tanguy-Schmidt (18).

(1) Hématologie, Hôpital Morvan, CHU Brest; (2) Hématologie, Hôpital Purpan, CHU Toulouse; (3) Hématologie, CHU Nantes; (4) Hématologie, Hôpital Necker, AP-HP; (5) Hématologie, Hôpital Edouard Herriot, CHU Lyon; (6) Hématologie, CHU Rennes; (7) Hématologie, CHU Clermont-Ferrand; (8) Hématologie, CH Dunkerque; (9) Hématologie, CHU Nancy; (10) Hématologie, CH Perpignan; (11) Hématologie, CHU Strasbourg; (12) Hématologie, CHU Besançon; (13) Hématologie, CHR Metz; (14) Hématologie, CHU Caen; (15) Hématologie, CHU Meaux; (16) Maladies du sang, CHU Angers; (17) Hématologie, Hôpital Saint-Louis, AP-HP; (18) Maladies du sang/INSERM U892 PRES LUNAM, CHU Angers, France.

Corresponding author: Aline Tanguy-Schmidt

Maladies du sang, INSERM U892 PRES LUNAM, CHU Angers 49000 FRANCE

AlSchmidt@chu-angers.fr, Tel: 33 2 41 35 44 75, Fax: 33 2 41 35 45 82

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Abstract

Central nervous system (CNS) thrombotic events are a well-known complication of acute lymphoblastic leukemia (ALL) induction therapy, especially with treatments including L-asparaginase (L-ASP). Data on risk factors and clinical evolution is still lacking in adult patients. We report on the clinical evolution of 22 CNS venous thrombosis cases occurring in 708 adults treated for ALL or lymphoblastic lymphoma (LL) with the Group for Research on Adult Acute Lymphoblastic Leukemia (GRAALL)-induction protocol, which included eight L-ASP (6000 IU/m²) infusions. The prevalence of CNS thrombosis was 3.1%. CNS thrombosis occurred after a median of 18 days (range: 11-31) when patients had received a median of three L-ASP injections (range: 2-7). Patients with CNS thrombosis exhibited a median antithrombin (AT) nadir of 47.5% (range: 36%-67%) at Day 17 (range: D3-D28), and 95% of them exhibited AT levels lower than 60%. There were no evident increase in hereditary thrombotic risk factors prevalence, and thrombosis occurred despite heparin prophylaxis which was performed in 90% of patients. Acquired AT deficiency was frequently detected in patients with L-ASP-based therapy, and patients with CNS thrombosis received AT prophylaxis (45%) less frequently than patients without CNS thrombosis (83%), $p = .0002$). CNS thrombosis was lethal in 5% of patients, while 20% had persistent sequelae. One patient received all planned L-ASP infusions without recurrence of CNS thrombotic whereas L-ASP injections were discontinued in 20 patients during the management of thrombosis without a significant impact on overall survival ($p = .4$).

Introduction

Venous thrombosis is a well-known complication of induction chemotherapy for acute lymphoblastic leukemia (ALL).[1-4] Thrombosis occurred during induction therapy at a rate of 4.8% in 1280 children included in 17 prospective studies, and of 5.9% in 323 adults included in 13 prospective studies.[2,3] Central nervous system (CNS) thrombosis is the most tragic thrombotic event (TE). Although its occurrence during ALL induction therapy is well-documented, little data is available regarding its clinical presentation, risk factors, and specific evolution.[4] L-asparaginase (L-ASP) is one of the major drugs used in ALL treatment.[5-7] This enzyme induces a relative deficiency in asparagine, which is essential for leukemic cell growth, thus leading to the death of human lymphoblasts. Because this drug inhibits the hepatic synthesis of L-asparagine-dependent hemostatic proteins, particularly plasma antithrombin (AT)[8,9], venous thrombosis, is highly associated with the administration of L-ASP during induction therapy in both children[10,11] and adults.[12] We therefore sought to explore this serious complication, and to analyze the impact of L-ASP on its incidence by reviewing every case of CNS venous thrombosis that occurred during induction chemotherapy of adult patients with ALL or lymphoblastic lymphoma (LL) included in the LL03[13], GRAALL 2003[14], and 2005[15] trials, with special attention to concomitant drugs and events.

Design and Methods

Patients

The occurrence of CNS thrombosis in patients included from April 2004 to July 2011 in the GRAALL 2003, GRAALL 2005, and LL03 protocols was prospectively reported to the GRAALL data management center as serious adverse events. Data from every patient with CNS thrombosis was reviewed retrospectively including risk factor, clinical presentation, coagulation data, thrombosis prophylaxis, and treatment. Clinical characteristics of patients with CNS thrombosis were compared to those without CNS thrombosis included in the GRAALL 2003 (n=225), GRAALL 2005 (n=347), and LL03 protocols (n=136). Compliance to thrombosis prophylaxis recommendations were not recorded and general or hereditary thrombosis risk factor were not systematically assessed and can not therefore be analyzed for patients without thrombosis. Patients without CNS thrombosis but with thrombosis occurring at other sites during induction were not included in the control group.

Chemotherapy regimen

The GRAALL 2003 evaluated the feasibility of a pediatric-based treatment in young adults (18-59 years old) with Philadelphia chromosome (Ph1)-negative ALL in a multicenter, prospective, phase II trial. GRAALL 2005 is a randomized trial evaluating the impact of high-dose cyclophosphamide and rituximab included in the induction therapy in patients with CD20+ ALL. The LL03 study was a multicenter, phase II trial evaluating the safety and effectiveness of intensive chemotherapy as used for the treatment of ALL in young adult patients with LL. In these three studies, induction therapy was very similar. All patients received a prephase with prednisone (60 mg/m²/day) for 7 to 10 days, then induction therapy combining daunorubicin (50 mg/m²/injection on Days 1-3, then 30 mg/m² on Days 15 and 16), weekly intravenous vincristin (2 mg/injection), eight L-ASP (6000 UI/m²/injection) intravenous injections and 2 weeks of daily oral prednisone (60 mg/m²/day). Cyclophosphamide (CPM) was administered at 750 mg/m² on Day 1, then at 300 mg/m²/12h on Days 15-17 in the GRALL 2003 and in the intensified arm of the GRAALL 2005 studies ("HyperC arm"); at 750 mg/m² on Day 15 in the standard arm of the GRAALL 2005 study and at 500 mg/m²/12h on Days 15 and 16 in the LL03 study. In order to prevent interaction with other drugs, L-ASP was administrated intravenously (IV) on Days 8, 10, and 12 during the first part of induction, and then on Days 20, 22, 24, 26, and 28, after the reintroduction of daunorubicin, cyclophosphamide and vincristin. In accordance with standard recommendations, *Escherichia coli* L-ASP (Kidrolase®) was switched for *Erwinia chrysanthemi* L-ASP (Erwiniase®) 12000 UI/m²/IV injections in case of allergic reactions. CNS prophylaxis included methotrexate 15 mg intra-thecal injections during the prephase and triple intra-thecal injections (methotrexate 15 mg, aracytine 40 mg and depomedrol 40 mg) at days 1 and 8.

Patients in complete remission (CR) received a consolidation course in quick succession of six alternating blocks with high-dose methotrexate, high-dose aracytine, and cyclophosphamide in conjunction with low doses of other cytotoxic drugs. In the three protocols, all patients in persistent CR for whom allogeneic stem cell transplantation was not indicated in first CR received a standard or HyperC late intensification with the same drugs used during the induction course, followed by a repetition of three consolidation blocks. In addition to the neuro-meningeal prophylaxis during prephase and induction, patients received 3 additional intra-thecal injections and cerebral irradiation (18Gy) at the end of consolidation (GRAALL 2003 and GRAALL 2005), or at the end of late intensification (LL03). Then, patients proceeded to maintenance therapy which included monthly vincristin infusions and long-term oral methotrexate and 6-mercaptopurine administration.

Thrombosis prophylaxis and diagnosis

Recommendations for thrombosis prophylaxis were based on CAPELAL study results[16]: Antithrombin (AT) and fibrinogen levels were evaluated prospectively before each L-ASP infusion (Days 8, 10, and 12, and then every other day from Day 20 to 28). No other coagulation factor was systematically evaluated. Fresh frozen plasma or fibrinogen concentrates were recommended if fibrinogen levels fell below 0.5 g/L, platelet transfusion support was recommended for platelets below 20.10^9/L and AT concentrate substitution therapy (Aclotine®, 25 U/kg) was recommended in order to maintain AT levels above 60%. AT levels were re-evaluated the day after AT infusion. L-ASP were re-administrated after correcting the acquired deficiency in AT. Recommendations also included the use of unfractionated heparin (UFH) at 100 UI/Kg/day in continuous infusion or low molecular weight heparin (LMWH) at prophylactic doses in subcutaneous injection during induction, without particular monitoring. Heparin prophylaxis was interrupted when platelets were below 20.10^9/L and resumed after platelet transfusion.

Cerebral imaging was performed in case of otherwise inexplicable neurological symptoms, including headaches: CNS venous thrombosis was confirmed by computed tomography [CT] or magnetic resonance imaging [MRI] showing the sino-venous thrombus. Cerebral angiography was performed in case of persisting doubt. The presence and nature of parenchymal lesion was reported, such as hemorrhagic infarcts or brain oedema. The localisation of thrombosis was classified as occurring in lateral, longitudinal, sagittal, transversal or cortical veins.

Statistical analysis

Statistical analyses were performed with the use of the GraphPad PRISM version 6.0e program. For univariate analysis, we performed Mann-Whitney nonparametric tests in case of quantitative variables, and a CHI^2 test to compare percentages. To compare survival data, we performed a log-rank test. A p-value <0.05 was considered statistically significant.

Results

Prevalence

From April 2004 to July 2011, 572 patients were included in the two GRAALL protocols, as well as 136 patients in the LL03 protocol. CNS thrombosis was reported in 22 of these 708 patients (3.1%), in 2.8% of ALL patients (16/572) and in 4.4% of LL patients (6/136). In patients included in the GRAALL 2005 study, the prevalence of thrombotic events during induction was 9.5% (33/347): 36% were CNS thromboses, 30% lower limb thromboses, 18% upper limb thromboses, 9% pulmonary embolisms, 3% portal system thrombosis, and only 3% arterial thromboses.

Patient characteristics

CNS thrombosis occurred in two patients prior to L-ASP infusions (2/708) and in 20/708 patients during or after L-ASP during induction therapy, i-e respectively 0.28% versus 2.8% (p= .0001). The 2 patients with CNS thrombosis occurring before L-ASP infusions are not included in the subsequent analysis but are detailed at the end of the results section. Of the 20 patients with CNS thrombosis occurring during L-ASP treatment, the median age was 29 (range: 18-50 years), and 75% were male (Table I). CNS thrombosis was more frequent in T cell-ALL (13/262, 5%) than in B cell-ALL (7/394, 1.8%**)** (p= .034). Poor-risk prognostic factors, such as steroid-resistance, early chemo-resistance, as well as failure to achieve complete remission, were similar between these 20 patients and those without CNS thrombosis (data not shown). There was no significant association between CNS thrombosis and hyperleucocytosis (> 30.10^9/L) or blastic CNS involvement (Table I). The distribution of karyotype abnormalities was roughly similar to that of patients without CNS thrombosis (data not shown); abnormalities were observed in more than half of the CNS thrombosis patients (12/20, 60%), including four poor cytogenetics with one t(1;14), one t(1;19), and two complex karyotypes. None of the patients with CNS thrombosis had allergic reaction to Escherichia coli L-ASP.

CNS thrombosis

CNS venous thrombosis was identified based on clinical signs and was confirmed by radiological imaging: cerebral CT in 8 patients, cerebral MRI in 5 patients, and both in 9 patients. The most common sites of CNS thrombosis included the longitudinal sinus (11), cortical vein (11), lateral sinus (5), sagittal sinus (4), and transverse (2). Intra-cerebral hemorrhage was associated with thrombosis in 7 patients (35%). At diagnosis, CNS clinical symptoms were headaches (8/20, 40%), which were isolated in 2 patients (10%), seizures (13/20, 65%), which were isolated in 2 patients (10%), or neurological changes (15/20, 75%) including motor deficits, paresthesias, or visual/ophthalmic signs. Half of the patients exhibited prodromal symptoms (10/20, 50%), consisting almost exclusively of headaches (9/10) (Table II). In this cohort, four patients (20%) experienced other venous thrombotic events during induction, as detailed in Table II. One patient died 5 days after CNS thrombosis due to cerebral hemorrhage associated with thrombosis. None of the other CNS thrombotic events were lethal, but 4 patients (20%) experienced neurological sequelae, recorded as persistent headaches (1 patient), epilepsy (1 patient), and motor or cognitive deficiency (2 patients).

Thrombotic risk factors

CNS thrombosis occurred from Day 11 through Day 31 (median: Day 18) after starting induction therapy, after a median of 3 L-ASP injections (range: 2-7) (Tables I and III). AT levels fell under the 60% threshold in almost all patients (95%), with levels falling below 50% in 15/20 (75%) patients. The median AT nadir was 47.5% (range: 36%-67%) and occurred at Day 17 (range: D3-D28); this was not significantly different with the median AT nadir of patients without CNS thrombosis (51%, p= . 25). At baseline, median platelet count was 91.10^9/L (range, 37-465) and median fibrinogen level was 3.3 g/L (range, 1.19-4.81), while when CNS thrombosis occurred, median platelet count was 101.10^9/L (range, 19-380) (p= .59) and fibrinogen level was 1.5 g/L (range, 0.8-7) (p= .1) which did not differ significantly from patients without CNS thrombosis (p= 0,9 and p=0,15, Table I). Despite recommendations, prophylactic AT infusions was only performed in 9 patients with CNS thrombosis (45%), whereas 83.2% of patients without CNS thrombosis received prophylactic AT concentrates (p= .0002) (Tables I and III). Fibrinogen concentrate was infused only once, at Day 9, in a patient whose CNS thrombosis occurred at Day 19 and Fresh-frozen plasma was infused in 2 patients, 3 and

14 days before the occurrence of CNS thrombosis. Prophylactic anticoagulation does not seem to protect from CNS thrombosis, as it was performed in 90% of patients with CNS thrombosis and surprisingly, in only 64.9% of the patients without CNS thrombosis (Tables I and III).

Intra-thecal injections were administered prior to CNS thrombosis in all but 1 patient. CNS thrombosis occurred after a median of 3 intra-thecal injections (range: 0-3), after a median of 11 days (range: 4-16) following the most recently received IT (Table II). In addition, the total number of intra-thecal injections administrated was higher in patients with CNS thrombosis than in those without (3 *vs* 2, $p< .0001$). Because the GRAALL protocols evaluated CPM dose intensification, we analyzed CPM doses as a potential thrombosis risk factor. The proportion of patients receiving high-dose CPM was similar in patients with and without CNS thrombosis (50% *vs* 45%) (Data not shown). The number of L-ASP infusions during induction was lower in patients with CNS thrombosis than in patients without thrombosis as the interruption of L-ASP infusions was recommended after CNS thrombosis.

None of the 20 patients reported a personal or familial history of thrombosis. Hereditary thrombotic risk factors (protein S deficiency, protein C deficiency, activated protein C, lupus anticoagulant) were evaluated at ALL/LL diagnosis according to the GRAALL2005 recommendations or at the time of thrombosis if not performed before. Two patients (10%) had been diagnosed as having hereditary protein S deficiency and heterozygous factor V Leiden mutations were found in 3 patients (15%). There were no prothrombin G20210A gene mutations. One patient was diagnosed as having, simultaneously, hereditary protein S deficiency, heterozygous factor V Leiden mutation, and a lupus anticoagulant. None of our patients were taking combined oral contraceptives, as this medication was stopped before induction therapy in one female patient, and progestin derivatives were administrated in all five female patients.

CNS thrombosis management

Of the 20 patients, anticoagulant treatment was not initiated in 1 patient presenting cerebral hemorrhagic lesions, rapidly resulting in death and 19 were initially treated with UFH or LMWH. The therapeutic goal level was a target APTT (activated partial thromboplastin time) between 1.5 and 2. There were no bleeding complications. Therapeutic anticoagulation was maintained for 6 months after thrombosis, or longer in case of recurrent thrombotic events. During anticoagulation, platelet levels were maintained above 50.10^9/L with platelet transfusion support. Concomitant surgical sinus thrombectomy was performed in 1 patient. No recurrence of CNS thrombosis occurred but despite therapeutic anticoagulation, recurrent thrombotic events occurred in 4 patients (20%) with 1 pulmonary embolism (PE), 1 lower limb thrombosis, and 2 concomitant pulmonary and limb thrombosis (table II). AT concentrates at curative doses were administered in 10 patients (50%), specifically in 80% and 33.3% of patients with and without neurological sequelae, respectively.

Impact on survival

The complete remission (CR) rate was 90% as 1 of the 20 patients died during induction because of intra-cerebral hemorrhagic complications of the CNS thrombosis and 1 failed to achieved CR at the end of induction. This is similar to the 90% CR rate obtained in patients without CNS thrombosis included in the GRAALL 2005 study.[15] Relapse occurred in 8 patients with CNS thrombosis. CNS thrombosis was associated with a non-significant reduced 3-year overall survival (55% *vs*. 75.9%, p=0.4) and 3-year disease-free survival (61.3% *vs*. 79.5%, p=0.18) (Table I, Figure 1).

CNS thrombosis occurring before L-ASP infusion

CNS thrombosis occurred prior to L-ASP infusion in 2 patients, at Day -5 and Day 2 from induction chemotherapy, respectively in a 22-year-old woman with B-ALL with normal karyotype who received oral combined contraceptive prior to ALL therapy and in an obese 28-year-old man with T-LL with normal karyotype. Both of them received thrombotic prophylaxis with LMWH. In these 2 cases, platelet count levels were stable. But in the first case, fibrinogen level was 3.3 g/L at baseline and decreased to 1.3 g/l during steroids when the diagnosis of CNS thrombosis was made, without FFP or fibrinogen supplementation. In the second case, fibrinogen levels were stable > 3 g/L. These 2 patients experienced headaches, without any other neurological modifications. CT brain showed thrombosis in the intern jugular vein, which extended in both sigmoid and lateral sinuses, without hemorrhagic intra-cerebral complications. The 2 patients received curative anticoagulation with UFH, with a complete resolution of thrombosis and without long-term sequelae. According to the physician's choice, L-ASP was not initiated in the patient with CNS thrombosis occurring at Day 2, despite a complete resolution of symptoms. In the remaining patient, whose CNS thrombosis occurred during the steroid prephase despite a curative anticoagulation with UFH started since Day -12, all eight L-ASP infusions were administered, even though 2 others thrombotic events (sub-clavicular thrombosis and CVC-related thrombosis) were experienced without any CNS thrombotic recurrence. To date, these 2 patients are still alive and in CR with OS of 71.8 and 80.9 months respectively.

Discussion

This study involving adult patients treated for ALL or LL aimed to evaluate the prevalence of cerebral venous thrombosis during induction courses with L-ASP, analyze clinical evolution, and discuss the management of asparaginase-associated CNS thrombosis. We report here a unique cohort of 20 CNS thrombosis occurring in adult ALL/LL during induction with L-ASP. However, thrombotic risk factors, especially hereditary thrombotic risk factors, were not prospectively evaluated in all patients without CNS thrombosis, limiting the power of this study to detect significant prothrombotic association.

In the GRAALL experience, CNS thrombosis occurred in 3.1% of adult patients during ALL induction therapy. This is higher than the 1 to 2% incidence rate reported in children, [2,17,18,19] but close to the 2.3% incidence observed during induction in 214 adults from the CAPELAL trial[16] or the 4.3% incidence recorded in 47 adults patients treated at the Dana Farber Cancer Institute.[20] Such a significant association between increased age and L-ASP-induced thrombosis has been previously reported.[20,21] Grace *et al* recently reported that age was the only significant predictor factor of venous thrombotic event, in multivariate analysis.[20]

Symptoms of CNS thrombosis (headaches, focal motor or sensory neurological deficiency, convulsive seizure, or loss of consciousness) were similar to those reported in patients without ALL.[4] However, in our cohort, 50% of patients exhibited prodromal signs--almost exclusively headaches--and headaches were the only sign of thrombosis for 10% of them. The same frequency (3/20) was reported in a recent cohort of children.[19] Therefore, CT scan, MRI, or MR angiography, depending on the physician's preference, should be performed in case of persistent headache without obvious explanation, especially if the headache is not modifified by a shift in posture and worsens with time.

In our cohort, the prevalence of genetic prothrombotic conditions was not higher than in healthy individuals,[22,23] contrary to the increased prevalence of genetic prothrombotic conditions observed in CNS thrombosis cases without ALL.[24,25]

None of the acquired, well-established risk factors for CNS thrombosis in the general population were present in our 20 cases, such as combined oral contraceptive use, obstetrical delivery, head injury, otitis, mastoiditis, meningitis or inflammatory disease.[4] An increased thrombotic risk has already been reported in T-ALL cases.[26] In our cohort, T-cell phenotype was observed in 65% patients in the CNS thrombosis group while it represented only 43% of patients without CNS thrombosis. This higher incidence of T-cell phenotype and male predominance is related to the inclusion of LL patients in our cohort, which were more frequently male with T-cell phenotype. In our study, data regarding platelet counts at diagnosis suggest that platelets are not a major factor for the occurrence of CNS thrombosis during ALL induction with L-ASP, as well as hyperleucocytosis, as shown in an Italian study where leucocytes or platelets were not found to increase the risk of thrombosis.[27] Lumbar puncture is a known risk factor for CNS thrombosis in the general population, as low cerebrospinal fluid pressure following lumbar puncture causes a downward shift of the brain, with traction on cortical veins and sinus walls that may induce thrombosis.[4] In our study, the 11-day delay between the last intra-thecal injection and CNS thrombosis occurrence appears too long to consider intra-thecal injection as an acquired thrombotic risk factor. However, corticosteroid therapy during the beginning of induction might increase the risk of thrombosis as previously reported.[2,3,28]

Our findings suggest that L-ASP infusion, AT level, and CNS thrombosis are closely related. Indeed, the AT nadir of 47.5% and CNS thrombosis occurred after a median of 17 and 18 days following the start of induction therapy, respectively, i.e., 4 days after the third L-ASP infusion. In addition, 83% of patients for whom data was available displayed AT <60% the day or the day before thrombosis. Likewise, in the CAPELAL study, thrombotic events occurred after a median of four (range: 1-6) L-ASP injections, with a median AT level of 53% at the time of thrombosis.[16] In general, AT dosages were performed by centers prior to each L-ASP infusion (Days 8, 10, 12, and then every two days from Day 20 to 28), as recommended but were not closely monitored in the time period (Days 13 to 20) without L-ASP infusions. The decreased AT monitoring, and resulting decreased AT substitution, might account for the occurrence of CNS thrombosis around Day 18. In the CAPELAL study, prophylactic replacement therapy with AT concentrates was associated with decreased thrombosis rates in ALL patients treated with L-ASP[16] (4.8% *vs* 12.2% in patients without AT prophylactic replacement, p= .04). The PAARKA trial in children showed safety and a trend towards efficacy with AT concentrates in the prevention of thrombosis but the study was not powered to answer this question.[29] In our study, AT prophylactic replacement was performed less frequently (45%) in patients with CNS thrombosis than in patients without CNS thrombosis (83.2%) (p= .0002) suggesting that AT prophylactic replacement might decrease the occurrence of CNS thrombosis and we highly recommend AT substitution for the future GRAALL2014 protocol.

In line with the CAPELAL study results,[16] 90% of our patients underwent thrombotic prophylaxis using continuous UFH infusion at 100 IU/Kg/day, which did not prevent CNS thrombosis. Prophylactic LMWH with AT infusion might be a better choice to prevent thrombosis, than a single prophylactic anticoagulation. Indeed, concomitant treatment with LMWH and AT was associated with reduced thrombosis rates (0/41) in children with ALL treated with L-ASP, as compared to a historical control cohort treated with AT alone (9/71, 12.7%) (p= .02)[30].

Finally, the optimal management of CNS thrombosis during ALL induction with L-ASP is not well-defined. In our cohort, no increased cerebral haemorrhages were reported after heparin treatment with platelet transfusion support. The 20% and 5% poor neurological recovery and death cases, respectively, were identical to those reported in the scientific literature in relation with CNS thrombosis in the general population.[4] However, no data is available for the use of AT replacement after thrombosis. Re-exposure to L-ASP after a venous thrombosis occurrence is subject to debate as L-ASP is a major drug in ALL therapy. Indeed, in pediatric and adults patients, early L-ASP discontinuation due to thrombosis was associated with reduced EFS rates.[16,20]

In our study, 1 patient with early CNS thrombosis received 8 L-ASP infusions without CNS thrombotic recurrence. Data about L-ASP re-exposure in this context are conflicting. One Indian child was re-exposed without thrombotic recurrence.[31] In the UKALL study, 38 children with thrombosis, including 10 CNS thrombosis cases, were re-exposed to L-ASP with concurrent heparin administration with neither recurrent thrombosis nor bleeding complications observed.[17] However in the DFCI experience, among 33 patients restarted on L-ASP after venous thrombosis, 33% (11/33) experienced a recurrence of thrombosis, more frequently in adults than in children (47% *vs* 17%, $p = .07$). There was no association between initial site of thrombosis and risk of clot recurrence.[20] Therefore, re-exposure to L-ASP can probably be performed in the setting of carefully monitored AT and anticoagulation with heparin or LMWH, provided that venous flow recovery is observed on CT scan or angioMRI but further studies are warranted to clarify this issue.

In conclusion, CNS thrombosis is a significant complication of ALL therapy in adults. Our findings suggest a predilection for this event in young male patients with T-ALL and LL treated as ALL - attributable neither to IT nor to thrombophilia - and an association with low-level AT that strengthens the causal link with L-ASP. However, L-ASP is a major drug in ALL therapy and the discontinuation of this drug might be detrimental to patients. AT concentrate replacement, heparin-based antithrombotic prophylaxis and L-ASP re-exposure still needs to be evaluated.

Author contributions

MAC collected the data, MAC, MH and ATS performed the research, analyzed data and wrote the paper, FH, PC, FS, XT, ME, VC, JMP, CB, LS, PB, ED, VD, OR and JF included patients, VL, NI, HD and MH designed the research study, HD and NI revised the manuscript.

Conflict of interest

None of the authors has any conflicts of interest to declare.

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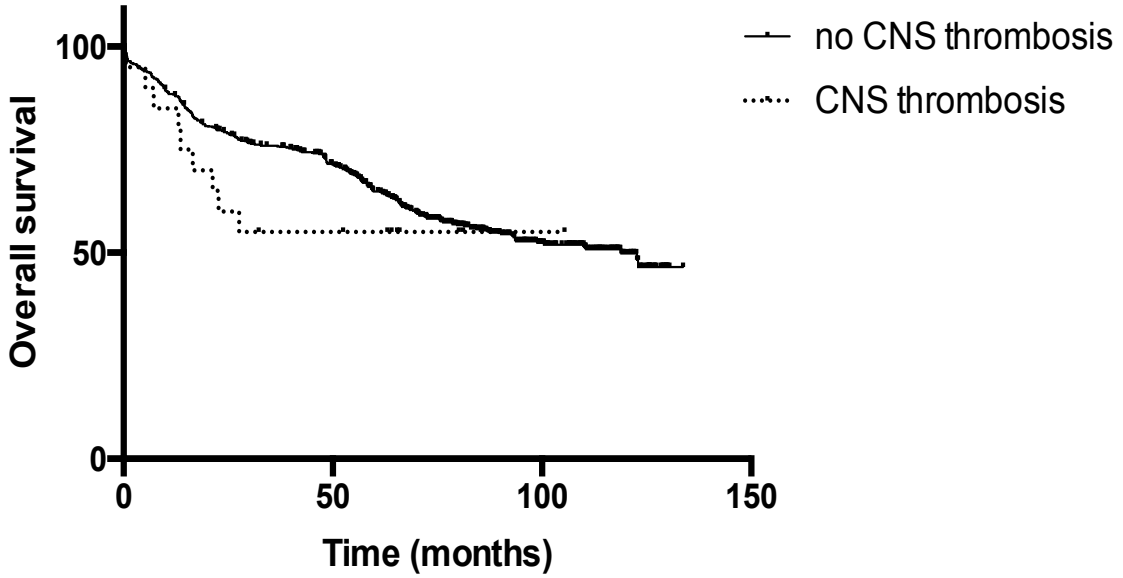

Figure 1: Kaplan-Meier overall survival curves in patients with and without central nervous system venous (CNS) thrombosis.

	Number of patients (available data)	CNS-Thrombosis n=20	No CNS-thrombosis n=686	*p*
Median of age (years)	706	29	33	.447
Number of male patients	706	15 (75%)	434 (63.3%)	.283
T-cell phenotype	705	13 (65%)	296 (43.2%)	.053
WBC > 30.10^9/L at diagnosis	706	5 (25%)	188 (27.5%)	.806
CNS involvement	706	1 (5%)	38 (5.9%)	.917
BMI	563	23	23 (n=540)	.839
Protocols type				
- GRAALL2003	225	2	223	
- GRAALL2005	346	13	333	.104
- LL03	135	5	130	
Median number of L-ASP infusions	696	3	7	**< .001**
Median number of IT	350	3	2	**< .001**
Platelet level at diagnosis (10^9/L)	482	91	123.5	.897
Fibrinogen level at diagnosis (g/L)	672	3,3	4	.149
Median AT nadir	327	47.5	51	.25
Median number of FFP	292	0	0	.83
Median number of fibrinogen infusions	318	0	0	.848
AT prophylaxis	353	9 (45%)	277 (83.2%)	**.0002**
Heparin (LMWH/UFH) prophylaxis	268	18 (90%)	161 (64.9%)	**.025**
3-year overall survival (%)	706	55	75.9	.399
3-years disease-free survival (%)	611	61.3	79.5	.182

Table I: Characteristics of the 706 patients with and without CNS thrombosis.

WBC: white blood cell count, BMI: body mass index, CNS: central nervous system, L-ASP: L-asparaginase, IT: intra-thecal injection, AT: antithrombin, FFP: fresh frozen plasma, LMWH: low molecular weight heparin, UFH: unfractionated heparin, OS: overall survival, DFS: disease-free survival.

Pt	Prodromal symptoms	Thrombosis symptoms	Site of thrombosis	Hemorrhagic associated	Other thrombosis	AT Curative	Curative anticoagulant	T-Evolution
1	no	deficit, seizure	lateral, longitudinal	no	PE, leg T	no	UFH	no
2	headaches	deficit, seizure	cortical veins	yes	leg T	yes	LMWH	sequelae
3	headaches	deficit	lateral, longitudinal	yes	no	yes	UFH	sequelae
4	no	deficit, seizure	sagittal, cortical	no	no	no	UFH	no
5	no	deficit, seizure	cortical, longitudinal	no	no	no	LMWH	no
6	no	deficit, seizure	cortical veins	no	no	yes	LMWH	sequelae
7	no	headaches, deficit	lateral, longitudinal, cortical	no	no	yes	UFH then LMWH	no
8	headaches	deficit, OPH tb	lateral, longitudinal	yes	no	no	UFH then LMWH	no
9	headaches	headaches	sagittal sup	no	PE	yes	UFH	no
10	headaches	headaches, deficit, seizure	lateral, longitudinal	no	no	no	UFH	no
11	headaches	headaches	cortical veins	no	no	yes	UFH	no
12	no	deficit	longitudinal sup	no	no	yes	UFH then LMWH	no
13	no	seizure	cortical, longitudinal	no	no	no	UFH then LMWH	no
14	no	deficit, seizure	cortical, longitudinal	yes	no	yes	UFH	no
15	headaches	headaches, deficit, seizure	longitudinal sup, transverse	yes	no	yes	UFH then LMWH	sequelae
16	UL paresthesia	headaches, deficit, seizure	sagittal sup, longitudinal	yes	PE, leg T	no	UFH	no
17	no	headaches, OPH tb	sagittal sup, transverse	no	no	no	UFH then LMWH	no
18	headaches	headaches, seizure	cortical veins	no	no	no	UFH then LMWH	no
19	no	deficit, seizure	cortical veins	yes	no	no	-	death
20	headaches	seizure	cortical veins	no	no	no	UFH	no

Table II: Prodromal symptoms, diagnosis, treatment, and evolution of CNS thrombosis

PE: pulmonary embolism, T: thrombosis, UFH: unfractionated continuous infusion heparin, LMWH: low molecular weight heparin., UL paresthesia: upper limb paresthesia, OPH tb: ophtalmic troubles, AT Curative : curative antithrombin infusion, T-Evolution: evolution of thrombosis (sequelae).

Pt	Age	Sex	Phenotype	High dose CPM	CNS+	WBC > 30 G/L	BMI	Day of CNS T	Nb of L-ASP before T	Nb of IT before T	Heparin prophylaxis	Day of AT Nadir (AT Nadir level)	AT level d-1/d0 before T	AT infusion prophylaxis
1	43	M	T-ALL	no	no	no	27.4	17	3	2	UFH	16 (45%)	45%	yes
2	42	M	B-ALL	no	no	no	28.7	19	3	3	no	18 (58%)	58%	no
3	46	F	B-ALL	yes	no	no	20.7	22	4	3	UFH	24 (36%)	43%	no
4	18	M	T-LL	yes	no	no	29.5	14	2	3	no	17 (55%)	65%	no
5	27	M	T-LL	no	no	no	23.6	21	4	3	UFH	12 (39%)	N/A	yes
6	38	F	T-ALL	yes	no	yes	20.4	25	6	2	UFH	25 (50%)	50%	no
7	24	M	T-LL	no	no	no	21	16	3	3	UFH	26 (47%)	N/A	no
8	26	M	B-ALL	yes	no	yes	21.9	31	7	3	UFH	22 (43%)	N/A	yes
9	19	F	T-LL	no	no	no	25.5	20	4	2	UFH	13 (56%)	N/A	yes
10	41	M	B-ALL	no	no	yes	22	18	3	3	UFH	14 (67%)	N/A	no
11	42	F	B-ALL	yes	no	yes	21.9	24	6	3	UFH	28 (41%)	56%	yes
12	27	M	T-ALL	yes	no	no	20.2	18	3	3	UFH	18 (42%)	41%	no
13	22	M	B-ALL	no	no	yes	23.9	21	4	3	UFH	21 (52%)	52%	yes
14	37	F	T-ALL	no	no	no	20.4	17	3	3	UFH	18 (43%)	N/A	no
15	40	M	T-ALL	yes	no	no	24.9	11	3	1	UFH	13 (47%)	58%	yes
16	23	M	T-ALL	yes	yes	yes	23.3	14	3	3	UFH	16 (50%)	78%	yes
17	20	M	T-ALL	yes	no	no	18.7	19	3	3	UFH	17 (48%)	51%	yes
18	23	M	T-ALL	yes	yes	yes	23	17	2	4	UFH	20 (59%)	N/A	no
19	50	M	T-ALL	no	no	no	24.9	12	3	3	UFH	12 (49%)	N/A	yes
20	31	M	B-ALL	no	no	no	23.3	15	3	2	UFH	3 (46%)	49%	no

Table III: Characteristics of patients and AT evolution

Pt: patient, CPM: cyclophosphamide, CNS +: blastic CNS involvement, BMI: body mass index, T: thrombosis, Nb: number, IT: intrathecal injection, UFH: unfractionated continuous infusion heparin, N/A: data not available.