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How I manage cerebral vasculopathy in children with sickle cell disease

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Summary

Sickle cell disease induces specific brain alterations that involve both the macrocirculation and the microcirculation. The main overt neurovascular complications in children are infarctive stroke, transient ischaemic attack and cerebral haemorrhage. Silent cerebral infarction, cognitive dysfunction and recurrent headache are also common. Cerebrovascular disease selectively affects children with the HbSS or HbS-β^0 genotypes (i.e. sickle cell anaemia). The incidence of stroke peaks between 2 and 5 years of age (1·02/100 patient-years) and increases with the severity of the anaemia. Most strokes can be prevented by annual transcranial Doppler screening from 2 to 16 years of age and providing chronic blood transfusion when this investigation shows elevated blood-flow velocities. The role for hydroxycarbamide in children with abnormal transcranial Doppler findings is under investigation. After a stroke, chronic blood transfusion is very strongly recommended, unless haematopoietic stem cell transplantation can be performed. Routine magnetic resonance imaging shows that more than one-third of children have silent cerebral infarction, which is associated with cognitive impairments. Screening for silent infarcts seems legitimate, since their presence may lead to supportive treatments. The role for more aggressive interventions such as hydroxycarbamide or chronic blood transfusion is debated.

Keywords: sickle cell anaemia, stroke, transcranial doppler, hydroxycarbamide transfusion.

Sickle cell disease (SCD), one of the most common genetic conditions, is a specific abnormality in the haemoglobin (Hb) molecule: the glutamic acid in the 6th position of the haemoglobin beta chain is replaced by valine, producing HbS instead of the normal HbA. The HbS mutation may be in a homozygous status, or combined on the other allele with another mutation in the β-globin gene (HBB). The sickle cell disease syndromes include the severe forms of the disease, the HbSS and HbS- β^0 genotypes [called also genotypes of sickle cell anaemia (SCA)], and the milder genotypes HbSC and HbS-β^+. Deoxygenated HbS molecules undergo polymerization, causing alterations in erythrocyte morphology and function, with rigid sickle-shaped cells generating impaired flow through blood vessels and oxygen delivery to tissues. These rigid erythrocytes are prone to haemolysis. Furthermore, excessive adherence of both erythrocytes and leucocytes to the vascular endothelium contributes to vaso-occlusion, most notably of the post-capillary venules. The resulting hypoxaemia can cause acute and chronic impairments in the function of all organ systems. Thus, most complications of SCD are related to small-vessel obstruction, which may involve the bones, kidneys, heart, liver, and lungs.

In the brain, however, both the large cerebral arteries and the microcirculation may show signs of occlusion or stenosis. The most commonly affected large cerebral arteries are the distal internal carotid artery (ICA), proximal middle cerebral artery (MCA) and proximal anterior cerebral artery (ACA). Overt neurovascular complications result in infarctive stroke, transient ischaemic attacks (TIAs) and cerebral haemorrhage. Silent cerebral infarction (SCI) and gradual cognitive dysfunction are also common. Whether recurrent headache should be classified as a neurovascular complication of SCD is under debate (DeBaun *et al*, 2014; Kossorotoff *et al*, 2014a).

Here, after a review of epidemiological data, we will focus on the management of cerebrovascular complications in children with SCD. We will not discuss the pathophysiology of these complications, as recent detailed reviews on this topic are available (Switzer *et al*, 2006; Connes *et al*, 2013). Multiple mechanisms are involved, including intimal proliferation, inflammation, a hypercoagulable state, vascular tone dysregulation and haemolysis. Neither the relative importance nor the timing of these mechanisms has been established.

Epidemiology

Overt stroke

A large American study assessed the rate and risk factors of overt cerebrovascular events in 4082 patients with SCD

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enrolled between 1978 and 1988 (Ohene-Frempong *et al*, 1998). Overall, the likelihood of experiencing an overt stroke before 20 years of age was 11% in children with the HbSS or HbS-β^0 genotypes. The incidence was highest before 10 years of age, with 1·02/100 patient-years between 2 and 5 years and 0·79/100 patient-years between 6 and 9 years. Infarctive stroke predominated. Intracranial haemorrhage was far less common and chiefly affected patients aged 20–29 years. Overall, the haemorrhage/infarction ratio has been estimated at 1/4 (Ohene-Frempong *et al*, 1998; Kossorotoff *et al*, 2014b). The HbSC genotype was associated with a 5-fold lower risk of overt stroke during childhood compared to the HbSS genotype. TIAs accounted for only one in ten cerebrovascular events in children (Ohene-Frempong *et al*, 1998). Unless specific treatment is given, the risk of recurrence after a first stroke is very high, at about 50% within 2 years (Powars *et al*, 1978; Pegelow *et al*, 1995).

Clinical risk factors for ischaemic stroke reported by Ohene-Frempong *et al* (1998) were a history of TIA [relative risk (RR), 56·0; 95% confidence interval (95%CI), 12·0–285], a lower steady-state Hb level (RR, 1·85 per 10 g/l decrease; 95%CI, 1·32–2·59), acute chest syndrome in the past 2 weeks (RR, 7·03; 95%CI, 1·85–26·7), a higher frequency of acute chest syndrome episodes (RR, 2·39 per event/year; 95%CI, 1·27–4·48) and higher systolic blood pressure (RR, 1·31 per 10 mmHg increase; 95%CI, 1·03–16·7). Risk factors for cerebral haemorrhage were a lower steady-state Hb level (RR, 1·61 per 10 g/l decrease; 95%CI, 1·11–2·35) and higher steady-state leucocyte count (RR, 1·94 per 5×10^9/l increase; 95%CI, 1·73–2·18).

Other studies identified additional risk factors for stroke, among which the most important is increased blood-flow velocity by transcranial Doppler (TCD) ultrasonography (Adams *et al*, 1992) (see below). Other risk factors include nocturnal hypoxaemia (Kirkham *et al*, 2001), a history of bacterial meningitis (De Montalembert *et al*, 1993), and SCI by magnetic resonance imaging (MRI) (Miller *et al*, 2001). Concomitant glucose-6-phosphate dehydrogenase deficiency was associated with an increased risk of stroke in one study (Bernaudin *et al*, 2008) but not in another (Miller *et al*, 2011). Several genetic factors have been shown to influence the risk of stroke, either by affecting total Hb and fetal Hb (HbF) levels or by influencing the inflammatory response (Adams *et al*, 1994; Styles *et al*, 2000; Sebastiani *et al*, 2005). Several studies demonstrated a protective effect of α-thalassaemia against stroke (Adams *et al*, 1994; Bernaudin *et al*, 2008).

Moyamoya syndrome

Moyamoya syndrome is defined as progressive stenosis of the distal ICAs combined with an abnormal network of collateral vessels (Scott & Smith, 2009). Moyamoya syndrome in children leads to recurrent strokes and TIAs and, sometimes, to cerebral haemorrhage. Progressive cognitive impairments may develop. A small case-series study of MRI/magnetic resonance angiography (MRA) in 30 patients with stroke (n = 23), headaches, or seizures (n = 7) showed moyamoya syndrome in 6 (20%) (Moritani *et al*, 2004). Moyamoya syndrome may be unilateral or bilateral in patients with SCA.

Silent cerebral infarction

Silent cerebral infarction is the most common neurological event in children with SCA. The SCI Transfusion trial defined SCI as visualization by brain MRI of a lesion measuring at least 3 mm along the greatest linear dimension and seen in at least two planes of T2-weighted images (axial and coronal) in children with no current focal neurological deficit consistent with the site of the lesion (Casella *et al*, 2010). In a cohort study of 217 children, regular screening showed SCI in one-third of patients by 8 years of age and 37·4% by 14 years of age (Bernaudin *et al*, 2011). In the SCI Transfusion trial, routine brain MRI in 1074 children with a mean age of 10 years showed SCI in 379 (35·3%) (DeBaun *et al*, 2014). Silent cerebral infarctions occur chiefly in the white matter of the border zones between the ACA and MCA territories, suggesting a pathophysiological role for haemodynamic variations (Debaun *et al*, 2012). DeBaun *et al* (2014) found that risk factors for SCI were male gender, lower baseline haemoglobin level, higher baseline systolic blood pressure and previous seizures. Silent cerebral infarction is associated with an increased risk of overt stroke (Miller *et al*, 2001), poor academic achievement and lower intelligence quotient (IQ) (Bernaudin *et al*, 2000). Thus, the term SCI is inappropriate, although it remains widely used.

Recurrent headache

Many children with SCA report recurrent headache. Whether this symptom should be considered a symptom of cerebrovascular disease and/or of chronic cerebral hypoxaemia is controversial. The recently published results of the SCI Transfusion trial described an association between recurrent headache and recurrent SCI [odds ratio (OR), 4·33; 95%CI, 1·5–13·6; P = 0·007] (DeBaun *et al*, 2014). In addition, our group recently demonstrated an imbalanced coagulation profile in SCA children with recurrent headache/migraine, in addition to the coagulation abnormalities often encountered in SCA (Kossorotoff *et al*, 2014a). Therefore, as described in other conditions in which enhanced thrombus formation/embolism and/or microvascular dysfunction is associated with increased migraine prevalence, in children with SCA the level of hypercoagulability might play a role in headache/migraine. Repeated ultra-transient ischaemic cerebral events and chronic cerebral hypoxaemia may relate to migraine in these children. Recurrent cephalalgia in children with SCA might, thus, represent a clinical marker of disease severity.

Cerebrovascular disease screening in children with sickle cell disease

Transcranial doppler: the reference-standard screening method

Transcranial Doppler measures blood-flow velocity in the large intracranial arteries of the circle of Willis. Arterial stenosis or blood-flow turbulence induces local blood-flow acceleration. Adams *et al* (1992) first established the effectiveness of TCD screening for assessing the risk of ischaemic stroke in asymptomatic children with SCA. An increased time-averaged mean maximal velocity (TAMMV) in the MCA was associated with an increased risk of stroke. The threshold MCA TAMMV value for a high risk of stroke was initially reported to be ≥170 cm/s (Adams *et al*, 1992). Subsequently, two stroke-risk subgroups were defined based on TAMMV values in the MCA and ICA, i.e., a high-risk group with TAMMV ≥200 cm/s and an intermediate-risk subgroup with TAMMV between 170 and 199 cm/s (Adams *et al*, 1997). Subsequently, the STOP (Stroke Prevention Trial in Sickle Cell Anaemia) study (Adams *et al*, 1998a,b) evaluated the efficacy of chronic transfusion in these subgroups at increased risk for stroke. The TCD categories used in the STOP trial were as follows: normal, velocities <170 cm/s; conditional, velocities > 170 cm/s and <200 cm/s; abnormal, velocities ≥200 cm/s; and inadequate, no information available on velocities in the MCA and/or ICA. These categories are currently used in clinical practice. Children undergoing randomization in the STOP trial had TAMMV ≥ 200 cm/s (see below).

Abnormal TCD findings mandate therapeutic intervention and further investigations (see Fig 1). If the TAMMV is normal (<170 cm/s), TCD screening should be performed once a year. Conditional TCD is an intermediate category that warrants repeated TCD and additional investigations, as conversion to abnormal TCD may occur. In a cohort of patients with SCA (median age, 7·1 years) not receiving hydroxycarbamide (also known as hydroxyurea) or chronic transfusion, 23% of patients converted from conditional to abnormal TCD within 18 months (Hankins *et al*, 2008a). In children enrolled in the STOP trial, the conversion rate to abnormal TCD increased from 29% after a single conditional TCD to 55% after two conditional TCDs (Adams *et al*, 2004). The optimal frequency of routine follow-up TCD is unclear but may depend on other known risk factors, such as age, severity of anaemia and genetic predisposition (see Fig 1).

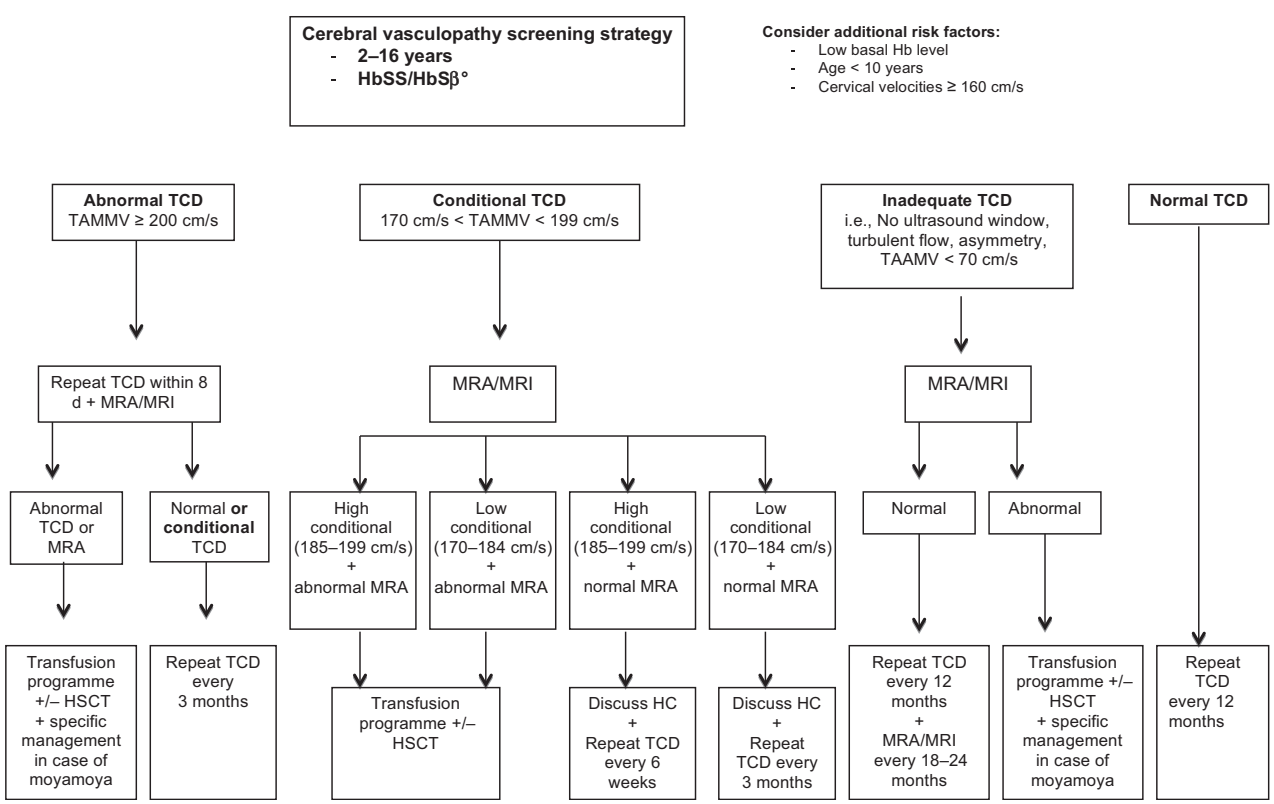

Fig 1. Suggested algorithm for cerebrovascular screening and management strategyTCD, transcranial doppler; MRA, magnetic resonance angiography; MRI, magnetic resonance imaging; HC, hydroxycarbamide; HSCT, haematopoietic stem cell transplantation [if human leucocyte antigen (HLA)-matched sibling]; TAMMV, time-averaged mean of the maximal velocity. MRA is considered abnormal when arterial stenosis is observed. MRI is performed to complete cerebrovascular work-up, allow subsequent follow-up and guide educational support if needed.

Current recommendations include annual TCD screening between 2 and 16 years of age (Yawn *et al*, 2014). Screening is not recommended before the age of 2 years. Among the 192 children included in the BABY HUG trial (age range, 7–17 months: mean age, 12·6 months), none had abnormal velocities and only 4 (2%) had conditional velocities. As with older children, TCD velocities were inversely associated with haemoglobin concentration and age (Pavlakis *et al*, 2010). Transcranial Doppler screening is not recommended after 16 years of age, as velocities ≥200 cm/s were not found in a study of 112 adults. Interestingly, adults with SCA had a higher mean TAMMV (110·9 ± 25·7 cm/s) than controls (71·1 ± 12 cm/s) (Valadi *et al*, 2006).

Low blood-flow velocities may also indicate a stroke risk. Blood-flow velocity is dramatically decreased downstream of stenoses and in nearly occluded arteries. Abnormally low TCD velocities may therefore reflect particularly severe arterial disease. Moyamoya should be suspected when velocities are very low (<70 cm/s), suggesting post-stenotic demodulation or near-occlusive arterial disease (Lee *et al*, 2004). Magnetic resonance angiography can confirm the diagnosis by showing severe stenoses of the ICA and its branches combined with abnormal collateral vessels.

More recent data suggest that elevated velocities in the anterior cerebral arteries increase the stroke risk in patients with SCA (Kwiatkowski *et al*, 2006).

Alterations in the extracranial portion of the ICA may also indicate an increased risk of overt stroke, including in patients who are free of intracranial arterial disease (Deane *et al*, 2010). Extracranial ICA velocity was recently evaluated in 435 stroke-free children with SCA (median age, 7·9 years). TAMMV values ≥160 cm/s in the cervical ICA were found in 10·3% of patients and were strongly associated with arterial stenosis (Verlhac *et al*, 2014). In this same cohort, isolated extracranial ICA stenosis was significantly and independently associated with SCI in 189 stroke-free SCA children (Bernaudin *et al*, 2015).

Finally, screening for cerebrovascular disease in children with the HbSC or HbS-β^+ genotype remains controversial. Overall, these subgroups have a low prevalence of stroke in childhood. Furthermore, all studies of transfusion efficacy were performed in patients with HbSS or HbS-β^0 disease. In a retrospective study of 46 children with HbSC disease, MCA TAMMV was significantly lower (mean, 129 cm/s) than the range reported in HbSS disease, suggesting that the threshold indicating an increased stroke risk might also be lower (Deane *et al*, 2008). At present, TCD screening is recommended only in patients with the HbSS or HbS-β^0 genotype (Yawn *et al*, 2014).

Contribution of other imaging studies to screening for cerebrovascular disease

Imaging techniques assessing the large intracranial vessels, such as MRA, computerized tomography angiography (CTA) and conventional angiography, showed good correlations with the TAMMV measured using TCD. Most children in the STOP trial underwent MRI/MRA (Abboud *et al*, 2004). Among patients who underwent MRA within 30 days of random assignment, those with severe stenosis had significantly higher TAMMV values compared to those with mild or no stenosis ($P < 0{\cdot}001$). Nevertheless, blood-flow alterations suggestive of a high stroke risk seemed detectable by TCD before the development of MRA abnormalities, further supporting the role for TCD as the reference-standard screening tool for lesions in large cerebral arteries.

Magnetic resonance angiography / Magnetic resonance imaging is needed in several situations (see Table I). At the acute phase, the sudden development of focal neurological manifestations in a child with SCD requires immediate MRI to look for signs of stroke. In the context of severe sudden-onset headache, emergency brain CT scan and CT angiogram is usually sufficient to diagnose intracranial haemorrhage. As paediatric cerebral infarction can be accompanied by severe headache and may not be recognized on CT scan in the first hours, in case of normal CT, emergency MRI/MRA is recommended. In the chronic setting, children with abnormal or conditional TCD findings must undergo MRA/MRI to evaluate potential arterial and cerebral lesions. MRA/MRI is also required in children with incomplete TCD assessments, as an inability to assess intracranial artery velocities using TCD is usually due to lack of a bone window but, alternatively, may be related to an underlying arterial occlusion. Finally, in children with chronic neurological symptoms, recurrent

Table I. Criteria for performing brain MRI/MRA in children with sickle cell disease.

Acute events
Sudden onset of a neurological abnormality, e.g., a focal deficit or seizures
Severe headache
Chronic manifestations
Routine MRI/MRA is recommended, at least once around 6 years of age
MRI/MRA is mandatory in patients who meet any of the following criteria:
TCD velocities >170 cm/s and <70 cm/s
Incomplete TCD
Asymmetric velocities
Turbulent flow
Recurrent headache
Cognitive delay
HSCT considered among the treatment options
Serial MRI/MRA is strongly recommended in children treated for cerebrovascular disease (hydroxycarbamide and/or chronic transfusion) and in those switched from chronic transfusion to hydroxycarbamide. The interval between imaging studies is determined on a case-by-case basis.

MRI, magnetic resonance imaging; MRA, magnetic resonance angiography; TCD, transcranial Doppler; HSCT, haematopoietic stem cell transplantation.

headache, and/or cognitive impairments, even when the TCD findings are normal, MRA/MRI should be performed to look for SCI and mild arterial stenosis. At present, routine MRI is not recommended in asymptomatic children with SCA (Yawn *et al*, 2014). However, SCI is associated with cognitive impairments and can be detected only using MRI. Patients with SCI may benefit from an early evaluation of their cognitive performance followed by appropriate educational and psychological support.

Regular neuropsychological assessments and monitoring may be highly helpful. In their recent review of SCI, DeBaun *et al* (2014) recommend performing MRI in all schoolchildren at least once. We agree with this recommendation, although we acknowledge that MRI imaging is expensive and that the cost-effectiveness of a systematic screening needs to be evaluated. Furthermore, it has been suggested that hydroxycarbamide stabilizes brain ischaemic lesions (Hankins *et al*, 2008b). Despite the lack of evidence concerning the ability of hydroxycarbamide to prevent the occurrence of SCI, an early diagnosis may also influence the decision to treat.

Longitudinal brain damage evaluation may be important, for instance when monitoring treatment effectiveness in children with cerebrovascular disease receiving hydroxycarbamide and/or chronic transfusion, particularly when switching from chronic transfusion to hydroxycarbamide (see below). Helton *et al* (2014a,b) recently used data from children enrolled in the SWiTCH (Stroke With Transfusions Changing to Hydroxyurea/hydroxycarbamide) trial to develop a scale for grading MRI/MRA findings based on the presence, size, location, and vascular territories of subcortical and/or cortical infarcts (see below for details on the SWiTCH trial). This new grading system awaits validation.

Conventional angiography is rarely appropriate. One of the two indications is cerebral aneurysm. The other is moyamoya syndrome, in which conventional angiography provides an assessment of cerebral perfusion and anastomoses between the external and internal carotid arteries.

Finally, imaging techniques that assess brain parenchymal perfusion can be useful to detect regional hypoperfusion, particularly as a few cases of stroke occur in patients with previously normal TCD findings (Adams *et al*, 1992). Arterial spin labelling (ASL) MRI involves the magnetic labelling of arterial blood protons, instead of the intravenous injection of a contrast agent (gadolinium). Arterial spin labelling provides a non-invasive quantitative assessment of cerebral blood flow in capillaries, which is dependent on the function of both large and small vessels. Arterial spin labelling studies in children with SCA showed blood-flow acceleration in the grey matter compared to normal controls, which was probably related to the increased cardiac output induced by chronic anaemia (Oguz *et al*, 2003). Interestingly, hydroxycarbamide may normalize grey matter blood flow without correcting the perfusion abnormalities in the white matter (Helton *et al*, 2009). Correlation studies between Transcranial doppler and techniques evaluating capillary blood flow, such as ASL and positron emission tomography are needed to validate tools evaluating the respective contributions of large and small vessels to the brain damage.

Preventing a first ischaemic stroke in children with cerebrovascular disease due to sickle cell disease

Prevention of a first stroke by chronic transfusion in children with abnormal TCDvelocities

STOP was a randomized trial of chronic transfusion versus standard care in children with abnormal TCD velocities (Adams *et al*, 1998a,b). Chronic transfusion decreased the risk of stroke by 92% (Adams *et al*, 1998a). This well-designed study prompted a recommendation to use chronic blood transfusion in children with abnormal TCD velocities National Heart, Lung, and Blood Institute, National Institutes of Health (NHLBI, 2014). In the STOP 2 trial, children receiving chronic transfusion because of abnormal TCD velocities were assigned at random to continuation or discontinuation of this treatment if they had received at least 30 months of transfusion therapy and had normal TCD velocities on repeat examination. Discontinuation of the chronic transfusion programme was followed by a high rate of stroke or reversion to abnormal TCD velocities (Adams & Brambilla, 2005). Based on STOP 2 results, the current recommendation for children with abnormal TCD velocities is to receive lifelong transfusion therapy.

There is no randomized study evaluating the impact of chronic transfusion or hydroxycarbamide *versus* observation in patients with abnormal velocity and/or stenosis in one or both extracranial portion of the internal carotid arteries. Considering the growing evidence on the relationship between cerebrovascular events and extracranial ICA stenosis (Telfer *et al*, 2011; Verlhac *et al*, 2014; Bernaudin *et al*, 2015) and extrapolating the experience acquired in patients with intracranial vasculopathy, we suggest recommending transfusions in the same way for patients with extracranial ICA stenosis as for those with intracranial ICA stenosis.

Is hydroxycarbamide an alternative to chronic transfusion in patients with abnormal transcranial Doppler (TCD) velocities?

Hydroxycarbamide is an inhibitor of the enzyme ribonucleotide reductase and has been used in children with severe SCD for almost two decades. Hydroxycarbamide increases the level of HbF, which prevents the elongation of polymerized deoxygenated HbS fibres, decreases blood-cell adherence to endothelial cells, and increases nitric oxide production (Hillery *et al*, 2000). In the BABY HUG study, compared to a placebo, hydroxycarbamide limited the age-related increase in cerebral blood-flow velocities in asymptomatic infants

(Wang *et al*, 2011), suggesting a protective effect on the brain. Few data exist on hydroxycarbamide therapy in children with increased TCD velocities. In a study from Belgium, 34 children with TCD velocities >200 cm/s, indicating a high-risk of first stroke, received hydroxycarbamide instead of chronic transfusion, which was contra-indicated by alloimmunization. A single patient experienced a neurological event (seizures) during the 96 patient-years of follow-up (Gulbis *et al*, 2005). In another study, 21 patients with conditional or abnormal TCD findings experienced a significant decrease in blood-flow velocities during hydroxycarbamide treatment, and only 1 had a stroke (Zimmerman *et al*, 2007). Finally, in November 2014, the National Institutes of Health (NIH) stopped the TWiTCH trial prematurely, as the early results showed that hydroxycarbamide therapy in children with abnormal TCD findings decreased blood velocities to the same extent as did chronic transfusion (www.nih.gov/news/health/nov2014/nhlbi-19.htm). However, an important point is that all children enrolled in the TWiTCH trial based on abnormal TCD velocities had received chronic transfusion, for a median of 4·6 ± 3·2 years, before switching to hydroxycarbamide, and were carefully screened out if they had any cerebral vasculopathy on MRA (Aygun *et al*, 2012). This point raises the possibility that, in patients with normal MRA receiving chronic transfusion, a switch to hydroxycarbamide may be safe albeit after a period of chronic transfusion, whose duration is unknown. In patients switched from chronic transfusion to hydroxycarbamide, we recommend regular TCD and MRA/MRI to look for cerebral and/or arterial lesions (Table I).

Management of silent cerebral infarction

Hydroxycarbamide therapy has been suggested to limit the extension or recurrence of SCI (Hankins *et al*, 2008b). It could be indicated in patients with SCI, but this indication has to be confirmed by controlled studies. Recently, a randomized single-blind study allocated 196 children with SCI to observation or chronic transfusion (DeBaun *et al*, 2014). Median follow-up was 3 years. In the transfusion group (n = 99), one child had a stroke and five new or enlarged SCIs, compared to seven and seven, respectively, in the observation group (n = 97) (P = 0·04). This study raises major questions. Up to one-third of children with SCA experience SCI during childhood (DeBaun *et al*, 2014), with an increased risk of stroke and neurocognitive impairments. Preventing these neurological complications is highly desirable. However, transfusing one third of patients with SCA may not be feasible, due to limitations in blood supplies and manpower, as well as concerns of patients, their families and providers regarding side effects, especially iron overload. Finally, it has been suggested that haematopoietic stem cell transplantation (HSCT) may be reasonable in patients with less severe forms of SCA and an HLA-matched sibling (Nickel *et al*, 2014).

Treating stroke in children with sickle cell disease: acute treatment and secondary prevention

Treating acute stroke

The sudden onset of a focal neurological abnormality in a patient with SCD suggests a stroke. When available immediately, MRI is recommended to confirm the diagnosis. The MRI protocol should include diffusion-weighted imaging to detect a recent ischaemic event; a perfusion sequence to detect focal hypoperfusion if the signs are very recent because diffusion-weighted imaging may be normal within the first 1 or 2 h of symptom onset; and vascular sequences to look for cerebral venous thrombosis. When emergency MRI is not available and the clinical manifestations are consistent with cerebral haemorrhage, CT is indicated. Finally, when brain imaging cannot be performed quickly, probabilistic treatment should be started without delay. Supportive measures, such as maintaining haemodynamic stability and monitoring glucose and oxygen levels are recommended in every case.

Despite the absence of controlled trials evaluating treatments for acute ischaemic stroke in paediatric patients with SCD and the uncertainty surrounding the potential impact of the initial management on long-term outcomes, emergency exchange transfusion is strongly recommended. The treatment targets are a decrease in HbS to less than 30% and an increase in total Hb to 100–120 g/l, without exceeding 120 g/l. Exchange transfusion produces immediate haemodynamic and rheological effects (Switzer *et al*, 2006; Webb & Kwiatkowski, 2013) and improves long-term neurological outcome (Hulbert *et al*, 2006). Extreme care must be directed at avoiding hypotension or hypertension and automated procedures are preferred (NHLBI, 2014). If a delay in the initiation of exchange transfusion is anticipated, a simple transfusion is recommended to increase the Hb level to about 100 g/l (Webb & Kwiatkowski, 2013).

To date, thrombolysis is not recommended in paediatric patients with SCD and acute ischaemic stroke, notably because thrombolysis is not among the recommended treatments in children outside the setting of a controlled trial (DeVeber & Kirkham, 2008).

After emergency treatment, complementary workup is suggested in order to screen for another cause(s) of stroke (thrombophilia workup, echocardiography).

Preventing recurrent stroke

Without secondary prevention, more than half the patients with a first stroke will experience a second stroke. Long-term observational studies have established that monthly blood transfusions greatly decrease the risk of recurrent stroke (Pegelow *et al*, 1995).

The optimal pre-transfusion HbS level for stroke prevention is unknown. The most widely used target is <30%. However, in 15 patients whose target HbS level was raised to 50% after at least 4 years with a target of 30%, no recurrences of ischaemic stroke were recorded over the median follow-up of 84 months. Two patients died from cerebral haemorrhage, but their HbS levels at the time of death were 30% and 29%, respectively (Cohen *et al*, 1992). Among 60 children treated with chronic transfusion and followed for 191·7 patient-years, 8 experienced a single recurrent stroke (six infarctive and two haemorrhagic strokes) and 13 experienced 15 transient neurological events. The HbS level was greater than 30% at the time of five of the six infarctions and 7 of the 15 transient events (Pegelow *et al*, 1995). In a retrospective study of 137 children receiving secondary stroke prevention at 14 centres, of which nine used an HbS target of 40–50%, the recurrence rate was 2·2 per 100 patient-years (Scothorn *et al*, 2002). However, these retrospective studies cannot establish a causal relationship between the HbS level and the risk of recurrent stroke. Recurrences are probably related not only to the HbS level, but also to the severity of the cerebrovascular disease and to concurrent medical events. Patients with severe arterial stenosis or moyamoya syndrome may experience stroke despite HbS levels well below 30% (Scothorn *et al*, 2002). In general, however, chronic transfusion protects most patients from clinical neurological events but does not always prevent worsening of the radiological abnormalities, even when the HbS levels are kept below 30% (Brousse *et al*, 2009; Hulbert *et al*, 2011; Helton *et al*, 2014a).

Transfusion protocols

Many teams use simple transfusion, which is easy to perform but may result in excessive Hb levels and high blood viscosity. Furthermore, repeated simple transfusions rapidly lead to iron overload (Singer *et al*, 1999), and exchange transfusion is therefore preferable. Exchange transfusion was associated with a lower risk of subsequent stroke compared to simple transfusion in a retrospective study (Hulbert *et al*, 2006). Exchange transfusion can be manual or automated (erythrocytapheresis). Erythrocytapheresis is better tolerated than manual exchange transfusion because it maintains isovolaemia and limits iron accumulation in chronically transfused patients (Hilliard *et al*, 1998). Limitations to the use of erythrocytapheresis are the need for two good venous access ports and the high cost of the procedure.

In our centre, for each transfusion, the patient arrives at about 8:30 AM. Blood is drawn for assays of Hb, HbS, ferritin, and hepatic and renal function parameters; as well as a direct anti-globulin test. When the Hb level is below 80 g/l, a simple transfusion of 15 ml/kg of packed red cells is given. In patients with Hb levels between 80 and 90 g/l, after arrival of the packed red-cell units at the day-treatment department, a phlebotomy is performed to remove 5 ml/kg of blood, and 5 ml/kg of saline is administered intravenously, followed by 15 ml/kg of packed red cells. Finally, in patients whose Hb level is above 90 g/l a phlebotomy is performed to remove 5 ml/kg of blood, a 10-ml/kg saline infusion is given, a second phlebotomy is performed to remove 5 ml/kg of blood and 15 ml/kg of packed red blood cells is administered, with all these steps being performed via the same venous access. The child usually leaves the day-treatment department at about 5 PM (Mirre *et al*, 2010).

Prevention and treatment of transfusion-related complications

The main complications of chronic transfusion fall into four categories: red-cell allo-immunization; iron overload; paucity of venous accesses and viral infections, which are the most feared by parents but are extremely rare in industrialized countries.

Red-cell allo-immunization is related to the discrepancy in blood group distribution between recipients of African descent and the donors, most of whom are of Caucasian origin in Europe and North America (Yazdanbakhsh *et al*, 2012). Some antigens are more immunogenic and clinically more relevant, leading to delayed haemolytic transfusion reactions. For sickle cell patients receiving chronic transfusion therapy, extended phenotyping should be performed and red blood cell units should be matched for Rh Cc, Rh Ee, and Kell antigens in addition to regular ABO and Rh D matching. Most centres reserve extensive matching of Kidd, Duffy and MNS blood groups for patients who have already developed antibodies.

Venous access may be limited. When the peripheral veins are insufficient, many teams use fully implantable venous access devices, such as Port-a-Cath, despite reports of an increased risk of sepsis (Jeng *et al*, 2002; Zarrouk *et al*, 2006). A few teams use a femoral catheter for each procedure (Billard *et al*, 2013).

Iron overload is an unavoidable complication of repeated transfusions. In contrast with thalassaemia, SCD seems to be associated with preferential iron accumulation in the liver and relative sparing of the heart (Darbari *et al*, 2006; Meloni *et al*, 2014). Iron chelation should be started if the ferritin level is greater than 1000 μg/l, the cumulative amount of transfused red-cell packs is greater than 120 ml/kg, or the liver iron content assessed using MRI is ≥7 mg/g dry weight (NHLBI, 2014). Until 2004, chelation relied on subcutaneous deferoxamine (Desferal®), 40 mg/kg/day. In 2006, deferasirox (Exjade®) was licensed for use over 2 years of age in children with SCD and transfusion-related iron overload. The starting dosage is 20 mg/kg/day and is subsequently adjusted according to the ferritin level and tolerance. Deferasirox is usually well tolerated in children with SCD (Vichinsky *et al*, 2011).

Blood leucodepletion and serological and genomic diagnostic tests for hepatitis viruses and the human immunodeficiency virus in blood products have dramatically reduced the

risk of transfusion-transmitted viral infections (Schreiber *et al*, 1996).

Is hydroxycarbamide an alternative to chronic transfusion in patients after a stroke?

The SWiTCH study (Stroke with Transfusions Changing to Hydroxyurea) was designed to determine whether hydroxycarbamide was as effective as chronic transfusion in preventing recurrent stroke in children with SCA and iron overload (Ware & Helms, 2012). Children with a past history of stroke were allocated at random to hydroxycarbamide/phlebotomy or standard transfusion/chelation treatment for secondary stroke prevention and control of iron overload. A composite endpoint combined stroke and iron removal. Stroke occurred in 7 of the 67 children receiving hydroxycarbamide *versus* none of the 66 children receiving transfusions while during the same time period there was no difference between the study arms for iron removal. The study was therefore closed prematurely and the authors concluded that transfusions and chelation remain a better way to manage children with SCA, stroke and iron overload. At present, chronic transfusion with a target HbS level <30%, when feasible and safe, remains the best option for stroke prevention in children with a history of overt stroke. Hydroxycarbamide is a treatment of second choice for patients with allo-immunization and those living in countries where the blood supply is limited.

Hydroxycarbamide combined with transfusions after a stroke

No randomized controlled trial has evaluated the potential benefits of adding hydroxycarbamide to chronic transfusion in patients with SCD and cerebrovascular disease. However, several studies reported a persistent risk of cerebrovascular disease progression in patients following well-conducted transfusion programmes (Brousse *et al*, 2009; Hulbert *et al*, 2011). This finding is probably ascribable to the adverse effects of residual sickle cells, high leucocyte counts, inflammation, a hypercoagulable state and severe vascular lesions. In a retrospective study in seven children, hydroxycarbamide combined with chronic blood transfusion was associated with a significant increase in pre-transfusion Hb levels combined with a decrease in HbS and an increase in HbF percentages. All seven children remained clinically stable on combined treatment and six of them experienced stabilization of their cerebrovascular disease (Brousse *et al*, 2013). We suggest that combining hydroxycarbamide with chronic transfusion may be helpful in children with persistent abnormal or conditional TCD velocities despite a well-conducted chronic transfusion programme.

Haematopoietic stem cell transplantation

Given the complications of chronic transfusion, especially iron overload, and the risk of cerebrovascular disease progression despite chronic transfusion, stroke is considered an indication for HSCT in children and adolescents who have an HLA-matched sibling (Walters *et al*, 1996; Bernaudin *et al*, 2007; Angelucci *et al*, 2014). This recommendation extends to children with impaired neuropsychological function and abnormal cerebral MRI findings. Neurological outcomes after HLA-identical HSCT have been assessed in two large but overlapping case-series studies (Bernaudin *et al*, 2007; Walters *et al*, 2010). Bernaudin *et al* studied 87 patients who received a transplant from an HLA-identical sibling between 1988 and 2004. Among them, 6 died, including 1 from cerebral haemorrhage. The event-free survival rate was 86·1% after a median follow-up of 6 years. Of the 36 patients with a history of stroke before transplantation, only two experienced recurrent neurological events (1 TIA and 1 fatal intracranial haemorrhage). The vascular occlusions persisted and, among the 23 stenoses, 5 resolved, 16 remained unchanged and 2 continued to worsen. In the work by Walters *et al*(2010), of 59 patients with a history of stroke who were transplanted between 1991 and 2000, 55 survived, including 50 who were free from SCD. Four patients died, two from intracranial haemorrhage and two from graft-versus-host disease. None of the survivors who had stable donor-cell engraftment experienced recurrent stroke after transplantation. Of the 55 survivors, 46 underwent brain MRI after transplantation, which usually showed stabilization of the cerebrovascular abnormalities. In both cohorts, seizures were common, but the seizure risk decreased after the introduction of routine anti-convulsant therapy. Finally, a literature review of neurological outcomes after HSCT in children with SCD has been recently reported (Bodas & Rotz, 2014). Of the 196 patients, 81 had radiological cerebrovascular abnormalities before transplantation. Two patients experienced a TIA during or after transplantation. Post-transplantation brain imaging performed in 45 patients showed no change in the cerebrovascular lesions in 71%, improvements in 13% and progression in 16%.

The appropriateness of HSCT in children with abnormal TCD velocities but normal MRI/MRA findings is more controversial (Khoury & Abboud, 2011). Haematopoietic stem cell transplantation is associated with severe complications, although the risk is relatively low in patients with SCD who receive stem cells from an HLA-identical donor (Bernaudin *et al*, 2007). Bernaudin *et al* (2007) considered that HSCT is indicated in patients with abnormal TCD velocities. Similarly, a recent study led the authors to suggest HSCT in patients with 'less severe' SCD, in order to prevent progression to severe SCD (Nickel *et al*, 2014).

Specific management of moyamoya syndrome

Moyamoya syndrome in children with SCD may require specific therapeutic interventions combined with chronic transfusion. Revascularization surgery may be indicated, depending on the angiography findings, previous stroke extent and

clinical examination. Direct or indirect revascularization procedures may be used, according to the protocols used in each centre (Scott & Smith, 2009). Patients with moyamoya should receive specific management if they require general anaesthesia or have severe acute anaemia (Scott & Smith, 2009).

Intracranial haemorrhage

Intracranial haemorrhage in children with SCD should be managed in the same way as acute ischaemic stroke. The aetiological work-up should look not only for an arteriovenous malformation, which is the most common cause of paediatric non-traumatic intracranial haemorrhage, but also for intracranial aneurysms, which have been reported in patients with SCD (Kossorotoff *et al*, 2014b) and moyamoya syndrome. The appropriateness of chronic transfusion is debated.

In conclusion, TCD screening has dramatically improved the prognosis of patients with SCD by identifying patients at high risk for stroke, who can then be enrolled in a chronic transfusion programme to decrease the risk. However, over one-third of patients have MRI evidence of SCI, whose management remains unclear. The extremely high prevalence of neurological abnormalities in children with SCD underlines the need for early detection and routine follow-up in centres with expertise in cerebrovascular disease. The heavy burden of cerebrovascular disease is SCA remains, to date, a challenging therapeutic issue.

Author contributions

Valentine Brousse, Manoelle Kossorotoff, and Mariane de Montalembert analysed the data and wrote the paper.

References

Abboud, M.R., Cure, J., Granger, S., Gallagher, D., Hsu, L., Wang, W., Woods, G., Berman, B., Brambilla, D., Pegelow, C., Lewin, J, Zimmerman, R.A. & Adams, R.J. STOP study (2004). Magnetic resonance angiography in children with sickle cell disease and abnormal transcranial Doppler ultrasonography findings enrolled in the STOP Study. *Blood*, **103**, 2822–2826.

Adams, R.J. & Brambilla, D. (2005) Optimizing Primary Stroke Prevention in Sickle Cell Anemia (STOP 2) Trial Investigators. Discontinuing prophylactic transfusions used to prevent stroke in sickle cell disease. *The New England Journal of Medicine*, **353**, 2769–2778.

Adams, R., McKie, V., Nichols, F., Carl, E., Zhang, D.L., McKie, K., Figueroa, R., Litaker, M., Thompson, W. & Hess, D. (1992) The use of transcranial ultrasonography to predict stroke in sickle cell disease. *The New England Journal of Medicine*, **326**, 605–610.

Adams, R.J., Kutlar, A., McKie, V., Carl, E., Nichols, F.T., Liu, J.C., McKie, K. & Clary, A. (1994) Alpha thalassemia and stroke risk in sickle cell anemia. *American Journal of Hematology*, **45**, 279–282.

Adams, R.J., McKie, V.C., Carl, E.M., Nichols, F.T., Perry, R., Brock, K., McKie, K., Figueroa, R., Litaker, M., Weiner, S. & Brambilla, D. (1997) Long term stroke risk in children with sickle cell disease screened with transcranial doppler. *Annals of Neurology*, **42**, 699–704.

Adams, R.J., McKie, V.C., Hsu, L., Files, B., Vichinsky, E., Pegelow, C., Abboud, M., Gallagher, D., Kutlar, A., Nichols, F.T., Bonds, D.R., Brambilla, D., Woods, G., Olivieri, N., Driscoll, C., Miller, S., Wang, W., Hurlett, A., Scher, C., Berman, B., Carl, E., Jones, A.J., Roach, E.S., Wright, E., Zimmerman, R.A. & Waclawiw, M. (1998a) Prevention of a first stroke by transfusions in children with sickle cell anemia and abnormal results on transcranial doppler ultrasonography. *The New England Journal of Medicine*, **339**, 5–11.

Adams, R.J., McKie, V.C., Brambilla, D., Carl, E., Gallagher, D., Nichols, F.T., Roach, S., Abboud, M., Berman, B., Driscoll, C., Files, B., Hsu, L., Hurlet, A., Miller, S., Olivieri, N., Pegelow, C., Scher, C., Vichinsky, E., Wang, W., Woods, G., Kutlar, A., Wright, E., Hagner, S., Tihe, F. & Waclawiw, M.A. (1998b) Stroke prevention trial in sickle cell anemia. *Controlled Clinical Trials*, **19**, 110–129.

Adams, R.J., Brambilla, D.J., Granger, S., Gallagher, D., Vichinsky, E., Abboud, M.R., Pegelow, C.H., Woods, G., Rohde, E.M., Nichols, F.T., Jones, A., Luden, J.P., Bowman, L., Hagner, S. & Morales, K.H. & Roach, E.S. STOP Study (2004) Stroke and conversion to high risk in children screened with transcranial Doppler ultrasound during the STOP study. *Blood*, **130**, 3689–3694.

Angelucci, E., Matthers-Martin, S., Baronciani, D., Bernaudin, F., Bonanomi, S., Cappellini, M.D., Dalle, J.H., Di Bartolomeo, P., deHeredia, C.D., Dickerhoff, R., Giardini, C, Gluckman, E., Hussein, A.A, Kamani, N., Minkov, M., Locatelli, F., Rocha, V., Sedlacek, P., Smiers, F., Thuret, I., Yaniv, I., Cavazzana, M. & Peters, C.; on behalf of the EBMT Inborn Error and EBMT Paediatric Working Parties (2014) Hematopoietic stem cell transplantation in thalassemia major and sickle cell disease: indications and management recommendations from an international expert panel. *Haematologica*, **99**, 811–820.

Aygun, B., Wruck, L.M., Schultz, W.H., Mueller, B.U., Brown, C., Luchtman-Jones, L., Jackson, S., Iyer, R., Rogers, Z., Sarnaik, S., Thompson, A.A., Gauger, C., Helms, R.W. & Ware, R.E. (2012) Chronic transfusion practices for prevention of primary stroke in children with sickle cell anemia and abnormal TCD velocities. *American Journal of Hematology*, **87**, 428–430.

Bernaudin, F., Verlhac, S., Freard, F., Roudot-Thoraval, F., Benkerrou, M., Thuret, I., Mardini, R., Vannier, J.P., Ploix, E., Romero, M., Casse-Perrot, C., Helly, M., Gillard, E., Sebag, G., Kchouk, H., Pracros, J.P., Kinck, B., Dacher, J.N., Ichowicz, V., Raybaud, C., Poncet, M., Lesprit, E., Reinert, P. & Brugieres, P. (2000) Multicenter prospective study of children with sickle cell disease: radiographic and psychometric correlation. *Journal of Child Neurology*, **15**, 333–343.

Bernaudin, F., Socie, G., Kuentz, M., Chevret, S., Duval, M., Bertrand, Y., Vannier, J.P., Yacouben, K., Thuret, I., Bordigoni, P., Fischer, A., Lutz, P., Stephan, J.L., Dhedin, N., Plouvier, E., Margueritte, G., Bories, D., Verlhac, S., Esperrou, H., Coic, L., Vernant, J.P. & Gluckman, E., for the SFGM-TC (2007) Long-term results of related myelo-ablative stem-cell transplantation to cure sickle cell disease. *Blood*, **110**, 49–56.

Bernaudin, F., Verlhac, S., Chevret, S., Torres, M., Coic, L., Arnaud, C., Kamden, A., Hau, I., Neonato, M.G. & Delacourt, C. (2008) G6PD deficiency, absence of alpha-thalassemia and hemolytic rate at baseline are significant independent risk factors for abnormally high cerebral velocities in patients with sickle cell anemia. *Blood*, **112**, 4314–4317.

Bernaudin, F., Verlhac, S., Arnaud, C., Kamden, A., Chevret, S., Hau, I., Coic, L., Leville, E., Lemarchand, E., Lesprit, E., Abadie, I., Medejel, N., Madhi, F., Lemerle, S., Biscardi, S., Bardakdjian, J., Galacteros, F., Torres, M., Kuentz, M., Ferry, C., Socie, G., Reinert, P. & Delacourt, C. (2011) Impact of early transcranial Doppler screening and intensive therapy on cerebral vasculopathy outcome in a newborn sickle cell anemia cohort. *Blood*, **117**, 1130–1140.

Bernaudin, F., Varlhac, S., Arnaud, C., Kamdem, A., Vasile, M., Kasbi, F., Hau, I., Madhi, F., Fournaux, C., Biscardi, S., Epaud, R. & Pondarré, C. (2015) Chronic acute anemia and extracranial internal carotid stenosis are risk factors for silent cerebral infarcts in sickle cell anemia. *Blood*, **125**, 1653–1661.

Billard, M., Combet, S., Hequet, O., Kebaili, K., Lorthois, S. & Pondarré, C. (2013) Short-term femoral insertion: a promising alternative to consistently allow long-term erythrapheresis

therapy in children with sickle cell anemia. *Journal of Pediatrics*, **162**, 423–426.

Bodas, P. & Rotz, S. (2014) Cerebral vascular abnormalities in pediatric patients with sickle cell disease after hematopoietic cell transplant. *Journal of Pediatric Hematology Oncology*, **36**, 190–193.

Brousse, V., Hertz-Pannier, L., Consigny, Y., Bresson, J.L., Girot, R., Mirre, E., Lenoir, G. & de Montalembert, M. (2009) Does regular blood transfusion prevent progression of cerebrovascular lesions in children with sickle cell disease? *Annals of Hematology*, **88**, 785–788.

Brousse, V., Gandhi, S.De, Montalembert, M., Height, S., Dick, M.C., O'Driscoll, S., Abihsera, G. & Rees, D.C. (2013) Combined blood transfusion and hydroxycarbamide in children with sickle cell anemia. *British Journal of Haematology*, **2013**, 259–261.

Casella, J.F., King, A.A., Barton, B., White, P.A., Noetzel, M.J., Ichord, R.N., Terrill, C., Hirtz, D., McKinstry, R.C., Strouse, J.J., Howard, T.H., Coates, T.D., Minniti, C.P., Campbell, A.D., Vendt, B.A., Lehmann, H. & Debaun, M.R. (2010) Design of the silent cerebral infarct transfusion (SIT) trial. *Pediatric Hematology Oncology*, **27**, 69–89.

Cohen, A.R., Martin, M.B., Silber, J.H., Kim, H.C., Ohene-Frempong, K. & Schwartz, E. (1992) A modified transfusion program for prevention of stroke in sickle cell disease. *Blood*, **79**, 1657–1661.

Connes, P., Verlhac, S. & Bernaudin, F. (2013) Advances in understanding the pathogenesis of cerebrovascular vasculopathy in Sickle Cell Anemia. *British Journal of Haematology*, **161**, 484–494.

Darbari, D.S., Kpile-Faget, P., Kwagyan, J., Rana, S., Goedeuk, V.R. & Castro, O. (2006) Circumstances of death in adult sickle cell disease patients. *American Journal of Hematology*, **81**, 858–863.

De Montalembert, M., Beauvais, P., Bachir, D., Galacteros, F. & Girot, R. (1993) Cerebrovascular accidents in sickle cell disease. Risk factors and blood transfusion influence. French Study Group on sickle cell disease. *European Journal of Pediatrics*, **65**, 201–204.

Deane, C., Goss, D., O'Driscoll, S., Mellor, S., Pohl, K.R., Dick, M.C., Height, S.E. & Rees, D.C. (2008) Transcranial Doppler scanning and the assessment of stroke risk in children with HbSC disease. *Archives of Disease in Children*, **93**, 138–141.

Deane, C.R., Goss, D., Bartram, J., Pohl, K.R., Height, S.E., Sibtain, N., Jarosz, J., Thein, S.L. & Rees, D.C. (2010) Extracranial internal carotid arterial disease in children with sickle cell anemia. *Haematologica*, **95**, 1287–1292.

Debaun, M.R., Amstrong, F.D., McKinstry, R.C., Ware, R.E., Vichinsky, E. & Kirkham, F.J. (2012) Silent cerebral infarcts: a review on a prevalent and progressive cause of neurologic injury in sickle cell anemia. *Blood*, **119**, 4587–4596.

DeBaun, M.R., Gordon, R.C., McKinstry, M.J., Noetzel, D.A., White, D.A., Sarnaik, S.A., Meier, E.R., Howard, T.H., Majumdar, S., Inusa, B.P.D., Telfer, P.T., Kirby-Allen, M., McCavit, T.L., Kamden, A., Airewele, G., Woods, G.M., Berman, B., Panepinto, J.A., Fuh, B.R., Kwiatowski, J.L., King, A.A., Fixler, J.M., Rhodes, M.M., Thompson, A.A., Heiny, M.E., Redding-Lallinger, R.C., Kirkham, F.J., Dixon, N., Gonzalez, C.E., Kalinyak, K.A., Quinn, C.T., Strouse, J.J., Miller, J.P., Lehmann, H., Kraut, M.A., Ball, W.S., Hirtz, D. & Casella, J.F. (2014) Controlled trial of transfusions for silent infarcts in sickle cell anemia. *The New England Journal of Medicine*, **371**, 699–710.

DeVeber, G. & Kirkham, F.J. (2008) Guidelines for the treatment and prevention of stroke in children. *The Lancet Neurology*, **7**, 983–985.

Gulbis, B., Haberman, D., Dufour, D., Christophe, C., Vermylen, C., Kagambega, K., Corazza, F., Devalck, M.F., Dresse, M.F., Hunninck, K., Klein, A., Le, P.Q., Loop, M., Maes, P., Philippet, P., Sariban, E., VanGeet, C. & Ferster, A. (2005) Hydroxyurea for sickle cell disease in children and for prevention of cerebrovascular events The Belgian experience. *Blood*, **105**, 2685–2690.

Hankins, J.S., Fortner, G.L., McCarville, M.B., Smeltzer, M.P., Wang, W.C., Li, C.S. & Ware, R.E. (2008a) The Natural history of conditional transcranial Doppler flow velocities in children with sickle cell anemia. *British Journal of Haematology*, **142**, 94–99.

Hankins, J.S., Helton, K.J., McCarville, M.B., Li, C.S., Wang, W.C. & Ware, R.E. (2008b) Preservation of spleen and brain function in children with sickle cell anemia treated with hydroxyurea. *Pediatric Blood Cancer*, **50**, 293–297.

Helton, K.J., Paydar, A., Glass, J., Weirich, E.M., Hankins, J., Li, C.S., Smeltzer, M.P., Wang, W.C., Ware, R.E. & Ogg, R.J. (2009) Arterial spin-labeled perfusion combined with segmentation techniques to evaluate cerebral blood flow in white and gray matter of children with sickle cell anemia. *Pediatric Blood Cancer*, **52**, 85–91.

Helton, K.H., Adams, R.A., Kesler, K.L., Lockhart, A., Aygun, B., Driscoll, C., Heeney, M.H., Jackson, S.M., Krishnamurti, L., Miller, S.T., Sarnaik, S.A., Schultz, W.H. & Ware, R.E. (2014a) Magnetic resonance imaging/angiography and transcranial Doppler velocities in sickle cell anemia: results from the SWiTCH trial. *Blood*, **124**, 891–898.

Helton, K.H., Roberts, D., Schultz, W.H., Davis, B.R., Kalfa, T.A., Pressel, S.L., Adams, R.J. & Ware, R.E. (2014b) Effects of chronic transfusion therapy on MRI and MRA in chidren with sickle cell anemia at risk for primary stroke: baseline imaging from the Twitch trial. *Blood*, **124**, 4052.

Hillery, C.A., Du, M.C., Wang, W.C. & Scott, J.P. (2000) Hydroxyurea therapy decreases the in vitro adhesion of sickle erythrocytes to thrombospondin and laminin. *British Journal of Haematology*, **109**, 322–327.

Hilliard, L.M., Williams, B.F., Lounsbury, A.E. & Howard, T.H. (1998) Erythrocytapheresis limits iron acculation in chronically transfused sickle cell patients. *American Journal of Hematology*, **59**, 28–35.

Hulbert, M.L., Scothorn, D.J., Panepinto, J.A., Scott, J.P., Buchanan, G.R., Sarnaik, S., Fallon, R., Chu, J.Y., Wang, W., Casella, J.F., Resar, L., Berman, B., Adamkiewicz, T., Hsu, L.L., Smith-Whitley, K., Mahoney, D., Woods, G., Watanabe, M. & DeBaun, M.R. (2006) Exchange blood transfusion compared with simple transfusion fort first overt stroke is associated with a lower risk of subsequent stroke: a retrospective cohort study of 137 children with sickle cell anemia. *Journal of Pediatrics*, **149**, 710–712.

Hulbert, M.L., McKinstry, R.C., Lacey, J.L., Moran, C.J., Panepinto, J.A., Thomson, A.A., Sarnaik, B.A., Woods, G.H., Casella, J.P., Inusa, B., Howard, J., Kirkham, F.J., Aniek, A., Mullin, J.E., Ichord, R., Noetzel, M., Yvan, Y., Rodeghier, M. & Debaun, M.R. (2011) Silent cerebral infarcts occur despite regular blood transfusion therapy after first stroke in children with sickle cell disease. *Blood*, **117**, 772–779.

Jeng, M.R., Feusner, J., Skibola, C. & Vichinsky, E. (2002) Central venous catheter complications in sickle cell disease. *American Journal of Hematology*, **69**, 103–108.

Khoury, R. & Abboud, M.R. (2011) Stem-cell transplantation in children and adults with sickle-cell disease: an update. *Expert Review in Hematology*, **4**, 343–351.

Kirkham, F., Hewes, D.K., Prengler, M., Wade, A., Lane, R. & Evans, J.P.M. (2001) Nocturnal hypoxemia and central -nervous-system events in sickle cell disease. *Lancet*, **357**, 1656–1659.

Kossorotoff, M., Lasne, D., Brousse, V., Desguerre, I., de Montalembert, M. & Gaussem, P. (2014a) Imbalanced cagulation profile as a biomarker of migraine in children with sickle cell: is this a link with cerebral ischemia? *The Journal of Pediatrics*, **165**, 645–646.

Kossorotoff, M., Brousse, V., Grevent, D., Naggara, O., Brunelle, F., Blauwblomme, T., Gaussem, P., Desguerre, I. & de Montalembert, M. (2014b) Cerebral haemorrhage risk in children with sickle-cell disease. *Developmental Medicine & Child Neurology*, **57**, 187–193.

Kwiatkowski, J.L., Granger, S., Brambilla, D.J., Brown, R.C., Miller, S.T., Adams, R.J. & STOP trial investiagtors (2006) Elevated blood flow velocity in the anterior cerebral artery and stroke risk in sickle cell disease: extended analysis from the STOP trial. *British Journal of Haematology*, **134**, 333–339.

Lee, Y.-S., Jung, K.-H. & Roh, J.-K. (2004) Diagnosis of Moya-Moya disease with transcranial Doppler sonography: correlation study with magnetic resonance angiography. *Journal of Neuroimaging*, **14**, 319–323.

Meloni, A., Puliyel, M., Pepe, A., Berdoukas, V., Coates, T. & Woods, J.C. (2014) Cardiac iron

overload in sickle cell disease. *American Journal of Hematology*, **89**, 678–683.

Miller, S.T., Macklin, E.A., Pegelow, C.H., Kinney, T.R., Sleeper, L.A., Bello, J.A., DeWitt, L.D., Gallagher, D.M., Guarini, L., Moser, F.G., Ohene-Frempong, K., Sanchez, N., Vichinsky, E.P., Wang, W.C., Wethers, D.L., Younkin, D.P., Zimmerman, R.A. & DeBaun, M.R. (2001). Silent infarction as a risk factor for overt stroke in children with sickle cell anemia: a report from the Cooperative Study of Sickle Cell Disease. *Journal of Pediatrics*, **139**, 385–390.

Miller, S.T., Milton, J. & Steinberg, M.H. (2011) G6PD deficiency and stroke in the CSSD. *American Journal of Hematology*, **86**, 331.

Mirre, E., Brousse, V., Berteloot, L., Lambot-Juhan, K., Verlhac, S., Boulat, C., Dumont, M.D., Lenoir, G. & de Montalembert, M. (2010) Feasibility and efficacy of chronic transfusion for stroke prevention in children with sickle cell disease. *European Journal of Haematology*, **84**, 259–265.

Moritani, T., Numaguchi, Y., Lemer, N.B., Rozans, M.K., Robinson, A.E., Hiwatashi, A. & Westesson, P.L. (2004) Sickle cell cerebrovascular disease. Usual and unusual findings on MR imaging and MR angiography. *Journal of Clinical Imaging*, **28**, 173–186.

NHLBI (2014) Evidence-based management of sickle cell disease: Expert Panel Report, 2014. National Heart, Lung, and Blood Institute, National Institutes of Health. http://www.nhlbi.nih.gov/sites/www.nhlbi.nih.gov/files/sickle-cell-disease-report.pdf

Nickel, R.S., Hendrickson, J.E. & Haight, A.E. (2014) The ethics of a proposed study of hematopoietic stem cell transplant for children with "less severe" sickle cell disease. *Blood*, **124**, 861–866.

Oguz, K. K., Golay, X., Pizzini, F.B., Freer, C.A., Winrow, N., Ichord, R., Casella, J.F., Van Zijl, P.C. & Melhem, E.R. (2003) Sickle cell disease: continuous arterial spin-labelingperfusion MR imaging in children. *Radiology*, **227**, 567–574.

Ohene-Frempong, K., Weiner, S.J., Sleeper, L.A., Miller, S.T., Embury, S., Moohr, J.W., Wethers, D.L., Pegelow, C.H., Gill, F.M. & the cooperative study on Sickle Cell Disease. (1998) Cerebrovascular accidents in sickle cell disease: rate and risk factors. *Blood*, **91**, 288–294.

Pavlakis, S.G., Rees, R.C., Huang, X., Brown, R.C., Casella, J.F., Iyer, R.V., Kalpatthi, R., Luden, J., Miller, S.T., Rogers, Z.R., Thornburg, C.D. & Wang, W.C., Adams, R.J. & BABY HUG investigators. (2010) Transcranial doppler ultrasonography (TCD) in infants with sickle cell anemia: baseline data from the BABY HUG trial. *Pediatric Blood Cancer*, **54**, 256–259.

Pegelow, C.H., Adams, R.J., McKie, V., Abboud, M., Berman, B., Miller, S.T., Olivieri, N., Vichinsky, E., Wang, W. & Brambilla, D. (1995) Risk of recurrent stroke in patients with sickle cell disease treated with erythrocyte transfusions. *Journal of Pediatrics*, **126**, 896–899.

Powars, D., Wilson, B., Imbus, C., Pegelow, C. & Allen, J. (1978) The natural history of stroke in sickle cell disease. *American Journal of Medicine*, **65**, 461–471.

Schreiber, G.B., Busch, M.P., Kleinman, S.H. & Korelitz, J.J. (1996) The risk of transfusion-transmitted viral infections. The Retrovirus Epidemiology Donor Study. *The New England Journal of Medicine*, **334**, 1685–1690.

Scothorn, D.J., Price, C., Schwartz, D., Terrill, C., Buchanan, G.R., Shunney, W., Sarnaik, I., Fallon, R., Chu, Y.P., Pegelow, C.H., Wang, W., Casella, J.F., Resar, L., Berman, B., Adamkiewicz, T., Hsu, L.L., Ohene-Frempong, K., Smith-Wjitley, K., Mahoney, D., Scott, J.P., Woods, L.M., Watanabe, M. & Debaun, M.R. (2002) Risk of recurrent stroke in children with sickle cell disease receiving blood transfusion for at least five years after initial stroke. *Journal of Pediatrics*, **140**, 348–354.

Scott, R.M. & Smith, E.R. (2009) Moyamoya disease and moyamoya syndrome. *The New England Journal of Medicine*, **360**, 1226–1237.

Sebastiani, P., Ramoni, M.F., Nolan, V., Baldwin, C.T. & Steinberg, M.H. (2005) Genetic dissection and prognostic modelling of overt stroke in sickle cell anemia. *Nature Genetics*, **37**, 435–440.

Singer, S.T., Quirolo, K., Nishi, K., Hackney-Stephens, E., Evans, C. & Vichinsky, E.P. (1999) Erythrocytapheresis for chronically transfused children with sickle cell disease: an effective method for maintaining low haemoglobin S level and reducing iron overload. *Journal of Clinical Apheresis*, **14**, 122–125.

Styles, L.A., Hoppe, C., Klitz, W., Vichinsky, E., Lubin, B. & Trachtenberg, E. (2000) Evidence for HLA-related susceptibility for stroke in children with sickle cell disease. *Blood*, **95**, 3562–3567.

Switzer, J.A., Hess, D.C., Nichols, F.T. & Adams, R.J. (2006) Pathophysiology and treatment of stroke in sickle –cell disease: present and future. *Lancet Neurology*, **5**, 501–512.

Telfer, P.T., Evanson, J., Butler, P., Hemmaway, C., Abdulla, C., Gadong, C., Whitmarsh, S., Kaya, B. & Kirkham, F.J. (2011) cervical carotid artery disease in sickle cell anemia: clinical and radiological features. *Blood*, **23**, 6192–6199.

Valadi, N., Silva, G.S., Bowman, L.S., Ramsingh, D., Vicari, P., Filho, A.C., Massaro, A.R., Kutlar, A., Nichols, F.T. & Adams, R.J. (2006) Transcranial doppler ultrasonography in adults with sickle cell disease. *Neurology*, **67**, 572–574.

Verlhac, S., Balandra, S., Cussenot, J., Kasbi, F., Vasile, M., Kheniche, A., Elmaleh-Berges, M., Ithier, G., Benkerrou, M., Bernaudin, F. & Sebag, G. (2014) Extracranial arteriopathy in stroke-free children with sickle cell anemia: detection by submandibular Doppler sonography. *Pediatric Radiology*, **44**, 587–596.

Vichinsky, E., Bernaudin, F., Forni, G.L., Gardner, R., Hassell, K., Heeney, M.M., Inusa, B., Kutlar, A., Lane, P., Mathias, L., Porter, J., Tebbi, C., Wilson, F., Griffel, L., Deng, W., Giannone, V. & Coates, W. (2011) Long term safety and efficacy of Deferasirox (Exjade) for up to 5 years in transfusional iron-overloadeded patients with sickle cell disease. *British Journal of Haematology*, **154**, 387–397.

Walters, M.C., Patience, M., Leisering, W., Eckman, J.R., Scott, J.P., Mentzer, W.C., Davies, S.C., Ohene-Frempong, K., Bernaudin, F., Matthews, D.C., Storb, R. & Sullivan, K.M. (1996) Bone marrow transplantation for sickle cell disease. *The New England Journal of Medicine*, **335**, 369–376.

Walters, M.C., Hardy, K., Edwards, S., Adamkiewicz, T., Barkovich, J., Bernaudin, F., Buchanan, G.R., Bunin, N., Dickerhoff, R., Giller, R., Haut, P.R., Horan, J., Hsu, L.L., Kamani, N., Levine, J.E., Margolis, D., Ohene-Frempong, K., Patience, M., Redding Lallinger, R., Roberts, I.A.G., Rogers, Z.R., Sanders, J., Scott, J.P. & Sullivan, K.M. (2010).Pulmonary, gonadal., and central nervous system status after bone marrow transplantation for sickle cell disease. *Biology Blood Marrow Transplant*, **16**; 263–272.

Wang, W.C., Ware, R.E., Miller, S.T., Iyer, R.I., Casella, J.F., Minniti, C.P., Rana, S., Thornburg, C.D., Rogers, Z.R., Kalpatthi, R.V., Barredo, J.C., Brown, R.C., Sarnaik, S.A., Howard, T.H., Wynn, L.V., Kutlar, A., Amstrong, F.D., Files, B.A., Goldsmith, J.C., Waclawiw, M.A., Huang, X., Thompson, P.W. & Baby HUG investigators. (2011) Hydroxycarbamide in very young children with sickle-cell anaemia: a multicentre, randomised, controlled trial (BABY HUG). *The Lancet*, **377**, 1663–1672.

Ware, R.E. & Helms, R.W. & SWiTCH investigators (2012) Stroke with transfusions changing to Hydroxyurea (SWiTCH). *Blood*, **119**, 3925–3932.

Webb, J. & Kwiatkowski, J.L. (2013) Stroke in patients with sickle cell disease. *Expert Review of Hematology*, **6**, 301–3016.

Yawn, B.P., Buchanan, G.R., Afenyi-Annan, A.N., Ballas, S.K., Hassell, K.L., James, A.H., Jordan, L., Lanzkron, S.M., Lottenberg, R., Savage, W.J., Tanabe, P.J., Ware, R.E., Murad, M.H., Goldsmith, J.C., Ortiz, E., Fulwood, R., Horton, A. & John-Sowah, J. (2014) Management of sickle cell disease: summary of the 2014 evidence-based report expert panel members. *Journal of American Medical Association*, **312**, 1033–1048.

Yazdanbakhsh, K., Ware, R.E. & Noizat-Pirenne, F. (2012) Red blood cell alloimmunization in sickle cell disease: pathophysiology, risk factors, and transfusion management. *Blood*, **120**, 528–537.

Zarrouk, V., Habibi, H., Zahar, J.R., Roudot-Thoraval, F., Bachir, D., Brun-Buisson, C., Legrand, P., Godeau, B., Galacteros, F. & Lesprit, P. (2006) Bloodstream infection in adults with sickle cell disease, association with venous catheters, staphylococcus aureus, and bone-joint infection. *Medicine*, **85**, 43–48.

Zimmerman, S.A., Schultz, W.H., Burgett, S., Mortirer, N.A. & Ware, R.E. (2007) Hydroxyurea therapy lowers transcranial Doppler flow velocities in children with sickle cell anemia. *Blood*, **110**, 1043–1047.