Endothelial-dependent vasodilation is impaired in children with sickle cell disease

Impairment of endothelial-dependent vasodilation has been demonstrated in adults with sickle cell anemia (SCA). We enrolled 21 SCA children, mean age 10.4±3.3 yrs, and 23 Afro-Caribbean controls. We examined flow-mediated (FMD) and nitroglycerine-mediated (GTNMD) dilation of the brachial artery, using echotracking techniques, and measured intima-media thickness (IMT) and mechanical properties of the common carotid artery. FMD was significantly decreased in SCA children *vs* controls (5.6±0.2 *vs* 8.0±0.2%, p=0.008), while IMT, stiffness of the common carotid artery, and GTNMD were comparable. In conclusion, endothelial dysfunction is present as early as childhood in SCA patients.

Haematologica 2007; 92:1709-1710. DOI: 10.3324/haematol.11253

Sickle cell anemia (SCA) is characterized by hemoglobin S polymerization, chronic hemolytic anemia, adherence of young red cells to the venular endothelium, chronic inflammation, and ischemia-reperfusion injury leading to endothelium damage and recurrent painful vasocclusion. Availability of the major endothelial vasodilator, nitric oxide (NO), may be reduced by the scavenging of cell-free hemoglobin and destruction by arginase released during hemolysis.[1] Sickle transgenic mice had a decreased arteriolar dilation in response to NO-mediated vasodilators.[2,3]

A reduction of the flow-mediated vasodilation, secondary to NO release from the endothelium following an increase in wall shear stress, has been reported in SCA adult patients.[4-6] No study of vascular tone had been performed in SCA children, and it is not known whether impairment of vasodilation could be observed early in childhood. We therefore studied arterial stiffness and endothelial- and non-endothelial-mediated vasodilation in SCA children, using non-invasive ultrasonographic techniques.

We enrolled 21 children with SCA (18 homozygous SS, 3 S-β^0 thalassemics) who were asymptomatic at the time of enrolment (12 males, 9 females, mean age 10.4±3.3 years, mean hemoglobin level 7.6±1.0 g/dL). None had undergone blood transfusion in the previous 3 months, or were treated with hydroxyurea. These SCA children were compared to a control group of 23 Afro-Caribbean AA and AS controls, matched for age and gender, selected from among siblings of our patients. Our institutional review board approved the protocol and written informed consent was obtained from all parents.

Blood pressure and vascular function parameters were measured after the child had been recumbent for at least 10 minutes. Change in right brachial artery diameter in response to reactive hyperemia (flow increase inducing endothelium-dependent vasodilation) and change induced by glyceryltrinitrate (GTN), an endothelium-independent vasodilator, were measured in SCA and in controls using echotracking techniques. The maximum flow-mediated dilation (FMD) and GTN mediated dilation (GTNMD) were calculated as the percent change in diameter compared with baseline resting diameter. We inflated a blood-pressure cuff to 300 mmHg for 4 min., the artery was scanned for 30 sec. before and 90 sec. after the cuff deflation. Ten minutes later, a resting scan was recorded. GTNMD was determined after the administration of glyceryltrinitrate sublingually. Additionally, intima-media thickness (IMT) and mechanical properties of the common carotid artery were measured using previously described methods.[7]

All statistical analyses were performed using StatView™ SE software. Blood pressure of SCA and control children was comparable. This contrasts with previous findings in SCA adults, who had significantly lower blood pressure.[8] There was no difference in SCA children (0.42±0.06 mm) and controls (0.42±0.03 mm). Systolic and diastolic diameters of the common carotid artery were significantly higher in SCA children than in controls (6.8±0.8 *vs* 5.7±0.4, p<0.001 and 5.7±0.7 *vs* 5.0±0.4 mm, respectively, p<0.005). However, there was no difference in the stiffness of the common carotid artery between SCA children and control. This leads us to hypothesize that the increase of the carotid artery diameters, without modification of the distensibility, was related to the higher cardiac output secondary to anemia.[9] Finally, FMD was significantly decreased in SCA children compared with controls (5.6±0.2 *vs* 8.0±0.2%, p=0.008) while GTNMD was comparable in SCA patients and controls (20±8 *vs* 21±8%). There was no correlation between FMD and either age or hemoglobin level. Reduced FMD and preserved nitrate-mediated vasodilation had also been observed in a group of 47 sickle-thalassemia adult patients.[6] However, these adult patients had, contrary to our children, an increased aortic stiffness, which would suggest that the endothelial dysfunction we observed in children could be the first step of a process which progressively modifies the mechanical properties of the arterial wall in adults. This study was designed before the evidence of the correlation between lactate deshydrogenase (LDH) elevation, NO resistance and endothelial dysfunction[10] was reported, so LDH levels were not assessed in our patients.

Interestingly, our observations were made on large vessels, such as common carotid and brachial arteries. This underlines the fact that SCA is not only a microvascular disease but also involves large vessels. This is seen by the occurrence of occlusion of the supraclinoid internal carotid or middle and anterior cerebral arteries, with a risk of stroke of 11% by the age of 20 and evidence of hyperplasia of intima of the supraclinoid internal carotid from autopsy findings.[11]

None of our patients had had a stroke. Transcranial Doppler was performed in 15 out of the 21 children enrolled in our study. Fourteen of them were normal, and one was pathological. This was observed 21 months after the cardiac examination of a 13-year-old child whose FMD was within the values observed in SCA patients.

To summarize, we concluded that the impaired flow-mediated dilation of the brachial artery, probably related to endothelial dysfunction, is observed in children with SCA who lack evidence of arterial stiffness or intimal thickening. This suggests that therapies intended to increase NO, such as arginine administration[12] should be considered in children with SCA.

Mariane de Montalembert, Yacine Aggoun,° Assa Niakate,* Isabelle Szezepanski,° Damien Bonnet°*

**Service de Pédiatrie Générale; °Service de Cardiologie Pédiatrique, Hôpital Necker, Paris, France*

Funding: this study was supported by the Institut National de la Santé et de la Recherche Médicale (RBM-02 12).

Correspondence: Mariane de Montalembert, Service de Pediatrie Generale, Hopital Necker, 149 rue de Sevres, 75015 Paris, France. E-mail: mariane.demontal@nck.aphp.fr

References

1. Reiter CD, Gladwin MT. An emerging role for nitric oxide in sickle cell disease vascular homeostasis and therapy. Curr Opin Hematol 2003; 10:99-107.
2. Kaul DK, Liu XD, Fabry ME, Nagel RL. Impaired nitric-oxide mediated vasodilation in transgenic sickle mouse. Am J Physiol Heart Circ Physiol 2000;278:H799-H1806.
3. Nath KA, Shah V, Haggard JJ, Croatt AJ, Smith LA, Hebbel RP, et al. Mechanisms of vascular instability in a transgenic mouse model of sickle cell disease. Am J Physiol Regul Integr Comp Physiol 2000;279: R1949-R1955.
4. Eberhardt E, McMahon L, Duffy SJ, Steinberg MH, Perrine SP, Loscalzo J, et al. Sickle cell anemia is associated with reduced nitric oxide bioactivity in peripheral conduit and resistance vessels. Am J Hematol 2003;74:104-11.
5. Belhassen L, Pelle G, Sediame S, Bachir D, Carville C, Bucherer C, et al. Endothelial dysfunction in patients with sickle cell disease is related to selective impairment of shear stress-mediated vasodilation. Blood 2001;97:1584-9.
6. Aessopos A, Farmakis D, Tsironi M, Diamanti-Kandarakis E, Matzourani M, Fragodimiri C, et al. Endothelial function and arterial stiffness in sickle-thalassemia patients. Atherosclerosis 2007;427-32.
7. Tounian P, Aggoun Y, Dubern B, Varille V, Guy-Grand B, Sidi D, et al. Presence of increased stiffness of the common carotid artery and endothelial dysfunction in severely obese children: a prospective study. Lancet 2001;358:1400-4.
8. Pegelow CH, Colangelo L, Steinberg M, Wright EC, Smith J, Phillips G, et al. Natural history of blood pressure in sickle cell disease: risks for stroke and death are associated with relative hypertension in sickle cell anemia. Am J Med 1997;102:171-7.
9. Varat MA, Adolph RJ, Fowler NO. Cardiovascular effects of anemia. Am Heart J 1972;83:415-26.
10. Kato GJ, McGowan V, Machado RF, Little JA, Taylor 6th J, Morris CR, et al. Lactate deshydrogenase as a biomarker of hemolysis-associated nitric oxide resistance, priapism, leg ulceration, pulmonary hypertension, and death in patients with sickle cell disease. Blood 2006;107:2279-85.
11. Switzer JA, Hess DC, Nichols FT, Adams RJ. Pathophysiology and treatment of stroke in sickle-cell disease: present and future. Lancet Neurol 2006;5:501-12.
12. Morris CR, Kuypers FA, Larkin S, Sweeters N, Simon J, Vichinsky EP, et al. Arginine therapy: a novel strategy to induce nitric oxide production in sickle cell disease. Br J Haematol 2000;111:498-500.