15. MacDonald JR, Ziman R, Yuen RK, Feuk L, Scherer SW. The Database of Genomic Variants: a curated collection of structural variation in the human genome. *Nucleic Acids Res.* 2014;42(Database issue):D986-D992.

16. Petit A, Trinquand A, Chevret S, et al. Oncogenetic mutations combined with MRD improve outcome prediction in pediatric T-cell acute lymphoblastic leukemia. *Blood.* 2018;131(3):289-300.

17. Bond J, Touzart A, Leprêtre S, et al. DNMT3A mutation is associated with increased age and adverse outcome in adult T-cell acute lymphoblastic leukemia. *Haematologica.* 2019;104(8):1617-1625.

18. Simonin M, Schmidt A, Bontoux C, et al. Oncogenetic landscape and clinical impact of IDH1 and IDH2 mutations in T-ALL. *J Hematol Oncol.* 2021;14(1):74-80.

19. Hientz K, Mohr A, Bhakta-Guha D, Efferth T. The role of p53 in cancer drug resistance and targeted chemotherapy. *Oncotarget.* 2016;8(5):8921-8946.

20. Mantovani F, Collavin L, Del Sal G. Mutant p53 as a guardian of the cancer cell. *Cell Death Differ.* 2019;26(2):199-212.

21. Lambert JMR, Gorzov P, Veprintsev DB, et al. PRIMA-1 reactivates mutant p53 by covalent binding to the core domain. *Cancer Cell.* 2009;15(5):376-388.

22. Bykov VJN, Issaeva N, Shilov A, et al. Restoration of the tumor suppressor function to mutant p53 by a low-molecular-weight compound. *Nat Med.* 2002;8(3):282-288.

23. Cluzeau T, Sebert M, Rahmé R, et al. Eprenetapopt plus azacitidine in TP53-mutated myelodysplastic syndromes and acute myeloid leukemia: a phase II study by the Groupe Francophone des Myélodysplasies (GFM). *J Clin Oncol.* 2021;39(14):1575-1583.

24. Demir S, Boldrin E, Sun Q, et al. Therapeutic targeting of mutant p53 in pediatric acute lymphoblastic leukemia. *Haematologica.* 2020;105(1):170-181.

25. Sallman DA, DeZern AE, Garcia-Manero G, et al. Eprenetapopt (APR-246) and azacitidine in TP53-mutant myelodysplastic syndromes. *J Clin Oncol.* 2021;39(14):1584-1594.

26. Mishra A, Tamari R, DeZern AE, et al. Eprenetapopt plus azacitidine after allogeneic hematopoietic stem-cell transplantation for TP53-mutant acute myeloid leukemia and myelodysplastic syndromes. *J Clin Oncol.* 2022;40(34):3985-3993.

https://doi.org/10.1182/blood.2022017755

TO THE EDITOR:

Diffuse myocardial fibrosis occurs in young patients with sickle cell anemia despite early disease-modifying therapy

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We read with great interest the recent report from Niss et al.[1] Our group has recently reviewed data from a similar population at our institution, which does not support the conclusion of Niss et al that diffuse myocardial fibrosis is prevented by early initiation of disease-modifying therapy (DMT).

Cardiovascular complications are the leading cause of early mortality in patients with sickle cell anemia (SCA).[2] Yet, the pathobiology of sickle cell cardiomyopathy is incompletely understood and likely multifactorial.[3] Chronic anemia leads to global cardiac enlargement and myocardial hypertrophy. Global cardiac chamber dilation is a common feature in patients with SCA, and enlarged left atrial (LA) volume is a marker of diastolic dysfunction in SCA.[4] Repetitive microvascular ischemic insults and associated reperfusion injury may contribute to diffuse myocardial fibrosis and myocardial remodeling in individuals with SCA.[5-7]

Cardiovascular magnetic resonance (CMR) imaging is the only noninvasive method capable of accurately and reproducibly assessing heart size and function as well as characterizing the myocardium. Emerging data suggest that patients with SCA develop diffuse myocardial fibrosis,[1,5-7] detected by quantifying the myocardial extracellular volume (ECV). Increased ECV fraction correlates strongly with histologically quantified myocardial fibrosis.[8] Our goal was to estimate the prevalence of diffuse myocardial fibrosis in a cohort of young individuals with SCA.

At our institution, all patients with SCA undergo cardiac surveillance starting at 16 years of age, which comprises a clinical assessment by a cardiologist and an echocardiogram. The presence of LA dilation (LA volume > 34 mL/m^2), left ventricular dilation, or prominent trabeculations seen by echocardiogram led to patients being referred for CMR (detailed echocardiographic data in supplemental Table 1, on the *Blood* website). We performed a retrospective chart review of pediatric and young adult patients with SCA who underwent CMR at our institution between 2020 and 2021. Demographic and clinical information was obtained from reviewing the electronic medical record. A waiver of the requirement for informed consent was granted by the institutional review board for use of deidentified data.

CMR was performed on a 1.5T scanner (Avanto Fit; Siemens Medical Solutions, Inc, Erlangen, Germany). The protocol

Table 1. Demographic and clinical characteristics overall and by use of early therapy (<6 years of age)

	Overall (n = 31), n (%) or median (IQR)	No early therapy (n = 8), n (%) or median (IQR)	Early therapy (<6 y) (n = 20), n (%) or median (IQR)	Normal values
Age at CMR (y)	16.7 (14.7-18.0)	16.8 (15.2-19.0)	17.0 (14.4-17.9)	
Sex				
Male	16 (51.6)	2 (25.0)	12 (60.0)	
Female	15 (48.4)	6 (75.0)	8 (40.0)	
Weight (kg)	62.0 (46.2-69.5)	64.8 (50.0-73.1)	58.2 (45.6-70.1)	
Height (cm)	162.5 (152.9-171.8)	161.9 (155.8-166.2)	164.4 (152.9-176.1)	
BSA (m^2)	1.7 (1.4-1.8)	1.7 (1.5-1.8)	1.6 (1.4-1.9)	
Received hydroxyurea in the past 2 y				
Yes	11 (35.5)	3 (37.5)	8 (40.0)	
No	20 (64.5)	5 (62.5)	12 (60.0)	
Received chronic transfusions in the past 2 y				
Yes	14 (45.2)	3 (37.5)	11 (55.0)	
No	17 (54.8)	5 (62.5)	9 (45.0)	
Receiving beta-blockers				
Yes	10 (32.3)	0 (0.0)	8 (40.0)	
No	21 (67.7)	8 (100.0)	12 (60.0)	
Laboratory data				
Hemoglobin (g/dL)*	8.9 (8.2-9.9)	9.1 (8.2-9.7)	9.0 (8.2-10.0)	12-16
Hematocrit (%)	25.6 (23.6-27.8)	26.0 (23.0-30.2)	25.8 (23.7-27.4)	36-46
Reticulocyte absolute ($/mm^3$)*	0.208 (0.139-0.338)	0.157 (0.125-0.236)	0.252 (0.155-0.358)	0.021-0.085
LDH*	473 (333-573)	452 (389-504)	464 (329-629)	94-260
Bilirubin (total)*	2.7 (1.9-3.5)	1.9 (1.5-2.5)	3.2 (2.1-3.6)	0.0-1.0
Cardiac MRI data				
RVEDVi (mL/m^2)	108.1 (95.3-121.3)	91.4 (84.2-101.2)	110.3 (104.4-123.4)	82 ± 17 (48-123)[11]
RVESVi (mL/m^2)	44.3 (36.7-54.9)	38.0 (35.3-41.0)	46.7 (39.1-56.4)	34 ± 11 (13-59)[11]
RVEF (%)	56.8 (53.7-61.4)	58.3 (55.5-61.8)	56.7 (53.9-61.6)	58 ± 8 (42-74)[11]
RVCi ($L/min/m^2$)	4.2 (3.7-5.4)	3.8 (3.3-4.2)	4.7 (4.1-5.7)	2.9 ± 0.7 (1.5-4.5)[11]
LVEDVi (mL/m^2)	109.2 (95.4-123.1)	93.8 (75.7-113.4)	118.7 (99.8-125.9)	74 ± 15 (47-107)[11]
LVESVi (mL/m^2)	48.2 (37.8-52.1)	38.2 (31.8-50.6)	50.3 (43.1-55.1)	27 ± 9 (10-47)[11]
LV mass indexed value (g/m^2)	64.5 (54.0-75.5)	56.2 (44.9-70.4)	65.5 (59.9-77.2)	51 ± 10 (30-75)[11]
LVEF (%)	57.4 (55.3-59.8)	58.0 (57.0-59.6)	57.2 (55.1-59.8)	64 ± 7 (51-79)[11]
LVCi ($L/min/m^2$)	4.2 (3.9-5.5)	3.9 (3.6-4.2)	4.8 (4.1-5.8)	3.1 ± 0.6 (1.9-4.3)[11]
LA volume (mL/m^2)	40.5 (30.0-51.5)	36.8 (24.8-46.0)	42.4 (32.4-59.0)	34 ± 10 (13-53)[12]
Native T1 (ms)	1029 (1004-1051)	1020 (986-1057)	1028 (1011-1036)	1005 ± 25 (959-1052)†
Native T2 (ms)*	46.5 (44.9-48.3)	46.6 (45.9-47.3)	46.6 (45.0-48.5)	44 ± 2 (40-50)†
Native T2 > 50	2 (6.7)	1 (12.5)	1 (5.3)	
ECV (%)	32.0 (29.0-34.0)	31.0 (29.8-34.0)	31.5 (29.0-34.0)	≥18 y = 25 ± 3[13]; <18 y = 21 ± 2[9]
ECV > 25	31 (100.0)	8 (100.0)	20 (100.0)	
ECV ≥ 30	22 (71.0)	6 (75.0)	14 (70.0)	
$T2^*$ (ms)	35.0 (32.2-39.0)	33.8 (31.2-34.8)	35.9 (33.6-40.4)	>20 ms

Three patients in the cohort did not receive any DMT.

BSA, body surface area; LDH, lactate dehydrogenase; LVCi, left ventricular cardiac index; LVEDVi, left ventricular end-diastolic volume (indexed); LVEF, left ventricle ejection fraction; LVESVi, left ventricular end-systolic volume (indexed); MRI, magnetic resonance imaging; RVCi, right ventricular cardiac index; RVEDVi, right ventricular end-diastolic volume (indexed); RVEF, right ventricle ejection fraction; RVESVi, right ventricular end-systolic volume (indexed).

*Two subjects are missing hemoglobin, LDH, and bilirubin; 3 subjects are missing reticulocyte absolute; 1 subject is missing native T2.

†St. Jude Children's Research Hospital 1.5 T normal references.

included ventricular long- and short-axis planes for chamber sizes, ejection fraction, and myocardial mass. ECV was measured from T1 maps acquired with a modified Look-Locker inversion recovery sequence in short-axis pre- and post-contrast.[9] T2 and T2* maps were obtained before contrast administration. Regions of interest for analysis were drawn in the interventricular septum at the midmyocardial level (supplemental Figure 1). All planimetric and region-of-interest analyses were performed with cvi42 (Circle Imaging; Alberta, Canada) by a single technologist (C.G.) and reviewed for accuracy by a cardiac imaging specialist (C.E.M., A.M., or J.N.J.). Myocardial MRI-T2* values were obtained from clinical reports and were measured as previously described.[10] Hematocrit was collected within 24 hours of the patients undergoing a cardiac MRI.

Thirty-one patients with SCA (SS genotype) underwent CMR during the study period (Table 1). The median age at the time of CMR was 16.7 years. At the time of CMR, 11 (36%) patients were receiving hydroxyurea for a median treatment duration of 3.3 years (interquartile range [IQR] 0.1-9.3 years), and 14 (45%) patients were receiving chronic monthly transfusions (CTXN) for a median treatment duration of 1.1 years (IQR 0.0-3.5 years). Some patients switched from hydroxyurea to CTXN or vice versa, therefore the collective median duration of any DMT was 9.6 years (IQR 2.2-11.8 years). In the 2 years preceding the CMR, the frequency of SCA acute complications was as expected for a DMT-exposed young population: 5 (16.1%) patients had 1 to 2 episodes of acute chest syndrome and 10 (32.3%) patients had 1 or more episodes of vaso-occlusive pain crises. The remaining participants had no vaso-occlusive events in the 2 years before undergoing CMR.

Many patients had left ventricle (LV, 25 [81%]) and right ventricle (20 [65%]) enlargement. Thirteen (42%) patients also had LA enlargement. Most patients had elevated LV cardiac index (24 [77%]). Both LA volume and LV cardiac index were elevated despite DMT, whether hydroxyurea, CTXN, or a combination of both, though this was not statistically significant (supplemental Figure 2).

All patients in our study had increased ECV with a median ECV of 32% (IQR 29%-34%, reference normal 20.8% ± 2.4% in children[9] and 25.9% ± 0.4% in adults[14]). This is consistent with prior studies in a similar population.[5,7] Duration of exposure to DMT was not associated with ECV (slope of ECV percent vs years on DMT: −0.07, P = .5). All patients had normal myocardial MRI-T2* values indicating the lack of significant iron overload of the myocardium.

Our study highlights that diffuse myocardial fibrosis is nearly universal in children and young adults with SCA, even those patients with normal cardiac size and function, confirming prior reports.[5,15] In contrast to Niss et al, we found no difference in the ECV in patients who began DMT early (less than 6 years of age) vs late in life (at least 6 years or after) despite the 2 cohorts being similar in demographics and treatment received. Although mean ECV was comparable in the early therapy

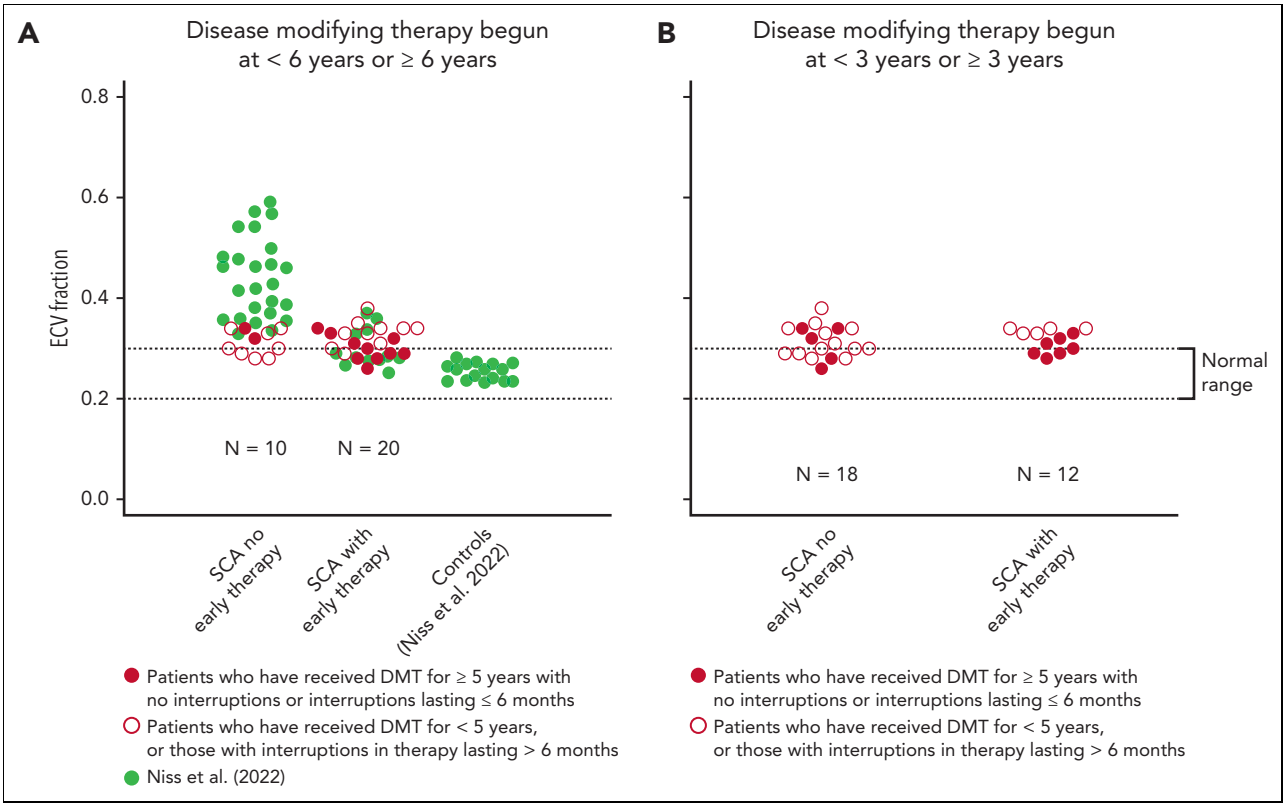

Figure 1. ECV fraction is abnormally elevated in nearly all patients in our study (reference normal 20.8% ± 2.4% in children[9] and 25.9% ± 0.4% in adults[14]). (A) ECV was not different among children who started DMT at less than 6 years of age (SCA with early therapy, n = 20) or after (SCA no early therapy, n = 8). Datapoints from our cohort are overlayed in solid red circles or open circles. Three patients in our cohort did not receive any DMT. Solid green circles represent data points from Niss et al for comparison.[1] (B) Starting DMT at an even earlier age (at less than 3 years of age) has no further protective effect, and the ECV of the 2 cohorts is comparable.

groups in our study and the one described by Niss et al (32% vs 30%), ECV in the no early therapy group was lower in our study compared with Niss et al (31% vs 44%). Participants in the Niss et al study were older (median age in the early therapy and no early therapy groups was 18 and 19 years respectively, compared with 17 and 16.8 years, respectively, in our study). In the cohort described by Niss et al, the median age of initiation of any DMT in the early therapy group was 2.9 years. In the no early therapy group, hydroxyurea was prescribed at a mean age of 11.3 years (patients receiving CTXN were excluded). In comparison, in our study, the median age of initiation of any DMT in the early therapy group was 2.6 years and in the no early therapy group was 9 years. The median duration of treatment with DMT in the Niss et al study was 13.7 years in the group that received early therapy. In our study, the median duration of treatment with DMT in the group that received early therapy was 10.5 years, which is slightly shorter but still comparable to the Niss et al study. It is important to note, though, that patients who subsequently received either hydroxyurea or CTXN were included in our no early therapy group, in contrast to exclusion of individuals who ever received CTXN in the no early therapy group in the Niss et al study. Even after exclusion of the patients who had received CTXN from the no early therapy group, our results remained the same—we found no significant differences in the ECV of those patients who received early therapy vs those who did not (supplemental Figure 3). We further explored whether starting DMT at an even earlier age would have a further protective effect and dichotomized our cohort into those who started DMT before or after 3 years of age and found no difference in the ECV of the 2 groups (Figure 1B, supplemental Figure 4). Lastly, even though 40% of patients in the early therapy group were receiving beta-blockers for management of diastolic dysfunction on echocardiogram and higher resting heart rates on 24-hour Holter monitors, the echocardiographic findings between the 2 groups were not significantly different. Our cohort of patients are younger with similar cardiac index and LV end-diastolic volumes but smaller LA volumes compared with the Niss et al cohort.[15] Only 8 (25.8%) patients had LA volume >50 mL/m^2 in our cohort, potentially reflecting less diastolic dysfunction compared with the Niss et al cohort.[15]

We cannot conclusively explain why our results differed from Niss et al. In general, increased ECV may be partially related to increased LV mass and may explain a portion of the elevated ECV seen in patients with SCA. The lower ECV observed in the no early therapy group compared with the Niss et al cohort may be related to smaller LA volumes, reflecting potentially less diastolic dysfunction. Although our no early therapy group included CTXN, excluding patients who had ever received CTXN from the analysis did not significantly change the results. Nearly all patients had abnormal ECV, suggesting that even CTXN is not completely protective. Our findings suggest that damage to the myocardium occurs before symptoms or other clinical markers of cardiac dysfunction and that our current screening protocols may require revision to include earlier cardiac assessments. Although we cannot conclude that early DMT is protective against the development of myocardial fibrosis, the presence of nearly universal diffuse myocardial fibrosis will require further longitudinal evaluation. CMR provides additional clinically relevant information that may help guide treatment in patients with SCA.

Acknowledgments

The authors thank Keith A. Laycock for scientific editing of the manuscript. The authors also thank their colleagues, advanced practice providers, nurses, and other health care professionals who participated in patient care. They also thank the parents who entrusted the care of their children to them.

This work was supported by the American Society of Hematology (Scholar Award to A.S.) and the American Lebanese Syrian Associated Charities (ALSAC).

Authorship

Contribution: C.E.M. and A.S. are the principal investigators of this study and developed the study design, contributed to data interpretation, and reviewed and edited the manuscript; J.N.J. contributed to the data collection, analysis, interpretation, and writing the manuscript; A.M. contributed to the data collection, analysis, interpretation, and editing the manuscript; S.S. contributed to the data analysis, interpretation, and editing the manuscript; C.G. contributed to data collection; G.B., J.S.H., P.R., and J.A.T. contributed to study design and editing the manuscript; and all authors have reviewed and approved the manuscript.

Conflict-of-interest disclosure: A.S. has received consultant fees from Spotlight Therapeutics, Medexus Inc, and Vertex Pharmaceuticals. He has also received research funding from CRISPR Therapeutics and honoraria from Vindico Medical Education. A.S. is the St. Jude Children's Research Hospital site principal investigator of clinical trials for genome editing of sickle cell disease sponsored by Vertex Pharmaceuticals/CRISPR Therapeutics (NCT03745287) and by Novartis (NCT04443907). The industry sponsors provide funding for the clinical trial, which includes salary support paid to A.S.'s institution. The remaining authors declare no competing financial interests.

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Footnotes

Submitted 26 August 2022; accepted 14 November 2022; prepublished online on *Blood* First Edition 28 November 2022

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The online version of this article contains a data supplement.

There is a *Blood* Commentary on this article in this issue.

REFERENCES

1. Niss O, Detterich JA, Wood JC, et al. Early initiation of disease-modifying therapy can impede or prevent diffuse myocardial fibrosis in sickle cell anemia. *Blood*. 2022;140(11):1322-1324.
2. Hamideh D, Alvarez O. Sickle cell disease related mortality in the United States (1999-2009). *Pediatr Blood Cancer*. 2013;60(9):1482-1486.
3. Niss O, Quinn CT, Lane A, et al. Cardiomyopathy with restrictive physiology in sickle cell disease. *JACC Cardiovasc Imaging*. 2016;9(3):243-252.
4. Nagueh SF. Diastology: 2020—a practical guide. *Echocardiography*. 2020;37(11):1919-1925.

5. Alsaied T, Niss O, Tretter JT, et al. Left atrial dysfunction in sickle cell anemia is associated with diffuse myocardial fibrosis, increased right ventricular pressure and reduced exercise capacity. *Sci Rep*. 2020;10(1):1767.

6. Bakeer N, James J, Roy S, et al. Sickle cell anemia mice develop a unique cardiomyopathy with restrictive physiology. *Proc Natl Acad Sci U S A*. 2016;113(35):E5182-E5191.

7. Alsaied T, Niss O, Powell AW, et al. Diastolic dysfunction is associated with exercise impairment in patients with sickle cell anemia. *Pediatr Blood Cancer*. 2018;65(8):e27113.

8. Sibley CT, Noureldin RA, Gai N, et al. T1 Mapping in cardiomyopathy at cardiac MR: comparison with endomyocardial biopsy. *Radiology*. 2012;265(3):724-732.

9. Pagano JJ, Yim D, Lam CZ, et al. Normative data for myocardial native t1 and extracellular volume fraction in children. *RadiolCardiothorac Imaging*. 2020;2(4):e190234.

10. Hankins JS, McCarville MB, Hillenbrand CM, et al. Ventricular diastolic dysfunction in sickle cell anemia is common but not associated with myocardial iron deposition. *Pediatr Blood Cancer*. 2010;55(3):495-500.

11. Kawel-Boehm N, Hetzel SJ, Ambale-Venkatesh B, et al. Reference ranges ("normal values") for cardiovascular magnetic resonance (CMR) in adults and children: 2020 update. *J Cardiovasc Magn Reson*. 2020;22(1):87.

12. Funk S, Kermer J, Doganguezel S. Quantification of the left atrium applying cardiovascular magnetic resonance in clinical routine. *Scand Cardiovasc J*. 2018;52(2):85-92.

13. Sado DM, Flett AS, Banypersad SM, et al. Cardiovascular magnetic resonance measurement of myocardial extracellular volume in health and disease. *Heart*. 2012;98(19):1436-1441.

14. Gottbrecht M, Kramer CM, Salerno M. Native T1 and extracellular volume measurements by cardiac MRI in healthy adults: a meta-analysis. *Radiology*. 2019;290(2):317-326.

15. Niss O, Fleck R, Makue F, et al. Association between diffuse myocardial fibrosis and diastolic dysfunction in sickle cell anemia. *Blood*. 2017;130(2):205-213.

https://doi.org/10.1182/blood.2022018209