4. Minniti CP, Zaidi AU, Nouraie M, et al. Clinical predictors of poor outcomes in patients with sickle cell disease and COVID-19 infection. *Blood Adv*. 2021;5:207-215.
5. Motta I, Migone de Amicis M, Pinto VM, et al. SARS-CoV-2 infection in beta thalassemia: preliminary data from the Italian experience. *Am J Hematol*. 2020;95:E198-E199.
6. Trafane LF, da Costa VA, da Silva Santos Duarte A, et al. Low SARS-CoV-2 seroprevalence in a cohort of Brazilian sickle cell disease patients: possible effects of emphasis on social isolation for a population initially considered to be at very high risk. *eJHaem*. 2021;2:478-482. doi:10.1002/jha2.254

Received: 2 November 2021 | Revised: 25 November 2021 | Accepted: 29 November 2021

DOI: 10.1002/ajh.26428

Risk of mortality from anemia and iron overload in nontransfusion-dependent β-thalassemia

To the Editor:

Ineffective erythropoiesis in patients with nontransfusion-dependent β-thalassemia (NTDT) leads to chronic anemia that does not necessarily require lifelong transfusion therapy for survival.[1] Nonetheless, chronic anemia in these patients is associated with significant morbidity, especially in patients with a hemoglobin level lower than 10 g/dL.[2] Hemoglobin variations greater than 1 g/dL have also been shown to modify morbidity risk.[3] Beyond the use of transfusions in specific clinical settings, there are currently no approved agents for the management of anemia in NTDT. Ineffective erythropoiesis can also lead to considerable iron overload due to hepcidin dysregulation and increased intestinal iron absorption.[4] A serum ferritin level greater than 800 ng/mL is also associated with an increased risk of morbidity and is an indication for the use of iron chelation therapy.[5] Such clinical complications in NTDT are often serious and involve various organ systems including hepatic, endocrine, and vascular disease.[6] Despite the abundance of reports highlighting anemia and iron overload as the hallmarks of morbidity in NTDT, data on their association with long-term mortality outcomes remain limited.

For this work, we used data from an International Health Repository (IHR) formed by 13 international thalassemia reference centers from the US and seven countries in Europe, the Middle East, and Asia.[7] The IHR was established and approved on May 25, 2017 by the Italian Ethical Committee (EudraCT and Sponsor's Protocol Code Numbers: 2017-004457-17 and 143AOR2017). All data were anonymized and added to the repository following informed consent by patients or their legal representatives in case of death. The IHR database includes all β-thalassemia patients who have attended participating centers from January 1, 1997 onward. We analyzed data from all patients with NTDT (defined as previously described[8]) who had not transitioned to regular transfusion programs and who had documented hemoglobin and serum ferritin levels. Patients were historically followed from birth up to December 31, 2019, death, or loss to follow-up. For each patient, we retrieved data on age and status at the last observation, sex, splenectomy status, iron chelation status, hemoglobin level, and serum ferritin level at the last observation. Hemoglobin and serum ferritin levels represented the average of all measurements done during the year of last observation.

A total of 415 patients (48.7% females) were included in the analysis (Table S1). The median age at last observation (follow-up time) was 30.1 years (interquartile range [IQR]: 23.6–44.2). The majority were splenectomized ($n = 243$, 58.6%) and received iron chelation therapy ($n = 379$, 91.3%). The median age at the start of iron chelation therapy was 7 years (IQR: 4.3–14). At last observation, the majority of patients were on deferoxamine (49%), followed by deferiprone (23.5%), deferasirox (22.5%), and deferoxamine + deferiprone combination (4.9%) therapy. The mean hemoglobin level was 9.2 ± 1.0 g/dL (range: 6–15) with 339 (81.7%) patients having a hemoglobin level ≤ 10 g/dL. The median serum ferritin level was 960 ng/mL (IQR: 500–2843) with 235 (56.6%) patients having a serum ferritin level > 800 ng/mL. A total of 185 patients (44.6%) had both a hemoglobin level ≤ 10 g/dL and a serum ferritin level > 800 ng/mL.

Thirty-two patients died during the observation period, giving a crude mortality rate of 7.7% (95% confidence interval [CI]: 5.3–10.7). Recorded causes of death included cardiovascular disease ($n = 17$), infection ($n = 2$), hepatic failure ($n = 1$), renal failure ($n = 1$), cancer ($n = 1$), and other disease complications ($n = 10$). The median age at death was 24.1 years (IQR: 28.3–61.9; 37.5% females). Survival was significantly worse in patients with a hemoglobin level ≤ 10 g/dL than those with > 10 g/dL (Log-rank test Chi-square: 4.259, $p = .039$, Figure 1A). Survival was also significantly worse in patients with a serum ferritin level > 800 ng/mL than those with ≤ 800 ng/mL (log-rank test Chi-square: 24.379, $p < .001$, Figure 1B). Finally, survival was significantly worse in patients with both a hemoglobin level ≤ 10 g/dL and a serum ferritin level > 800 ng/mL than those with either a hemoglobin level ≤ 10 g/dL or a serum ferritin level > 800 ng/mL and those with both a hemoglobin level > 10 g/dL and a serum ferritin level ≤ 800 ng/mL (Log-rank test Chi-square: 33.728, $p < .001$, Figure 1C).

We constructed a multivariate Cox regression analysis including hemoglobin level (≤ 10 vs. > 10 g/dL), serum ferritin level (> 800 vs. ≤ 800 ng/mL), sex, splenectomy, and iron chelation status. A hemoglobin level ≤ 10 g/dL was independently associated with a 7.6-fold increase in the risk of mortality (hazard ratio [HR]: 7.632, 95% CI: 1.036–56.219, $p = .046$). A serum ferritin level > 800 ng/mL was also independently associated with a 9.8-fold increase in the risk of mortality (HR: 9.755, 95% CI: 3.368–28.257, $p < .001$).

Our study furthers our understanding of the detrimental effects of anemia and iron overload in NTDT and highlights an increased risk of mortality in patients with clinically relevant thresholds. Our work is limited by our ability to only analyze a subset of patients with documented hemoglobin and serum ferritin levels, which could have introduced a selection bias for patients with severe disease requiring regular follow-up, as is also evident from a higher and earlier mortality in this subset of patients compared to our overall cohort.[8] Prospective studies are merited in such context, as they could also assess the

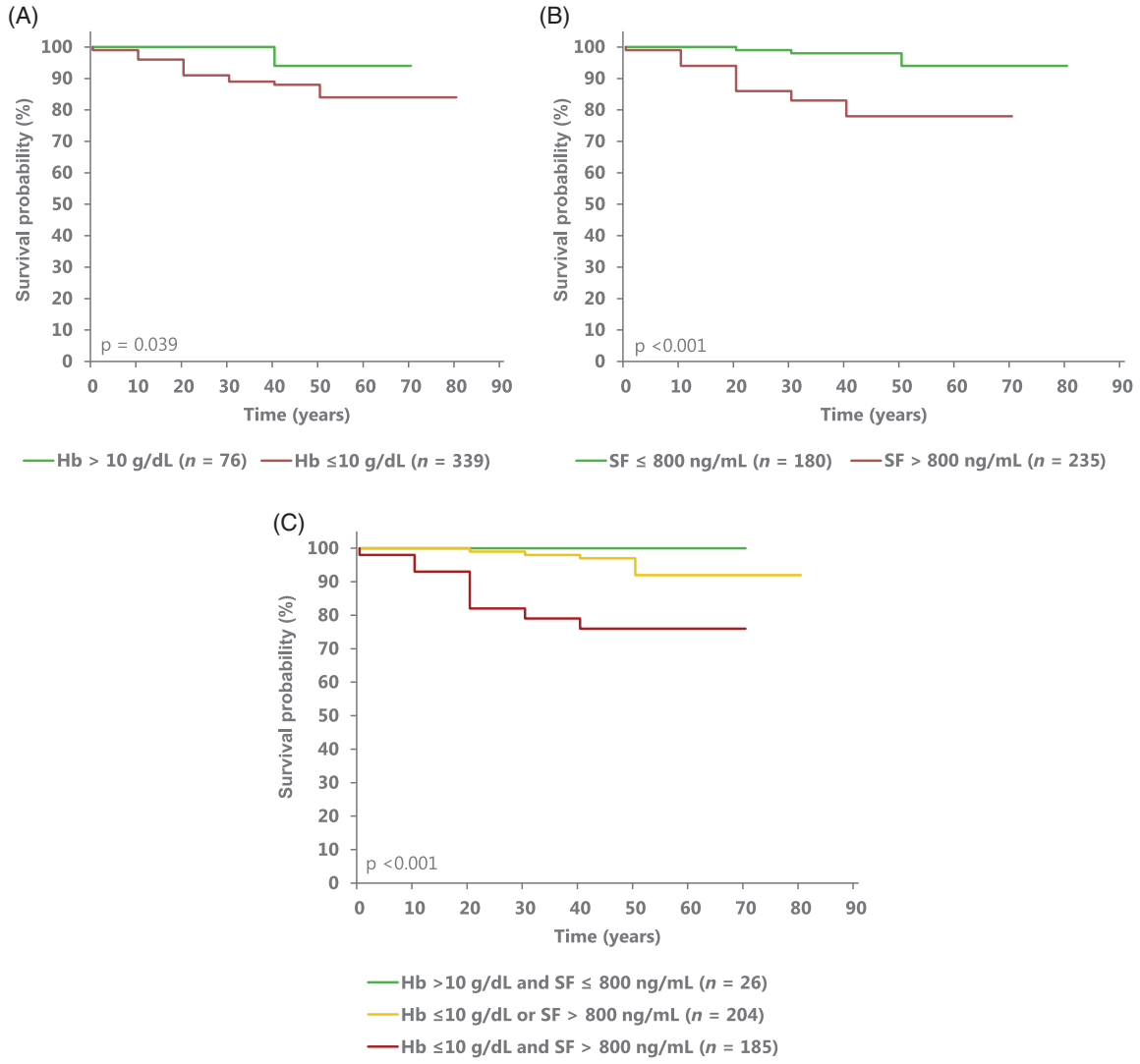

FIGURE 1 Kaplan–Meier survival curves. (A) Mortality according to hemoglobin level, (B) mortality according to serum ferritin level, and (C) mortality according to both hemoglobin and serum ferritin levels. Hb, hemoglobin; SF, serum ferritin

effects of longitudinal variations over time and not only spot measurements which may not be ideally representative. The risks of mortality from low hemoglobin and high serum ferritin levels seem to be additive, which suggests a need to address both factors with prompt and comprehensive conventional management. We have previously highlighted a role for regular transfusion and iron chelation therapy in this patient population,[8] but data from novel agents targeting anemia and iron overload are eagerly awaited.[9]

ACKNOWLEDGMENTS

The authors would like to thank all patients for agreeing to participate in this study. The support by Foundation Franco and Piera Cutino is appreciated.

CONFLICT OF INTEREST

KMM has been or is a consultant for Novartis, Celgene Corp (Bristol Myers Squibb), Agios Pharmaceuticals, CRISPR Therapeutics, and Vifor Pharma. AMe received speakers' honoraria from Chiesi Farmaceutici S.p.A. EVl received honoraria from DEMO S.A. Pharmaceutical Industry and Novartis. AP is the principal investigator of the MIOT project that receives "nonprofit support" from industrial sponsorships (Chiesi Farmaceutici S.p.A. and Bayer) and she received speakers' honoraria from Chiesi Farmaceutici S.p.A. ATT has been or is consultant for Novartis, Celgene Corp (Bristol Myers Squibb), Vifor Pharma, Silence Therapeutics, and Ionis Pharmaceuticals; and received research funding from Novartis, Celgene Corp (Bristol Myers Squibb), La Jolla Pharmaceutical Company, Roche, Protagonist Therapeutics, and Agios Pharmaceuticals. VGS serves as an advisor to and/or has equity in Novartis, Forma, Cellarity, Ensoma, and Branch Biosciences. AMa has been or is a member of advisory boards for Novartis, Celgene Corp (Bristol Meyers Squibb), and Bluebird Bio. The remaining authors have no conflicts of interest to disclose.

AUTHOR CONTRIBUTION

Study design: Khaled M. Musallam, Angela Vitrano, Aurelio Maggio. *Data collection*: Angela Vitrano, Antonella Meloni, Sebastiano Addario Pollina, Mehran Karimi, Amal El-Beshlawy, Mahmoud Hajipour, Vito Di Marco,

Saqib Hussain Ansari, Aldo Filosa, Paolo Ricchi, Adriana Ceci, Shahina Daar, Efthymia Vlachaki, Sylvia Titi Singer, Zaki A. Naserullah, Elliott Vichinsky. *Data analysis*: Khaled M. Musallam, Angela Vitrano. *Manuscript Drafting*: Khaled M. Musallam, Angela Vitrano, Aurelio Maggio. *Data interpretation and manuscript review for intellectual content*: all authors. All authors gave final approval for submission.

DATA AVAILABILITY STATEMENT

Data were collected and stored on the IHR electronic platform (http://www.sanitasicilia.eu/IWG), and can be available upon request from the corresponding author.

Khaled M. Musallam[1], Angela Vitrano[2], Antonella Meloni[3], Sebastiano Addario Pollina[4], Mehran Karimi[5], Amal El-Beshlawy[6], Mahmoud Hajipour[7], Vito Di Marco[8], Saqib Hussain Ansari[9], Aldo Filosa[10], Paolo Ricchi[10], Adriana Ceci[11], Shahina Daar[12], Efthymia Vlachaki[13], Sylvia Titi Singer[14], Zaki A. Naserullah[15], Alessia Pepe[3], Salvatore Scondotto[4], Gabriella Dardanoni[4], Fedele Bonifazi[11], Vijay G. Sankaran[16,17,18], Elliott Vichinsky[14], Ali T. Taher[19], Aurelio Maggio[2], International Working Group on Thalassemia (IWG-THAL)

[1]*Thalassemia Center, Burjeel Medical City, Abu Dhabi, United Arab Emirates*
[2]*Campus of Haematology Franco and Piera Cutino, AOOR Villa Sofia-V, Cervello, Palermo, Italy*
[3]*MRI Unit, Fondazione G. Monasterio CNR-Regione Toscana, Pisa, Italy*
[4]*D.A.S.O.E., Regione Siciliana, Palermo, Italy*
[5]*Haematology Research Center, Shiraz University of Medical Sciences, Shiraz, Iran*
[6]*Department of Pediatric Haematology, Faculty of Medicine, Cairo University, Cairo, Egypt*
[7]*Pediatric Gastroenterology, Hepatology and Nutrition Research Center, Research Institute for Children's Health, Shahid Beheshti University of Medical Sciences, Tehran, Iran*
[8]*Sezione di Gastroenterologia e Epatologia, Dipartimento Biomedico di Medicina Interna e Specialistica, University of Palermo, Palermo, Italy*
[9]*Children's Hospital Karachi (CHK), Karachi, Pakistan*
[10]*Rare Blood Cell Disease Unit, "Cardarelli" Hospital, Naples, Italy*
[11]*Fondazione per la Ricerca Farmacologica Gianni Benzi Onlus, Valenzano (BA), Italy*
[12]*Department of Haematology, College of Medicine and Health Sciences, Sultan Qaboos University, Sultanate of Oman; Wallenberg Research Centre, Stellenbosch Institute for Advanced Study, Stellenbosch University, Stellenbosch, South Africa*
[13]*Thalassaemia Unit, Ippokratio University Hospital, Thessaloniki, Greece*
[14]*Division of Hematology-Oncology, Department of Pediatrics, University of California San Francisco, UCSF Benioff Children's Hospital Oakland, Oakland, California, USA*
[15]*Dammam Maternity and Child Hospital, Dammam, Saudi Arabia*
[16]*Division of Hematology/Oncology, Boston Children's Hospital and Department of Pediatric Oncology, Dana-Farber Cancer Institute, Harvard Medical School, Boston, Massachusetts, USA*
[17]*Broad Institute of MIT and Harvard, Cambridge, Massachusetts, USA*
[18]*Harvard Stem Cell Institute, Cambridge, Massachusetts, USA*
[19]*Department of Internal Medicine, American University of Beirut Medical Center, Beirut, Lebanon*

Correspondence
Aurelio Maggio, Campus of Haematology Franco and Piera Cutino, AOOR Villa Sofia-V. Cervello, Palermo, Italy.
Email: md.amaggio@gmail.com

Khaled M. Musallam and Angela Vitrano contributed equally as co-first authors.

This study did not receive any funding.

ORCID

Khaled M. Musallam https://orcid.org/0000-0003-3935-903X
Antonella Meloni https://orcid.org/0000-0002-2284-8585
Vito Di Marco https://orcid.org/0000-0001-6397-4206
Saqib Hussain Ansari https://orcid.org/0000-0002-4209-6560
Paolo Ricchi https://orcid.org/0000-0001-7361-3308
Ali T. Taher https://orcid.org/0000-0001-8515-2238
Aurelio Maggio https://orcid.org/0000-0002-9601-900X

REFERENCES

1. Taher AT, Musallam KM, Cappellini MD. beta-Thalassemias. *N Engl J Med*. 2021;384:727-743.
2. Musallam KM, Cappellini MD, Daar S, Taher AT. Morbidity-free survival and hemoglobin level in non-transfusion-dependent beta-thalassemia: a 10-year cohort study. *Ann Hematol*. 2021.
3. Musallam KM, Cappellini MD, Taher AT. Variations in hemoglobin level and morbidity burden in non-transfusion-dependent beta-thalassemia. *Ann Hematol*. 2021;100:1903-1905.
4. Rivella S. Iron metabolism under conditions of ineffective erythropoiesis in beta-thalassemia. *Blood*. 2019;133:51-58.
5. Musallam KM, Cappellini MD, Daar S, et al. Serum ferritin level and morbidity risk in transfusion-independent patients with beta-thalassemia intermedia: the ORIENT study. *Haematologica*. 2014;99:e218-e221.
6. Vichinsky E. Non-transfusion-dependent thalassemia and thalassemia intermedia: epidemiology, complications, and management. *Curr Med Res Opin*. 2016;32:191-204.
7. Vitrano A, Meloni A, Addario Pollina W, et al. A complication risk score to evaluate clinical severity of thalassaemia syndromes. *Br J Haematol*. 2021;192:626-633.
8. Musallam KM, Vitrano A, Meloni A, et al. Survival and causes of death in 2,033 patients with non-transfusion-dependent beta-thalassemia. *Haematologica*. 2021;106:2489-2492.
9. Musallam KM, Bou-Fakhredin R, Cappellini MD, Taher AT. 2021 update on clinical trials in beta-thalassemia. *Am J Hematol*. 2021;96:1518-1531.

SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.