

Anemia and thalassemia in the Kui (Suay) elderly living in Sisaket Province located at the lower Northeastern Thailand

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KEYWORDS

Anemia;
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ABSTRACT

This study investigated the prevalence of anemia and thalassemia in the Suay elderly population. The study conducted on 63 apparently healthy individuals with an age range of 60-85 years. Blood samples were collected to determine the following laboratory tests; red blood cell (RBC) parameters, hemoglobin analysis, and globin genotyping. Based on established WHO criteria, the overall prevalence of anemia was found to be 47.6%. The astoundingly high prevalence of thalassemia was found in non-anemic subjects for 84.8% and anemic subjects for 93.3%. As expected, the high frequency of Hb E and α^+ -thalassemia was observed in this population, consistent with the evidence previously known in the region. A high prevalence of α^0 -thalassemia (SEA deletion) was found in Suay ethnic group. Also, α -thalassemia and Hb E related diseases like AEBart's and EEBart's were found in this population. These data indicated that health burden resulting from Hb Bart's hydrops fetalis may be serious in Suay ethnic group and complex thalassemia disease was found to be health problem in the elderly. Our findings provide not only baseline information, potentially useful for implementing appropriate control measures, but also an enhanced awareness and understanding of anemia and health care advice among the Suay elderly population.

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Introduction

Anemia in the elderly is common and increasing as the population ages. In older patients, anemia of any degree contributes significantly to morbidity and mortality and has a significant effect on the quality of life. Part of the problem here relates to its definition, which is based on WHO-criteria established in 1968⁽¹⁾. The WHO definition of anemia is hemoglobin (Hb) less than 130 g/L (13 g/dL) in men, Hb less than 120 g/L (12 g/dL) in non-pregnant women, and less than 110 g/L (11 g/dL) in pregnant women. Hb levels decline with age, and there has been a debate as to whether these values are applicable to older people, although there is no acceptable alternative definition of anemia in this age group. The prevalence of anemia in the elderly has been found to range from 8 to 44%, with the highest prevalence in men 85 years and older⁽¹⁻³⁾. Clinical symptoms of anemia vary, including mild pallor, weakness, shortness of breath, fatigue, and reduced work or productivity^(4, 5). Anemia has multiple etiologies, including iron deficiency (ID), chronic inflammatory disorders, micronutrient deficiencies, parasitic infections, excessive bleeding, and congenital defects of Hb production such as thalassemia and hemoglobinopathies⁽⁶⁾. From several studies, it has been clear that anemic subjects with combined thalassemia and iron deficiency (ID) are apparently associated with more severe hematological phenotypes than those with thalassemia or ID alone⁽⁷⁻¹⁰⁾. Ascertaining anemia and defining its underlying causes are essential for providing appropriate care and management of the patients as well as the establishment of a control program. From this context, the survey of thalassemia prevalence in the endemic areas of thalassemia and hemoglobinopathies such as the Northeastern (NE) Thailand is necessary. The community-based study of anemia in the elderly in NE Thailand revealed that the overall prevalence of anemia was found to be 47.7% and thalassemia was considered to be significant contributors to anemia⁽¹¹⁾. As far as we know, community-based surveys on anemia and its relationship to thalassemia within ethnic populations distributed in many areas of Thailand

are limited, despite the information that ethnic minorities are prone to several health problems including anemia. In 2018, the study of anemia, iron deficiency, and thalassemia in a group of hill tribe, Pakakayo-Karen in Northern Thailand revealed that anemia in the minority is more likely due to thalassemia than ID⁽¹²⁾. To the best of our knowledge, the survey of anemia and thalassemia in the ethnic minority in NE Thailand has never been reported. Our study aimed to survey the prevalence of anemia and thalassemia in the older population of Kui or Suay ethnic group. Suay is a Mon-Khmer language of the West Katuic branch, spoken in the southeastern part of the Northeastern (NE) Thailand, especially in the provinces of Surin and Sisaket and the adjoining areas of Cambodia and Laos⁽¹³⁾. In NE Thailand, Suay minority, about 273,570 people, resides in 686 villages in Surin, Sisaket, Buriram, and Ubon Ratchathani⁽¹⁴⁾. This ethnic minority has been previously observed as having the high prevalence of Hb E⁽¹⁵⁾. The findings herein will serve as baseline information and a better understanding of health situation in the region. This data will be useful for appropriate health care advice to this population.

Materials and methods

Subjects

The study protocol was approved by the Institution Review Board (IRB) of Ubon Ratchathani University, Ubon Ratchathani, Thailand (UBU-REC-36/2561). A total of 63 samples belong to Suay ethnic group distributed in seven villages in two districts (Muang Chan and Kantharalak) of Sisaket Province were selected as the study population. The criteria for volunteer enrollment in this study were as follows; look healthy, over 60 years of age, unrelated, and recognized as a member of the Suay ethnic population for at least three generations with no admixture from other populations. Venous blood specimens (EDTA as an anticoagulant) of volunteers were obtained after signing informed consents.

Laboratory investigation

Complete blood count (CBC) was performed in routine practice at Ubon Ratchathani University Hospital using the Sysmex XE-2100 Hematology

Analyzer (Sysmex, Kobe, Japan) within 24 hours. A Hb level of less than 13.0 g/dL in males and 12.0 g/dL in females was identified as anemia according to the WHO criteria⁽¹⁶⁾. Hemoglobin analysis was done using capillary zone electrophoresis (Minicap: Sebia, Lisses, France). For globin gene genotyping, DNA was extracted from peripheral blood leukocytes using the standard method and identifications of common α -thalassemia including α^0 -thalassemia (SEA & THAI deletions) and α^+ -thalassemia ($-\alpha^{3.7}$ & $-\alpha^{4.2}$) were performed using the gap-PCR methods described elsewhere^(17,18). Identifications of Hb Constant Spring and Hb Pakse' were done using allele-specific PCR based methods^(19,20).

Statistical analysis

Data processing was done using IBM SPSS Statistic 20 software (SPSS Inc, Chicago, IL, USA)

Descriptive statistics, i.e., percentage, mean and standard deviation, were used to describe the prevalence of anemia and thalassemia as well as hematological features.

Results

Among 63 Suay elderly subjects aged 60-85 years, non-anemia and anemia were detected in 33 (52.4%) and 30 (47.6%) subjects, respectively. The overall MCV results suggested that most cases expressed microcytic red blood cells. The mean of MCV in non-anemic males and females was 76.3 and 78.0 fL, respectively. The mean of MCV in anemic males and females was 71.2 and 68.4 fL, respectively. The characteristics of subjects including sex, age, and RBC parameters were summarized in Table 1.

Table 1 Characteristics of 63 Suay elderly subjects classified as non-anemic and anemic subjects (data expressed as mean \pm SD and min-max in parentheses)

| | Sex | RBC Parameters | | | | | | |
|------------------------------|-------------|--------------------------------|-------------------------------|-------------------------------|-------------------------------|-------------------------------|-------------------------------|-------------------------------|
| | | RBC [$10^6/\mu\text{L}$] | HGB [g/ dL] | HCT [%] | MCV [fL] | MCH [pg] | MCHC [g/ dL] | RDW-CV [%] |
| Non-Anemia n = 33 (52.4%) | Male (14) | 5.37 \pm 0.52 (4.61-6.53) | 14.0 \pm 0.8 (13.1-16.2) | 40.7 \pm 2.3 (38.7-47.5) | 76.3 \pm 6.9 (61.7-86.8) | 26.2 \pm 2.5 (21.4-30.0) | 34.3 \pm 0.6 (33.2-35.5) | 14.9 \pm 1.7 (13-19.6) |
| | Female (19) | 4.97 \pm 0.36 (4.18-5.54) | 13.0 \pm 0.8 (12-14.3) | 38.7 \pm 2.1 (36.2-43.3) | 78.0 \pm 4.8 (67.9-88.5) | 26.1 \pm 1.6 (23.1-30.4) | 33.5 \pm 0.7 (32.6-34.5) | 14.6 \pm 1.0 (12.7-17.2) |
| Anemia n = 30 (47.6%) | Male (11) | 4.97 \pm 0.59 (4.16-6.05) | 11.8 \pm 0.8 (10.3-12.9) | 35.1 \pm 2.9 (29.3-38.2) | 71.2 \pm 7.6 (58.2-85.5) | 24.0 \pm 2.5 (20.5-28.1) | 33.8 \pm 1.5 (32.2-36.6) | 16.4 \pm 2.1 (13.8-20.8) |
| | Female (19) | 4.72 \pm 0.56 (3.4-5.73) | 10.6 \pm 1.3 (6.7-11.9) | 32.2 \pm 4.1 (19.2-36.1) | 68.4 \pm 7.9 (52.8-81.7) | 22.6 \pm 2.4 (16.8-27.4) | 33.0 \pm 1.0 (31.5-34.9) | 16.7 \pm 3.6 (12.9-29.5) |

Table 2 described the thalassemia types in non-anemic and anemic subjects. The extremely high prevalence of thalassemia was observed in the Suay elderly population, with the prevalence of 84.8 and 93.3% in non-anemic and anemic subjects, respectively. The most common thalassemia found in Suay elderly was heterozygous Hb E, followed by heterozygous Hb E with α -thalassemia and heterozygous α^+ -thalassemia. Out of the 63 Suay elderly, the overall prevalences of Hb E, and α^0 -thalassemia

were 76.2, and 7.9%, respectively, whereas, no β -thalassemia was found in this population. The heterozygous α^0 -thalassemia was found in 6.7% of the anemic group. Thalassemia diseases were also found in three cases including two cases of AEBart's disease ($--^{SEA}/-\alpha^{3.7}$, β^A/β^E) and a case of EEBart's disease ($--^{SEA}/-\alpha^{3.7}$, β^E/β^E). The homozygous Hb E with α -thalassemia 2 female subjects had the lowest Hb level (6.7 g/dL) and subject with AEBart's disease had the lowest MCV (52.8 fL) and MCH (16.8 pg) values in this study.

Table 2 Thalassemia types and globin genotypes of 63 Suay elderly subjects in this study

| Thalassemia type | Globin genotype | Non-Anemia | Anemia |
|---|--|-----------------|-----------------|
| | | n (%) | n (%) |
| Non-thalassemia | | 5 (15.2) | 2 (6.7) |
| Thalassemia | | 28 (84.8) | 28 (93.3) |
| 1. Heterozygous α^+ -thalassemia | $\alpha\alpha/-\alpha^{3.7}, \beta^A/\beta^A$ | 3 (9.1) | 2 (6.7) |
| 2. Homozygous α^+ -thalassemia | $-\alpha^{3.7}/-\alpha^{3.7}, \beta^A/\beta^A$ | - | 1 (3.3) |
| 3. Heterozygous α^0 -thalassemia | $--^{SEA}/\alpha\alpha, \beta^A/\beta^A$ | - | 2 (6.7) |
| 4. Heterozygous Hb E | $\alpha\alpha/\alpha\alpha, \beta^A/\beta^E$ | 14 (42.4) | 8 (26.7) |
| 5. Heterozygous Hb E with α -thalassemia | $\alpha\alpha/-\alpha^{3.7}, \beta^A/\beta^E$ | 5 (15.2) | 3 (10) |
| | $\alpha\alpha/\alpha^{CS}\alpha, \beta^A/\beta^E$ | 3 (9.1) | - |
| | $\alpha\alpha/\alpha^{PS}\alpha, \beta^A/\beta^E$ | 1 (3.0) | - |
| | $\alpha^{CS}\alpha/-\alpha^{3.7}, \beta^A/\beta^E$ | 1 (3.0) | 2 (6.7) |
| | $\alpha^{PS}\alpha/-\alpha^{3.7}, \beta^A/\beta^E$ | - | 1 (3.3) |
| | $\alpha^{CS}\alpha/\alpha^{PS}\alpha, \beta^A/\beta^E$ | - | 1 (3.3) |
| | $\alpha\alpha/-\alpha^{3.7}, \beta^E/\beta^E$ | 1 (3.0) | 2 (6.7) |
| 6. Homozygous Hb E with α -thalassemia | $\alpha\alpha/\alpha^{CS}\alpha, \beta^E/\beta^E$ | - | 2 (6.7) |
| | $\alpha^{CS}\alpha/\alpha^{PS}\alpha, \beta^E/\beta^E$ | - | 1 (3.3) |
| | $--^{SEA}/-\alpha^{3.7}, \beta^A/\beta^E$ | - | 2 (6.7) |
| 7. Thalassemia disease | $--^{SEA}/-\alpha^{3.7}, \beta^E/\beta^E$ | - | 1 (3.3) |
| | | | |
| Total | | 33 (100) | 30 (100) |

Note: Data are numbers with percentages in parentheses.

Discussion

This study is the first report that focuses on the prevalence of anemia and thalassemia in the Suay elderly. Suay ethnic population in this study is one of the most common minorities in the lower NE Thailand⁽¹⁴⁾. This survey revealed a high prevalence of anemia in approximately 47.6% of Suay elderly people living in Sisaket Province which is located near the border region of Thailand, Lao PDR, and Cambodia. The prevalence of anemia found in this study was equal to that in a previous study in the elderly living in Udon Thani Province of NE Thailand⁽¹¹⁾. Unlike this study, several community-based studies conducted in Western and Northeastern Asian countries reported a relatively low prevalence of anemia in the elderly people (11-18%)⁽²¹⁻²³⁾. Conversely, a relatively higher prevalence of anemia (21-34%) has been reported in a group of African-American

elderly^(21,24,25). Concerning the variations in genetic background, it is noteworthy that the higher anemia prevalence has been observed within those ethnicities having high carrier rates of thalassemia. Previous studies conducted in this area suggested that the most likely causative factor in more than half of the anemic individuals identified may be the presence of thalassemia^(8,10,26). The study in a group of hill tribe, Pakakayo-Karen in Northern Thailand also confirmed that anemia in the minority is more likely due to thalassemia than ID⁽¹²⁾. This study investigated all common forms of thalassemia (α^0 -thal, β -thal, Hb E, and α^+ -thal) in the Suay elderly population. Surprisingly, astoundingly high prevalence of thalassemia was found in non-anemic (84.8%) and anemic (93.3%) Suay elderly population. As expected, the finding of a high frequency of α^+ -thalassemia in this study

was consistent with the evidence in the southern part of NE Thailand⁽²⁷⁾. The high prevalence of Hb E herein confirmed the study of Fucharoen G, et al⁽¹⁵⁾ which revealed the high frequency of Hb E in Suay and Khmer, and close relation was observed in these two ethnic groups. No occurrence of β -thalassemia was found in the Suay elderly population and this seemed to be consistent with the evidence of the low prevalence of β -thalassemia in the lower part of NE Thailand⁽²⁷⁾. High prevalence of α^0 -thalassemia (SEA deletion) was found in Suay ethnic group, heterozygous α^0 -thalassemia was found in two cases and also, α -thalassemia and Hb E related diseases like AEBart's and EEBart's were found in this study. In conclusion, the prevalence of major thalassemia was 76.2, 7.9, and 0% for Hb E, α^0 -thalassemia, and β -thalassemia, respectively. These data indicated that health burden resulting from Hb Bart's hydrops fetalis may be serious in Suay ethnic group and complex diseases like AEBart's and EEBart's were important health problems in Suay elderly. Moreover, since the extremely high prevalence of thalassemia was found in the Suay elderly, the awareness of ID should be concerned in this group because of the information that anemic subjects with combined thalassemia and ID are apparently associated with more severe hematological phenotypes⁽⁷⁻¹⁰⁾. Unfortunately, this study has some limitations, particularly, the small sample size and the lack of finding out other causes of anemia. The understanding of the cause of the low Hb in female subject with homozygous Hb E/ α^+ -thalassemia has been limited; thereby, ID or other causes may result in this appearance. Further study focusing on the cause of anemia in this population will be of benefit to public health effort to provide better health care. However, the findings in this study might suggest that thalassemia and hemoglobinopathies are one of the causes that should be taken into account in any proposed anemia control program conducted within the Suay population. It is therefore essential that health workers understand this outcome so that they can provide informed advice and/or more effective management strategies for the affected individuals.

Conclusion

This study is the first report that focuses on the prevalence of anemia and thalassemia in the Suay elderly. The high prevalence of anemia (47.6%) was found in this population. The overall prevalence of Hb E, and α^0 -thalassemia were 76.2, and 7.9%, respectively, whereas, no β -thalassemia was found in this population. Thalassemia diseases were also found in three cases including two cases of AEBart's disease and a case of EEBart's disease. This study might suggest that thalassemia and hemoglobinopathies are one of the causes that should be considered in any proposed anemia control program conducted within the Suay population.

Take home messages

The prevalence of anemia in the Suay elderly was 47.6%. The finding of a high prevalence of Hb E, α^+ -thalassemia, and α^0 -thalassemia indicated that complex thalassemia diseases like AEBart's and EEBart's were health problems in the elderly and Hb Bart's hydrops fetalis should be concerned in the Suay population.

Conflicts of interest

The authors declare no conflict of interest.

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References

1. Blanc B, Finch CA, Hallberg L, Lawkowitz W, Layrisse M, Mollin DL, et al. Nutritional anaemias. WHO Tech Rep Ser 1968; 405: 1-40.
2. Cheng CK, Chan J, Cembrowski GS, van Assendelft OW. Complete blood count reference interval diagrams derived from NHANES III: stratification by age, sex, and race. Lab Hematol 2004; 10: 42-53.
3. Zakai NA, Katz R, Hirsch C, Shlipak MG, Chaves PHM, Newman AB, et al. A Prospective study of anemia status, hemoglobin concentration, and mortality in an elderly cohort: The Cardiovascular Health Study. Arch Intern Med 2005; 165: 2214-20.
4. McLean E, Cogswell M, Egli I, Wojdyla D, de Benoist B: Worldwide prevalence of anemia, WHO vitamin and mineral nutrition information system, 1993-2005. Public Health Nutr 2009; 12: 444-54.
5. Lasch KF, Evans CJ, Schatell D: A qualitative analysis of patient-reported symptoms of anemia. Nephrol Nurs J 2009; 36: 621-44.
6. Evatt BL, Gibbs WN, Lewis SM, McArthur JR: Fundamental Diagnostic Hematology: Anemia, ed 2. Geneva, World Health Organization, 2002.
7. Pansuwan A, Fuchareon G, Fuchareon S, Himakhun B, Dangwiboon S. Anemia, iron deficiency and thalassemia among adolescents in Northeast Thailand: results from two independent surveys. Acta Haematol 2011; 125: 186-92.
8. Panomai N, Sanchaisuriya K, Yamsri S, Sanchaisuriya P, Fucharoen G, Fucharoen S, et al. Thalassemia and iron deficiency in a group of northeast Thai school children: relationship to the occurrence of anemia. Eur J Pediatr 2010; 169: 1317-22.
9. Thurlow RA, Winichagoon P, Green T, Wasantwisut E, Pongcharoen T, Bailey KB, et al. Only a small proportion of anemia in northeast Thai schoolchildren is associated with iron deficiency. Am J Clin Nutr 2005; 82: 380-87.
10. Sanchaisuriya K, Fucharoen S, Ratanasiri T, Sanchaisuriya P, Fucharoen G, Dietz E, et al. Thalassemia and hemoglobinopathies rather than iron deficiency are major causes of pregnancy-related anemia in northeast Thailand. Blood Cells Mol Dis 2006; 37: 8-11.
11. Deeruska L, Sanchaisuriya K. Anemia in the elderly in Northeastern Thailand: A community based study investigating prevalence, contributing factors, and hematologic features. Acta Haematologica 2017; 138: 96-102.
12. Deeruska L, Raksasang M, Sanchaisuriya K. Anemia, Iron Deficiency and Iron Deficiency Anemia in a Group of Hill-Tribe, Pakakayo-Karen. KKU Journal for Public Health Research 2018; 11: 18-27.
13. Smith, KD. A lexico-statistical study of Mon-Khmer language. In: Gonzalez AB, Thomas DD, Editors. Linguistics Across Continents: Studies in Honor of Richard S. Pittman. Manila (Philippines): The Linguistic Society of the Philippines, 1981: 180-205.
14. van der Haak F, Woykos B. Kui dialect survey in Surin and Sisaket. Mon-Khmer Studies 1990; 16: 109-142.
15. Fucharoen G, Fucharoen S, Sanchaisuriya K, Sae-Ung N, Suyasunanond U, Sriwilai P, et al. Frequency Distribution and Haplotypic Heterogeneity of beta(E)-Globin Gene among Eight Minority Groups of Northeast Thailand. Hum Hered 2002; 53: 18-22
16. Iron deficiency anemia: assessment, prevention and control. A guide for program managers. Document WHO/NHD/01.3. Geneva, World Health Organization, 2001.
17. Sae-ung N, Fucharoen G, Sanchaisuriya K, Fucharoen S. α^0 -Thalassemia and related disorders in northeast Thailand: a molecular and hematological characterization. Acta Haematol 2007; 117: 78-82.
18. Charoenwijitkul T, Singha K, Fucharoen G, Sanchaisuriya K, Thepphitak P, Wintachai P, et al. Molecular characteristics of α^+ -thalassemia (3.7 kb deletion) in Southeast Asia: Molecular subtypes, haplotypic heterogeneity, multiple founder effects and laboratory diagnostics. Clin Biochem 2019; 71: 31-7.

19. Fucharoen S, Fucharoen G, Fukumaki Y. Simple non-radioactive method for detecting haemoglobin Constant Spring gene. *Lancet* 1990; 335(8704): 1527.
20. Sanchaisuriya K, Fucharoen G, Fucharoen S. Hb Paksé [(α2) codon 142 (TAA→TAT or Term→Tyr)] in Thai patients with EAbart's disease and Hb H disease. *Hemoglobin* 2002; 26: 227-35.
21. Guralnik JM, Eisenstaedt RS, Ferrucci L, Klein HG, Woodman RC: Prevalence of anemia in persons 65 years and older in the United States: evidence for a high rate of unexplained anemia. *Blood* 2004; 104: 2263-8.
22. Choi CW, Lee J, Park KH, Yoon SY, Choi IK, Oh SC, et al. Prevalence and characteristics of anemia in the elderly: cross-sectional study of three urban Korean population samples. *Am J Hematol* 2004; 77: 26-30.
23. Yamada M, Wong FL, Suzuki G: Longitudinal trends of hemoglobin levels in a Japanese population - RERF's Adult Health Study subjects. *Eur J Haematol* 2003; 70: 129-35.
24. Wang JL, Shaw NS. Iron status of the Taiwanese elderly: the prevalence of iron deficiency and elevated iron stores. *Asia Pac J Clin Nutr* 2005; 14: 278-84.
25. Denny SD, Kuchibhatla MN, Cohen HJ: Impact of anemia on mortality, cognition, and function in community-dwelling elderly. *Am J Med* 2006; 119: 327-34.
26. Carnley BP, Prior JF, Gilbert A, Lim E, Devenish R, Sing H, et al: The prevalence and molecular basis of hemoglobinopathies in Cambodia. *Hemoglobin* 2006; 30: 463-70.
27. Tritipsombut J, Sanchaisuriya K, Phollarp P, Bouakhasith D, Sanchaisuriya P, Fucharoen G, et al. Micromapping of thalassemia and hemoglobinopathies in different regions of Northeast Thailand and Vietaine, Laos People's Democratic Republic. *Hemoglobin* 2012; 36: 47-56.