Prevalence of anaemia and its associated factors among HIV-infected adults at the time of ART initiation at Debre Markos Comprehensive Specialized Hospital, Northwest Ethiopia: a retrospective cross-sectional study

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INTRODUCTION

Anaemia is a serious global public health problem that affects all age groups of the population. It affects up to one-third of the global population, and if it is undiagnosed or left untreated for a prolonged period of time, it can lead to multiorgan failure and even death. 1 2 It can also have a negative effect on the quality of life and adversely impact the social and economic development of a patient and the country at large. 3 4

There are varieties of haematological abnormalities associated with HIV infection, of which anaemia remains a public health challenge in HIV-positive patients around the world, particularly in sub-Saharan Africa, including Ethiopia. Anaemia is a well-known complication and the most common haematological abnormality associated with HIV infection, especially among patients with advanced HIV disease. 5 6 In addition to the

STRENGTHS AND LIMITATIONS OF THIS STUDY

⇒ This study provided baseline information about anaemia status before starting antiretroviral therapy (ART).
⇒ To make a representative sample, reviewed charts were selected randomly.
⇒ Due to the retrospective nature of the study, the current study lacks some variables like smoking, alcohol consumption, chat chewing, and type of anaemia.
⇒ It is also difficult to determine the temporal link between the outcome and exposure variables.
⇒ Additionally, this study was conducted among ART-naive adults, which lacks comparison with anaemia after ART initiation.
weakening of the immune system, HIV infection has a negative impact on the haematopoietic system of infected individuals, which results in a decreased concentration of haemoglobin in the blood. On the other hand, anaemia contributes to the progression of HIV infection to the AIDS stage and this in turn accelerates progression to mortality.\textsuperscript{3–10}

Globally, in 2019, a total of 1.74 billion anaemia cases with an overall prevalence of 22.8\% were reported.\textsuperscript{11} Anaemia is more prevalent in developing countries, particularly sub-Saharan Africa, which accounts for more than 89\% of overall anaemia.\textsuperscript{12–14} This anaemia prevalence increased more among HIV-infected patients who did not start antiretroviral therapy (ART).\textsuperscript{15} In different study settings, the prevalence of anaemia at the time of ART initiation among HIV-infected patients was estimated to be 55.8\%, 51.9\%, and 25.8\% in Nepal, China, and Johannesburg, South Africa,\textsuperscript{10–14} respectively.

Ethiopia is one of the countries most seriously affected by HIV, and anaemia is a known predictor of disease progression and death among HIV-infected patients.\textsuperscript{3–6} Studies conducted in Ethiopia showed that the prevalence of anaemia at the time of ART initiation ranged from 21.2\% to 52.6\%.\textsuperscript{16–22} Identifying risk factors for anaemia prevalence is crucial for developing effective interventions and monitoring anaemia control programmes among HIV-infected patients. A cross-sectional survey conducted in China showed that advanced age, low CD4+ T cell count and ethnicity were significant factors in increased anaemia.\textsuperscript{16} Another study conducted in Tanzania revealed that female gender, low body mass index (BMI), lower CD4+ T cell count and concurrent tuberculosis (TB) treatment were associated with an increased risk of anaemia.\textsuperscript{8} Studies conducted in Ethiopia reported that female sex, WHO clinical stage III/IV, TB/HIV coinfection, lower CD4+ T cell counts, presence of opportunistic infection (OI), lower BMI and history of TB treatment were independent predictors of anaemia occurrence at baseline.\textsuperscript{16–21}

Ethiopia is strongly committed to promote health and well-being among HIV-infected patients and the community at large, and anaemia control is among its priorities. However, the relative contribution of different risk factors to anaemia among HIV-infected patients in Ethiopia still varies from one setting to another. So, knowing the factors at local levels is important to intervene accordingly. Therefore, this study aimed to assess the prevalence of anaemia and its associated factors at the time of ART initiation among HIV-infected patients at Debre Markos Comprehensive Specialized Hospital.

METHOD AND MATERIALS

Study design, setting and period

An institution-based retrospective cross-sectional study was conducted at Debre Markos Comprehensive Specialized Hospital from 1 January 2014 to 31 December 2018, among HIV-infected adults. Patients’ charts were reviewed to collect the data from randomly selected charts. Debre Markos Comprehensive Specialized Hospital is located in Debre Markos town in the East Gojjam Zone of Amhara Nation Regional State (ANRS). It is located 299 km from Ethiopia’s capital, Addis Ababa, and 265 km from Bahir Dar, the capital city of ANRS.\textsuperscript{24}

Source and study population

Source population

All adult people living with HIV aged 15 years old and above attended the ART clinic at Debre Markos Comprehensive Specialized Hospital in Northwest Ethiopia.

Study population

Randomly selected HIV-positive adults aged 15 years old and above who were newly enrolled in the ART clinic at Debre Markos Comprehensive Specialized Hospital between 1 January 2014 and 31 December 2018, in Northwest Ethiopia.

Inclusion and exclusion criteria

Adults aged 15 years and above who were newly enrolled in the ART clinic at Debre Markos Comprehensive Specialized Hospital at the time of the study were included in this study. Patients’ charts with transferred record information were excluded because these charts may lack baseline information. Additionally, being pregnant at baseline was excluded from the study.

Sample size and sampling procedure

The sample size was determined by using a formula to estimate single population proportion with the assumption of a 95\% level of confidence, 23.4\% proportion\textsuperscript{19} and a 4\% marginal error.

\[
n_1 = \frac{(z_{\alpha/2})^2 \times p(1-p)}{w^2}
\]

Where \(n_1\)=initial sample size, \(\alpha\)=precision level or level of significance, \(p\)=population proportion of anaemia, \(w\)=marginal error, and \(Z_{\alpha/2}\)=the value under the standard normal table.

Finally, the sample size was calculated using the Epi Info statistical package V.7.2.2.6 and, with consideration of 10\% expected incomplete records, the final sample size was 473. During the study period, a total of 1264 patients were newly enrolled in the Debre Markos ART clinic. Of these, 1117 patient charts fulfilled the inclusion criteria. Then, by generating a random number on the computer, a total of 473 patients’ charts were selected through a simple random sampling technique, and data were collected from them.

Operational definition of variables

Anaemia in this study was defined as anaemic or non-anaemic based on WHO criteria: haemoglobin concentration <130 g/L for males and <120 g/L for females.\textsuperscript{25}

Functional status was classified based on WHO criteria as: working (W)=capable of going out of home and doing routine activities, including daily work; ambulatory
(A)=capable of self-care and going to the toilet unsupported; bedridden (B)=cannot go even to the toilet unsupported.26

**Baseline WHO clinical stages**: taken from the chart record at enrolment to ART based on WHO classification criteria, labelled as stage I–IV.

**BMI**: defined as the weight of the individual in kilograms divided by their height in metres squared.

**Data collection tools and procedures**

First, an extraction tool was developed from HIV/AIDS care monitoring and evaluation tools. Forms used for laboratory requests and ART intake were also incorporated into the development of the extraction tool. The tool contains sociodemographic and related baseline clinical variables. Prior to data collection, training was given to three nurses regarding the tool and the way they extract the data from the chart. With the help of medical record numbers (MRNs), patients’ charts were selected by a computer-generated simple random sampling technique. Patient charts were picked up from the chart room using MRN and then data were extracted from the patient’s medical charts using the tool. A common code was given for each selected chart after the data were extracted, so that there was no chance of recollection of data from a similar chart. In this way, all selected patients’ charts that fulfil the inclusion criteria were reviewed and data extraction was completed.

**Data quality assurance**

A pretested data extraction tool was used to maintain data quality. Quality is also maintained by extracting data using trained nurses and close monitoring of the procedure by the supervisor. Data clerks were involved in the selection of patients’ charts from the computer as well as from the chart room. Before returning the chart to the shelf, the completeness of the data extraction tool was checked and a necessary correction was made.

**Statistical analysis**

After the data were extracted from the chart, it was first checked for consistency and completeness. After that, it was coded and entered into Epi Info V.7.2.2.6 and exported to STATA V.14.0 for analysis. A frequency table was used to describe the sociodemographic and clinical variables of the study. The prevalence of anaemia with a 95% CI at the time of ART initiation was estimated. Model fitness was checked by using the Hosmer-Lemeshow goodness-of-fit test (p=0.6106) and the model was fitted well. Bivariate analysis was executed for each variable, and those variables with a p value of <0.2 were entered into multivariate binary logistic regression to identify factors associated with anaemia prevalence at the time of ART initiation. An OR with a 95% CI was computed, and variables having a p value of <0.05 in the multivariate logistic regression were considered as statistically independent factors for anaemia at ART initiation.

**Patient and public involvement**

Since the study was based on chart review without active involvement of patients, no patients were involved.

**RESULT**

**Sociodemographic characteristics of HIV-infected adults**

Out of all the patients enrolled in the ART clinic from 1 January 2014 to 31 December 2018, a total of 473 charts were selected and reviewed based on the inclusion criteria. From these reviewed charts, 468 charts were included in the analysis, and only 5 charts were excluded due to data incompleteness. Of all the patients’ charts included in the analysis, 281 (60.04%) were female, and about 190 (40.60%) of the participants were grouped under the category of age 25–34 years (table 1).

**Clinical and immunological-related characteristics of HIV-infected adults**

Of all the study participants included in the analysis, about 136 (29.06%) had started ART 6 months or above after HIV status had been confirmed, and 78 (16.67%) of the study subjects had past OIs. From the group, about 121 (25.85%) and 29 (6.20%) of patients were grouped under baseline WHO clinical staging of III and IV, respectively. One-third of patients included in the analysis had a baseline CD4+ T cell count of less than 200 cells/µL and 134 (28.63%) of patients were grouped under the category of BMI of less than 18.5 kg/m². Regarding the functional status of HIV-infected patients in this study, around 98 (20.84%) had ambulatory or bedridden functional status at enrolment into highly active ART (table 2).

**Anaemia prevalence among HIV-infected adults at the time of ART initiation**

The overall prevalence of anaemia at the time of ART initiation in this study was 35.04% (95% CI: 30.84% to 39.49%). Of these, about 74 (45.12%) were grouped under the category of moderate anaemia level (figure 1).

**Factors that determine anaemia prevalence at the time of ART initiation**

After applying bivariate logistic regression, variables with a p value of 0.2 or less were taken into multivariate logistic regression. Variables included in multivariate analysis were sex, age at enrolment, level of education, residence, family size, pre-ART duration, past OIs, CD4+ T cell count at enrolment, BMI, WHO clinical stage and functional status at enrolment. After multivariate logistic regression, five variables, including sex, level of education, CD4+ T cell count, BMI and functional status at enrolment, were found to be statistically independent predictors of anaemia prevalence at the time of ART initiation at a p value of less than 0.05.

The current study revealed that the odds of being anaemic at the time of ART initiation among males were 2.45 times that of females (adjusted OR (AOR)=2.45; 95% CI: 1.51 to 3.98). Similarly, the odds of developing...
anaemia among patients who had not attended formal education were 2.38 times that of those with a college degree and above educational level (AOR=2.38; 95% CI: 1.12 to 5.05). The CD4+ T cell count was also another significant factor of anaemia at the time of ART initiation. The likelihood of developing anaemia at the time of ART initiation among patients with a CD4+ T cell count of less than 200 cells/µL was 4.67 times that of those with CD4+ T cell count of 200 cells/µL or more (AOR=4.67; 95% CI: 2.78 to 7.85). In the same manner, the odds of being anaemic at enrolment into ART were 2.43 times higher among patients with a BMI of less than 18.5 kg/m² than among patients with a normal BMI (AOR=2.43; 95% CI: 1.42 to 4.16). Lastly, the odds of being anaemic among ambulatory and/or bedridden patients were 2.69 times that of working functional status.

| Characteristics          | Frequency (%) | Anaemia status | P value |
|--------------------------|---------------|----------------|---------|
|                          |               | Anaemic (%)    | Not anaemic (%) |
|                          |               | 86 (45.99)     | 101 (54.01)    |
|                          | 77 (27.40)    | 204 (72.60)    |
| Age in years             | 15–24         | 51 (10.90)     | 11 (21.57)     |
|                          | 25–34         | 190 (40.60)    | 65 (34.21)     |
|                          | 35–44         | 157 (33.55)    | 61 (38.85)     |
|                          | ≥45           | 70 (14.96)     | 27 (38.57)     |
| Religion                 | Orthodox      | 447 (95.51)    | 158 (35.35)    |
|                          | Muslim        | 20 (4.27)      | 6 (30.00)      |
|                          | Protestant    | 1 (0.21)       | 0              |
| Marital status           | Single        | 56 (11.97)     | 18 (32.14)     |
|                          | Married       | 251 (53.63)    | 85 (33.86)     |
|                          | Divorced/separated | 125 (26.71) | 47 (37.60)     |
|                          | Widowed       | 36 (7.69)      | 14 (38.89)     |
| Educational level        | No education  | 171 (36.54)    | 62 (36.26)     |
|                          | Primary       | 109 (23.29)    | 38 (34.86)     |
|                          | Secondary     | 113 (24.15)    | 44 (38.94)     |
|                          | College+      | 75 (16.03)     | 20 (26.67)     |
| Residence                | Rural         | 101 (21.58)    | 45 (44.54)     |
|                          | Urban         | 367 (78.42)    | 119 (32.43)    |
| Occupation               | Employed      | 153 (32.69)    | 50 (32.68)     |
|                          | Unemployed    | 315 (67.31)    | 114 (36.19)    |
| Family size              | <2 persons    | 187 (39.96)    | 55 (29.41)     |
|                          | 3–4 persons   | 213 (45.51)    | 82 (38.50)     |
|                          | >4 persons    | 68 (14.53)     | 27 (39.71)     |
| HIV disclosure status    | Disclosed     | 426 (91.03)    | 149 (34.98)    |
|                          | Not disclosed | 42 (8.97)      | 15 (35.71)     |

ART, antiretroviral therapy.
The overall prevalence of anaemia at the time of ART initiation in the current study was 35.04% (95% CI: 30.84% to 39.49%), which is higher than studies conducted at the University of Gondar Referral Hospital (21.2%)\textsuperscript{20} and Hawassa University Referral Hospital (23.4%).\textsuperscript{19} However, anaemia prevalence in the current study is lower than in studies from Addis Ababa\textsuperscript{16,17} and Arba Minch town.\textsuperscript{18} Similarly, anaemia prevalence in the current study is lower than in a similar study at Black Lion Specialized Hospital (41.9%).\textsuperscript{21} This could be due to a higher mean of baseline CD4+ T-cell count in the current study (289.5 cells/µL±167.8) than that of Black Lion Specialized Hospital.

\textbf{DISCUSSION}

The overall prevalence of anaemia at the time of ART initiation in the current study was 35.04% (95% CI: 30.84% to 39.49%), which is higher than studies conducted at the University of Gondar Referral Hospital (21.2%)\textsuperscript{20} and Hawassa University Referral Hospital (23.4%).\textsuperscript{19} However, anaemia prevalence in the current study is lower than in studies from Addis Ababa\textsuperscript{16,17} and Arba Minch town.\textsuperscript{18} Similarly, anaemia prevalence in the current study is lower than in a similar study at Black Lion Specialized Hospital (41.9%).\textsuperscript{21} This could be due to a higher mean of baseline CD4+ T-cell count in the current study (289.5 cells/µL±167.8) than that of Black Lion Specialized Hospital.
Anaemia prevalence is also higher than a study done in Johannesburg, South Africa, with a prevalence of 25.8%. The variation may be due to sociodemographic differences. However, it is also lower than studies conducted in China and Nepal, with a prevalence of 51.9% and 55.8%, respectively. This lower prevalence of anaemia in the current study could be related to differences in sociodemographic characteristics, or it could be due to differences in the time period in which better interventions to reduce anaemia have been applied recently than in previous times. Additionally, the majority of patients (72.3%) from China had a CD4+ T cell count of less than 200 cells/mm³ compared with the current study, which is about 34.83%. This might have an important biological implication in that the lower CD4+ T cell count was associated with an increased risk of anaemia.

In this study, the male sex was found to be an independent predictor of increased anaemia at the time of ART initiation. This is in line with studies conducted in Zewditu Memorial Hospital and Arba Minch town, Ethiopia. More alcohol consumption among males than females might contribute to this difference between males and females. As males consume more alcohol, the rate of vitamin B₁₂ absorption into the circulatory system becomes lower and may result in anaemia. In contrast to the current study, the odds of being anaemic at the time of ART initiation at Hawassa University Referral Hospital were higher in females than in males. This may be explained by the fact that patients who have not attended formal education are less aware of better nutrition and better healthcare. When HIV infection is observed among those who have not attended formal education, the risk of poor nutrition and the occurrence of anaemia will be double burdened. Additionally, non-educated patients are not fully aware of anaemia symptoms, so they will come to the hospital quite late with high anaemic grades.

The current study revealed that the odds of being anaemic among patients with a CD4+ T cell count of less than 200 cells/mm³ were 4.67 times that of a CD4+ T cell count greater than 200 cells/mm³. This is congruent with studies from the University of Gondar Referral Hospital, Arba Minch town, and Black Lion Specialized Hospital. It could be due to the fact that bone marrow abnormalities are found at all stages of HIV disease and increase in frequency and severity as the disease advances. So, the risk of anaemia occurrence increases with progressive immunological deterioration and a CD4+ T cell count of less than 200 cells/mm³ is related to the development of anaemia.

Anaemia was 2.43 times more likely in patients with a lower BMI (18.5 kg/m²) than in those with a normal BMI. This is congruent with a study from Wolaita Sodo University Teaching Referral Hospital, Ethiopia. This might be as a result of deficiencies in micronutrients such as iron, folate and vitamin B₁₂ in patients who were undernourished. This deficiency of micronutrients directly contributes to the development of anaemia among those with a BMI of less than 18.5 kg/m². Lastly, being in an ambulatory or bedridden functional status increased the likelihood of developing anaemia among patients with HIV. This might be explained by the fact that being in an ambulatory or bedridden functional status could be an indicator of HIV infection advancement and the occurrence of other OIs that may also make them lose their appetite, expose them to malnutrition and result in anaemia.

In this study, we noted a few limitations. Since the cause of anaemia in HIV-infected adults is multifactorial and the study was based on chart review, we noted that a few variables were missed. Furthermore, this study did not collect data on dietary habits, substance use status, menstrual habits in females or the type of anaemia. Additionally, this study was conducted among ART-naïve adults, which lacks comparison with anaemia after ART initiation.

CONCLUSION
The current study showed that a significant proportion of HIV-infected adults developed anaemia at the time of ART initiation. Male sex, lack of formal education, a CD4+ T cell count <200 cells/mm³, decreased BMI, and ambulatory or bedridden functional status were discovered to be independent predictors of anaemia. Hence, a college degree and above educational status. This is supported by a retrospective cross-sectional study from Mizan-Aman General Hospital, Ethiopia. This may be explained by the fact that patients who have not attended formal education are less aware of better nutrition and better healthcare. When HIV infection is observed among those who have not attended formal education, the risk of poor nutrition and the occurrence of anaemia will be double burdened. Additionally, non-educated patients are not fully aware of anaemia symptoms, so they will come to the hospital quite late with high anaemic grades.
Table 3  Bivariate and multivariable logistic regression analyses of factors associated with anaemia at the time of ART initiation among HIV infected adults at Debre Markos Comprehensive Specialized Hospital from 1 January 2014 to 31 December 2018 (n=468)

| Characteristics                      | Frequency | Anaemic | Not anaemic | COR (95% CI) | AOR (95% CI) | P value |
|--------------------------------------|-----------|---------|-------------|--------------|--------------|---------|
|                                      |           |         |             |              |              |         |
| Sex of the patient                   |           |         |             |              |              |         |
| Male                                 | 86        | 101     | 2.30 (1.56 to 3.40) | 2.45 (1.51 to 3.98) | <0.001*     |
| Female                               | 77        | 204     | 1.00        | 1.00         |              |         |
| Age at enrolment (in years)          |           |         |             |              |              |         |
| 15–24                                | 11        | 40      | 1.00        | 1.00         |              |         |
| 25–34                                | 65        | 125     | 1.89 (0.91 to 3.93) | 1.96 (0.79 to 4.85) | 0.145       |
| 35–44                                | 60        | 97      | 2.31 (1.10 to 4.85) | 1.65 (0.65 to 4.22) | 0.295       |
| 45+                                  | 27        | 43      | 2.28 (1.003 to 5.19) | 1.65 (0.58 to 4.67) | 0.346       |
| Level of education                   |           |         |             |              |              |         |
| No education                         | 62        | 109     | 1.57 (0.88 to 2.82) | 2.38 (1.12 to 5.05) | 0.024*      |
| Primary                              | 37        | 72      | 1.32 (0.70 to 2.51) | 1.69 (0.76 to 3.77) | 0.195       |
| Secondary                            | 44        | 69      | 1.87 (1.01 to 3.45) | 1.88 (0.87 to 4.04) | 0.107       |
| College+                             | 20        | 55      | 1.00        | 1.00         |              |         |
| Patient’s residence                  |           |         |             |              |              |         |
| Rural                                | 118       | 249     | 1.67 (1.07 to 2.62) | 1.58 (0.91 to 2.75) | 0.105       |
| Urban                                | 45        | 56      | 1.00        | 1.00         |              |         |
| Family size                          |           |         |             |              |              |         |
| ≤2 persons                           | 55        | 132     | 1.00        | 1.00         |              |         |
| 3–4 persons                          | 81        | 132     | 1.50 (0.99 to 2.28) | 1.30 (0.76 to 2.22) | 0.333       |
| ≥5 persons                           | 27        | 41      | 1.58 (0.89 to 2.82) | 1.41 (0.68 to 2.94) | 0.352       |
| Pre-ART duration                     |           |         |             |              |              |         |
| <6 months                            | 122       | 210     | 1.00        | 1.00         |              |         |
| ≥6 months                            | 42        | 94      | 0.77 (0.50 to 1.17) | 0.93 (0.55 to 1.60) | 0.807       |
| Presence of past OI                  |           |         |             |              |              |         |
| Yes                                  | 34        | 44      | 1.55 (0.94 to 2.53) | 0.88 (0.46 to 1.69) | 0.708       |
| No                                   | 130       | 260     |              |              |              |         |
| CD4+ T cell count at enrolment (cells/μL) |       |         |             |              |              |         |
| <200                                 | 103       | 60      | 6.86 (4.49 to 10.49) | 4.67 (2.78 to 7.85) | <0.001*     |
| ≥200                                 | 61        | 244     | 1.00        | 1.00         |              |         |
| Body mass index (kg/m²)              |           |         |             |              |              |         |
| <18.5                                | 78        | 56      | 3.85 (2.49 to 5.93) | 2.43 (1.42 to 4.16) | 0.001*      |
| 18.5–24.9                            | 76        | 210     | 1.00        | 1.00         |              |         |
| >24.9                                | 10        | 38      | 0.73 (0.35 to 1.53) | 1.26 (0.54 to 2.93) | 0.593       |
| WHO clinical staging                  |           |         |             |              |              |         |
| Stage I                              | 43        | 158     | 1.00        | 1.00         |              |         |
| Stage II                             | 35        | 86      | 1.49 (0.89 to 2.51) | 0.94 (0.51 to 1.73) | 0.850       |
| Stage III/IV                         | 86        | 60      | 5.27 (3.29 to 8.44) | 1.78 (0.96 to 3.30) | 0.068       |
| Functional status at enrolment       |           |         |             |              |              |         |
| Working                              | 93        | 277     | 1.00        | 1.00         |              |         |
| Ambulatory/bedridden                 | 71        | 27      | 7.83 (4.74 to 12.93) | 2.69 (1.41 to 5.12) | 0.003*      |

*Significance at a p value of <0.05.

AOR, adjusted OR; ART, antiretroviral therapy; COR, crude OR; OI, opportunistic infection.
giving special attention to patients not attending formal education, males and late presenters is crucial to reduce anaemia occurrence and its health impact. Finally, the findings of this study will provide baseline information to healthcare providers in order to select ART drugs accordingly, and may provide additional insight into the development of new anaemia preventive strategies.

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Patient consent for publication Not required.

Ethics approval Ethical clearance was obtained from the Institutional Review Board (IRB) of the University of Gondar (reference no. S/N/1600/06/2011). Upon the ethical clearance, a letter of cooperation was obtained from the school of nursing to collect data. Permission was also obtained from Debre Markos Comprehensive Specialized Hospital medical director and ART focal person. Confidentiality was maintained by avoiding the registration of personal identifiers like names to the extraction tool. Also, no raw data were given to anyone other than the investigator, and these were fully anonymised.

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Data availability statement Data are available upon reasonable request.

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