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Pattern of anemia and associated parameters in HIV positive patients taking highly active retroviral therapy (HAART)

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***Abstract*---**Objective: This study was designed to determine the pattern of anaemia among HIV positive patients taking HAART. Study Design: Cross-sectional study. Place and Duration: Diagnostic and

research Laboratory Liaquat university of Medical and health sciences Jamshoro, Hyderabad. January 2022-December 2022. Methods: Total 250 HIV patients (aged 18 years and above) of both genders taking HAART for at least the past six months. After taking written informed consent, data was recorded with the help of a structured questionnaire that contained questions regarding basic bio-data and medical history, along with the interpretation of general physical and systemic examination. A blood complete picture was done to evaluate baseline haematological parameters. Data were analyzed using Statistical Package for Social Sciences (SPSS) version 22.0. Results: The mean age of the patients was 32.09 (±SD12.165). Most of the individuals, i.e., 160 (64%), were males, and the number of females was 90 (36%). The mean viral load was 2674.60. Anaemia was present in 210 (84%) patients and 40 (16%)non-anemic with standard parameters. Among anaemic patients, 130 52% had mild disease, moderate anaemia was found in 105 (42%), and only a few (n=15) 6% had severe anaemia. A highly significant association (p-value <0.001) was found between HIV and the different age groups (p-value<0.001), gender (p-value=0.006) and there was a borderline significant association was found between the presence of anaemia and HIV (p-value=0.091). Conclusion: Anemia is the frequently occurring haematological parameter in HIV positive patients.

***Keywords*---**anemia, highly active retroviral therapy (HAART), prevalence, severity, HIV.

Introduction

Haematological abnormalities are the second leading cause of morbidity and mortality in HIV-positive individuals.[1] Anemia is a frequent haematological abnormality that is an independent predictor of HIV disease progression in patients living with HIV.[2] Among various research settings, the prevalence of anaemia in HIV positive patients has been estimated to range from 63% to 95%.[3] Reduced blood haemoglobin levels below the W.H.O.-defined threshold (i.e., less than 12 g/dL for females and less than 13 g/dL for males) are indicative of anaemia, which lowers the red blood cells' ability to carry oxygen.1 According to WHO standards, anaemia is defined22 as having a haemoglobin concentration (Hb) of less than 13 g/dl in men and less than 12 g/dl in women. Haemoglobin levels between 8 and 13 g/dl for men and 8 to 12 g/dl for women are considered mild anaemia. A haemoglobin level of less than 8 g/dl for both males and females was considered severe anaemia.[4] This disease condition is a global health problem that affects all ages. The global prevalence of all-ages anaemia was 22.8% in 2019, which has been improved since 1990, when it was reported to be 27.0%. Internationally, 3.4% of anaemia cases were severe, 42.5% were moderate, and 54.1% were mild.[5]

Iron deficiency anaemia takes the lead among the many variants and manifestations of anaemia. It is the most prevalent anaemia, accounting for more than half of all anaemia cases worldwide.[6] Other etiologies of anaemia include:[7]

Blood loss, Infections (parasitic [Hookworm], chronic [tuberculosis], viral Human Immunodeficiency Virus – HIV), malaria and genetic blood disorders (sickle cell anaemia) `In persons with HIV, anaemia presents an additional burden and can lead to an increase in mortality and reduced quality of life.[8] Studies done in low-income settings stated that the anaemia in HIV-infected persons ranges from 40% to 90%, depending on geographical location and definition of anaemia.[9] HIV infected women are highly afflicted, ranging from 50% to 80%.[10]

Anaemia in HIV is caused by a reduction in red blood cell synthesis in the bone marrow from HIV infection; opportunistic infections (OIs) such as Kaposi's sarcoma and Hodgkin's lymphoma; and vitamin B12 malabsorption from HIV-related gastric pathologies. Risk factors of anaemia in HIV include female gender, advancing HIV disease (CD4 counts<200 cells/microliter), pregnancy, injection drug use, and coinfection with illnesses that cause anaemia.[11] Anaemia is a significant clinical concern in HIV-positive patients, with prevalence estimates ranging from 10% in asymptomatic HIV-positive patients to 92% in individuals with AIDS.[12] Human immunodeficiency is linked with haematology anomalies such as anaemia. Many causes may be attributed to this association [18-19], yet anaemia is deemed the strongest predictor of death.[13]

Materials and Methods

This was a cross-sectional study including the patients with confirmed diagnosis of HIV and taking HAART therapy were identified from the HIV clinic and Medical Out patients Department at Liaquat University of Medical & Health Sciences, Jamshoro and Hyderabad, and they provide their blood sample. HIV infected patients who were HAART for a minimum period of 6-months.Either gender with age more than 18 years were included. Patients taking any antiviral medicines other than HAART and pregnant women with diagnosed hematological disorders were excluded.

All these patients were invited and informed consent was taken. Laboratory work for blood complete picture was carried out at Department of Pathology, Liaquat University of Medical and Health Sciences Jamshoro and Daignostic and Research Laboratory Hyderabad. Initial Sample size calculated was 243 and 7 more patients were added to avoid any missing data influence. Thus a total of 250 patients were recruited. Fresh blood samples were collected (while adhering to strict aseptic procedures) in tubes devoid of anticoagulant and in tubes containing anticoagulant EDTA. An automated analyzer was used to perform a complete blood count, Blood was drawn into a small tube that was linked to the needle. A bandage was wrapped around the location of the blood draw. The blood sample was transferred to LUMHS's D & R laboratory for examination. World Health Organization (WHO) defined cut-offs of the parameters were taken as standard for comparison with HIV positive patients in our study.

Utilising SPSS 22.0, data was examined. Quantitative information was presented as a number and a percentage (No &%). To determine the relationship between anaemia and HIV-infected individuals on HAART, quantitative data was reported as mean and standard deviation (X SD) and analysed using Pearson's correlation

coefficient and the chi square test. A P value of 0.05 or lower was regarded as statistically significant.

Results

The mean age of the patients was 34 (SD ± 5) years ranging from 19 years to 62 years (Figure 1).

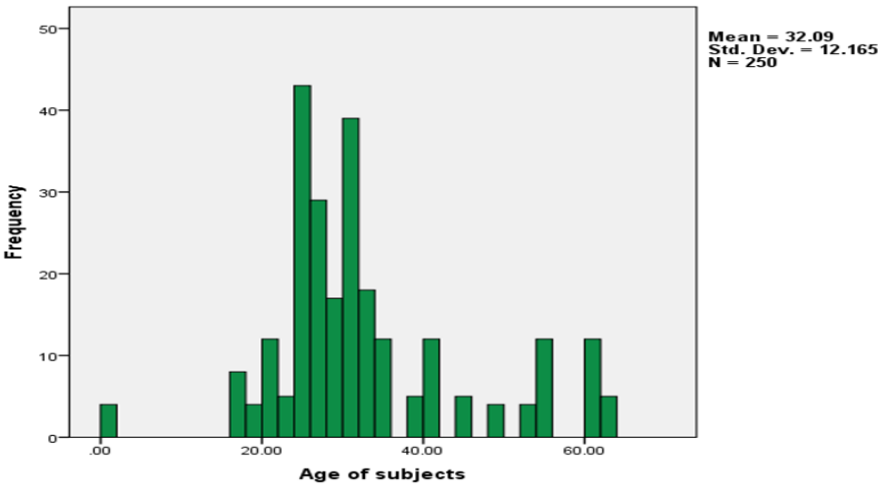

Figure 1. Distribution of the age of the study subjects

Out of 250 enrolled subjects, most of the individuals i.e., 160 (64%) were males, and the number of females was 90 (36%). As presented in Figure 02.

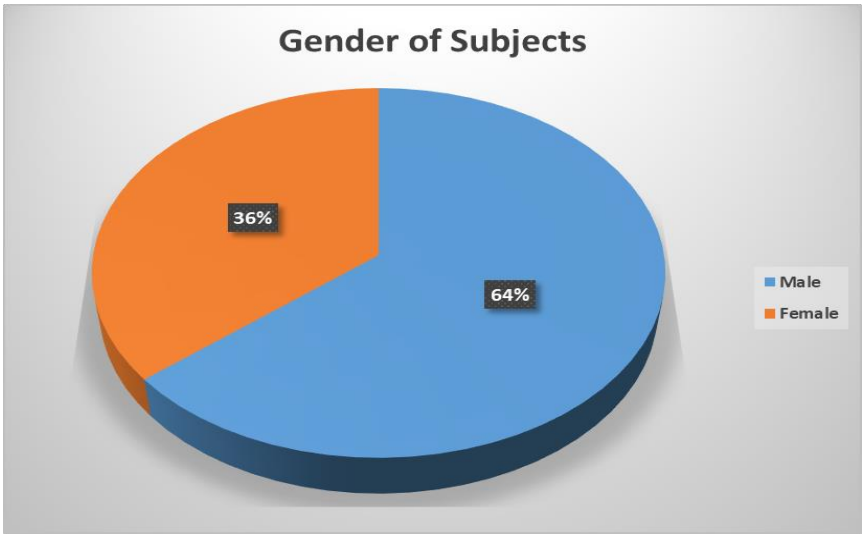

Figure 2. Distribution of the gender of the study subjects.

Hematological parameters

Anemia was present among a cumulative total 210 (84%) of the sample, on the contrary 40 subjects (16%) were non-anemic as demonstrated in Figure 03. Figure 04 exhibits that most of the individuals suffering from mild (130) 52%, and moderate (105) 42% anemia, and only a few (15) 6% facing severe anemia.

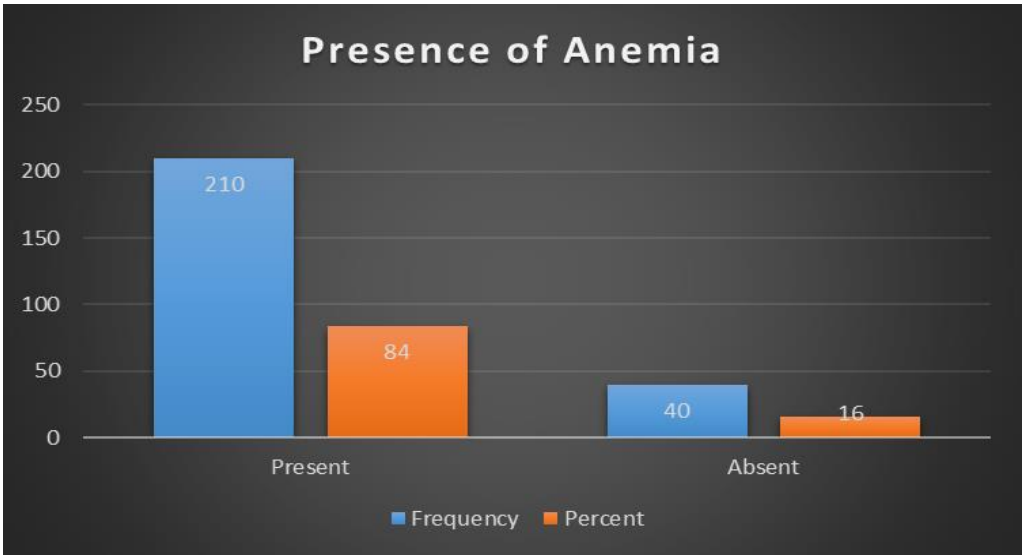

Figure 3 shows the presence of anemia in the study subjects.

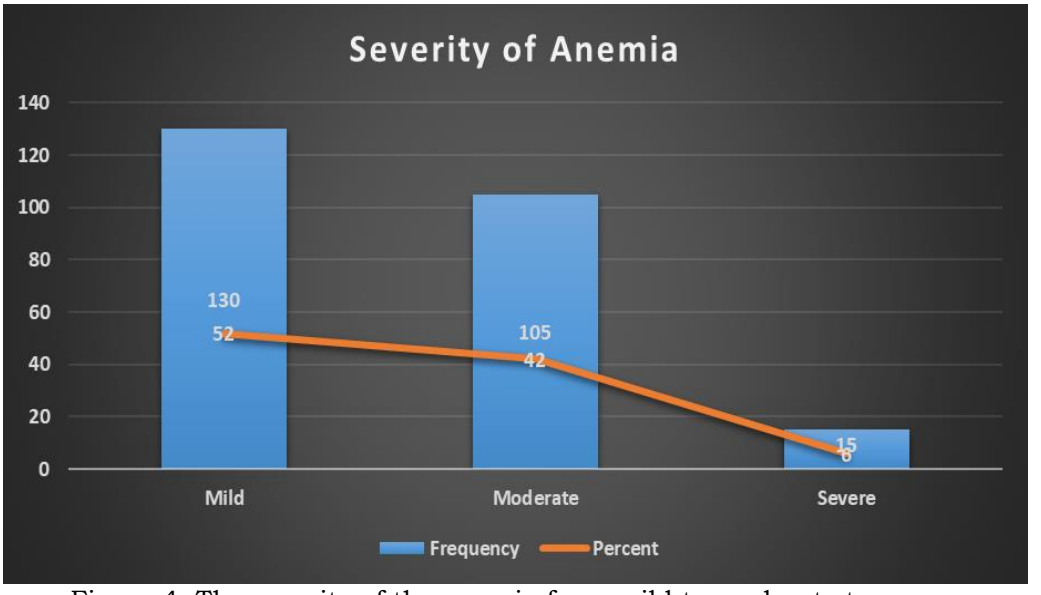

Figure 4. The severity of the anemia from mild to moderate to severe

All hematological paraemeters including hematocrate, white cell count, and pletelets were reduced as compared to the WHO defined criteria of normal limits. A summary is given in Table 1.

Table 1. Summary of descriptive statistics of blood parameters

Variable	Mean	Std. Deviation	Range	Normal Standard according to WHO criteria
Hemoglobin	10.13	1.79	7.0- 15.1	Male:14-18g/dl Female:12-16g/dl
Hematocrit	31.03	4.61	22.9- 43.0	Male:38.3-48.6% Female:35.5-44.9%
Red Blood Cells of subject	4.14	0.52	3.35-5.33	Male:4.35-5.65(mcL) Female:3.92-5.13(mcL)
Mean Corpuscular Volume	75.27	9.02	54.9-90.6	78-98fL
Mean Corpuscular Hemoglobin	24.71	4.00	16.7-32.1	27-35pg
Mean Corpuscular Hemoglobin Concentration	32.53	1.71	29.1-35.7	31-37%/g/dL
White Blood Cells	9.00	5.52	1.93-29.05	4.6-10.8X10E9/L
Neutrophils	66.71	11.68	46.7-94.6	34.9-76.2%
Lymphocytes	24.66	10.82	0.9-43.8	17.5-45%
Monocytes	6.24	1.63	3.5-9.9	3.9-10%
Eosinophils	2.22	1.95	0-8.0	0.3-7.4%
Basophils	0.18	0.30	0-1.1	0-1%
Platelet Count	245.47	95.86	80.0-480	154-433X10E9/L
erythrocyte sedimentation rate	42.99	31.72	5-145	Male:0-22mm/hr Female:0-29mm/hr

Derangemnets were observed in the mean value of direct bilirubin, SGPT, Alkaline phosphatase and Gamma-glutamyl Transferase in our sample population. A summary of the liver function test parameters is given in Table 2.

Table 2. Summary of descriptive statistics of Lipid profile, Plasma Glucose random and Serum creatinine

Variable	Mean	Std. Deviation	Range	Normal standard value
Serum Bilirubin (Total)	0.65	0.24	0.17-0.93	0.1-1.2mg/dL
Serum Bilirubin (Direct)	0.30	0.04	0.05-0.62	0.1-0.3mg/dL
Serum Bilirubin (Indirect)	0.35	0.20	0.12-0.53	0.2-0.8mg/dL
Serum Glutamic-pyruvic Transaminase (Alanine Aminotransferase)	62.00	53.450	10-112	7-56units/litr of serum
Alkaline phosphatase	82.50	3.74	79-338	30-120(IU/L)
Gamma-glutamyl Transferase	16.50	8.02	9-122	5-40U/L

Discussion

The study showed a decline in blood complete picture parameters with wide variation among HIV positive patients taking HAART therapy. Anemia is appears to be a predominant feature. Anemia is a key source of concern for HIV/AIDS patients since it is a crucial driver of disease progression. Around 64% of the subjects enrolled in this study were male and 36% were females. This study showed that majority of the HIV/AIDS patients 68% were from urban area and only 32% were from rural residents. Present study indicated that a large proportion 82% of the HIV/AIDS patient were anemic on the contrary 16% of the HIV/AIDS patients were non-anemic. Aynalem et al. performed a research from 2013 to 2018 at Debre Berhan Referral Hospital in Ethiopia,[14] reported 26.2% of the AIDS/HIV positive adults had anemia. Many other studies revealed similar results in South Africa (25.8%), Nigeria (24.3%), and Rawanda (29%).[15] However, compared to studies conducted in other countries, the prevalence of anemia in HIV-positive patients was higher in this study. Our results, on the other hand, were significantly greater than those observed in investigations of patients from China (39.2%)[16], Nepal (55.8%)[17], Bayamón, Puerto Rico (41%)[18], Nigeria (60.61%), and Tanzania (56%) . Nevertheless, amongst the patents of HIV, the overall prevalence of anemia was higher than that found in adults from Ghana (23.8%), India (16.2%), and Jimma (23.1%).[20-23]

The prevalence of mild, moderate, and severe anemia in the present research was 52 percent, 42 percent, and 6 percent, respectively, which is greater than the findings of a Chinese study. These inequalities in the burden of anemia among HIV-positive patients may be due to differences in sample size (for example, the sample size was high in Nigeria), changes in treatment regimens, research participant characteristics, and sociodemographic disparities. Furthermore, every participant in this study was HIV-positive and on antiretroviral therapy (ART). This might also indicate that HAART helps to reduce HIV-related anemia by lowering the prevalence of chronic diseases, OIs, and increasing patients' nutritional status.

Anemia was shown to be significantly associated to OI, WHO clinical stage, low BMI, and anti-TB treatment.[97] Advanced WHO clinical staging has been identified as a separate risk factor for anemia in HIV/AIDS patients. Adults with advanced WHO clinical stage HIV/AIDS were three times less likely to develop anemia at the commencement of ART than those with WHO clinical illness stages I and II. A recent study was out in Ethiopia supports this theory.[24] Anemia is a common health disease that affects people of all ages, genders, and nationalities. It affects about a third of the global population.[25] 109 Anemia is linked to higher morbidity and mortality in women and children, as well as worse birth outcomes, lower work productivity in adults, and poorer cognitive and behavioral development in children, according to the research. The most vulnerable age groups are preschoolers and women of reproductive age, although no age group is exempt.[26]

According to local literature, more than half of the population (51 percent) of women of reproductive age in Pakistan are anemic.[27] Non-pregnant women had a lower anemia burden than pregnant women. According to the most recent National Nutritional Survey in Pakistan, around 41.7 percent of women of

reproductive age were anemic, with a slightly higher percentage in rural (44.3 percent) settings than in urban (40.2 percent).[28] It is vital to mention that iron deficiency is the most common kind of anemia in Pakistan. Sindh has the highest frequency of iron deficiency anemia in Pakistan, followed by Balochistan (19.0 percent) and Punjab (18.7 percent). According to recent data, 77 percent of reproductive-aged women in Sindh are anemic, with 7.8 percent, 48.7 percent, and 20.8 percent classified as severely, moderately, and mildly anemic, respectively.[29]

The mean age of the sample in this research stood at 34 (SD ± 5) years. Though seemingly this contradicts the literary findings, which claim that the condition is more common at extremes of age and not among middle aged individuals such as in this research, however, it is important to note that the inclusion criteria of this research included a mandatory condition of being HIV positive and taking HAART.[30] Hence the age difference is justified. Research suggests that HIV is more common among sexually active men and women, and since according to the Pakistan Demographic Health Survey the mean age of marriage in Pakistan is 24, it is only likely that a pattern of age distribution such as ours may be produced, wherein the prevalence increases till middle age and then declines in the extremes of age.[31]

The five largest and most comprehensive studies, conducted in the United States and Europe, examined the association between anaemia and survival. The largest of these studies, by Sullivan et al.[32], examined the impact of anaemia on survival using the medical records of 32,867 HIV-positive individuals enrolled in the multistate Adult & Adolescent Spectrum of HIV Illness Surveillance Project. Patients who acquired anaemia prior to the first measured CD4 count, patients who did not monitoring after the date of the first recorded CD4 count, patients without a recorded haemoglobin level or a CD4 count after base review, and patients who did not acquire anaemia were eliminated from the study.

The authors used Kaplan-Meier methods to estimate median survival, and proportional hazards regression to assess the effect of anaemia on survival after the first CD4 count while managing for CD4 measure, AIDS diagnosis, age, gender, HIV exposure mode, neutropenia and a condition called antiretroviral therapy, and Pneumocystis carinae the illness prophylaxis. Individuals who had or did not have anaemia were compared for their risk of dying, as well as those who had recovered or failed to recover from anaemia (3,203 individuals were available for the recovery study). Regardless of starting CD4cell count, those with anaemia had a considerably shorter median life than those who did not have anaemia (P= 0.0001).[33]

Mocroftet al.[34] studied the connection between haemoglobin levels with mortality in the planned EuroSIDA undertaking, involving a European cohort of 6,725 HIV patients in 52 locations. Kaplan-Meier methods were used to compare the proportion of deaths in the following categories of individuals: (1) those who did not have anaemia; (2) those who had mild anaemia, characterised by haemoglobin levels that varied from 80 g/L to 140 grammes per litre in men and 80 g/L to 120 g/L in women; and (3) those who had severe anaemia, stated as haemoglobin levels of 80 g/L in both sexes.

The death rates in the three groups were noticeably different at the 12-month follow-up: Patients with mild anaemia died at a rate of 3.1%, those with moderate anaemia at 15.9%, and those having severe anaemia died at 40.8% (P=0.0001). After adjusting for gender, age, risk group, therapy, and Hiv status using a Cox multivariate model and stratifying for clinical centre, it was discovered that both haemoglobin level & CD4 cell count were independently connected to death risk. The risk ratio (HR) of mortality associated with a 10 g/L decrease in haemoglobin was 1.39 (P = 0.0001)35.

A majority of the sample was noted to be hailing from a middle socioeconomic status and a rural background. This is synonymous with published literature that suggest high prevalence of HIV among the rural areas of Sindh. Hence most of the study participants may have hailed from such areas. Anemia was found in 85.8% of the participants, with the majority of them suffering from mild to moderate anemia and just a handful suffering from severe anemia. Anemia was shown to be more common and severe in various genders, age groups, and socioeconomic levels.[6] Elderly individuals reported a greater severity of anemia, and likewise patients from lower socioeconomic status reported a higher prevalence and greater severity of anemia. Women too suffered from severe anemia more commonly that their male counterparts.[36]

The prevalence of severity is higher than the evidence-based values (of normal cohorts) from literature owing to the fact that the research participants were HIV infected. Similarly, when compared to cohorts of HIV positive patients (in literature), the prevalence and severity are somewhat mild. This is attributable to the fact that all patients in the sample were undergoing treatment (HAART).[37] According to published research from Indonesia, "anemia is less prevalent among HAART patients (7.4 percent), but ZDV usage is related with anemia" (20.3 percent).[38] They also found 14.3 percent of individuals to have mild anemia, 1.9 percent to have moderate anemia, and no patients to have severe anemia." An observational study done in Ethiopia revealed a prevalence of anemia of 23%. Being HAART-naive, having a history of anti-tuberculosis drugs, utilizing a ZDV-containing HAART regimen, and having CD4 counts below 200 cells/L were all linked to anemia in adult HIV patients.[119]

According to a larger cross-sectional research comprising 54 HIV patients in Ethiopia, the overall prevalence of anemia is 23.1 percent, with anemia observed in 16.2 percent of HAART experienced persons and 29.9 percent of HAART naïve individuals. According to the research, 1.9 percent, 25.9 percent, and 72.2 percent of all patients had severe, moderate, or mild anemia. According to their results, the kind of HAART regimen and the duration of HAART are significantly related to anemia.[37]

Anemia was the most common cytopenia among HAART patients, accounting for 43.1 percent (56/130) in another research. Given this context, the goal of this study was to assess the severity of anemia and its predictors, as well as the severity and morphological features of HIV/AIDS patients receiving a combination of first-line antiviral treatments. [38]This experiment will also give information on the anemia features of individuals who received ZDV and TDF-containing HAART. This study will aid health care providers in analyzing and treating

differences in the hematologic profiles of HIV-infected people on ZDV and TDF-containing HAART.

Conclusion

According to the findings of this study, the prevalence of anemia in HIV/AIDS patients is greater than in most prior investigations. As a consequence, the government should prioritize regularly monitoring HIV patients with low Hgb levels and opportunistic diseases, as well as improving proper TB medication and nutritional diversification.

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