

**Original Article**

Clinico-Haematological profile of Anaemia in Diabetic patients: One year study from a region of Jammu and Kashmir

Authors

Bilal Musharaf¹, Tazeen Jeelani*², Danish Rafiq³, Nausrat Ali¹, Manzoor Ahmad Sofi¹

¹Senior Resident Dept of Pathology Govt Medical College Srinagar Jammu and Kashmir India 190010
Email: bilalbanday@gmail.com, nausratali@gmail.com, dr.manzoor123@yahoo.com

²Lecturer Dept of Pathology Government Medical College Srinagar Jammu and Kashmir India 190010

³Assistant Professor Dept of Pathology Govt Medical College Chamba Himachal Pradesh India 176310
Email: drdanishkhan19@gmail.com

*Corresponding Author

Tazeen Jeelani

Lecturer Dept of Pathology Govt Medical College Srinagar Jammu and Kashmir India 190010

Phone: +919070129836, +919018267086, Email: tazeenjeelani@gmail.com**Abstract**

Introduction: Type 2 diabetes has recently escalated in all age groups and is now being seen in younger age groups, including adolescents, especially in high risk populations. Anaemia is one of the common finding in diabetes, particularly in patients with albuminuria or renal impairment. Recent studies have shown that at least 1 in 5 out patients with type 1 or type 2 diabetes in tertiary clinics have anaemia, in which it constitutes a significant additional burden.

Material & Methods: Study was conducted in the Postgraduate department of Pathology and department of Medicine, Government Medical College and associated hospitals, Jammu. It was prospective extending over a period of one year from 1st November 2011 to 31st October 2012. Observations comprised of: History, General physical examination, Systemic examination, haematological Investigations Biochemistry including urine examination.

Results: The main study was done on 87 patients. Out of 87 cases 41(47.12%) patients showed microcytic blood picture, 24(27.58%) patients showed dimorphic blood picture, 19(21.83%) patients showed predominantly macrocytic blood picture and 3(3.44%) patients showed normocytic normochromic blood picture. The prevalence of anaemia among diabetics in our study came out to be 58%.

Conclusion: Anaemia is an important cause of physical and mental impairments in diabetic patients including malaise, fatigue, weakness and other symptoms. Although anaemia represents an unwelcome additional burden in patients with diabetes. Fortunately, anaemia can be treated, and improved quality of life can be achieved.

Keywords: Anaemia; Diabetes; Iron deficiency; Albuminuria; Megaloblastic anaemia.

Introduction

Diabetes was recognized in antiquity and its clinical features were recorded over 3500 years

ago in the Egyptian 'Elbers' papyrus. Type 2 diabetes has recently escalated in all age groups and is now being seen in younger age groups,

including adolescents, especially in high risk populations^[1]. The significant impact of anaemia on the lives of people with diabetes was highlighted nearly a decade ago when it was suggested that anaemia in people with diabetes is unrecognised, undetected and untreated^[2]. Anaemia is one of the common finding in diabetes, particularly in patients with albuminuria or renal impairment. Recent studies have shown that at least 1 in 5 outpatients with type 1 or type 2 diabetes in tertiary clinics have anaemia, in which it constitutes a significant additional burden^[3]. A WHO expert group proposed that anaemia should be considered to exist when haemoglobin is below the cut-off levels as shown in Table-1. Anaemia is associated strongly with an increased risk of diabetic complications including nephropathy, retinopathy, and heart failure^[4]. Anaemia can cause recognised forms of morbidity such as fatigue and exertional breathlessness, but in patients with diabetes, anaemia has also been found to be an independent risk factor for diabetic retinopathy^[5]. Some of the Potential contributors to the development of anaemia in diabetes, include: Chronic blood loss, Iron deficiency, Vitamin B12 or folate deficiency, relative erythropoietin deficiency, Erythropoietin resistance in association with chronic infection or inflammation, Nephrotic syndrome, Autonomic neuropathy, ACE inhibitors/Angiotensin II receptor blockers. Reduced red cell survival revolve around either impaired production and/or response to EPO, with or without concomitant haematinic deficiency. In patients with diabetes it was found that EPO deficiency was present in 34% of anaemic, and abnormal haematinics in 40%, but anaemia was unexplained in 26 % of the cases^[6]. There is evidence that erythropoietin levels in patients with diabetes and anaemia are inappropriately low^[7]. Anaemia is associated with a poorer prognosis in diabetic-associated co-morbid conditions, but targeted correction of anaemia has improved diabetic patient's quality of life^[8].

Material & Methods: The present study was conducted in the Postgraduate department of Pathology and department of Medicine, Government Medical College and associated hospitals, Jammu. The study was prospective extending over a period of one year from 1st November 2011 to 31st October 2012. Various observations were made in all the diabetic patients who were the part of this study. Observations comprised of:-

A} Clinical presentation

- 1) History
- 2) General physical examination
- 3) Systemic examination

B} Investigations

- 1) Haematological
- 2) Biochemistry including urinary examination.

The Inclusion criteria for the study consisted of: All patients diagnosed as a case of diabetes mellitus irrespective of age and gender.

The exclusion criteria for the study consisted of: Patients with other systemic disorders, thalassemia and drugs that could result in anaemia were not included in this study.

Haemoglobin estimation was done using the Cyanmet haemoglobin method/Automated cell counter. Red cell indices comprising of mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC) was estimated using automated cell counter. Differential leucocyte count (DLC) was done both manually and by using automated cell counter. Platelets were examined for adequacy on PBF. Count was given in all cases by using automated cell counter. Reticulocyte count was done by staining the blood film with supra-vital stain and reticulocytes were counted amongst 1000 red cells and expressed as percentage. Detailed PBF examination was carried out after staining the blood film by Romanowsky stains (Leishman/May Grunwald Giemsa stains). Bone marrow aspiration (wherever indicated) was performed on posterior superior iliac spine under all aseptic precautions by using salah's aspiration needle.

Perl's Prussian blue staining was done for estimation of marrow iron stores.

Blood glucose estimation comprised of evaluating fasting, random and postprandial glucose levels in all the diabetic patients. HbA1c was measured primarily to identify the average plasma glucose concentration over prolonged periods of time. Renal function test and liver function test was done in all the patients using the automated analyser.

Urinary blood sugar estimation was done using the dipstick method. The level of albumin protein produced by micro-albuminuria were detected by special albumin-specific urine dipsticks.

Results

In our one year study, we had a total of 150 diabetic patients, 117 patients came out to be anaemic and out of these 117 anaemic patients, 30 patients had various underlying disorders known to cause anaemia like anaemia of chronic disease, myelodysplastic syndrome and leukemias. So the main study was done on remaining 87 patients. Anaemia was classified on the basis of peripheral blood film and red cell indices on cell counter which were supported by history and other biochemical investigations. These were also confirmed on bone marrow studies in patients on whom this procedure was carried. The age distribution of the diabetic patients studied, ranged from 25 to 85 years. However most of patients belonged to age group 65 ± 5 years. Among 87 anaemic patients the proportion of females (50 patients) was more than that of males (37 patients) with male female ratio of 1:1.3 Majority of anaemic patients had long standing history of diabetes ranging from 16-20 years and almost all the patients were having type 2 diabetes mellitus and there was only a single case of type 1 diabetes. 8 (8.04%) patients gave a history of having received blood transfusions in the past for severe anaemia (<6 gm% Hb).

Clinical presentation in diabetic patients showed, pallor and generalized weakness was commonest

presentation in 74 (85.05%) and 20 (22.98%) patients respectively.

Over all grading of anaemia in all 87 patients showed that most of the patients were in the category of moderate and severe anaemia. Majority of patients with severe degree of anaemia (Hb <7.9 g/dl) comprised of cases having diabetes mellitus with megaloblastic anaemia, iron deficiency anaemia (Fig-1). More than 50% of the patients included in the study had normal platelet counts. Decreased platelet counts were observed in the patients of severe megaloblastic anaemia. Increased platelet count was seen in patients of iron deficiency anaemia. ANC (absolute neutrophil count) done in all the diabetic patients showed that 80% patients had normal ANC while as 0.66% patient had neutropenia and 19.33% patients had neutrophilia. Packed cell volume was low in most of the patients and ranged from 18% to 45%. The values between 20% to 30% were observed in more than 50% of the total cases.

ESR was seen to be raised in almost all the patients. MCV, MCH and MCHC values varied from case to case. MCV value was a useful parameter in making diagnosis of iron deficiency anaemia (which was 70fl in a patient of iron deficiency anaemia) and megaloblastic anaemia (Fig-2) (highest value was 115.2fl in a patient of megaloblastic anaemia) where MCV values were low and high respectively. In most of the patients with dual deficiency anaemia MCV value was normal.

The reticulocyte count was done in all the patients. Normal reticulocyte count was observed in 71 (81.6%) patients, increased count in 14 (16.09%) patients and decreased count in 2(2.29%) patients. Reticulocytosis was found in those patients who either received haematinics or blood transfusions. The lowest reticulocyte count of 0.3% was observed in a case of severe megaloblastic anaemia.

Out of 87 cases 41(47.12%) patients showed microcytic blood picture, 24(27.58%) patients showed dimorphic blood picture, 19(21.83%) patients showed predominantly macrocytic blood

picture and 3(3.44%) patients showed normocytic normochromic blood picture (Table-2).

It was observed that HbA1c levels were in control range in 43.67 % (38) of diabetic patients.9.19% (8) patients had HbA1c levels less than 5.5 and majority of them comprised of newly diagnosed cases of diabetes. 24.13% (21) of patients had poorly controlled HbA1c levels owing to their continuous high blood sugar levels. Over all the HbA1c levels ranged from 5.2% to 8.6% with the average of 6.54%.

Among 87 anaemic patients, 51 (58.62%) patients had micro-albuminuria, 10 (11.49%) patients had albuminuria of less than 30mg/litre and 26 (29.88%) patients had overt proteinuria.

Urinary protein estimation was done in all 87 anaemic patients and it was observed that overt proteinuria was more common in males than females while as micro-albuminuria was common in females. Overt proteinuria was mostly observed in elderly diabetic patients.

Table-1; Cut-off haemoglobin levels for diagnosis of anaemia

CATEGORY	Hb (gm/dl) (Venous blood)	MCHC
Adult male	13	34
Adult female (non-pregnant)	12	34
Adult female (Pregnant)	11	34
Children (6 months-6 years)	11	34
Children (6years-14 years)	12	34

Table-2: Blood picture of the diabetic patients suffering from anaemia

RBC morphology	No of patients (n=87)	Male	Female	%age
Microcytic hypochromic predominance	41	16(18.39%)	25(28.73%)	47.12%
Dimorphic blood picture	24	13(14.94%)	11(12.64%)	27.58%
Macrocytic predominance	19	10(11.49%)	9(10.34%)	21.83%
Normocytic normochromic picture	3	1(1.14%)	2(2.29%)	3.44%

Table-3: Prevalence of anaemia in diabetes in different studies

Studies	Prevalence (%)
Thomas MC et al (2003)	23%
Almoznino et al (2010)	40.2%
Jones SC et al (2010)	15%
Ahmad AT et al (2010)	34.7%
Bonakdaran et al (2011)	19.6%
Present study	58%

Fig-1: PBF in a case of iron deficiency anemia showing anisopoikilocytosis with cigar cell. (Leishman’s stain X 1000)

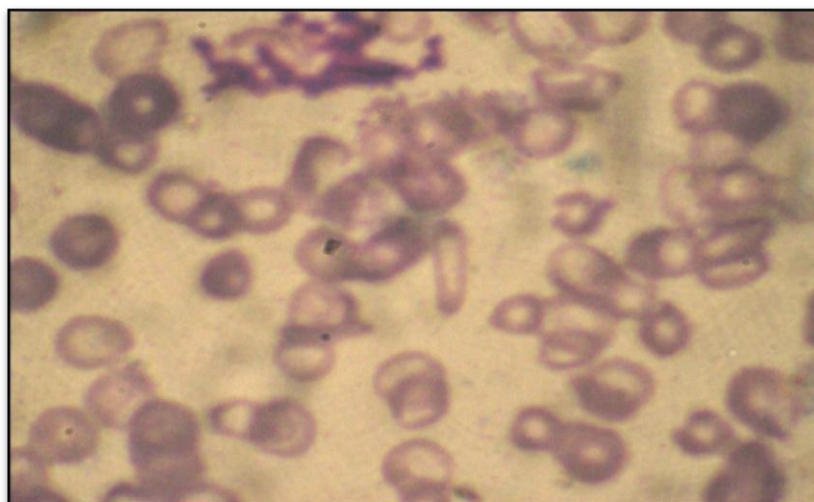


Fig-2: PBF showing macro-ovalocytes and eosinophils in a case of megaloblastic anemia. (Leishman's stain X 400)

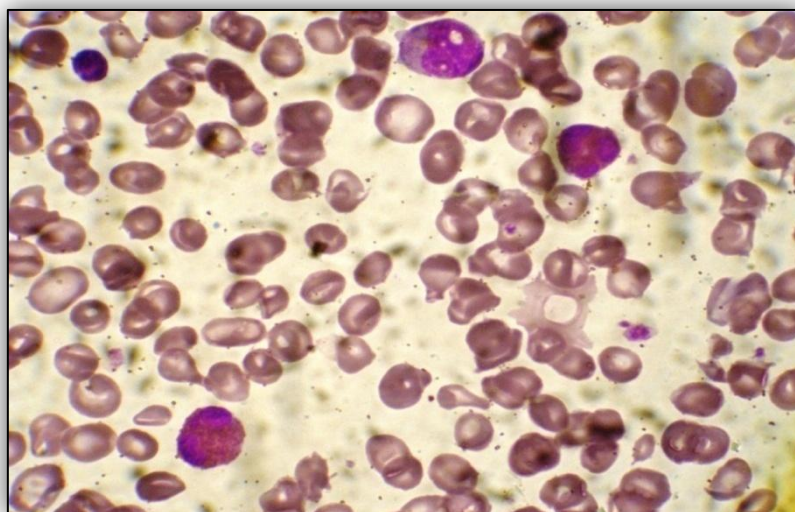
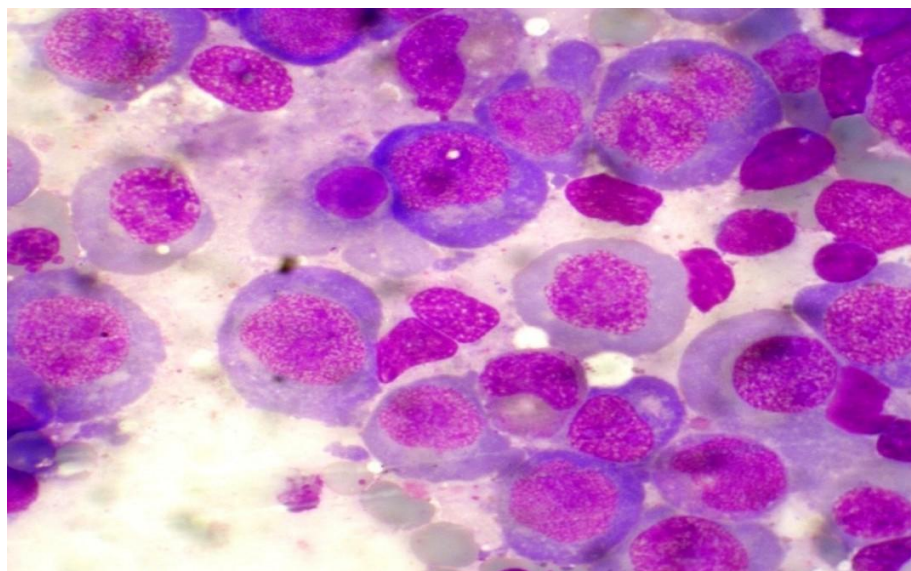


Fig-3: Bone marrow smear from a case of megaloblastic anemia showing classical megaloblasts. (MGG X 1000)



Discussion

There are plenty of studies on the prevalence of anaemia in high risk groups like pregnant and lactating females, elderly and children. Prevalence of anaemia in these groups has been found to vary from 50-90% in different parts of India^[9]. Stevens *et al* in their study suggested that anaemia in people with diabetes goes unrecognized, undetected and untreated^[10]. In our study, the mean age of patients was 55.10 years. This was in accordance with the study conducted by Bonakdaran *et al* in 2011^[11]. The Male female ratio in our study was comparable to the studies

done by Bonakdaran *et al*⁽¹¹⁾ and Ahmad AT *et al*^[12]. A total of 87 patients comprised the main study group and thus the prevalence of anaemia in diabetics was evaluated to be 58%. This prevalence is much higher when compared to other studies (Table-3).

The high prevalence of anaemia observed in our study in comparison to other studies can be explained by the fact that anaemia is rampant in our region owing to poor nutrition, low Socio-economic status, unhealthy dietary habits and alcoholism.

Among 87 anaemic patients studied 86 (98.85%) patients were having type 2 diabetes while 1(1.14%) patient was having type 1 diabetes. Dimkovic N et al in their study included a total of 539 patients with 20% type I and 80% type II diabetic patients^[13]. In another study conducted by Thomas MC et al showed 80% of patients were having type 2 diabetes^[14].

In our study reticulocyte count was done in all patients and the mean reticulocyte count was 3.56%, this was in accordance with the study carried out by Waggiallah H et al in 2011, where they found mean reticulocyte count was 4.4%^[15].

The detailed examination of peripheral blood smears yielded important diagnostic clues and confirmatory evidence. Brill JR and Baumgardner DJ mentioned in their study that the diagnostic accuracy was enhanced when PBF and bone marrow findings were combined with clinical examination and other relevant investigations^[16].

In severe megaloblastic anaemia, circulating megaloblasts (Fig-3) were also seen in the peripheral blood smears of the patients. Dimorphic blood picture with presence of both microcytes and macrocytes was suggestive of dual deficiency anaemia. Serum ferritin and vitamin B₁₂ /folic acid levels were low in these patients. The red blood cells morphology in cases of dual deficiency anaemia was usually dimorphic^[17]. Presence of microcytes as well as macrocytes in the peripheral smear is indicative of dual deficiency anaemia^[18]. Glycosylated haemoglobin (HbA1c) concentration is an indicator of average blood glucose concentration over three months and has been suggested as a diagnostic or screening tool for diabetes^[17]. Thomas MC et al reported that the prevalence of albuminuria (micro or macro) varied between 27% and 43% in the diabetic patients and concluded that patients with persistent micro-albuminuria had 4 times the risk of anaemia and patients with persistent macro-albuminuria had 12 times the risk of anaemia^[14]. In a study Jones SC et al found that most common hematological abnormality was low levels of vitamin B₁₂ and/or ferritin, occurring in

40% of diabetic patients with anaemia^[19]. Anaemia in diabetics is more complex and multifactorial and in addition to erythropoietin deficiency other causes can be inflammation, nutritional deficiencies, autoimmune disease, drugs or hormonal changes^[20].

Conclusion

The prevalence of anaemia among diabetics in our study came out to be 58%. This high prevalence can be explained by the fact that anaemia is very common in our region owing to poor nutrition, low socio-economic status, unhealthy dietary habits and alcoholism. Nutritional deficiency was the most common cause of anaemia in the diabetic patients in our setup. Among the causes iron deficiency anaemia was the commonest one in diabetics (47.12%). Anaemia is an important cause of physical and mental impairments in diabetic patients including malaise, fatigue, weakness and other symptoms. Although anaemia represents an unwelcome additional burden in patients with diabetes. Fortunately, anaemia can be treated, and improved quality of life can be achieved.

References

1. WHO Technical Report Series No 916 (TRS-916): 2003.
2. Stevens PE, O'Donoghue DJ, Lameire NR. Anaemia in patients with diabetes: unrecognised, undetected and untreated? *Curr Med Res Opin.* 2003; 19(5):395-401.
3. Merlin Thomas C. High prevalence of anaemia in Diabetes linked to erythropoietin deficiency, 2006: vol. 26, no4; 275-282.
4. David C Holland, Miu Lam. Predictors of hospitalization and death among pre-dialysis patients: *Nephrol Dial Transplant*, 2000: 15; 650-658.
5. David MD, Fisher MR et al. Risk factor for high risk proliferative diabetic retinopathy and severe visual loss: *Invest Ophthalmol Vis Sci*, 1998 Feb; 39(2); 233-252.

6. Jones SC, Smith D, Nag S. Prevalence and nature of anaemia in a prospective, population-based sample of people with diabetes: Teesside anaemia in diabetes (TAD) study. *Diabet Med.* 2010 Jun; 27(6):655-9.
7. Bosman DR, Winkler AS, Marsden JT et al. Anaemia with erythropoietin deficiency occurs early in diabetic nephropathy. *Diabetes Care.* 2001 Mar; 24(3):495-9.
8. David R Thomas. Anaemia in Diabetic patients: *Clinics in Geriatric Medicine*, Aug 2008: Vol. 24, Issue 3, 529-540.
9. P Malhotra, Savita Kumari et al. Prevalence of Anaemia in Adult Rural population of North India: *J. Assoc Physicians India*, Jan 2004: 52; 18-19.
10. Stevens PE, O'Donoghue DJ, Lameire NR. Anaemia in patients with diabetes: unrecognized, undetected and untreated? *Curr Med Res Opin.* 2003; 19(5):395-401.
11. Bonakdaran S, Gharebaghi M, Vahedian M. Prevalence of anaemia in type 2 diabetes and role of renal involvement. *Saudi J Kidney Dis Transpl.* 2011 Mar; 22(2):286-90.
12. Ahmed AT, Go AS, Warton EM et al. Ethnic differences in anaemia among patients with diabetes mellitus: the Diabetes Study of Northern California *Am J Hematol.* 2010 Jan; 85(1):57-61.
13. Dimković N; Studijske Grupe. Anaemia in patients with diabetes mellitus. *Med Pregl.* 2007 May-Jun; 60(5-6):225-30.
14. Thomas MC, MacIsaac RJ, Tsalamandris C et al. Unrecognized anaemia in patients with diabetes: a cross-sectional survey. *Diabetes Care.* 2003 Apr; 26(4):1164-9.
15. Waggiallah H, Alzohairy M. The effect of oxidative stress on human red cells glutathione peroxidase, glutathione reductase level, and prevalence of anaemia among diabetics. *N Am J Med Sci.* 2011 Jul; 3 (7):344-7.
16. Brill JR, Baumgardner DJ. Normocytic anaemia. *Am Fam Physician.* 2000 Nov 15; 62(10):2255-64.
17. Kovács F, Szakál I, Révész K, Lévai A, Dobos A. Anaemia and iron metabolism in patients with type-2 diabetes mellitus. *Orv Hetil.* 2006 Feb 26; 147(8):345-9.
18. McCance DR, Hanson RL, et al. Comparison of tests for glycated haemoglobin and fasting and two hour plasma glucose concentrations as diagnostic methods for diabetes. *BMJ* 1994; 308: 132-38.
19. Jones SC, Smith D, Nag S. Prevalence and nature of anaemia in a prospective, population-based sample of people with diabetes: Teesside anaemia in diabetes (TAD) study. *Diabet Med.* 2010 Jun; 27(6):655-9.
20. Kengne AP, Czernichow S, Hamer M, et al. Anaemia, haemoglobin level and cause-specific mortality in people with and without diabetes *PLoS One.* 2012; 7(8):e41875. Epub 2012 Aug 2.

Abbreviations

WHO: World health organisation, EPO: Erythropoietin, ANC: Absolute neutrophil count, ESR: Erythrocyte sedimentation rate, MCH: Mean corpuscular haemoglobin, MCV: Mean corpuscular volume, MCHC: Mean corpuscular haemoglobin concentration, Hb: Haemoglobin.