ISSN Print: 2394-7500
ISSN Online: 2394-5869
Impact Factor: 5.2
IJAR 2019; 5(10): 344-347
www.allresearchjournal.com
Received: 24-08-2019
Accepted: 29-09-2019

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A study of lipid profile in iron deficiency anemia at a tertiary care teaching hospital

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Abstract
This study was done on 50 anaemic cases and 50 non anaemic controls to study the clinical presentation and effect on lipid profile of anaemia.
Younger individuals are more likely to have severe anaemia. Cases with severe anaemia have more symptoms. They have higher mean pulse rate, lower mean blood pressure and mean BMI. Vegetarians are more likely to have severe anaemia. Cases with severe anaemia also have more signs on examination. Anaemia is associated with significant hypercholesterolemia, with lowering in all lipid subfractions. The extent of hypercholesterolemia is proportional to the severity of anaemia. The type of anaemia has no effect on the hypercholesterolemia seen in anaemia.
Further studies are required to study the long term effect of anaemia on the risk of developing atherosclerosis and to study the long term effect of treatment of anaemia on lipid levels and cardiovascular morbidity and mortality.

Keywords: anaemia, hypercholesterolemia, demographic data, lipid profile

Introduction
Current data indicate that serum lipid levels are significantly correlated with the risk of atherosclerosis, which causes coronary artery disease, cerebrovascular disease and peripheral vascular disease, important causes for mortality and morbidity worldwide [1, 2]. The burden of coronary artery disease (CAD) in India is increasing and addressing the risk factors of the same is the need of the hour to reduce its prevalence. Dyslipidemia being one of the important risk factors for CAD, effective preventive measures, including lifestyle modification should be adopted to achieve recommended lipid goals. Another equally important public health concern is the prevalence of anemia [3-8]. Dyslipidemia may be more prevalent in an affluent society, whereas anemia obviously more common in people with low socioeconomic status. Anemia could be associated with low levels of all the subfractions of lipoproteins.
Anaemia is a common disorder in India. Although it may be due to various causes, iron deficiency is most commonly responsible. Anaemia has been reported to have a beneficial effect on the lipid profile. The lowering of lipid levels is not related to the type of anaemia. The decrease in serum cholesterol is not due to a specific lowering of any of the serum lipoprotein families; hypocholesterolemia is caused by a proportional reduction in all the major lipoprotein families. This may have a beneficial effect on the risk of developing coronary artery disease, a disease to which Indians are particularly susceptible [10-14].
The exact mechanism by which anaemia causes a fall in serum lipids is not known. The simplest explanation is a dilution effect (the increased volume of serum in anaemia carrying the same total load of cholesterol). Other possibilities are increased utilization of cholesterol by proliferating cells, decreased endogenous synthesis of cholesterol by the liver due to decreased liver oxygenation, elevated levels of granulocyte-macrophage colony stimulating factor and enhanced receptor mediated removal of LDL in the bone marrow. Correction of anaemia is associated with a rise in serum lipids. In this study, we documented and evaluated the demographic, lipid profile and clinical features of anaemia patients.

Materials and Methods

This is a study which has been carried out in the Department of Medicine, Sriram Chandra Bhanj Medical College & Hospital (S.C.B Medical College and hospital). The data for this study was collected from patients who presented to Sriram Chandra Bhanj Medical College & Hospital, Department of General Medicine either on inpatient or outpatient basis. A total of 50 cases and 50 controls were enrolled in this study for the period of August 2018 to July 2019. All proven cases of anaemia. Men: Hb < 13 gm%, Women: Hb < 12 gm% were participated in this study.

A detailed history was obtained from the subjects of the study, with special emphasis on age, sex and occupation; nonspecific symptoms of anaemia like fatigability, dyspnoea, giddiness, palpitations and angina; symptoms suggestive of a specific cause for anaemia like pica, dysphagia, abdominal pain pain, bony pain, fever, loss of appetite, weight loss, jaundice, bleeding, malaena, haemoglobinuria, menorrhagia, pregnancy and post menopausal bleeding. Past history of disorders associated with dyslipidemia or anaemia was obtained, including diabetes mellitus, hypertension, ischemic heart disease, cerebrovascular accident, AIDS, recent blood loss and gall stones. Dietary habits and habits like alcoholism and tobacco smoking was ascertained. History of intake of drugs affecting lipid levels, such as oral contraceptives, beta blockers, diuretics, steroids and NSAIDs was obtained. Family history of anaemia, jaundice and gallstones was also obtained.

Each patient was subjected to a detailed general physical examination, with special emphasis on pallor, koilonychias, icterus, pedal edema, lymphadenopathy, glossitis, angular stomatitis, petechiae, haemolytic facies, ankle ulcers, perioral pigmentation and knuckle pigmentation. Pulse, blood pressure, weight, height and body mass indexwas measured.

Thorough systematic examination was made of the cardiovascular system to look for the presence of elevated JVP, venous hum, cardiomegaly, S3 and flow murmur. The respiratory system was examined to look for evidence of pulmonary oedema. Abdomen was examined to look for organomegaly. The central nervous system was examined for confusion, muscular weakness, deep tendon reflexes, vibration sense, position sense and romberg's sign.

Venous blood was drawn for investigations like complete haemogram, random blood sugar, blood urea, serum creatinine, liver function tests, and thyroid stimulating hormone levels. A urine sample was obtained for urine analysis, including albumin, sugar and microscopy. Fasting venous blood sample (> 12 hours) was obtained for estimation of lipid profile. T3 and T4 levels, fasting and post prandial (two hours after an oral dose of 75gms of glucose) blood sugar levels, and bone marrow aspiration cytology was done in selected cases based on clinical assessment.

Complete haemogram was performed using the Sysmax automated analyzer. Haemoglobin levels were confirmed by the colorimetric method. Differential count and peripheral smear was done manually using Leishmann's stain by a qualified pathologist. Urine albumin and sugar was estimated by dipstick method. Urine microscopy was done manually by a qualified pathologist. Biochemical analyses were done using the fully automated Technicon RA-XT system by Bayer. TSH, T4 and T3 were estimated using the chemiluminescence method on the fully automated ADVIA Centaur system by Bayer.

Estimation of total cholesterol, HDL and triglycerides was done with the commercially available Autopak cholesterol kit on Technicon RA-XT system. VLDL was calculated using the formula, VLDL = Triglyceride/5. LDL cholesterol was calculated using the Friedewald's equation. LDL = Total cholesterol – (Triglycerides/5) + HDL] mg/dl. All the data documented in excel sheet and analyzed by MS excel 2007.

Results

In order to study the lipid profile among anemic cases and their association with severity of anemia, observations on 50 anaemia patients and 50 non-anaemic control group were taken and data were analysed following statistical procedures as elaborated under material and methods. The results along with interpretations are presented in this chapter. Section1 deals with the demographic profile of cases and their association with level of anaemia. Section 2 presents associated symptoms, risk factors and their relationship with the severity and type of anaemia. Section 3 analyses the association of the severity of anaemia with blood parameters and comparison of lipid profile by anaemic and non-anaemic cases.

Out of 50 cases in group 1,the middle age group, 31-50 years, constituted nearly one-third (34%) of the sample, elderly age group constituted approximately one-third (34%) and the younger age group i.e. 30 years or lower had about one-third (32%) of the cases. In the control group the age distribution is not significantly different from the case group (p=0.997) (Table 1).

Table 1: Age distribution by groups

Age group	Group 1 (Case)		Group 2 (Control)		Total		□□, p
	No.	%	No.	%	No.	%	
≤20	5	10	4	8	9	9	p=0.997
21-30	11	22	13	26	24	24	
31-40	10	20	9	18	19	19	
41-50	7	14	7	14	14	14	
51-60	10	20	10	20	20	20	
61-70	7	14	7	14	14	14	
Total	50	100	50	100	100	100	

In the total sample there were 51 males and 49 females. In the case and control group the males and females were evenly distributed (p=0.841) (Fig 1).

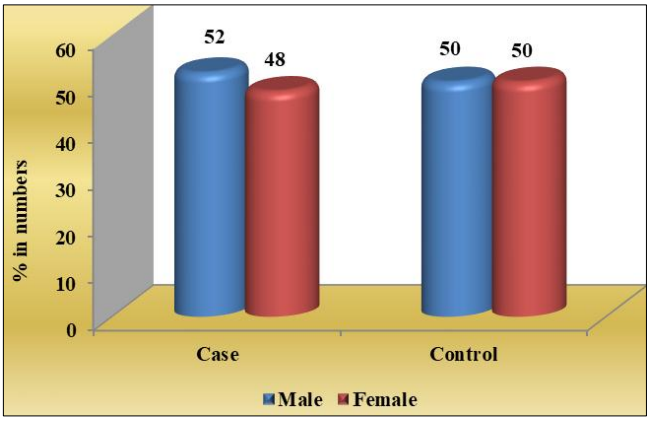

Fig 1: Gender distribution by groups

Table 2 present association of age and severity of anaemia levels. Out of 50 anaemic cases 11 (22%) were having severe, 20 (40%) moderate and 19 (38%) mild levels of anaemia. In each of the levels of severity the proportions of cases among different age groups were not significantly different (p=0.722). This implied the severity of anaemia didn't have any association with age (Table 2).

Table 2: Age distribution with Hemoglobin levels in case group

Age group	Hemoglobin						Total		□□, p
	Severe		Moderate		Mild				
	No.	%	No.	%	No.	%	No.	%	
≤20	2	22.2	3	13.6	0	0	5	10	
21-30	3	33.3	4	18.2	4	21.1	11	22	
31-40	1	11.1	5	22.7	4	21.1	10	20	□□□□
41-50	1	11.1	4	18.2	2	10.5	7	14	□□□□
51-60	1	11.1	4	18.2	5	26.3	10	20	p=0.722
61-70	1	11.1	2	9.1	4	21.1	7	14	
Total	9	100	22	100	19	100	50	100	

Table 3 present severity of anaemia and lipid profile**.** It is seen that the mean TC has the lowest value for severe anaemic cases and highest value for mild anaemic cases. This implied the TC level decreases with the severity of anaemia (p=0.000). The mean HDL has the lowest value for severe anaemic cases and highest value for mild anaemic cases. This implied the TC level decreases with the severity of anaemia (p=0.000). The mean LDL has the lowest value for severe anaemic cases and highest value for mild anaemic cases. This implied the LDL level decreases with the severity of anaemia (p=0.000). The mean VLDL has the lowest value for severe anaemic cases and highest value for mild anaemic cases. This implied the VLDL level decreases with the severity of anaemia (p=0.001). The mean TG has the lowest value for severe anaemic cases and highest value for mild anaemic cases. This implied the Tg level decreases with the severity of anaemia (p=0.001). The mean TC; HDl ratio didn't vary significantly with the severity of anaemia (p=0.972). The mean LDL: HDL significantly decreases with the severity of anaemia (P=0.031) (Table 3).

Table 3: Severity of Anaemia and Lipid profile

Lipid Profile	Severe (n=9)		Moderate (n=22)		Mild (n=19)		Total (n=50)		ANOVA 'p' value
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	
TC	104.0	16.6	125.4	15.2	145.1	16.0	129.0	21.4	0.000
HDL	25.1	4.0	30.4	3.6	34.8	4.6	31.1	5.3	0.000
LDL	61.6	12.3	78.3	9.1	99.5	8.8	83.3	17.0	0.000
VLDL	18.3	4.6	21.9	3.8	24.6	3.9	22.3	4.5	0.001
TG	94.1	20.3	110.5	15.1	119.8	15.7	111.1	18.4	0.001
TC: HDL Ratio	4.2	0.9	4.2	0.6	4.2	0.3	4.2	0.6	0.972
LDL: HDL Ratio	2.5	0.6	2.6	0.4	2.9	0.3	2.7	0.4	0.031

Table 4 presents comparison of mean lipid profile across types of anaemia within levels of anaemia. Lipid profile like TC, HDL, TG, TC: HDL ratio did not differ significantly among types of anaemia like DM, MH, NH and NN. Within each of the 3 levels of anaemia i.e severe, moderate and mild (p>0.05). The mean LDL level varies significantly among anaemia types within severe level of anaemia (p=0.041). But the mean LDL level didn't differ significantly by anaemia types within moderate and mild levels of anaemia (p>0.05). The mean VLDL level didn't differ significantly by anaemia types with severe and moderate levels of anaemia (p>0.05). However, within mild levels of anaemia the mean VLDL varies significantly by anaemia types (p=0.05). This aberration may be due to sampling fluctuation because of small samples in each group. Thus by and large the lipid profile did not vary significantly among the anaemia type with each of the three levels of anaemia (Table 4).

Table 4: Type of Anaemia and Lipid Profile

Lipid profile	Hb (in gm/dl)	Type of Anaemia												Total			ANOVA 'p' value
		DM			MH			NH			NN						
		Mean ± SD of Lipid profile															
		N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	
TC	Severe	7	102.4	18.6	2	109.5	7.8							9	104.0	16.6	0.629
	Moderate	8	123.9	18.2	6	127.2	15.7	5	126.8	17.3	3	123.3	4.2	22	125.4	15.2	0.973
	Mild	6	148.3	9.9	4	141.5	25.1	4	147.5	17.1	5	142.0	17.0	19	145.1	16.0	0.887
HDL	Severe	7	24.3	3.5	2	28.0	5.7							9	25.1	4.0	0.271
	Moderate	8	30.3	3.0	6	31.7	4.5	5	30.4	3.8	3	28.0	3.0	22	30.4	3.6	0.581
	Mild	6	35.8	4.5	4	33.0	5.7	4	35.0	5.8	5	34.8	4.0	19	34.8	4.6	0.846
LDL	Severe	7	57.3	10.3	2	76.5	2.1							9	61.6	12.3	0.041
	Moderate	8	79.5	9.2	6	78.5	11.7	5	80.6	5.2	3	71.0	8.7	22	78.3	9.1	0.524
	Mild	6	100.2	4.9	4	96.8	9.4	4	97.3	14.9	5	102.6	7.8	19	99.5	8.8	0.762
VLDL	Severe	7	17.3	4.2	2	22.0	5.7							9	18.3	4.6	0.223
	Moderate	8	23.3	3.2	6	21.5	5.4	5	20.6	2.1	3	21.0	4.6	22	21.9	3.8	0.645
	Mild	6	25.8	4.0	4	24.0	2.2	4	20.3	2.9	5	27.0	3.2	19	24.6	3.9	0.039
TG	Severe	7	91.7	18.7	2	102.5	31.8							9	94.1	20.3	0.544
	Moderate	8	103.4	18.5	6	115.5	13.2	5	121.0	5.5	3	102.3	6.4	22	110.5	15.1	0.115
	Mild	6	109.8	12.5	4	119.3	20.5	4	130.3	12.5	5	124.0	14.3	19	119.8	15.7	0.213
TC: HDL Ratio	Severe	7	4.3	1.0	2	4.0	0.5							9	4.2	0.9	0.682
	Moderate	8	4.1	0.7	6	4.1	0.8	5	4.2	0.5	3	4.4	0.4	22	4.2	0.6	0.892
	Mild	6	4.2	0.4	4	4.3	0.4	4	4.2	0.3	5	4.1	0.3	19	4.2	0.3	0.81
LDL: HDL Ratio	Severe	7	2.4	0.6	2	2.8	0.6							9	2.5	0.6	0.456
	Moderate	8	2.7	0.4	6	2.5	0.4	5	2.7	0.4	3	2.5	0.1	22	2.6	0.4	0.849
	Mild	6	2.8	0.3	4	3.0	0.4	4	2.8	0.3	5	3.0	0.4	19	2.9	0.3	0.788

Discussion

The observations made in 50 cases of anaemia and 50 non anaemic controls, who presented to Department of Medicine, SCB Medical College and Hospital, Cuttack from august 2018 to july 2019 is discussed here and results have been compared with other similar studies.

All cases in this study were between 14 and 75 years. Majority of the cases were middle aged (30-60 years). Anaemic cases younger than 50 years were more likely to have more severe anaemia, as compared to cases older than 50 years, who were more likely to have less severe anaemia. This is probably due to younger individuals having a higher risk of worm infestation and also the onset of menopause with cessation of menstrual blood loss after the age of 50 years.

The cases consisted of 26 males and 24 females. There was no correlation between sex and severity of anaemia.

The results of this study confirm the findings of previous investigations that the mean serum total cholesterol, HDL, LDL, VLDL and triglyceride levels are decreased in anaemia.

The mean total cholesterol was found to be lower in anaemic cases when compared to controls. The decrease in mean serum cholesterol was not due to a specific lowering of any of the serum lipoprotein families; hypercholesterolemia was caused by a reduction in all the major lipoprotein families, including mean HDL, LDL, VLDL and triglycerides. There was a very large decrease in mean total cholesterol and LDL levels, and a large decrease in mean HDL levels, resulting in a mild fall in mean TC/HDL and LDL/HDL ratios. There was a mild decrease in mean VLDL and triglyceride levels.

Rifkind and Gale [4, 5] in 1967 showed that anaemia was associated with hypercholesterolemia and the decrease in serum cholesterol was not due to a specific lowering of any of the serum lipoprotein families and that hypercholesterolemia was caused by a proportional reduction in all the major lipoprotein families.

Elwood and Mahler [6], in 1970, conducted a study 4070 women, and demonstrated a significant difference in cholesterol between women with haemoglobin levels above and below 10.5 g/dl.

Patients with more severe anaemia were found to have a larger fall in mean total cholesterol and all the lipid sub fractions. This suggests that the severity of anaemia is responsible for the hypocholesterolemia seen in anemia.

A study conducted by Choi [15] *et al*. in 2001 showed that lipid levels in patients with iron deficiency anaemia were directly related to the hemoglobin levels.

The type of anaemia did not have a significant effect on the mean lipid levels. This suggests that it is anaemia per se, and not the type of anaemia that is responsible for the lowering of lipid levels in anaemia.

A study by Westerman [7] in 1975 examined the relationship between hypercholesterolemia and various types of anaemia, including megaloblastic anaemia, hereditary spherocytosis, homozygous sickle cell disease, aplastic anaemia and liver associated anaemia. The study showed that the plasma cholesterol level is closely related to haematocrit levels, both initially and throughout the course of the anaemias associated with hypercholesterolemia. This association was maintained regardless of the cause of changes in haematocrit levels. The authors concluded that low haematocrit, not the type of anaemia, is the cause of low cholesterol levels.

Seip and Skrede [16], in 1967, found an association between serum cholesterol and haemogloin in all cases, regardless of cause of anaemia.

Conclusion

With this study it is concluded that clinical presentation and biochemical changes with special reference to lipid profile presents key role for severity and type of anaemia.

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