A Study Of The Hematological Profile In Relation To Some Allergic Diseases (A Hospital Based Study)

Ava Dihingia[*], Wasima Jahan[*]

*Demonstrator, **Professor, Department of Physiology, , Assam Medical College and Hospital, Dibrugarh -786002, Assam

Abstract: Background: Eosinophils and neutrophils play major roles in pathogenesis of allergy.However the relation between peripheral blood cell counts of other major leukocyte groups, Hb%, ESR in allergic diseases is less clear.Therefore this study was conducted to find out if there is any variation in the haematological profile in subjects having allergic disease. Method: 50 cases of bronchial asthma, 50 cases of some allergic skin disorders and 50 cases of allergic rhinitis and 50 controls were chosen for this study. The study design was cross-sectional. Total Leucocyte Count, Absolute Eosinophil Count were done using Neubauer's chamber, Differential Leucocyte Count by Leishman's staining and Erythrocyte Sedimentation Rate using Westergren's method. Haemoglobin estimation was done by cyanmethaemoglobin method. The statistical analysis was done using one way analysis of variance (ANOVA) followed by t test. Result: The results showed a significant increase ($p<0.05$) in the Total Leucocyte Count, Absolute Eosinophil Count, Erythrocyte Sedimentation Rate and the differential counts of neutrophil and eosinophil, and a significant decrease ($p<0.05$) in the lymphocyte count among the cases as compared to the controls. Conclusion: It is seen that in allergic condition besides an increase in the eosinophil count, other haematological parameters also change.

Key Words: Allergy, Absolute Eosinophil Count (AEC), Differential Leucocyte Count (DLC), Total Leucocyte Count (TLC)

Author for correspondence: Ava Dihingia, Demonstrator in Department of Physiology, Assam Medical College and Hospital, Dibrugarh -786002, Assam, India. Email-ID:- ava.dihingia2011@gmail.com

Introduction: Allergy is a disorder of the immune system which is a form of hypersensitivity. Allergic reactions occur to normally harmless environmental substances known as allergens. These reactions are acquired, predictable and rapid. Strictly, allergy is one of four forms of hypersensitivity and is called type-1 (or immediate hypersensitivity). It is due to the combination of an antigen with an antibody (IgE) bound to mast cells in individuals previously sensitized to the antigen. This may occur as a local or systemic reaction.

The allergic diseases are a severe problem for the community as well as for the national economy[1]. About 20% of the world populations are estimated to suffer from one allergic disease or another. According to Aggarwal et al., the total estimated burden of Asthma is an overall prevalence of 3% (30 million patients), and among adults over the age of 15, a median prevalence of 2.4%. They have also found higher prevalence rates among school children.[1] A recent survey carried out in India shows that 20-30% of the population suffer from allergic rhinitis and that 15% developed asthma[2]. Therefore there is an impending need to create awareness regarding allergic diseases and help thousands of people who suffer from the disease. It is well known that eosinophils and neutrophils play major roles in the pathogenesis of allergy. However the relation between peripheral blood cell counts of other major leukocyte groups, Hemoglobin%, Erythrocyte Sedimentation Rate in allergic diseases is less clear.

Diseases like bronchial asthma, allergic rhinitis and allergic skin disorders like eczema, urticaria are very common in the north-eastern region of India due to the geo-climatic conditions. In spite of this no previous study was done on the haematological profile of subjects having these disorders in this part of the country. Therefore this study was conducted with the following Aim and objectives:-

1. To estimate the haematological profile in normal healthy individuals.
2. To estimate the haematological profile in individuals having some allergic disorders.
3. Comparative analysis of the haematological profile in subjects having some allergic disorders with that in normal subjects.

Material and Method: In this study 50 cases of bronchial asthma, 50 cases of some allergic skin disorders and 50 cases of allergic rhinitis, altogether 150 subjects were chosen for this study from the inpatient and outpatient departments of, medicine, dermatology and ENT of Assam Medical College & Hospital, Dibrugarh. Due approval was taken from the ethical committee of our institution before proceeding with the study.

Inclusion criteria: Only the diagnosed cases of the respective diseases attending the Medicine, Dermatology and ENT OPD or admitted in the inpatient ward without having other ailment were included in the study.

Exclusion criteria*:*

1. Those having worm infestation or parasitic infestation.
2. Those on steroid therapy or any other medication.
3. Those having haematological disorders like bleeding disorders

50 apparently healthy individuals were selected as controls. The subjects for control group were taken from the healthy population of Assam Medical College and Hospital campus and also from the healthy attendants accompanying the diseased persons. Efforts were made to match each case with normal individual by age and sex and if possible by community also. The patients who fulfilled the inclusion of criteria were subjected to a detailed history, clinical examination and ancillary investigations following a pre-designed proforma.

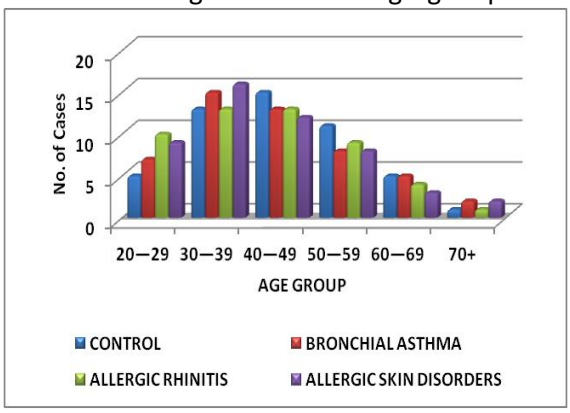
Method: Total Leucocyte Count was done manually using Turk's Diluting Fluid and Neubauer's chamber, DLC was done by examination of blood films stained with Leishmain's stain under the oil immersion objective. Haemoglobin estimation was done by the standard Cyanmethaemoglobin method using a spectrophotometer. Erythrocyte Sedimentation Rate (ESR) was examined by the Westergren method using Westergren's pipette. Absolute Eosinophil Count was done by the Direct counting method using Neubauer's chamber and Dunger's solution. Indirect counting method was simultaneously used to check the result of direct counting. All the Samples were collected at 10 am to avoid diurnal variations in Absolute Eosinophil Count (AEC). Statistical analysis was done using t-test and One Way Analysis Of Variance (ANOVA).

Result: Table–1 and Fig -1 shows that the maximum number of cases of control group belongs to the age group 40—49 years (30%) followed by age group 30-39 years (26%).

Table 1: Showing Distribution Of Controls And Cases According To Different Age Groups

AGE GROUP	CONTROL		BRONCHIAL ASTHMA		ALLERGIC RHINITIS		ALLERGIC SKIN DISORDERS	
	No	%	No.	%	No.	%	No.	%
20—29	5	10.00	7	14.00	10	20.00	9	18.00
30—39	13	26.00	15	30.00	13	26.00	16	32.00
40—49	15	30.00	13	26.00	13	26.00	12	24.00
50—59	11	22.00	8	16.00	9	18.00	8	16.00
60—69	5	10.00	5	10.00	4	8.00	3	6.00
70+	1	5.00	2	4.00	1	2.00	2	4.00
TOTAL	50	100.00	50	100.00	50	100.00	50	100.00

Fig 1: Showing distribution of controls and cases according to different age groups



The maximum number of cases with allergic rhinitis were in the age groups of 30—39 and 40—49 years (26% each).The maximum number of cases with allergic skin diseases were from the age group of 30—39 (32%).

Table–2 shows that in controls the maximum number were male (68%), with female being 32%, similarly the maximum number of cases having asthma were males (56%), whereas the maximum number of cases having allergic rhinitis and allergic skin diseases were females, (64%) and (52%) respectively.

From the above tables and histograms it is seen that there was a significant difference in the Absolute Eosinophil Count, Total Leucocyte Count, Erythrocyte Sedimentation Rate and the differential leucocyte counts of Neutrophil, Lymphocyte and Eosinophil between cases and controls.

While doing paired t test it was found that:
There was a significant difference in the Absolute Eosinophil Count, Total Leucocyte Count, ESR and differential leucocyte counts of neutrophil and eosinophil between the controls and cases with bronchial asthma and allergic skin disorders

Table 2 : Showing distribution of controls and cases according to gender

GENDER	CONTROL		BRONCHIAL ASTHMA		ALLERGIC RHINITIS		ALLERGIC SKIN DISORDERS	
	No.	%	No.	%	No.	%	No.	%
Male	34	68	28	56	18	36	24	48
Female	16	32	22	44	32	64	26	52
TOTAL	50	100	50	100	50	100	50	100

Table 3 : Comparison of different haematological parameters among cases and controls

Parameters		A	B	C	D	p value	t test
TLC (cells/cumm)		7474 ±1128.44	8609.60 ±1892.27	8267.80 ±2118.63	9102.80 ±2179.38	p<0.05	A vsB p<0.05 A vs C p<0.05 A vs D p<0.05
Hb(gm/dl)		11.4420 ±1.61322	11.2520 ±1.68985	10.72 ± 1.64	11.14 ± 1.63	p>0.05	A vs B p> 0.05 A vs C p>0.05 A vs D p> 0.05
ESR(mm AEFH)		5.72 ± 2.13	8.28 ± 2.73	9.14 ± 3.49	7.88 ± 2.95	p<0.05	A vs B p< 0.05 A vs C p< 0.05 A vs D p< 0.05
Differential Leucocyte Count	Neutrophil(%)	64.7 ± 2.14	65.26 ± 3.84	66.82 ± 4.23	65.30 ± 4.12	p<0.05	A vs B p <0.05 A vs C p> 0.05 A vs D p<0.05
	Lymphocyte(%)	30.52 ± 2.18	27.38 ± 4.20	25.12 ± 3.78	26.74 ± 4.07	p<0.05	A vs B p>0.05 A vs C p< 0.05 A vs D p>0.05
	Monocyte(%)	2.9 ± 1.11	2.14 ± 1.08	2.38 ± 1.19	2.16 ± 1.11	p>0.05	A vs B p>0.05 A vs C p>0.05 A vs D p>0.05
	Eosinophil(%)	1.8 ± 1.08	5.16 ± 1.73	5.66 ± 2.20	5.54 ± 1.84	p<0.05	A vs B p<0.05 A vs C p<0.05 A vs D p <0.05
	Basophil(%)	0.06 ± 0.23	0.1 ± 0.3	0.08 ± 0.27	0.08 ± 0.27	p>0.05	A vsB p>0.05 A vs C p>0.05 A vs D p>0.05
Absolute Eosinophil Count		193.28± 19.23	656.72 ± 97.94	553.22 ± 81.07	571.50 ± 131.15	p<0.05	A vs B p<0.05 A vs B p<0.05 A vs C p<0.05

A=Control, B= Bronchial Asthma, C= Allergic Rhinitis , D=Allergic Skin Disease

Fig 2: Showing differential leucocyte count in cases and controls

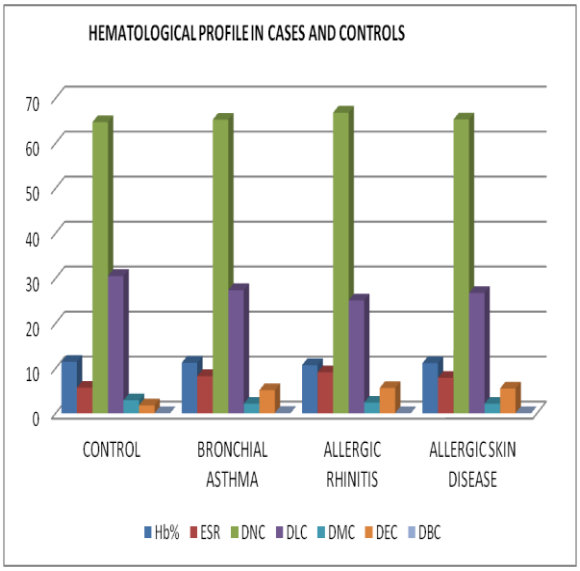


Fig 3: Total leucocyte count in cases and control

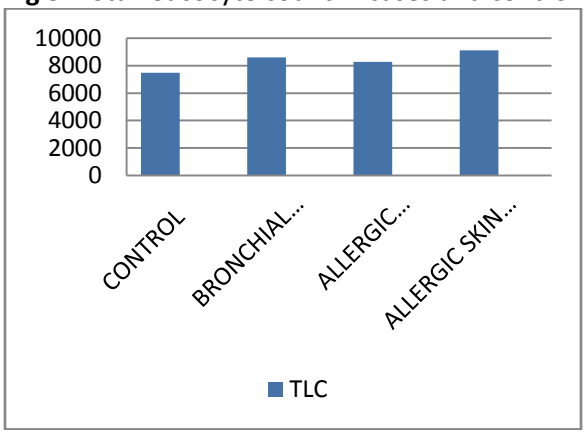


Fig 4: Showing absolute eosinophil count in cases and controls

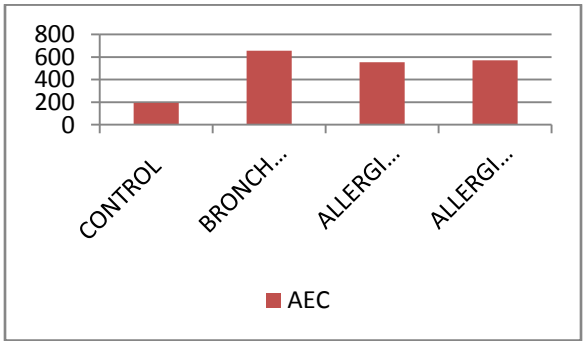
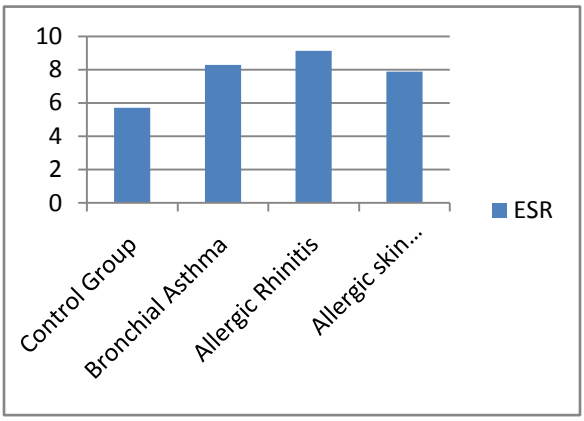


Fig 5: Showingthe erythrocyte sedimentation rate in cases and controls



There was a significant difference in the differential leucocyte count of lymphocyte and eosinophil along with the Absolute Eosinophil Count, Total Leucocyte Count and ESR between the controls and cases with allergic rhinitis.Other parameters showed no significant difference.

Discussion: From the study we can observe that the Total leukocyte counts were higher in the diseased groups than in controls. This finding is consistent with the findings of Sarah A Lewis et al[3]. The level of Hb% was slightly lower in the diseased groups as compared to controls, but there was no statistical difference. In the present study it is seen that neutrophils, lymphocytes and eosinophils were higher in the diseased groups having bronchial asthma and allergic skin disorders than the control groups which is consistent with the findings of Sarah A. Lewis et al[3].

However they have found that there was a significant decrease in the lymphocyte count between the controls and cases with allergic rhinitis, while in our study we have found that lymphocyte counts decrease in the cases having allergic rhinitis without any statistical difference. Neutrophil influx and the subsequent neutrophil activation involve IL-8 mediation. Although the stimuli that trigger this response may vary, the features point to the activation of innate immune mechanisms rather than IgE mediated activation of acquired immunity.[4]

In the last few years strong evidence has accumulated to suggest that lymphocytes, specially allergen-reactive type-2 T helper (T_h2) cells play an important role in the induction and maintenance of the allergic inflammatory cascade. First, cytokines and chemokines produced by T_h2 cells and those produced by other cell types in response to T_h2 cytokines or as a reaction to T_h2-related tissue damage account for most pathophysiologic aspects of allergic disorders.[5] The present study reveals that maximum number of cases having asthma , allergic rhinitis and allergic skin disorders had high absolute eosinophil count in the range of 601-800 (68%), 401-600 (76%), and of 401-600 (82%) respectively , while in the controls the count was in the range <200 cells/cumm (58%).

Dhar S. Et al[6] in their study have also found that the average absolute eosinophil count was significantly higher in diseased group than in controls. Knott FA, Pearson RsB reported eosinophilia in allergic conditions in Guy's Hospital Report (1934-35)[7]. LowelFc also reported clinical aspects of eosinophilia in atopic diseases[8].

The increase in eosinophils suggest increase activation of allergic response. This isconsistent with the study of Tulinska*et al.*[9](2004), where they observed an increase in activation markers on eosinophil (CD 66b and CD 69) and Ogunbileje*et al*[10] where they also reported significantincrease in IgE. Circulating eosinophils are elevated in patients with allergic conditions. It has been reported that eosinophils recruitment is being induced by eotaxin(Conroy and Williams, 2001)[11] via IL-4 and IL-13 involvement (Rothenberg and Hogan, 2006)[12]. BotHomocysteinetokines (IL-4 and IL-13) are known to be potent pro-fibrotic mediators.

In the present study ESR was found to be increased in the diseased groups compared to controls. CanozM *et al*[13] also found that ESR was raised in asthmatic patients compared to that of controls. Daniel Reichmuthet al[14] also found that ESR was raised in asthmatic patients as compared to that of controls. The study did not find significant changes in monocytes and basophils. However they play important roles in the allergic response.

Monocytes migrate from the bone marrow to the inflamed tissue through the peripheral blood system. In tissues monocytes differentiate and mature into macrophages, the functional cell of the lineage. Monocytes can produce a complex repertoire of cytokines; are regulated by cytokines; and can actively participate in the pathogenesis of infection and several inflammatory diseases.[15] A study found evidence for activation of alveolar macrophages, but not peripheral blood monocytes, in subjects with allergic rhinitis and asthma.[16]

Basophils circulate in the peripheral blood under homeostatic conditions and are often recruited to the affected tissues in allergic disorders, including asthma, atopic dermatitis and rhinitis. Basophils may function as initiators as well as effectors of the allergic inflammation and contribute to the recruitment of other proinflammatory cells such as eosinophils and neutrophils.

Basophils were thought to be precursors of mast cells in the immunologic response. However a recent study discussed the active immunomodulatory role of basophils. They showed IgE involved in a delayed-onset allergic inflammation in the skin, where basophils but not mast cells play an essential role in the development of inflammation. After encountering the corresponding antigens, IgE associated basophils are activated to secrete soluble factors, including cytokines which attract proinflammatory cells such as eosinophils and neutrophils, leading to chronic allergic inflammation.[17]

Another study by the authors recently demonstrated that basophils, unlike mast cells, have no significant contribution to IgE-mediated systemic anaphylaxis in a mouse model. Instead, basophils are involved in IgG-mediated systemic anaphylaxis under our experimental conditions.[18]

Conclusion: There is a variation in the haematological profile in subjects having allergic disorders. Therefore routine blood investigations should be made mandatory in all allergic conditions even with minimal or no symptoms which will further help in the early diagnosis and treatment.

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