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Research Article

Anemia in Asthmatic Females Exaggerates the Severity of Inflammation in Asthma by Inducing Dyslipidemia, High levels of IgE and Absolute Eosinophil count

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Abstract: The objective of the study was to investigate the effects of anemia on severity of inflammation in asthmatic females with the assessment of dyslipidemia, IgE and absolute Eosinophil count. Study was performed at Pathology Lab of Asia Diagnostic Center, Blue area Islamabad Pakistan. Total 100 asthmatic Females (Group 1) and 100 normal females as control (Group 2) were enrolled in this study. Hemoglobin (Hb) levels, Total leukocyte count, Lipid profile, serum IgE and Eosinophil count were analyzed in Group 1 and Group 2. Group 1 was sub divided in to two groups. Group 1a asthmatic females with anemia and Group1b asthmatic females without anemia. Triglycerides (TG) and very low density lipoprotein (VLDL) with mean value 139.8 ± 27.9 and 23.6 ± 6 in Group 1a were not significantly different with p-value >0.05 from Group1bwith mean value of TG 137 ± 25.5 and VLDL 25.9 ± 5.8. Cholesterol and low density lipoprotein (LDL) were significantly high in Group 1a than Group1b with p-value <0.05. HDL levels in Group 1a were significantly lower than Group1b with the p-value 0.017. Markers of severity of inflammation in asthma, IgE and Absolute Eosinophil Count were also high in Group 1a with p value 0.0297 and <0.0001 respectively from Group1b. The strongest negative correlations ware observed for LDL - 0.60. IgE - 0.61 and absolute eosinophil count -0.99 with Hb levels in Group 1a ,which indicate that anemia exaggerate the severity of inflammation in asthma as well as dyslipidemia in anemic asthmatic females. From our study it was concluded that when anemia is present in asthmatic females it will exaggerate the severity of inflammation in asthma by inducing dyslipidemia, high eosinophil count and high level of IgE.

Keywords: Asthma, Anemia, Dyslipidemia, Female asthmatic patients, IgE, eosinophil.

1. INTRODUCTION

Asthma is a chronic disease. More than 300 million people are asthmatic worldwide [1]. The prevalence of asthma is high in females after the age of 18 years but the boys in childhood are more prone for asthma than young girls. Sex hormones plays important role in this switchover. Overall prevalence, severity, hospitalization and mortality rate is high in females than the males due to asthma [2]. Asthma is an inflammatory condition, associated with hyper responsiveness of airways and leads to repeated incidence of chest tightness, wheezing, breathlessness and coughing specially at night and morning [3]. T-helper cell immune response to various allergic substances like dust, pollens, molds and non-allergic substances like smoke, cold air and infections are associated with airways tightness and asthma. Increased levels of T-helper cell in airways due to these allergic and non-allergic substances secret various cytokines which exaggerate eocinophilic inflammation and increase the production of IgE. Histamine and cysteinyl leukotrienes are produced due to inflammation in airway that cause bronchospasm and mucus production which leads towards asthma [4]. Prevalence of asthma is 4.3% in children and adults. Around 255,000 are died per year due to asthma. In Pakistani females, the prevalence of asthma is 12% which is going to be increased [5]. Serum IgE levels, eosinophil's counts, Fractional Exhaled Nitric Oxide (FENO) levels, and

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sputum eosinophil's counts are used to diagnose asthma [6]. Iron deficiency is also associated with asthma [7]. Iron deficiency (ID) and low serum ferritin have suspicious effects on lipids and sometime iron deficiency is associated with dyslipidemia that may cause atherosclerosis [8, 9]. ID is widespread nutritional disorder and also the common cause of anemia, regardless of age, gender and socioeconomic status. It is the most common cause of anemia. Iron deficiency anemia (IDA) is the one of major public health problem due to its complication. According to WHO about a third of the population of world is suffering from anemia with IDA [10]. Anemia is a condition in which l hemoglobin level is low below the reference range according to age and sex. According to WHO, the Hb levels in adult males <13g/dl, non-pregnant females <12g/dl and pregnant females <11g/dl are considered as anemic [11]. According to the Hb concentration in blood, anemia is classified in to mild, moderate and severe state. For females the range of mild anemia is 10-11.9 g/dl, moderate is 7-9.9g/dl and <7g/dl is consider as severe state of anemia [12]. Variation in lipid profile and risk of endothelial dysfunction are relatively high in asthmatic patients. The incidences of asthma are high in the anemic children and anemic females [13]. Eosinophil's and serum immunoglobulin E (IgE) are considered a good marker of airway inflammation and severity of inflammations in asthma [14]. Various studies also describe that prevalence of asthma is high in anemia. However the separate analysis of lipids, IgE and eosinophil is required to describe the contribution of anemia in severity of air ways inflammation and cardiovascular complications in asthmatic patients [15, 16]. Our study was an effort to fulfill this gape and to provide the knowledge about the variation in lipid profile, IgE and neutrophil levels in the anemic patients having asthma with respect to non-anemic asthmatic patients.

2. MATERIALS AND METHODS

Cross sectional comparative study was carried out at Pathology lab of Asia Diagnostic center Blue area Islamabad. From October 2018 to Jan 2019 total 100 asthmatic females and 100 normal control females of same age group were enrolled in this study after getting their informed consents. EDTA and Clotted samples were collected by using the sops of Phlebotomy. EDTA samples were saved at 2-8^{o}C and after centrifugation of clotted samples, Serum were saved at -20^{o}C. Hb levels and Total leukocyte count was performed by hematology analyzer Abacus 380 and eosinophil count was performed by manual counting on glass slide after stain the blood smear with giemsa stain and % of eosinophil was converted in to absolute eosinophil count by the formula % eosinophil* total leukocyte count *103/ul. TG, Total Cholesterol, HDL were measured on semiautomatic Chemistry analyzer Micro Lab 300 by using the Reagents of Merck. VLDL was calculated by TG/5 and LDL-C ware calculated by Fried Ewald equation. Serum IgE levels were performed by ELISA method.

2.1 Inclusion Criteria

Asthmatic Females were selected as cases and Non asthmatic females were selected as control.

2.2 Exclusion Criteria

Females with the history of parasitic infections, Diabetes and metabolic syndrome, and pregnant females were also excluded in this study.

3. RESULTS

This study was carried out among 100 asthmatic females and 100 normal control females of same age group. The frequency of different age group of cases and control is given in the Table 1. Asthmatic group was further divided in to two groups, Anemic asthmatic females (Group 1a) and Non Anemic asthmatic females (Group 1b). Among the 100 asthmatic females that were enrolled in our study, there were 42 females were anemic having their Hb level less than 12 g/dl. Table 2 shows the frequency of anemia in different age groups and percentage of anemic and non-anemic asthmatic females enrolled in this study. The comparison of lipid profile, IgE and absolute eosinophil count was performed between total asthmatic females (n=100 Asthmatic females Group 1) and control group (n=100 Group 2) females, and P-Value was calculated by two-tailed T-test with 95% confidential interval and p-value <0.05 was considered clinically significant. Table 3 describes the comparison of lipid profile, IgE and absolute eosinophil between asthmatic and non-asthmatic females. All the parameters of lipid

profile have p-value less than 0.05, which indicate that there are significant variations in triglyceride, Total Cholesterol, HDL, LDL and VLDL in Asthmatic females (Group 1) and Control (Group 2). Asthmatic females were divided in two groups anemic asthmatic females n=42 (Group 1a) and non-anemic asthmatic (n=58) (Group 1b) as shown in Table 2 and comparison of lipid profile, IgE and Absolute eosinophil count were performed by using two Tailed t-test with 95% confidential interval and P-Value <0.05 was used as clinically significant. Table 4 shows the comparison of Lipid profile, IgE and eosinophil count between Group 1a and Group 1b. There was no difference observed in triglyceride concentration between both Groups as the p vale is greater than 0.05 for TG, but the other parameters of lipid profile were significantly high in Anemic (42) Group 1a. IgE and absolute eosinophil count were also significantly high in Group 1a than the Group 1b. Pearson correlation coefficient was also performed to investigate the relationship between Hb levels and variation in lipid profile, IgE and absolute Eosinophil count. Shown in Table 5. Hb levels were negatively associated with triglyceride. Total Cholesterol, LDL, VLDL, IgE and eosinophil count and positively correlated with HDL in

Table 1. Frequency distribution of different age groups of Asthmatic females (Group 1) and Control (Group 2)

Age groups	19-29	30-39	40-49	>50	Total
Control (Group 2)	27	26	29	18	100
Asthmatic females (Group 1)	28	25	30	17	100

Table 2. Frequency distribution table of Anemic asthmatic females (Group 1a) and Non Anemic asthmatic females (Group 1b)

Group	19-29	30-39	40-49	>50	Total	percentage
Non Anemic asthmatic females (Group 1b)	16	15	17	10	58	58%
Anemic asthmatic females (Group 1a)	12	10	13	7	42	42%

anemic asthmatic females patients. Strong negative correlation was observed in the concentration of IgE and absolute eosinophil count with Hb.

4. DISCUSSION

In the present study 100 asthmatic females and 100 normal controls were enrolled. In asthmatic females there were 42 anemic and 58 were non anemic females. The prevalence of anemia in asthmatic females were 42%, Table 2 without the consideration of urban and rural distribution and that is higher than the prevalence of anemia in general population that is 26% in urban areas and 47 % in rural areas [17]. In bronchial asthma there is pulmonary hypoxia which may contribute in increase of Hb in asthmatic patients [18] but in our study there is high prevalence of anemia in asthmatic patients which indicates that anemic population has higher risk to develop asthma that is why females are at higher rate to have asthma than the males. High Incidents of asthma were reported in child bearing age of females as they have lower Hb than other age groups of females [19]. Another study [20] describes that females having low iron store were higher incidence of asthma and severity of inflammation that supports our findings. Asthma has association with hypocholesteremia and it has been reported that high LDL level in asthma contributes the airways obstruction by inducing inflammation and HDL has negative correlation with the severity of inflammation in asthma. Triglyceride has also positive association with severity of inflammation and airway blockage in bronchial asthma [21]. A positive correlation was observed between dyslipidemia and asthma in large scale study of adolescents. Triglyceride, Total Cholesterol, LDL-C and Low HDL level were associated in high prevalence of asthma [22]. Similar associations were observed in our study Table 3. All the components of lipid profile TG, Total Cholesterol, LDL-C,HDL-C and VLDL-C were significantly different than the controls having P-Value <0.05.HDL-C levels were markedly low in asthmatic females with elevated TG, Total Cholesterol, LDL-C and VLDL-C. Serum IgE and absolute Eosinophil count were markedly high than the controls. The association of lipid profile with anemia is different

Table 3. Comparison of Lipid profile in Asthmatic females (Group 1) and Control (Group 2) by using s-t-test.

Test	Asthmatic females (Group 1)	Control (Group 2)	P- value
Triglyceride	138.2+_23.4	130.4+_22.5	0.0172
Total Cholesterol	164.2+_22.1	157.6+_23.5	0.0377
HDL	38.8+_5.4	40.3+_5.1	0.0448
LDL	100.2+_29.5	92.5+_24.3	0.0453
VLDL	26.6+_5.8	25.8+_5.4	0.3140
IgE	2032+_590	345.8+_170.8	<0.0001
Eosinophil Count	0.27*103/ul	0.05*103/ul	<0.0001

Table 4. Comparison of Lipid profile, IgE AND eosinophil count between Group 1a and Group 1b.

Test	Anemic (42) Group 1a	Non anemic (58) and Group 1b	P-value
Trig	139.8+_27.9	137.0+_25.5	0.6036
T.Chol	171.5+_20.7	159.0+_25.3	0.0100
HDL	36.7+_4.9	39.6+_5.9	0.0107
LDL	104.3+_30.9	97.2+_28.6	0.2391
VLDL	26.3+_6	25.9+_5.8	0.7380
IgE	2241+_672	1987+_480	0.0297
Eosinophil	0.32+_0.08*103/ul	0.25+_0.06*103/ul	<0.0001

Table 5. Pearson correlation coefficient of test parameters with HB in Anemic Asthmatic patients

Test	Correlation coefficient
Trig	-0.26
T.Chol	-0.41
HDL	0.33
LDL	-0.60
VLDL	-0.36
IgE	-0.61
Eosinophil	-0.99

in different studies. Some studies support the statement that in anemia, there is low level of TG, Cholesterol and LDL-C [23]. In present study there was high concentration of lipid profile parameters in Asthmatic Group1 than non-asthmatic Group 2. When we compared the values of lipid parameters between the anemic and non-anemic asthmatic Groups, there were considerable differences in levels of Total Cholesterol, LDL-C, HDL-C and VLD-C between both Groups. High levels of LDL and Low levels of HDL were associated with anemia in asthmatic patients (Table 4). Pearson correlation coefficient was used to determine the relationship between Hb levels in anemic asthmatic females and Lipid parameters. Here we observed a negative correlation among all the parameters of lipid profile except HDL which was positively associated with Hb levels and strong negative correlation was observed for Total cholesterol and LDL-C (Table 5). This association of dyslipidemia with anemia in asthmatic patients can be described when we look upon the association of asthma with lipids and anemia separately. Prevalence of asthma is at higher rate in iron deficient anemic females [7] and dyslipidemia is also positively associated with asthma [9]. When anemia is positively associated with asthma and dyslipidemia is also positively associated with asthma then in anemic asthmatic females with high levels of lipids as in the results of our study have possibilities to have high severity of inflammation in anemic asthmatic females. Serum IgE and Eosinophil count were also high in anemic asthmatic females than non-anemic asthmatic female with p-value < 0.05. Serum IgE and Eosinophil count showed strong negative correlation with Hb levels in anemicasthmatic females, shown in (Table 5). Iron deficiency is correlated with High eosinophil count [24] and

IgE levels were also associated with iron status and Hb levels in asthmatic patients [25]. Main cause of anemia in females is iron deficiency and iron deficiency induces eosinophilia and elevated IgE levels in anemic asthmatic females.

5. CONCLUSIONS

From our study it was concluded that when anemia is present in asthmatic females, it exaggerates systemic inflammatory markers by inducing dyslipidemia, high eosinophil count and high level of IgE, suggesting possibility of worsening of asthma severity due to anemia.

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7. REFERENCES

1. Sharifi, L., R. Dashti., Z. Pourpak., M.R. Fazlolihai., M. Movahedi., Z. Chavoshzadeh., H. Soheili., S. Bokaie., A. Kazemnejad, & M. Moin, Economic Burden of Pediatric Asthma: Annual Cost of Disease in Iran. *Iranian Journal of Public Health*, 47(2): 256 (2018).
2. Zein, J.G, & S.C. Erzurum. Asthma is different in women, *Current allergy and asthma reports* 15(6): 28 (2015).
3. Bateman, E.D. Global strategy for asthma management and prevention: GINA executive summary. *European Respiratory Journal* 31(1): 143-178 (2008).
4. Hammad, M.A., K.M. Alakhali., M. Hattan., D.A.M. Noor. Asthma in Saudi arabia: risk factors and pharmacotherapy, *Indo American Journal of Pharmaceutical Research*, 6(11): 6814-6821 (2016).
5. Aslam, S. K., S. Zaheer., M.S. Qureshi., S.N. Aslam, & K. Safique, Socio-economic disparities in use of family planning methods among Pakistani women: findings from Pakistan demographic and health surveys, *PloS one* 11(4): e0153313 (2016).
6. Szefler, S. J., et al. Asthma outcomes: biomarkers, *Journal of Allergy and Clinical Immunology*, 129(3) :S9-S23 (2012).
7. Brigham, E.P., F. Kolahdooz., N. Hansel., P.N. Breysse., M. Davis., S. Sharma., E.C. Matsui., G. Diette, & M.C. McCormak. Iron status is associated with asthma and lung function in US women, *PLoS One*, 10(2) (2015).
8. Brigham, E. P., et al, Association between Western diet pattern and adult asthma: a focused review. *Annals of allergy, asthma & immunology*, 114 (4): 273-280 (2015).
9. Su, X., Y. Ren., M. Li., X. Zhao., L. Kong, & J. Kang, Association between lipid profile and the prevalence of asthma: a meta-analysis and systemic review, *Current medical research and opinion*, 34 (3) :423-433 (2018).
10. Abbaspour, N., R. Hurrell, & R. Kelishadi. Review on iron and its importance for human health. *The official journal of Isfahan University of Medical Sciences*, 19(2):164-174 (2014).
11. Chen, M.H., T.P. Su., Y. Chen., J.W. Hsu., K.L. Huang., W.H. Chang., T.J. Chen, & Y.M. Bai. Association between psychiatric disorders and iron deficiency anemia among children and adolescents: a nationwide population-based study. *BMC psychiatry*, 13(1) :161 (2013).
12. Jadhav, B.N. A critical review on iron deficiency anaemia in female population of developing India. *International journal of fauna and biological studies*, 3(5) :116-119 (2016).
13. Tuleta, I., D. Skowasch., F. Aurich., N. Eckstein., R. Schueler., C. Pizzaro., N. Schahab., G. Nickenig., C. Schaefer, & S. Pingel. Asthma is associated with atherosclerotic artery changes. *PLoS One*, 12(10) (2017).
14. Lin, J, & D. Yang. Chinese expert consensus on diagnosis and management of severe asthma. *Journal of Thoracic Disease*, 10(12) :7020 (2018).
15. Seaton, A, & G. Crompton. Asthma: clinical features. Seaton A, Seaton D, Leitch AG. Crofton and Douglas's *Respiratory Diseases* 5: 922-972 (2008).
16. Lunt, A., S.S. Sturrock, & A. Greenough."Asthma and the outcome of sickle cell disease". *Expert Opinion on Orphan Drugs*, 6 (12) :733-740 (2018).
17. Baig-Ansari, N., S.H. Badruddin., R. Karmaliani., H. Harris., I. Jehan., O. Pasha., N. Moss., E.M. McClure, & R.L. Goldenberg. Anemia prevalence and risk factors in pregnant women in an urban area of Pakistan. Food and nutrition bulletin, 29 (2) :132-139 (2008).
18. Ejaz, S., F. U. H. Nasim, & M. Ashraf. Hematological and Biochemical Profile of Patients Suffering from Non-Atopic Asthma. *Insights Chest Dis*, 2 (2) :6 (2017).
19. Brauer, M., G. Hoek., H.A. Smit., J.C. De Jongste.,

J. Gerritsen., D.S. Postma., M. Kerkhof, & B. Brunekreef. Air pollution and development of asthma, allergy and infections in a birth cohort. *European Respiratory Journal* 29, (5) :879-888 (2007).

20. Brigham, E. P., C. Meredith., M.C. McCormack., M. C.Takemoto, & E. C. Matsui. Iron status is associated with asthma and lung function in US women. *PLoS One*, 10(2) (2015).
21. Vinding, R.K., J. Stokholm., B.L. Chawes, & H. Bisgaard. Blood lipid levels associate with childhood asthma, airway obstruction, bronchial hyperresponsiveness, and aeroallergen sensitization. *Journal of Allergy and Clinical Immunology*, 137 (1) :68-74 (2016).
22. Johnston, R. A, & A. S. Shore. Obesity and asthma: What have we learned from animal models?. In Mechanisms and Manifestations of Obesity in Lung Disease. *Academic Press*, 111-142 (2019).
23. Zhang, L., L. He., J. Gong, & C. Liu. Risk factors associated with irreversible airway obstruction in asthma: a systematic review and meta-analysis. *BioMed research international*, 2016 (2016).
24. Cabada, M.M., M.R. Goodrich., B. Graham., P.G Villanueva-Meyer., E.L. Deichsel., M. Lopez., E. Arque, & J.R.C. White. Prevalence of intestinal helminths, anemia, and malnutrition in Paucartambo, Peru. *RevistaPanamericana de SaludPública*, 37: 69-75 (2015).
25. Brigham, E.P., F. Kolahdooz., N. Hansel., P.N. Breysse., M. Davis., S. Sharma., E.C. Matsui., G. Diette, & M.C. McCormack. Association between Western diet pattern and adult asthma: a focused review. *Annals of allergy, asthma & immunology*, 114(4): 273-280 (2015).