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Exposure to Textile Chemicals Leads to Microcytic Anemia and Hypersensitivity in Textile Workers

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Abstract.- This study was aimed at investigating the toxic effects of dyeing chemicals on various haematological parameters of textile workers. A total of 62 workers involved in dyeing processes were compared with 95 control subjects. The findings revealed significant decrease in haemoglobin content, haematocrit, haematological indices and neutrophil whereas leucocytes, eosinophils and lymphocytes counts were observed to increase in the exposed workers. It is concluded that haematological parameters are as useful physiological indices for hazards caused by toxic chemicals used in the textile industry.

Keywords: Toxic chemicals; textile dyes; occupational exposure; hypersensitivity; haematology.

INTRODUCTION

A wide range of chemicals are used by the textile processing industry for dyeing and printing operations. These include bleaching agents, vat dyes, azo dyes, sulphur dyes, disperse dyes and colour pigments (Vutukuru, 2005). Workers involved in dyeing process get directly or indirectly exposed to these chemicals which are health hazardous (Mathur and Bhatnagar, 2007).

Toxic effect of azo dyes, reactive dyes and direct dyes causing asthama, contact dermatitits, have already been reported in textile industry (Hassamanova *et al*., 2000; Vutukuru, 2005; Li *et al*., 2006). Moreover, association between employment-exposure to textile industry and cancer have also been detected for bladder, nasal, oesophagus, stomach, colon, rectal, nasopharyngeal and lung cancers (De-Roos *et al*., 2005; Singhi *et al*., 2005; Wernli *et al*., 2006; Busch *et al*., 2008), but less is known about its influence on haematological parameters of workers. Haematological indices have often been used to determine the sub lethal effects of various pollutants as well as the physiological status of mammals under stress (Bhagwant and Bhıkajee, 2000; Keith, 2000; Rebecca, 2000; Vutukuru, 2005).

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Present study was therefore undertaken to determine the haematoxicity of dyeing chemicals in textile workers. The haematological parameters including haemoglobin concentration, total erythrocytes counts, total leucocytes counts, erythrocytes sedimentation rate, haematocrit value, haematological indices and differential leucocytes counts were recorded to assess the toxic effects of textile dyes on allergic manifestations and hypersensitivity among textile workers.

MATERIALS AND METHODS

Data collection and blood sampling

The data regarding age, salary, smoking habit, job duration, exposure time/day and history of any disease or allergy of workers involved in dyeing process and control subjects was recorded. Only healthy workers were included in the study.

Sixty two workers exposed to textile dusts/ and fume of dyes, having direct contact with dyeing chemicals in two textile factories in and around Lahore were included in this study. They were sex and age matched with 95 control workers from a village, a textile industry free area and had never been directly exposed to these dyes. With the help of sterilized disposable syringes, blood samples were collected into lithium heparin specimen tubes. After thorough mixing, samples were transferred to laboratory under maximum possible precautionary measures. This blood was used for haematological

studies.

Study of haematological parameters

Estimation of haemoglobin (Hb) was done following the procedure of Van Kampen and Zijlstra (1961). Total erythrocyte count (TEC) and total leucocyte count (TLC) were done according to Dacie and Lewis (1991). Haematocrit value (PCV) and haematological indices were recorded according to Swarup *et al*. (1986). Erythrocyte sedimentation rate (ESR) was determined by the Westergren's method (Dacie and Lewis, 1991). Thin blood smear on glass slides fixed in ethanol and stained in 25% giemsa stain (Merck) for 30 minutes were used for DLC.

Data analysis

Comparison between exposed and control groups was carried out using Student's t test in the SPSS personal computer statistical package (Version 12, SPSS Inc, Chicago). Overall, a comparison was made between exposed group (62) and control group (95). Age specific changes were analysed by dividing exposed and control workers into 5 age groups (21-25; 26-30; 31-35; 36-40 and 40-45 years). In contrast, to study the effect of experience on work related changes, subjects were divided into five groups (≥ 1; 1-5; 6-10; 11-15 and 16-20 years) on the basis of job duration. In smoking habit comparison, data was divided into two groups and smokers (28) were compared with non smokers (34). Results were expressed as Mean±SEM. p values ≤ 0.05 were considered significant.

RESULTS

Comparison of haematological parameters of 62 exposed workers with 95 control exhibited a significant decrease in neutrophil, and increase in eosinophil counts in workers (Table I). The duration of job of textile workers varied from 2 months to 15 years. So the data was arranged into 5 classes with respect to duration of exposure to dyeing chemicals. Significant decrease in Hb, PCV, MCH, MCHC, neutrophil, was observed in workers having < than 1 year experience, however, eosinophil count was observed to be high. Similar changes were observed in workers upto 1-5 years job experience, moreover a rise in lymphocytes was also observed. In workers with 10-15 years job duration, decrease in neutrophil and MCH level, whereas increase in eosinophil was recorded in exposed group (Table II).

Table I.- Comparison of various haematological parameters of occupationally exposed textile workers and control population.

Parameters[a]	Control (n=95)	Exposed (n=62)
Hb (g/dl)	14.63±1.43[b]	12.90±0.28
TEC ($x10^6$/µl)	4.14±0.44	4.44±0.15
TLC ($x10^3$/µl)	4.76±0.48	6.01±0.18
PCV (%)	45.09±1.96	42.70±0.53
MCV (fl)	109.40±12.18	105.79±3.23
MCH (pg)	35.47±5.76	30.92±1.23
MCHC (g/dl)	32.29±3.46	30.49±0.78
ESR (mm/hr)	8.95±2.77	11.08±0.52
Neutrophil (%)	53.14±3.83	44.24±1.44*
Eosinophil (%)	3.78±1.41	7.27±0.29*
Basophil (%)	0.67±0.15	0.87±0.01
Lymphocyte (%)	37.14±3.83	40.63±1.57
Monocytes (%)	5.82±2.02	6.95±0.27

[a]Abbreviations used: Hb, Haemoglobin; TEC, total erythrocyte count; TLC, total lecucocyte count; PCV, packed cell volume; MCV, mean corpuscular volume; MCH, mean corpuscular haemoglobin concentration; ESR, erythrocyte sedimentation rate.
[b]Mean±SEM; Student's `t' test, *p<0.05.

The age of textile workers ranged from 20-45 years. Alterations in various parameters were observed in different age groups. In workers having 20-24 years, levels of Hb, MCH, MCHC, and neutrophil percentage were recorded to be decreased significantly whereas an increase in ESR level and eosinophil count was observed in exposed group. Similarly in workers with 25-29 years of age, in addition to differences in above mentioned parameters, depletion in PCV, MCV and elevation in lymphocyte count was also observed. Similar to group II, in age group IV, Hb, PCV, MCV, MCHC and neutrophil count showed significant depletion except for TLC where an increase was observed (35-39 years). Decrease in neutrophil and increase in eosinophil count was also observed in age group with 40-44 years (Table III).

Table II. Comparison of various haematological parameters of textile industry workers and control population of various age groups.

Parameters	Age group (Years)									
	21-25		26-30		31-35		36-40		40-50	
	Control (n=20)	Exposed (n=19)	Control (n=18)	Exposed (n=21)	Control (n=19)	Exposed (n=7)	Control (n=18)	Exposed (n=6)	Control (n=20)	Exposed (n=9)
Hb (g/dl)	15.0± 1.56	11.59± 0.46*	14.4± 1.47	11.49± 0.59*	14.7± 1.38	13.12± 0.58	14.42± 1.38	11.21± 0.63*	14.44± 1.38	12.0± 0.84
TEC ($x10^6/\mu l$)	4.20± 0.52	5.68± 0.28	4.19± 0.43	4.99± 0.22	4.02± 0.33	4.80± 0.44	4.15± 0.42	5.51± 0.35	4.20± 0.50	4.66± 0.31
TLC ($x10^6/\mu l$)	4.99± 0.53	5.88± 0.34	5.0± 0.519	6.28± 0.40	5.0± 0.51	5.67± 0.37	4.60± 0.46	6.57± 0.40*	4.20± 0.42	5.65± 0.27
PCV (%)	45.99± 2.22	43.14± 0.89	45.26± 1.98	41.67± 0.71*	44.52± 1.74	40.53± 1.65	44.73± 1.86	40.17± 2.52*	44.93± 1.86	44.93± 0.48
MCV (fl)	109.56± 17.07	104.59± 6.19	110.46± 13.17	100.28± 4.68*	111.36± 9.27	105.54± 14.06	109± 11.23	97.487± 9.94*	106.64± 6.22	102.47± 2.03
MCH (pg)	36.05± 8.11	25.31± 0.99*	35.98± 5.88	32.43± 1.96	35.91± 3.65	32.20± 7.18	35.11± 4.94	34.49± 2.99	34.31± 6.22	33.25± 2.03
MCHC (g/dl)	32.39± 3.47	26.48± 0.95*	32.38± 3.34	29.01± 1.305*	32.37± 3.21	32.89± 3.28	32.24± 3.49	27.12± 2.54*	32.22± 3.78	31.15± 1.76
ESR (mm/hr)	8.25± 2.95	12.91± 0.31*	8.36± 2.68	9.98± 0.75	8.47± 2.41	9.12± 0.37	9.47± 2.24	11.96± 2.93	10.20± 3.08	11.44± 2.59
Neutrophil (%)	53.65± 3.73	44.37± 2.16*	52.61± 2.37	43.43± 2.50*	52.16± 3.61	45.72± 4.88	53.31± 4.37	42.4± 8.03*	54± 5.07	47.33± 3.81*
Eosinophil (%)	3.10± 1.37	7.89± 0.35*	3.61± 1.47	6.57± 0.58*	4.12± 1.56	8± 0.58*	4.06± 1.7	6.20± 1.07	4± 1.84	7.55± 1.08*
Basophil (%)	0.52± 0.13	0.79± 0.14	0.63± 0.14	0.76± 0.15	0.59± 0.18	0.71± 0.29	0.64± 0.15	0.60± 0.24	0.69± 0.19	1.67± 0.29
Lmphocyte (%)	37.10± 3.65	39.84± 2.43	37.53± 3.62	42.90± 2.87*	37.95± 3.60	38.29± 4.72	36.87± 3.75	43.60± 7.77	35.80± 3.90	3.91± 3.38
Monocytes (%)	5.30± 2.08	7.11± 0.57	5.59± 6.33	6.33± 0.42	5.89± 1.94	7± 0.31	6.07± 2.01	7.20± 0.86	6.25± 2.07	8.33± 0.73

[a] n = number of samples; * $p < 0.05$

Table III.- Comparison of various haematological parameters of control population and workers employed in the textile industry from one year to 20 years.

Parameters	Control (n =95)[a]	Job duration of exposed groups (Years)[a]				
		≥ 1 (n=4)	1-5 (n =22)	6-10 (n =8)	11-15 (n =18)	16-20 (n =10)
Hb (g/dl)	14.79± 1.43	12.11±0.49*	13.75±0.422	11.21±0.49*	13.24±1.11	13.04±0.85
TEC ($x10^6/\mu l$)	4.14±0.44	5.82±0.77	4.99±0.24	4.82±0.29	4.72±0.28	4.76±0.29
TLC ($x10^3/\mu l$)	4.76±0.49	5.95±0.54	5.93±0.38	6.12±0.60	6.40±0.43	5.76±0.35
PCV (%)	44.53±1.74	40.69±0.95*	41.39±0.94	42.93±0.69*	40.95±1.46	42.82±1.72
MCV (fl)	111.36±9.27	101.06±11.25	115.22±6.58	99.53±5.62*	102.53±2.74	100.86±7.28
MCH (pg)	35.91±3.65	28.43±2.0*	30.74±2.64	30.51±2.03*	29.57±2.74*	34.16±2.54
MCHC (g/dl)	32.37±3.21	27.0±3.85*	29.59±0.96	28.04±1.47*	27.58±2.86	31.15±2.01
ESR (mm/hr)	9.42±2.47	9.94±0.12	10.68±0.68	10.57±0.94	11.96±0.44	12.54±2.38
Neutrophil (%)	52.16±3.61	44.0±3.67*	44.18±2.49*	40.055±2.44*	49.75±4.49	47.20±3.41*
Eosinophil (%)	4.12±1.56	9.25±1.32*	7.50±0.45*	6.50±0.57	8.13±0.39*	6.7±0.83
Basophil (%)	0.66±0.16	0.52±0.13	0.63±0.14	0.59±0.18	0.64±0.15	0.69±0.19
Lymphocyte (%)	37.95±3.60	37.25±2.66	40.77±2.76	46.0±2.60*	39.63±4.90	37.60±3.63
Monocytes (%)	5.89±1.94	7.25±1.70	6.64±0.49	6.78±0.42	7.50±0.68	7.40±0.76

[a] n = number of samples; * $p < 0.05$

No significant difference was observed in any of haematological parameter among the workers when comparison was made with respect to smoking habit and data of exposed group of smokers was compared with non smoker workers (Table IV).

Table IV.- Effects of smoking habit on various haematological parameters of textile industry workers.

Parameters[a]	Nonsmoker (n=28)	Smokers (n=34)
Hb (g/dl)	12.98±0.41[b]	12.83±0.40
TEC ($x10^6$/µl)	4.45±0.19	4.43±0.22
TLC ($x10^3$/µl)	6.02±0.27	6.02±0.24
PCV (%)	45.76±0.86	45.07±0.66
MCV (fl)	103.38±5.28	102.84±4.19
MCH (pg)	31.65±2.13	30.33±1.41
MCHC (g/dl)	30.85±1.01	30.21±1.17
ESR (mm/hr)	10.53±0.78	19.47±0.74
Neutrophil (%)	44.82±2.25	43.76±1.89
Eosinophil (%)	7.61±0.45	7.0±0.38
Basophil (%)	1.11±0.15	0.68±0.11
Lymphocyte (%)	39.14±2.29	41.852±2.15
Monocytes (%)	7.25±0.39	6.75±0.39

[a]Abbreviations used: Hb, Haemoglobin; TEC, total erythrocyte count; TLC, total lecucocyte count; PCV, packed cell volume; MCV, mean corpuscular volume; MCH, mean corpuscular haemoglobin concentration; ESR, erythrocyte sedimentation rate. [b]Mean±SEM; Student's `t' test, *$p<0.05$; n = number of samples

DISCUSSION

The haematological study of workers can provide an insight into potentially hazardous exposures experienced in the textile industries. Blood is an excellent indicator of metabolic disorder in the body and various tissues. Functional state of many tissues, thus can be assessed by analyzing changes in blood. Recently, we have reported the toxic effect of dyes on biochemical parameters of textile workers (Liaqat *et al*., 2009).

In the present study, significant changes in various haematological parameters were observed in exposed group compared to the control group, hence pointing towards the toxic effect of textile dyes on the health of workers. Erythrocytes and particularly Hb have been postulated to function as O_2 sensors and controllers of local blood flow and O_2 delivery (González-Alonso *et al*., 2002). A significant decrease observed in Hb level may be due to decreased synthesis of Hb in bone marrow or decreased concentration of Hb/cell (Pradip *et al*., 2005). Present findings are in agreement with Singhi *et al*. (2005), who reported similar findings with respect to Hb in a study from India.

Erythrocytes itself functions as an O_2 sensor, contributing to the control of O_2 delivery by releasing the vasodilators ATP and S-nitrosohaemoglobin with the offloading of O_2 from the haemoglobin molecule (González-Alonso *et al*., 2006). Elucidating the role of erythrocytes number on the control of blood flow could provide new insights into the regulation of vascular tone in patients suffering from anemia and polycythaemia (González-Alonso *et al*., 2006). In present investigation, no significant change in erythrocytes was recorded in any of the comparisons. Indeed, the capacity of the circulatory system to respond to changes in TEC appears to be enormous. There are various compensatory mechanisms in the body which work in stress conditions and keep TEC in normal range. For examples, in anaemic conditions, rate of erythropoiesis increases as a result of that immature erythrocytes in circulation. This condition is also evident in our study as indicated by low PCV. Our findings are consistent with Kokila *et al.* (2005), who reported similar results following exposure to chemicals in zinc electroplating industry workers.

Haematocrit value, also called as packed cell volume (PCV), is the determination of volume of a given unit of blood that is composed of blood cells and plasma. Direct relationship exists between TEC count and haematocrit value. An increase in one causes increase in other and vice versa (Hegazy *et al*., 2008). Significant decrease in PCV value in experience specific comparison and age specific comparison was observed. This low level of PCV indicates anaemic condition, also evident by low Hb concentration in the same groups (Jude *et al*., 2002).

Mean corpuscular volume is the ratio of haematocrit to the erythrocytes count. In healthy persons, the correlation between MCV and concentration of blood erythrocytes is described by a parabola with alternating zones of positive and

negative relationships (Matyushichev *et al*., 2000). In our study, a significant decrease in MCV was observed in age specific and experience specific comparison, which are conventional of microcytic anemia. Decreased haemoglobin, TEC and haematocrit, and increased TLC has also been reported in the zinc electroplating industry workers (Kokila *et al.,* 2005). Similar findings were reported by Jude *et al*. (2002) in workers occupationally exposed to cement dust. MCH is the relationship determined from haemoglobin concentration and the red blood cells count. An increase in TEC or decrease in Hb concentration causes the MCH to become lower. Significant decrease in MCH was observed in workers having 21-25 years age and upto 10 years experience which also points to induction of microcytic anemia in industrial workers. Markedly reduced levels of haemoglobin content, TEC, TLC, haematocrit, MCV, MCH have been reported by other workers in freshwater fish, *Cirrhinus mrigala* on exposure to environmental contaminants (lead acetate) (Pradip *et al*., 2005). Mean cell haemoglobin concentration (MCHC) is the relationship between the haemoglobin and packed cell volume. Significant depletion observed in workers at various instances of age and exposure to dyes may be due to hypochromic condition of erythrocytes thus again reflecting a condition of microcytic anemia (Kokila *et al.,* 2005).

In the present study a significant increase in ESR was observed only in workers upto 25 years of age, thus indicating the insensitivity of this parameter on exposure to textile dyeing chemicals, hence can be excluded while planning the future study in this respect. Chemical exposure also decreases the chemotaxis and phagocytosis of macrophages and neutrophils. It affects the absolute numbers and proportions of leucocytes and lymphocytes (Bhagwant and Bhikajee, 2000; González-Alonso *et al*., 2002; Kobayashi *et al.*, 2006). Leukocytes respond to toxic, infectious, and inflammatory processes to defend tissues and eliminate disease process or toxic challenges. Accurate and prompt counting and differentiation of leukocytes is critical for differentiating between many clinical conditions like bacterial and viral infectious disease, recognizing malignant disease such as leukaemia, the presence of or staging of HIV, assessing allergic conditions, and monitoring bone marrow function or the body's response to various treatments (Zhenga *et al*., 2007). An increase in TLC observed in workers upto 40 years of age may be associated upper respiratory tract infections (El-Banna *et al*., 2006). Elevation in TLC as a result of exposure to many other toxicants (industrial chemicals, heavy metals etc), has also been reported by other authors (Kokila *et al*., 2005; Pradip *et al*., 2005).

Neutrophils also known as polymorphs are the body’s first line of defense against invasion by bacteria (Barton *et al*., 2000). Significant depletion observed in exposed group of all comparisons indicate that exposure to the dyes may lead to leukopenia due to high rate of destruction of neutrophils and makes body more vulnerable to infections (Barton *et al*., 2000). Alterations in neutrophil function by exposure to (environmental) contaminant may affect inflammatory/infectious conditions and thereby contribute to chronic inflammatory diseases of the respiratory tract (Nguyena *et al*., 2001).

Eosinophils, characterized by their large granules and bilobed nucleus destroy bacteria intracellularly to any great extent. A significant increase was observed in eosinophil count in workers upto 40 years of age and 15 years experience. It is showing that workers may be experiencing hypersensitivity or allergic diseases. Since raised level of eosinophils has been associated with parasitic infections and allergic responses (Gleich, 2000). Significantly higher number of eosinophils has been reported in patients positive for NGF (nerve growth factor), which plays an important role in allergies and lung respiratory diseases (El-Banna *et al*., 2006; Nassenstein *et al*., 2003).

Lymphocytes are the second numerous class of leukocytes, accounting for 20-40% of the total leukocytes. Changes observed in lymphocytes count at few instances, may be due to altered immune responses/reactions resulting in lymphocytosis in the exposed workers (Jude *et al*., 2002; Goyarts *et al*., 2006). In conclusion, the results obtained in the present study and previously published reports, indicate that exposure to textile chemicals induce microcytic anemia, leads to hypersensitivity and

makes body more vulnerable to infections. Furthermore, significant haematological changes observed in this study are in need to be elucidated so that the after-effects of toxicant contamination may be monitored and eliminated. This will not only help in the protection of the surrounding environment, but also in the prevention of malignant diseases in the present populations working in the factories as well as those residing in the industrial area.

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