AN OBSERVATIONAL STUDY TO ASSESS THE ASSOCIATION OF HEMOGLOBIN WITH BLOOD PRESSURE IN ADOLESCENT’S STUDENTS.

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ABSTRACT... Objectives: To assess the association of hemoglobin with blood pressure in adolescent’s students. Study Design: Observational study. Setting: Physiology Department of Baqai Medical College Karachi. Period: 6 months from February 2017 to August 2017. Sample Size: A total of 500 adolescents students of MBBS, BDS and DPT were enrolled in this study. Sampling Technique: Non probability consecutive sampling. Material & Method: A total of 500 students of MBBS, BDS and DPT were enrolled in this study after fulfilling the inclusion/exclusion criteria and was conducted after taking ethical approval from Baqai medical university. Complete Blood Count examination was carried out by Sysmex (XS-1000i) as to determine and record hemoglobin levels, blood pressure (BP) was measured by using sphygmomanometer with stethoscope and Anthropometric measurements were done. Data of study were analyzed by SPSS version 22.0. Results: The comparison of anemia prevalence in study participants for different categories of systolic and diastolic blood pressure had showed significant association for both systolic and diastolic blood pressure ($X^2$=12.036, p=0.002 and $X^2$=8.300, p=0.016 respectively). Conclusion: It showed positive association of hemoglobin with blood pressure (systolic and diastolic) in this study.

Key words: Anemia, Blood Pressure, Diastole, Systole.

INTRODUCTION

Anemia is a condition categorized by low hemoglobin level or decreased red blood cell count in the human body. Worldwide low hemoglobin (Hb%) level or anemia, is considered as major public health problem with prevalence in developing countries (43%) and in developed nations (9%). Anemia is caused by 3 basic mechanisms in human body: (i) decreased erythropoiesis or decreased production of erythrocytes (RBCs); (ii) loss of blood (internal or external); (iii) hemolysis. Nutritional anemia’s are caused by due to an inadequate storage of specific nutrients (iron i-e iron deficiency anemia, folic acid i-e megalobalstic anemia and vitamin b12 i-e pernicious anemia) in body that is needed for hemoglobin synthesis. According to world health organization (WHO) report in 2001, around two billion individuals had anemia and 50% of all sufferers were having Iron Deficiency Anemia (IDA). Iron plays an essential role for life, as it is the agent that carries oxygen. Oxygen is used in metabolism and provides the energy to the body for its normal activities. Only red blood cells (RBCs) or erythrocytes are capable of carrying oxygen to tissue cells in the body. According to world health organization (WHO), in 2008, 17.3 million people died from cardiovascular disease, which is responsible for 30% of deaths globally. Hypertension (HTN) or high blood pressure (BP) is considered as one of the most important risk factor for cardiovascular (CVD) disease. The high blood pressure or hypertension was observed after administration of erythropoietin hormone in hemodialysis patients for the treatment of anemia and also in orthostatic hypotension or postural hypotension, erythropoietin elevates the blood pressure while standing. Furthermore, it was observed that free hemoglobin (Hb%) exerts vasoconstriction due to release of nitric oxide (NO) scavenger and it limits the availability of nitric oxide (NO) for vessels,
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which elevates the blood pressure (BP).\textsuperscript{11,12} Nitric oxide (NO) acts as a vasodilator, so that increasing blood supply to tissue cells and decreases blood pressure. It helps in protection of tissue cells from hypotension.\textsuperscript{13} Nitric oxide (NO) also plays a role in vascular homeostasis by inhibiting contraction of vascular smooth muscles and growth. It also inhibits the platelet aggregation and leukocyte attachment with endothelium of vessels. Impaired nitric oxide pathways are associated with atherosclerosis, diabetes mellitus, or hypertension.\textsuperscript{14} It was observed in previous studies, that blood pressure (systolic and diastolic) may be increasing with increase in concentration of hemoglobin (Hb\%) levels in healthy individuals\textsuperscript{15,16} as well as in hypertensive individuals.\textsuperscript{17} The objective of this study was to assess the association of hemoglobin (Hb\%) with blood pressure in adolescent’s students.

**OPERATIONAL DEFINITION**

**Anemia**
It is defined as hemoglobin (Hb\%) concentration of less than 100 g/L or less than 11g/dl in venous blood.

**Hypertension**
It is defined as sustained rise of blood pressure (BP) in arteries (systolic blood pressure (SBP) i-e persistently at or above 140 mm of Hg or diastolic blood pressure (DBP) i-e at or above 90 mm of Hg).

**MATERIAL & METHODS**

**Set Up**
This observational study was done in students of MBBS, BDS, DPT of Baqai Medical University, who were attending the physiology department during classes, practical and tutorial timing.

**Study Subjects**
In this study, 500 (250 males and 250 females) adolescents’ medical students; with age ranges from 18-25 years were included. Ethical clearance was obtained from the Ethical review Committee of Baqai Medical University (BMU) Karachi. All the participants of the study were briefed about the research procedure and its significance.

**Duration**
Duration of study was 6 months from February 2017 to August 2017.

**Study Design**
Observational, Cross-sectional study in adolescents’ medical students.

**Sampling Technique**
Non probability consecutive samplig was used.

**Inclusion Criteria**
All students of MBBS, BDS and DPT were enrolled, after getting written consent.

**Exclusion Criteria**
All students were excluded from this study after getting positive history of blood disorder (thalassemia minor or major), history of blood transfusion or blood donation in last six months, history of intake of drugs (iron or folic acid or vitamin B12), history of passing worms, history of malabsorption syndrome, history of ileal surgery that leads to macrocytic anemia.

**Data Collection**
Data of study had been gathered with pre-tested questionnaire. Anemia is classified on the basis of world health organization (WHO) guidelines into mild anemia (Hb\% between 9.5 gm/dl and 11.5 gm/dl), moderate anemia (Hb\% between 7.5 gm/dl and 9.5 gm/dl) and severe anemia (Hb\% < 7.5 gm/dl).\textsuperscript{18} Blood pressure (BP) or arterial blood pressure was measured by standard methodology.\textsuperscript{19} Blood pressure (BP) or arterial blood pressure was measured in sitting position after giving rest to students for 5–10 minutes. Arterial blood pressure or Blood pressure (BP) was recorded to the nearest 2 mmHg. All blood pressures (BP) or Arterial blood pressures were recorded at the same time of the day, i.e., during afternoon hours. Blood pressure (BP) or Arterial blood pressure was measured by using sphygmomanometer with stethoscope. It was recorded by the same person and by the same instrument. Student’s data were verified for missing or incomplete entries and then entered in computer excel sheets. Statistical Package for Social Sciences (SPSS) version 22.0 was installed in computer for analyzing the collected data.
Frequency, percentages and Chi-square test were applied for qualitative variables. This test was used to assess the significance for categorical variables. The tables and graphs of this study were generated by Microsoft word and excel.

RESULTS

It was seen in Table-I of evaluation of Blood Pressure (BP) and Hemoglobin (Hb%) level in adolescent’s medical students. Figure-1(A) showed the Systolic Blood Pressure (SBP) readings (Normotensive, Pre-hypertensive & Hypertensive; 63.4%, 33.4% & 3.2% respectively), whereas Figure-1(B) showed the Diastolic Blood Pressure (DBP) readings (Normotensive, Pre-hypertensive & Hypertensive; 56.4%, 33.4% & 10.4% respectively) and Figure-1(C) showed the hemoglobin (Hb%) level readings (low, normal & high; 36.4%, 61.0% & 2.6% respectively). In Table-II showed the comparison of anemia prevalence in study participants for different categories of blood pressure (systolic blood pressure and diastolic blood pressure) and it was observed that results showed significant different for blood pressure including systolic and diastolic pressures ($X^2=12.036$, $p<0.002$ and $X^2=8.300$, $p<0.016$ respectively). An increase in blood pressure (systolic and diastolic) levels from normal to high was followed by an increasing percentage of study participants who had either low, normal or high hemoglobin level (58.0%, 72.5%, 81.2% and 59.1%, 66.5%, 78.8% respectively).

| Variables (n=500)                                      | Frequency | Percentage (%) |
|--------------------------------------------------------|-----------|----------------|
| Systolic Blood Pressure (SBP)                          |           |                |
| Normotensive                                           | 317       | 63.4%          |
| Pre-hypertensive                                       | 167       | 33.4%          |
| Hypertensive                                           | 16        | 3.2%           |
| Diastolic Blood Pressure (DBP)                         |           |                |
| Normotensive                                           | 281       | 56.2%          |
| Pre-hypertensive                                       | 167       | 33.4%          |
| Hypertensive                                           | 52        | 10.4%          |
| Hemoglobin (Hb%)                                       |           |                |
| Low                                                    | 182       | 36.4%          |
| Normal                                                 | 305       | 61.0%          |
| High                                                   | 13        | 2.6%           |

| Table-I. Evaluation of blood pressure (Systolic and Diastolic) and hemoglobin (Hb%). |

Figure-1 (A). It indicates the Systolic Blood Pressure (SBP).

Figure-1 (B). It indicates the Diastolic Blood Pressure (DBP).

Figure-1 (C). It indicates the Hemoglobin (Hb%) level.
Table-II. Association between Blood Pressure (BP) and Anemia.

| Variable (n=500) | Hemoglobin Level | Low Frequency (%) | Normal High Frequency (%) | P-Value |
|-----------------|------------------|-------------------|--------------------------|---------|
| Systolic Blood Pressure (SBP) | Low | 133(42.0%) | 184(58.0%) | <0.002 |
|                | Normal | 46(27.5%) | 121(72.5%) |         |
|                | High   | 3(18.8%)  | 13(81.2%)  |         |
| Diastolic Blood Pressure (DBP) | Low | 115(40.9%) | 166(59.1%) | <0.016 |
|                | Normal | 56(33.5%) | 111(66.5%) |         |
|                | High   | 11(21.2%) | 41(78.8%)  |         |

P < 0.05 shows significant values and P > 0.05 shows non significance values.

DISCUSSION

This observational study was done in department of physiology in Baqai Medical College (BMC) of Baqai Medical University (BMU) of Karachi, in which 500 (250 male and 250 female) adolescents medical student were enrolled of different departments like MBBS, BDS, DPT. The blood samples were drawn for measuring hemoglobin (Hb%) level and blood pressure (BP) was measured by using sphygmomanometer with stethoscope of all participating students of study, after fulfilling the inclusion and exclusion criteria. The objective of this study was to assess the association of hemoglobin (Hb%) with blood pressure in adolescent’s students.

According to our study results comparison of anemia prevalence is categorized into low frequency and normal high Frequency (%) in study participants for different categories of systolic blood pressure (Normotensive, Prehypertensive and Hypertensive) & diastolic blood pressure (Normotensive, Prehypertensive and Hypertensive). It was observed that that it was significantly different for blood pressure (systolic and diastolic) (X²=12.036, p<0.002 and X²=8.300, p<0.016 respectively). An increase in blood pressure (systolic and diastolic) level from normal to high was followed by an increasing percentage of study participants who had either low, normal or high hemoglobin level (58.0%, 72.5%, 81.2% and 59.1%, 66.5%, 78.8% respectively). This present observational study showed positive association between hemoglobin (Hb%) level with systolic blood pressure (SBP) and diastolic blood pressure (DBP) in students. Many studies have been done in various countries, which showed association in results, so that it support our results too. According to Atsma et al, there was a positive association between hemoglobin with systolic blood pressure (SBP) and diastolic blood pressure (DBP) in both genders (men & females). Göbel et, also observed similar results in healthy persons. Lianxiang Ren et al, also concluded that the blood pressure is increased with the increasing of hemoglobin (Hb%) level both in SBP and DBP in normal individuals and the exact mechanisms for hemoglobin (Hb%) leading to an increased blood pressure (BP) are not entirely known, but having the viewpoints that hemoglobin (Hb%) had a direct or indirect effects on vascular system. The mechanisms that may leads to association of hemoglobin with blood pressure are not entirely known. So proposed mechanisms behind this association may be like hemoglobin (Hb%) may be associated with arterial stiffness, that ultimately increases blood pressure (systolic and diastolic) and free hemoglobin (Hb%). Nitric oxide acts as a scavenger in the human body and endothelial cells of blood vessels are responsible for production in the human body. It controls blood pressure (BP) by relaxing muscle cells of vessels. Another obvious physiological mechanism lying behind the association of blood pressure and...
Hemoglobin level would be due to elevation in hematocrit (Hct%) level, and it increases blood viscosity. Blood viscosity exerts effects on blood pressure and can worsen cardiovascular function. Blood viscosity is also responsible for production of nitric oxide (NO) and causes vasodilation via the induction of shear stress, so blood pressure increases. Erythropoietin, hormone is used in treatment of anemia, it would leads to elevation of hemoglobin (Hb%) and blood pressure (BP). Renin-angiotensin-aldosterone system (RAAS) may be related with hemoglobin (Hb%) and blood pressure (BP).20

Since the autonomic nervous system especially sympathetic nervous system mainly regulates the Renin-angiotensin-aldosterone system (RAAS), which affects the production of erythropoietin and so hemoglobin and blood pressure is related to this system.22

CONCLUSION
This present study showed positive significant (p-value<0.05) association b/w hemoglobin (Hb%) level with systolic and diastolic blood pressure in adolescent’s students.

RECOMMENDATION
To extend this study on large scale and participation of multi-centers.

STUDY LIMITATIONS
As this study was carried on university students, as a requisite of M-phil degree in subject of Physiology, so there were limitations of included students and fundings.

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CONFLICT OF INTEREST
Authors declare no conflict of interest

REFERENCE
1. Habibzadeh F. Anemia in the Middle East. Lancet. 2012;379(1).
2. Arshad K, Azam M, Munir T, Khan MI, Abbas HN, Butt MA. Frequency of Anemia in Healthy School Age Population in Faisalabad. APMC 2017; 11(2):132-135.
3. Who U. UNU. Iron deficiency anaemia: assessment, prevention and control, a guide for programme managers. Geneva: World Health Organization. 2001 Oct:1-14.
4. Zahra Khan et al, Hemoglobin, Red Blood Cell Count, Hematocrit and Derived Parameters for Diagnosing Anemia in Elderly Males. Proceedings of the Pakistan Academy of Sciences 50 (3): 217–226 (2013).
5. Lawes CM, Vander Hoorn S, Rodgers A, et al. Global burden of blood-pressure-related disease, 2001. Lancet 2008; 371:1513-8.
6. Brook RD, Appel LJ, Rubenfire M, et al. Beyond medications and diet: Alternative approaches to lowering blood pressure: A scientific statement from the American heart association. Hypertension 2013; 61:1360-83.
7. Cheng HM, Chuang SY, Sung SH, et al. Derivation and validation of diagnostic thresholds for central blood pressure measurements based on long-term cardiovascular risks. J Am Coll Cardiol 2013; 62:1780-7.
8. Chambers JK. Case study of the anemic patient: Epoetinalfa--focus on blood pressure. ANNA J. 1994; 21:154–157.
9. Kanbay M, Akcay A, Delibasi T, Uz B, Kaya A, Koca C, Turgut F, Bavybek N, Uz E, Duranay M, Yigitoglu R. Comparison of effects of darbepoetinalfa and epoetinalfa on serum endothelin level and blood pressure. Adv Ther. 2007; 24:346–352.
10. Hoeldtke RD, Streiten DH. Treatment of orthostatic hypotension with erythropoietin. N Engl J Med. 1993; 329:611–615.
11. Cabrales P, Han G, Nacharaju P, Friedman AJ, Friedman JM. Reversal of hemoglobin-induced vasoconstriction with sustained release of nitric oxide. Am J Physiol Heart Circ Physiol. 2011; 300:H49-H56.
12. Cabrales P, Sun G, Zhou Y, Harris DR, Tsai AG, Intaglietta M, Palmer AF. Effects of the molecular mass of tense-state polymerized bovine hemoglobin on blood pressure and vasoconstriction. J Appl Physiol. 2009; 107:1548–1558.
13. Van Faassen EE, Bahrami S, Feelisch M, Hogg N, Kelm M, et al., “Nitrite as regulator of hypoxic signaling in mammalian physiology”, Med Res Rev, 2009 Sep; 29(5):683–741.
14. Dessy C, Feron O. Pathophysiological roles of nitric oxide: in the heart and the coronary vasculature. Current Medicinal Chemistry-Anti-Inflammatory & Anti-Allergy Agents. 2004 Sep 1;3(3):207-16. doi:10.2174/1568014043355348.

15. Göbel BO, Schulte-Gobel A, Weisser B, Glanzer K, Vetter H, Dusing R. Arterial blood pressure: correlation with erythrocyte count, hematocrit, and hemoglobin concentration. Am J Hypertens. 1991; 4:14–19.

16. Kawamoto R, Tabara Y, Kohara K, Miki T, Kusunoki T, Katoh T, Ohtsuka N, Takayama S, Abe M. A slightly low hemoglobin level is beneficially associated with arterial stiffness in Japanese community-dwelling women. Clin Exp Hypertens. 2012; 34:92–98.

17. Atsma F, Veldhuizen I, de Kort W, et al. Hemoglobin level is positively associated with blood pressure in a large cohort of healthy individuals. Hypertension 2012; 60:936-41.

18. WHO | Haemoglobin concentrations for the diagnosis of anaemia and assessment of severity [Internet]. WHO. [cited 2014 Sep 4]. Available from: http://www.who.int/vmnis/indicators/haemoglobin/en/.

19. National High Blood Pressure Education Program. Working Group on High Blood Pressure in Children and Adolescents. The fourth report on the diagnosis, evaluation, and treatment of high blood pressure in children and adolescents. Pediatrics, 2004; 114:555–76.

20. Femke Atsma, Ingrid Veldhuizen, Wim de Kort, Marian van Kraaij, Pieteren Pasker-de Jong, et al; Hemoglobin level is positively associated with blood pressure in a large cohort of healthy individuals, hypertension. 2012; 60:936-941.

21. Ren L, Gu B, Du Y, Wu X, Liu X, Wang H, Jiang L, Guo Y, Wang J. Hemoglobin in normal range, the lower the better?—Evidence from a study from Chinese community-dwelling participants. Journal of thoracic disease. 2014 May;6(5):477.

22. Shimizu Y, Nakazato M, Sekita T, Kadota K, Arima K, Yamasaki H, Takamura N, Aoyagi K, Maeda T. Association between the hemoglobin levels and hypertension in relation to the BMI status in a rural Japanese population: the Nagasaki Islands Study. Internal Medicine. 2014;53(5):435-40.

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**AUTHORSHIP AND CONTRIBUTION DECLARATION**

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