



INVESTIGATION THE ROLE OF SERUM LEVEL OF HAPTOGLOBULIN (HP) IN PATIENTS SUFFERED FROM CHRONIC OBSTRUCTIVE PULMONARY DISEASE (COPD)

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ABSTRACT

Chronic obstructive pulmonary disease (COPD) is the most common lung disorders in adults with progressive obstruction, defuse irreversible airway narrowing is associated with increased resistance to air. Many inflammatory and immunological activates have role in induction of disease. Among of them haptoglobin (Hp) as an acute phase protein may has key role in induction of disease. As there is no available report in connection with the study of serum haptoglobin in COPD patients so we decided to investigate the role of serum Hp in COPD patients. This case-control study was done on 60 patients with COPD and 60 healthy controls in the clinic of lung at Ali Ibn Abi Talib (AS) hospital of Zahedan. 5mls of peripheral blood were collected from patients and healthy control and serum was prepared and haptoglobin concentration was measured by nephelometry method. Among individuals who were diagnosed with COPD, the haptoglobin serum levels was calculated as 1.89 ± 0.80 grams per liter and in people without COPD mean haptoglobin concentration was 1.19 ± 0.76 grams per liter. This difference was statistically significant ($p < 0.0001$). Thus it was concluded that ,high concentration of haptoglobin has relation with COPD disease.

Key words:- Haptoglobin - COPD.

INTRODUCTION

Chronic obstructive pulmonary disease (COPD) or progressive lung obstructive refers to a condition characterized by progressive obstruction, increased resistance to air flow and airflow limitation in the lungs with increasing frequency and severity of attacks intensified somewhat reversible but not fully reversible (Taffet GE *et al.*, 2014). Obstruction caused by inflammation and increased production of mucus in the respiratory tract of the bronchi which is called bronchioles caused reduce the air flow (Bade G *et al.*, 2014).

Disease is mainly caused by smoking so that 10 to 15 percent of smokers are infected (Yoshimoto D *et al.*, 2014; Salvi S *et al.*, 2014). Other factors such as genetic, occupational and environmental factors such as exposure to dust exposure are also effective (Wang X *et al.*, 2014; Atkinson RW *et al.*, 2015). Symptoms are usually silent at the beginning and gradually progressive and include shortness of breath and cough, excessive sputum production, wheezing, increased carbon dioxide levels in blood, reduced oxygen levels and physical intolerance activity (fatigue) accompanied with systemic manifestations like secondary polycythemia, anxiety and impaired skeletal muscle function and increased frequency

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and severity of attacks (Barisione G *et al.*, 2014; Yamashiro T *et al.*, 2010; Livermore N *et al.*, 2012).

It includes a range of different types of diseases such as emphysema which is characterized by the destruction and enlargement of alveolar, chronic bronchitis that is accompanied with cough and sputum while there is obstruction or chronic airflow obstruction is a component of COPD disease and finally bronchiolitis that in this disease bronchioles can be tight (Kurashima K *et al.*, 2012; Langer D *et al.*, 2014). Comorbidities of COPD have been reported include hypertension, asthma, heart disease, anemia and depression (Kristan SS, 2013; Blakemore A *et al.*, 2014).

COPD is one of the most common respiratory disorders in elderly that would be third cause of death and the fifth leading cause of chronic disability (Taffet GE *et al.*, 2014) and increases risk of death as a result of lifestyle changes and urbanization (Valero C *et al.*, 2009). In Iran country this disease with pulmonary heart disease are the twelfth cause of life lost due to premature death or disability (Naghavi M *et al.*, 2009).

Preventive policies include: control increasing number of respiratory attacks, hemoptysis, pulmonary rehabilitation and surgery (Overington JD *et al.*, 2014). Treatment are dependent to the smoking give up by 90%, improves environmental conditions hospitalization, using a combination bronchodilator drugs, inhaler steroids and antibiotics to keep the airways open, reduce inflammation and treat possible infections. Oxygen therapy at night, pulmonary rehabilitation, exercise and pneumococcal influenza vaccination may improve lung function (Müllerová H *et al.*, 2014). On physical examination the patient's height and weight and muscles, especially in the thighs and look for signs of hyperinflation of the chest (barrel-shaped), loss beat at the apex, resonance exorbitance and reduce diaphragm motion that can be seen in disease intensity, should be considered (Langer D *et al.*, 2014; Cielen N *et al.*, 2014).

For the detection and measurement of respiratory volumes chest X-Ray, spirometry and arterial blood gas measurement by pulse oximetry system, which reflects the amount of oxygen in the bloodstream, are used. Recently other diagnostic methods are used to diagnose better the disease by the measurement of other parameters such as blood serum, including measurement of serum CRP, cytokines, immunoglobulin, complement, alpha-1 antitrypsin and especially haptoglobulin (Johns DP *et al.*, 2014; Chen C *et al.*, 2014; Molloy K *et al.*, 2014; Quayle IK, 2008).

Haptoglobulin (HP) is a plasma α -2-sialoglycoprotein that belongs to the family acute phase proteins in human serum that synthesis In response to the

release of cytokines such as interleukin-6 (IL-6), IL-1, tumor necrosis factors (TNF) in the liver and lung epithelial and alveolar macrophage cells and increases in inflammation and infections (Jelena A *et al.*, 2013, Arredouani M *et al.*, 2005). This protein plays a role in the regulation of acute phase proteins and equilibrium between Th1 and Th2 immune responses and thus will contribute to immune regulation (Arredouani M *et al.*, 2008).

This protein was identified for the first time in 1940 by Jayle and Polonovski as a natural bacteriostatic has role in diseases such as tuberculosis, malaria, major depressive disorder, epilepsy, rheumatoid arthritis and systemic lupus erythematosus (Hzecka J *et al.*, 1996). Its serum concentration is 0.32-2.08 g/L and is associated with various forms (phenotype) of this protein as Hp1-1, Hp2-1 and Hp2-2 as based on kind of phenotype, its concentration getting low respectively. Its serum concentration of this protein is usually lower than normal in tissues, umbilical cord and blood of newborns and varies with age (Koch W *et al.*, 2002; Tseng CF *et al.*, 2004).

Changes in serum levels of this protein in asthmatic patients after allergen skin test trials have been reported in children (Kim CK *et al.*, 1998).

Due to the presence of this protein in serum, its main role is to protect the lungs against inflammatory agents and its presence in the lung is the most common prevention cause of lung injury. Its deficiency leads to lung tissue destruction caused by natural bacteriostatic neutrophil function, leading to the creation of emphysema and COPD. The protein deficiency leads to early onset of emphysema usually the incidence of disease is exacerbated by smoking and in some cases becomes asthma (Wobeto VPA *et al.*, 2008; Khazaei HA *et al.*, 2013).

Low levels of this protein has been found in the serum of patients with bone marrow transplant rejection that was due to pathogenicity of immune complexes as reported by Shichishima (2010).

Due to the lack of this protein almost is "a risk factor for COPD disease and also in connection with the study of serum protein levels of Hp in COPD patients, there is no available report, therefore, this study aimed to examine the association of Hp with COPD disease in order to make better diagnosis and treatment of disease.

MATERIALS AND METHODS

This case-control study was carried out on 60 COPD patients and 60 healthy controls without affection of systemic by non-probability sampling method.

The patients with a previous diagnosis by pulmonologist referred at lung clinic of Ali Ibn Abi Talib hospital (AS) of Zahedan-Iran, were selected based on inclusion criteria included patients who complain of having COPD symptoms, confirmation of the disease and the symptoms of obstructive spirometer. At the same time, 60 healthy individuals without risk of systemic disease and COPD and chronic diseases, non-cancer diseases and systemic diseases, lack of infectious pulmonary diseases, pulmonary emphysema, asthma and COPD, no addiction to smoking, no being older, BMI normal, appropriate socio-economic status, job status, as well as selected After completion of the informed consent form, demographic data were collected and then 5 ml of peripheral blood were taken and after separation of serum, the serum was stored in the fridge -70 ° C until the time of use.

The measurement of serum Hp was determined by *nephelometry* method.

RESULTS

The distribution of patients by age and sex were, 19 patients (31.7%) were male and 41 (68.3%) were female. The mean age of these patients was 59.76 with the age range of 40 to 78 years old. The average amount of haptoglobin serum levels obtained was 1.89 ± 0.80 gram per liter whereas in non COPD individuals was 1.19 ± 0.76 gram per liter. Based on statistical T independent test, the significant difference between the average amount of haptoglobin in two groups was found ($p < 0.0001$) (below table).

Table 1. The comparison of serum haptoglobin levels in COPD patients and non COPD individuals

| Amount of haptoglobin g/liter | Group |
|-------------------------------|---------------------|
| 80.89±0.1 | COPD patients |
| 76.19±0.1 | Healthy individuals |
| <0.0001 | P value |

DISCUSSION

In this study, the serum concentration of haptoglobin levels in COPD patients and healthy people, were evaluated. The results showed that in cases compared with healthy individuals, haptoglobin level was higher which was statistically significant.

In relation to the role of haptoglobin in the pathogenesis of the COPD disease, few studies have been done in this case. but as this protein is a family of acute phase protein, there is a report issued by Engström *et al* in 2009 with the aim of evaluation the inflammatory markers in COPD, their results indicated that those who had higher

plasma inflammatory proteins such as fibrinogen, ceruloplasmin, alpha-1 antitrypsin, haptoglobin and orosomucoid, were associated with a higher risk of hospitalization (Engström G *et al.*, 2009). The results of our study are the same as in our study group had higher levels of haptoglobin.

In another study done by Verrills *et al* (2011), their results showed that alpha-2-microglobulin, haptoglobin, ceruloplasmin and hemopexin in patients with asthma and COPD significantly increases which is consistent with our results.

In report of Varadi *et al* (2013), they showed that changing of haptoglobin glycolysiation could be as a biomarker used in patients with lung cancer and inflammatory diseases. Their results are also consistent with our results.

And more recently Pedersen *et al* (2014) investigated multi-analyte profiling of inflammatory mediators in COPD sputum. Their results indicated that significantly higher levels of some inflammatory mediators such as vitamin D binding protein (VDBP), α -2-Macroglobulin, haptoglobin, α -1-antitrypsin, vascular cell adhesion molecule- 1 (VCAM-1) and fibrinogen were observed following dithiothreitol (DTT) treatment induced sputum samples of 20 patients with stable COPD (Pedersen F *et al.*, 2015). The results of our study are the same as Pedersen *et al* study as in our study group there was higher levels of haptoglobin.

Regarding the relationship between serum concentration of haptoglobin and inflammatory diseases such as COPD, as haptoglobin is an acute phase proteins with immune regulation stimuli, increased serum levels of these proteins has been reported in infectious and inflammatory processes as well as in cancer, collagen disease, tissue damage, encephalitis, nephritis, pyelonephritis, ulcerative colitis, peptic ulcer disease, myocardial infarction. Whereas decreased levels of this protein has been observed in hemolytic anemia transfusion reactions, prosthetic heart valves, systemic lupus erythematosus (SLE), primary liver disease without associated with hemolytic anemia, erythroblastosis fetalis, *tissue* bleeding and chronic liver disease.

Because in COPD, the processes of inflammation, infectious processes and tissue destruction can be seen and also haptoglobin is a member of acute phase protein, we expect its concentration in serum of patient's increases. On the other hand increase high amount of this protein, indicates the presence of inflammatory processes in the body. Furthermore, due to the presence and effect of serum haptoglobin that indicates the activity of immune system, especially "balance between the innate and acquired immune activities

(Arredouani M *et al.*, 2008), thus low amount of this protein in serum can also cause immune deficiency, we expect in people who have lesser amount of this protein than usual, they are susceptible to getting numerous infection.

CONCLUSION

Our results showed that the amounts of serum Haptoglobin levels are higher in patients with COPD. It is suggested that similar studies needs to be done in future on haptoglobin levels and disease severity. It is also

recommended that molecular studies such as phenotyping and genotyping of this protein in COPD patients need to be done in the future.

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