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THE RELATION WITH GLUTATHIONE PEROXIDASE, TRACE ELEMENTES IN PATIENTS WITH ACNE VULGARIS

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ABSTRACT: Acne vulgaris is a chronic inflammatory skin condition common in adolescence, but occasionally occurs intermittently throughout life. It is characterized by skin eruption on the face, chest, neck and back. This disease is more common in males than in female. It happens when the oil gland in the skin become plugged for many reasons. When oil backs up, it becomes infected by bacteria P.acne that is normally present on the skin and acne vulgaris develops. In this work seventy three individuals with acne vulgaris classified into 3 groups, 24 patients with mild acne, 19 patients with moderate acne and 30 patients with severe acne were studied. In addition, included forty-two healthy individuals considered as a control group. The study revealed that the zinc shows significantly decreased level in the patients and an increase in the level of copper by comparison with the control group. Also results showed that glutathione peroxidase is significantly decreased in patients with severe acne patient group while no difference in other types.

Key Words: Acne, P.acne , glutathione peroxidase , zinc ,copper

INTRODUCTION

Acne is described as a disease of pilosebaceous units (PSUs). Those are found over most of the body skin, PSUs consists of a sebaceous gland connected to a canal, called a follicle, which contains a fine hair. These units are most numerous on the face, upper back, and chest. The sebaceous glands make an oily substance called sebum that normally empties onto the skin surface through the opening of the follicle, commonly called a pore, and the cells which line the follicle called keratinocytes (Ayres S.Jr., Mihan R., 1981). People with acne frequently have a variety of lesions, and basic acne lesion, called the comedone (KOM-e-do), which is simply an enlarged and plugged hair follicle. If the plugged follicle, or comedone, stays beneath the skin, it is called a closed comedone and produces a white bump called a whitehead (Degitz K., Ochsendorf F., 2008). A comedone that reaches the surface of the skin and opens up is called an open comedone or blackhead because it looks black on the skin's surface. This black discoloration is due to changes in sebum as it is exposed to air and both whiteheads and blackheads may stay in the skin for a long time (William D., et.al. 2006).Copper is a mineral found in trace amounts in all tissues and widely distributed in the body and occurs in liver, muscle and bone. Copper is transported in the bloodstream on a plasma protein called ceruloplasmin. When copper is first absorbed in the gut it is transported to the liver to albumin. Copper metabolism and excretion is controlled through delivery of copper to the liver by ceruloplasmin, where it is excreted in bile (Wapnir RA, 1998).

Copper is found in a variety of enzymes, including the copper centers of cytochrome oxidase which is involved in mitochondria for reduction of O2 to water and produce energy in form of ATP. Also the enzyme superoxide dismutase (containing copper and zinc), also in addition to its enzymatic roles, copper is used for biological electron transport (Turnlund JR., et.al. 2005).
Zinc is an essential trace element which is necessary for plant (Broadley MR., et.al. 2007) and animal life, including microorganisms (Prasad AS., 2008). The human body has 2-4 grams of zinc distributed throughout the body. Most zinc is in the brain, muscle, bones, kidney, and liver, with the highest concentrations in the prostate and parts of the eye (Hershfinkel M., et.al. 2007). Serum zinc has a rapid turnover rate and it represents only about 0.l percent of total body zinc content. This level appears to be under close homeostatic control. High concentrations of zinc are found in the choroid of the eye 4.2 µmol/g (274 µg /g) and in prostatic fluids 4.6-7.7 mmol/l (300-500 mg/l) (Wapnir RA., 1990). Both zinc and selenium are intimately involved in protecting the body against oxidant stress. Zinc combined with copper is found in the cytoplasmic form of superoxide dismutation (SOD) whereas zinc and magnesium occur in the mitochondrial enzyme. SOD occurs in all aerobic cells and is responsible for the dismutation of superoxide:

$$O_2.^- + O_2.^- + 2H^+ \longrightarrow H_2O_2 + O_2$$

Glutathione peroxidase EC 1.11.1.9 is one of the most important lines of defence against the oxidative damage by hydrogen peroxide or lipid peroxide produced in various cells of the body. It has been suggested that glutathione peroxidase may be able to break the autocatalytic chain reaction of lipid peroxidation protecting the cell membrane from oxidative damage (Whitin JC, et.al. 2002). Humans have five selenoprotein glutathione peroxidases, including GPx1, gastrointestinal GPx2, plasma GPx3 and its close homolog GPx6, and phospholipid hydroperoxide glutathione peroxide se, known as PHGPX or GPx4 (Meimaridou E., et.al. 2006).
The aim of this paper is to study the changes occurs in serum level of trace element such as zinc, copper [zinc as antioxidant and copper as contributor to oxidative stress] and Glutathione peroxidase in patients with acne vulgaris compared to control groups.

MATERIAL AND METHODS

PATIENTS

The study was conducted in Hilla city, from December 2009 to September 2010. Seventy three patients [38 males with mean age + SD (20 + 4)years and 35 females with mean age + SD (20 + 2.8) years] with Acne vulgaris, whom were collected from Merjan Teaching Hospital in Hilla city, have been subjected to present study and these selected patients were divided into three groups according to type of disease:-
- The first group includes 24 patients with mild acne vulgaris
- The second group includes 19 patients with moderate acne vulgaris
- The third groups include 30 patients with severe acne vulgaris

CONTROL PATIENTS

The control group includes forty-one apparently healthy individuals, after having been asked about their health. PCOS female have been excluded from the control group and divided into two groups:-
- The first one includes 22 females with mean age + SD (18 + 2.1) years.
- The second one includes 19 males with mean age + SD (20+ 2.6) years.

1. Collection of Blood and Serum Preparation

Five to eight millilitres of blood were obtained from patients and healthy persons, then collected in tubes without anticoagulants and were left for 15 minutes at room temperature to clot. After that, the blood samples were centrifuged at 1000 - 2000 x g for approximately 10 minutes. The sera were aspirated and stored at -20 C until time of use.
For hormone samples serum may be stored at 2-8 C for up to 24 hours, and should be frozen at -10 C or lower for longer periods. Grossly hemolyzed or grossly lipemic specimens were not used.

2. Determination of serum zinc by using zinc kit. LTA s.r.l(Italy)

3. Determination of serum copper level by using commercial kit LTA s.r.l(Italy)

4. Determination of serum glutathione peroxidase (GPx) according to the procedure of Rotruck et al (Rtruck JT., et.al. 1973).

RESULTS AND DISCUSSIONS

Zinc and Copper in Acne Vulgaris

Zinc and copper are two important trace elements in biological system. The changes in level of these trace elements are studied in this work, as well as the effects on each others. The study shows significant decrease in the mean zinc level in the female patients group by, and also significant decreases in the mean of male patients in comparison with the control group. Table (1) (Michaelsson G., et.al. 1977). Also in Table (2) there was significantly decrease in mean serum level of zinc in patients by comparison between the female and male groups of control and female and male groups in mild, moderate and severe acne vulgaris groups (Michaelsson G., et.al. 1977). Table (2).

Table (1): The mean serum levels of zinc in female & male patient and control

ZINC (µg /dl)	Female				Male			
	M	SD	No.	*P-value*	M	SD	No..	*P-value*
patients	206	37.54	25	0.04	196	48.19	25	0.02
control	215.6	76.42	21		262	77.77	11	

The differences in mean of serum level of copper in acne patients and control were shown in the Table (3). The study shows significant increase in mean copper level in the female and male patients group in comparison with control group, which is shown in Table (3), while in Table (4) there is significant decreases in the copper mean level of female and male control group by comparison with female and male groups in mild, moderate as well as severe acne groups.
In the present study there is significant difference in zinc levels between patients and control groups. In acne patients, the serum level of zinc is significantly lower than that in control group p-value < 0.01. In contrast to this, the serum level of copper is significantly elevated in acne patients in relation to control, p value < 0.01, however result of this study agrees with the previous studies like Mchaelsson G (Michaelsson G., et.al. 1977), where he found that the zinc is reduced and copper is elevated in acne vulgaris state compared to control group.

Table (2): The mean serum levels of zinc in severe, moderate & mild male & female patients and control

Female Zinc (µg /dl)	**severe**				**moderate**				**mild**			
	M	**SD**	**No.**	***P-value***	**M**	**SD**	**No**	***P-value***	**M**	**SD**	**No.**	***P-value***
Patients	**174**	**30.2**	**7**	**0.002**	**201**	**8.6**	**8**	**0.034**	**201**	**35.2**	**11**	**0.048**
Control	**215**	**76.4**	**21**		**215**	**76.4**	**21**		**215**	**76.4**	**21**	
Male Zinc (µg /dl)	**severe**				**moderate**				**mild**			
	M	***SD***	**No.**	***P-value***	***M***	***SD***	**No**	***P-value***	***M***	***SD***	**No.**	***P-value***
Patients	**211**	**52.1**	**15**	**0.026**	**200**	**3.4**	**4**	**0.031**	**206.5**	**16.9**	**6**	**0.049**
Control	**262**	**77.7**	**11**		**262**	**77.7**	**11**		**262**	**77.7**	**11**	

Table (3): The mean serum levels of copper in female & male patient and control

Cupper **(µg /dl)**	Female				Male			
	M	**SD**	**No.**	***P-value***	**M**	**SD**	**No.**	***P-value***
patients	**173**	**22.08**	**35**	**0.019**	**185**	**45.23**	**38**	**0.004**
control	**154**	**25.94**	**22**		**138**	**26.87**	**19**	

The copper is one of the metals that have been demonstrated to catalyze the formation of OH and other radicals from H_2O_2 and O_2 through Fenton reaction. Through these reaction trace elements, mainly iron and copper in the presence of reducing agents (AH2) such as ascorbate can catalyze the formation of hydroxyl radical, this reaction called Haber-Weiss reaction or Fenton reaction as follow, (Micheal D., Jonathan H., 2004) (Thanaa M., 2008) and this study suggest that zinc concentration may be considered a biochemical marker of oxidative stress associated with acne and other studies included copper concentration analysis of patients with acne disease, where high levels were shown when compared with healthy controls.

$2AH_2+2O_2 \longrightarrow (Cu/Fe)\ 2A+2H_20_2$

$AH_2+2Cu^{+2} \longrightarrow A+ 2Cu^{+1}+2H^{+}$

$2Cu^{+1}+2H_2O_2 \longrightarrow 2Cu^{+2}+2OH+2OH$

Table (4): The mean serum levels of copper in severe, moderate & mild Male & Female patient and control

Female Copper (µg /dl)	**severe**				**moderate**				**mild**			
	M	**SD**	**No.**	***P-value***	**M**	**SD**	**No**	***P-value***	**M**	**SD**	**No.**	***P-value***
patients	**180.5**	**25.99**	**16**	**0.044**	**178**	**18.4**	**9**	**0.000**	**161**	**16.6**	**10**	**0.017**
control	**138**	**22.3**	**22**		**138**	**22.3**	**22**		**138**	**22.3**	**22**	
Male Copper (µg /dl)	**severe**				**Moderate**				**Mild**			
	M	**SD**	**No.**	***P-value***	**M**	**SD**	**No**	***P-value***	**M**	**SD**	**No.**	***P-value***
patients	**195**	**31.8**	**20**	**0.001**	**182**	**29.6**	**9**	**0.040**	**179**	**10.0**	**9**	**0.002**
control	**138**	**26.8**	**19**		**138**	**26.8**	**19**		**138**	**26.8**	**19**	

Table (5): The mean serum levels of glutathione peroxidise in Female & Male patient and control.

GPx **(mlU/ml)**	Female				male			
	M	SD	No.	***P-value***	M	SD	No.	***P-value***
patients	548	310	25	0.106	614	332.78	27	0.13
control	773	379.4	11		804	760	11	

Glutathione Peroxidase

The result shows no significant difference in the activity of GPx in patient and control in both male and female which is obvious in Table (5). Also Table (6) shows that there are no significantly differences between groups of control and groups in mild acne groups as well as in the moderate acne groups while there is significant decrease between groups of control and groups patient with severe acne (Michaelsson G., Edgvist LE., 1984).

This decrease can be attributed that the skin synthesizes hydrogen peroxide to fight each acne inflammation and this can continue few weeks, until the inflammation is resolved. Hydrogen peroxide is from free radical which causes skin harm and skin aging. As time passes the volume of hydrogen peroxide acts just like prolonged sun exposure, damaging skin elements such as collagen and causing the skin to sag and wrinkle (Baynes JW and Dominiczak HM, 2004). The primary natural defense against free radicals is to prevent their formation by various enzyme, especially GPx which regulating hydrogen peroxide (H_2O_2) level in catalyzing the dismutation of H_2O_2 to H_2O +CO_2.

Table (6): The mean serum levels of Glutathione Peroxidise in severe, moderate & mild Male & Female patients and control

Female GPx **(mlU/ml)**	**Severe**				**moderate**				**Mild**			
	M	**SD**	**No.**	***P-value***	**M**	**SD**	**No**	***P-value***	**M**	**SD**	**No.**	***P-value***
patients	**421**	**361.9**	**9**	**0.048**	**540**	**245.6**	**9**	**0.147**	**750**	**231**	**7**	**0.706**
control	**773**	**379.4**	**11**		**773**	**379.4**	**11**		**773**	**379.4**	**11**	
Male GPx **(mlU/ml)**	**Severe**				**moderate**				**Mild**			
	M	**SD**	**No.**	***P-value***	**M**	**SD**	**No**	***P-value***	**M**	**SD**	**No.**	***P-value***
patients	**459**	**174.8**	**12**	**0.034**	**545**	**228**	**7**	**0.085**	**906**	**418.9**	**8**	**0.776**
control	**804**	**760**	**11**		**804**	**760**	**11**		**804**	**760**	**11**	

The results of the present study agree with those obtained result (Michaelsson G., Edgvist LE., 1984) where they attributed the decrease in the activity of this enzyme to the increase of hydrogen peroxide level by the neutrophils from (Rana Abd. Al-Aly, 2008) acne inflammation in those patients, sebum produced by sebaceous gland, content changes and reactive oxygen species (ROS). All these may be released from the impacted damaged follicular walls; at the same time it is thought this may be the reason for the progress of the inflammation in the pathogenesis of the disease (Briganti S & Picardo M., 2003) (Akamatsu H., Horio T. & Hattori K., 2003).

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