Evaluation of Serum Zinc Level in Patients with Chronic Telogen Effluvium in Premenopausal Adult Females in Kirkuk City.

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Abstract:
Chronic telogen effluvium (CTE) was defined as a primary idiopathic disease entity in 1996. Women suffering from CTE present with an abrupt onset of generalized shedding of telogen hair from the scalp, with or without an identifiable trigger that persists for more than 6 months. Therefore, this study is conducted to detect the association of serum zinc with CTE. The study included one hundred adult menstruating female patients with age ranged between 18-49 years, complaining of hair loss from the scalp and fifty ages matched healthy female controls. Both groups were evaluated for serum zinc levels. There was strong statistically significant reduction (p= 0.01) in the level of serum zinc in patients compared to that in control group. This indicating that there is a definite association between decreased serum zinc levels and CTE in adults menstruating females.

تقيق مستوى الخاصين في مصل دم النساء البالغات اللاتي يعانين من تساقط الشعر التيولوجي
المزمن في مدينة كركوك.

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فرع الأمراض الجلدية والتناسلية/كلية الطب/جامعة تكريت.
فرع الكيمياء الحياتية الطبية/كلية الطب/جامعة تكريت.

مفتاح البحث: زمن، التيولوجين المزمن، كريبتولوجين

الخلاصة:
عرف تساقط الشعر التيولوجي المزمن على أنه مرض مجهول السبب الرئيسي في سنة 1996. حيث أن النساء المصابات بهذا المرض يعاني من تساقط مقاس من شعر الرأس سواء كان بسبب مشروط أو مجهول وتشتهر الحالات لدة تتجاوز السنة أشهر. لذلك أجريت هذه الدراسة لتققيم العلاقة بين تساقط شعر التيولوجي المزمن ونقصان الخاصين في النساء البالغات. تضمنت الدراسة إدراج حالة من حالة من النساء البالغات اللاتي يعانين من تساقط الشعر المزمن. وتراحت أعمارهن بين 18-49 سنة. تم مقارنهم بـ 50 من النساء الأسحاء.
Introduction:
Telogeneffluvium is a form of non-scarring diffuse scalp hair loss; it results from the synchronous transition of hair follicles from the growth stage (anagen) of the hair cycle to the resting stage (telogen) of the hair cycle. Shedding does not occur, however, until the new anagen hair begins to grow 3 to 4 months after the inciting event.  

Telogen effluvium can be divided into:
1- Acute telogen effluvium which lasts less than 6 months.
2- Chronic telogen effluvium which lasts longer than 6 months.

Chronic telogen effluvium, in some women is an early feature of androgenic alopecia. Its onset is often insidious and it can be difficult to identify a triggering factor. Nutritional deficiencies can contribute to increased hair shedding by weakening hair shafts that cause breakage to the hair and slow regrowth, hair loss that are caused by nutritional deficiencies can be corrected by a proper nutrition. Trace elements have a much important role in the growth and health of hair such as zinc.

Zinc is a trace mineral with widespread roles to sustain human health. It is a component of many enzymes as a cofactor and is involved in cellular division (DNA and protein synthesis) that promotes cell reproduction and tissue growth and repair. It also acts in the maintenance of the oil-secreting glands attached to hair follicles. The total daily requirement of zinc is about 15 milligrams for the average adults. Zinc is mainly excreted in the stool and in a lesser degree in the urine, skin, seminal fluid, menstrual blood, hair and nail. Serum zinc is a good determinant for predicting the zinc level of the body; it ranges from (70-140 µg/dl) in adults.

Zinc deficiency may cause hair loss. Harrison and Sinclair found that zinc deficiency both hereditary and acquired, leads to sparse, dry, and brittle hair. Garcia et al. showed that the combined use of a shampoo and hair lotion, formulated with vitamins and zinc, shortens the course of acute telogen effluvium. Therefore this study is conducted to detect the association of serum zinc with CTE.

Patients and methods:
One handed and fifty adult menstruating females attending the outpatient clinic of dermatology of Azadi teaching hospital (ATH) in Kirkuk city were included in a case
control hospital based study during the period from 25th October of 2011 till the end of March of 2012.

Their age ranged from 18-49 years. They were classified into two groups:

1- The case group included 100 females complaining of hair loss. They were diagnosed as CTE by history and clinical examination (Hair pull test: It done by tightly grasping 50-60 hairs firmly between thumb and forefinger followed by a pull of sufficient intensity to cause slight discomfort, there should be fewer than six normal club hairs. Several pulls may be required to test different areas of the scalp). Which is done by supervision of a dermatologist in ATH (figure 1).

2- The control group: Fifty healthy volunteers were investigating to serve as a control group. None of them had clinical or laboratory evidence of a disease that would affect the parameters to be measured.

**Inclusion criteria:**
All patients fitted in the definition of chronic telogen effluvium, included subjects with diffuse hair loss of at least 6 months; the diagnosis was done based on increase hair shedding by medical history and physical examination, and confirmed by a positive hair pull test.

**Exclusion criteria:**
Women were not included if they had a history of hair loss less than 6 months duration, surgical operation, lactation, chronic systemic disease or used medication that may be associated with hair loss in the last 6 months, and abnormal laboratory studies (except serum zinc).
Laboratory investigation

Included serum zinc level measurement which was carried out in all cases and controls, whereas the following tests were performed whenever necessary.

1- Complete blood count by automatic hematology analyzer.
2- Iron studies (iron, ferritin, TIBC, transferrin saturation) manually by colorimetric method.
3- Thyroid function tests by minividas kit using minividas instrument.
4- Renal function tests manually by colorimetric method.
5- Liver function tests manually by colorimetric method.

Methods:

Serum zinc was measured by colorimetric method on a spectrophotometer, using S.zinc kit manufactured by Randox laboratories, United Kingdom. As shown in table 1 and 2.

The Principle of the Method

Zinc present in the sample is chelated by 5-Br-PAPS, (5-bromo-2-pyridylazo)-5-(N-propyl-N-sulfopropylamino)-phenol in the reagent. The formation of this complex is measured at a wavelength of 560nm.

PROCEDURE 1: DEPROTEINIZATION

Table (1): DEPROTEINIZATION.

Pipette into test tube:

<table>
<thead>
<tr>
<th></th>
<th>Blank</th>
<th>Standard</th>
<th>Test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>H2O</td>
<td>STD</td>
<td>Sample</td>
</tr>
<tr>
<td>Test Specimen</td>
<td>0.5(0.2) ml</td>
<td>0.5(0.2) ml</td>
<td>0.5(0.2) ml</td>
</tr>
<tr>
<td>Deproteinizing Reagent (R1)</td>
<td>0.5(0.2) ml</td>
<td>0.5(0.2) ml</td>
<td>0.5(0.2) ml</td>
</tr>
</tbody>
</table>

Mix, well then centrifuge for 10 mins at 10,000 g. Use supernatant in the zinc assay within 2 hours.
Table (2) Zinc assay.

**PROCEDURE 2: ZINC ASSAY**

<table>
<thead>
<tr>
<th>Wavelength:</th>
<th>560 nm (550 - 570 nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Incubation Temperature:</td>
<td>20/25°C</td>
</tr>
<tr>
<td>Cuvette:</td>
<td>1 cm light path</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>Blank</th>
<th>Standard</th>
<th>Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>H2O</td>
<td></td>
<td>STD</td>
<td>Sample</td>
</tr>
<tr>
<td>Supernatant</td>
<td>0.5(0.2) ml</td>
<td>0.5(0.2) ml</td>
<td>0.5(0.2) ml</td>
</tr>
<tr>
<td>Working Reagent (R2)</td>
<td>2.5(1.0) ml</td>
<td>2.5(1.0) ml</td>
<td>2.5(1.0) ml</td>
</tr>
</tbody>
</table>

Mix, incubate for 5 min at 25°C. Measure the absorbance of the standard (A standard) and the sample (A sample) against the reagent blank within 60 minutes.

**Results:**

The mean age of both groups who included in the study was 30.3 years. Statistically there was no difference in mean age between both case and control groups (30.3±8.5) vs. (30.4 ± 7.0), respectively; p value=0.9 (statistically non-significant). as shown in table (3).

<table>
<thead>
<tr>
<th>No. of cases</th>
<th>Min</th>
<th>Max</th>
<th>Mean</th>
<th>S.D</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>100</td>
<td>18</td>
<td>49</td>
<td>30.3</td>
</tr>
<tr>
<td>Controls</td>
<td>50</td>
<td>19</td>
<td>47</td>
<td>30.4</td>
</tr>
</tbody>
</table>

Student t test showed that the mean serum zinc levels to be statistically significantly low in patients with CTE hair loss than in those (control group) normal subjects without CTE. The mean of serum zinc levels in CTE cases and controls were (9.9±7 and 13.3±8.5 µmol/l, respectively). This difference in mean of serum zinc levels found to be highly significant (p value=0.01) as shown in table (4).
Table (4): Comparison of Serum Zinc Level between Cases and Controls.

<table>
<thead>
<tr>
<th>S.Zinc µmol/l</th>
<th>Cases</th>
<th>Controls</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>9.9± 7.0</td>
<td>13.3± 8.5</td>
<td>0.01</td>
<td></td>
</tr>
</tbody>
</table>

Dietary status have a role in serum zinc level, in this study it was appeared that the effect of the type of diet on serum zinc level was statistically significant (p<0.05), as shown in table (5).

Table (5): The effect of the type of diet on serum zinc level.

<table>
<thead>
<tr>
<th>of diet</th>
<th>nts</th>
<th>rol group</th>
<th>ue</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mixed diet</td>
<td>10.7 ± 2.0</td>
<td>12.41 ± 1.5</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Vegetarian</td>
<td>9.5 ± 0.5</td>
<td>11.62 ± 9.8</td>
<td>0.01</td>
</tr>
<tr>
<td>Crash</td>
<td>11.5 ± 2.0</td>
<td>14.8 ± 2.0</td>
<td>0.07</td>
</tr>
</tbody>
</table>

According to menstrual duration the patients divided into periods that extend from 2-4 days, 5-7 days, and > 7 days. Only seven patients had low serum zinc level from eighteen patients who had menstruation period more than seven days and this was not significant (p>0.05). There was no significant relation with occupation, and marital status.

Discussion:

Telogen effluvium is an abnormality of hair cycling that result in excessive shedding of telogen hair; it is one of the most common causes of diffuse nonscarring hair loss. Many cases of telogen effluvium are subclinical, so the true incidence in the community is unknown. (3,12)

Frausto da Silva and Williams in 2001 stated that there were many trace elements in the human body that directly or indirectly participate in metabolism and may play key roles in its modulation. Some of these metals are calcium, copper, iron, magnesium, potassium, sodium, and zinc which have been identified as essential for human health. (13)

The present study demonstrated that serum zinc levels were significantly decreased in CTE patients compared to control group. This result is in agreement with that of Sinclair and Prasad. (8, 14)

On the other hand there were some studies that disagreed with this result, Rhushton (15) and Yacoub et al. (16), who found that there was no evidence to support low serum zinc concentration in telogen effluvium patients.
The most common causes of zinc deficiency are the inhibitory effect of dietary fibers and phytates on zinc absorption (only 20% of the dietary zinc is absorbed), so its deficiencies is more common in regions with high consumption of rice and unleavened bread.\(^{(4,5)}\)

Some studies believed that long menstruation periods had effect in low serum zinc levels.\(^{(17)}\)

It has been found in this study that only seven patients had low serum zinc level from eighteen patients who had menstruation period was more than seven days and this was not significant. This agreed with Haghollahi et al.\(^{(5)}\) Moreover, this study showed that dietary status played an important role in serum zinc level, vegetarian patients had low serum zinc level which is statistically significant p value 0.01..This finding agreed with Haghollahi et al.\(^{(5)}\) Yokoi K, et al.\(^{(18)}\)

The significant difference in the level of serum zinc in both groups might also have caused a distortion in the role of zinc in gene expression as well as in stabilizing the structure of proteins and nucleic acids that is necessary for the proper functioning of all cells, including the hair follicles. Specifically it was highlighted that the gene SOX21 appears to be responsible for hair loss in human subjects, although a link has not been identified between zinc and this gene.\(^{(19)}\)

**Conclusion:**

It was concluded that the low serum zinc is prevalent in patients with chronic telogen effluvium.

Appreciating the importance of zinc as a factor in hair loss may be important both in designing new therapies and in generating hypotheses to better elucidate the biochemical underpinning of these disorders.

**References:**


