Effect of Aqueous Extract of *Vitis Vinifera* Leaf on Some Immnoglobulin in Levothyroxine Sodium Induced Hyper Thyroidism Rabbit Females

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Abstract
The Aqueous Extract of *Vitis vinifera* leaves was investigated for its effect on some immunoglobulin in induced hyperthyroidism rabbits. The aqueous extract of leaves at does level of 100 mg ml showed significant increase (p<0.05) on level of IgG and significant decrease (p < 0.05) on level of IgA , IgM to normal level .

Keywords : vitis vinifera , L- thyroxin , IgG , IgA, IgM .

INTRODUCTION
*Vitis vinifera* L.(grape) is native to the Mediterranean region , central Europe and south western Asia and cultivated widespread in Europe , Asia and America . The plant is alina with flaky bark and the leaves are alternate palmately and board . The fruit isaberry and can be green red or purple [1]. Grape leaves with antioxidant activity [2] have been reported to treat chronic venous insufficiency in human [3] and nephrotoxicosis induced by citrinin [4] .
It has also been demonstrated that the grape leaf hydro-alcoholic extract induces spasmolytic effect on rat uterus precontracted by oxytocin [5].

Effect of specific medicinal herbs on Immune system and Immune cells :
Systemic studies on the effect of specific medicinal herbs on immune system are designed to obtain evidence – based scientific knowledge on the appropriate use of traditional medicinal herbs . The development of immunology has resulted in further complexity by combining external (environment and pathogens ) and internal (neroendocrine – immune system) factors in the pathogenesis of infectious diseases .
The most important thing is to learn how to modulate the immuneresponse to external conditions with powerful new techniques and drugs [6-8] .

Traditional herbal medicine provides several remedies for strengthening the body s resistance toilness through effects on immune system components . such as dedritic cells , T cell . macrophages , etc [9].
Inflammation is the body's protetive reaction to controlling infections and promoting tissue repair however , uncontrolled and excessive inflammation results in tissue damage and diseases including rheumatoid arthritis , inflammatory bowel diseases psoriasis , cancer etc .

Recently many laboratories have focued on the indentificantion of immunomodulatory phytocompounds from herbal medicine that are reported to modulate immunity .

Several biochemical, cellular immunological, and molecular biological techniques and mouse model have been used to investigate the immunomodulatory function of phcto compound in regul immunity and in modulating human cells including T cells , macrophages and dendritic cell functions .

Several plant compounds are know to be able to bind T cell components and to regulate T cell function . the indentification of genes involved in T cell function is also very important . In T cell differentiation several genes play very important . These genes products can be important for screening the phytocompounds to which the gene products can bind . Also an understanding of important signaling molecules in T cells helps us to screen their intraction partners from plants [10-14].
The aim of the present study was to investigate the effect of vitis vinifera leaves extract on some immunoglobulin in hyperthyroidism rabbits .

MATERIAL AND METHODS
The fresh and healthy leaves of grape (vitis vinifera) were collected in April 2012 from Iraqi farms in Baghdad . the leaves were dried and powdered . The powered mixed with distilled water (25g:250mL) and were incubated for 3 hrs at (60)℃ than incubated overnight at room temperature .
Suspension was than filtered . water extracts were prepared daily just before administration orally to the experimental in does of (5 mL/Rabbit of 1.5 – 2.5 kg)
Preparation of thyroxin : fresh solution of levothyroxine sodium was prepared (tablet dissolve in water ) just before feeding . for the animal given the thyroxin (50µg/kg body weight).Levothyroxine sodium was purchased from Al-Sophee Pharma, Al-Adamia, Baghdad, Iraq.experimental
animal = Twenty female ORYCIOLAGUS CUNTCULUS (1.5 – 2.5 kg each) were kindly supplied by city of 
medicine (in Baghdad) for the period from September 2012 to May 2013. And were used in this research 
rabbits were maintained with free access to water and diet (containing multivitamins protein, vegetable, bread). 
Experimental animals were divided into two groups (10 rabbits each).
1. control group: rabbits were orally administered (using a feeding solution) with a daily dose of 5 ml distilled 
   water for 2 months.
2. plant-treated levothyroxine sodium group: 50 µg of L-thyroxin was orally administered daily to each rabbit 
   for one month, then 5 mL of the plant extract (100 mg/mL) was orally administered daily to each 
   rabbit in this group for two months.

Blood sampling: Blood samples were collected from the heart of rabbits using heparinized capillary tubes. Serum 
was separated from the blood sample, then frozen until used.
The levels of immunoglobulin (IgG, IgA, IgM) in serum were measured by ELSA (Human Germany) using 
special kits for each globulin provided from Monobind Inc [15-16].

Preliminary phytochemical screening: The tests were done to check the presence of the active chemical 
constituents such as alkaloids, phenolic content, carbohydrate, reducing sugar, amino acid, protein, and tannin 
by the following procedures:
1. Test for alkaloids [17].
2. Test for phenolic content [18].
3. Test for carbohydrates [19].
4. Test for reducing sugar [20].
5. Test for tannins [21].
6. Test for amino acid and protein [22].

Statistical analysis: All statistical analysis of the study were done using SPSS version 15.0 for windows (Statistical 
package for social science, Inc, Chicago, IL, USA).

Descriptive analysis used to show the mean ± standard deviation of variable. The significance of difference 
between mean values was estimated by student T-test. The probability p < 0.05 = significant.

RESULTS AND DISCUSSION
The daily orally treatment with doses 50 µg/kg resulted in the development of l-thyroxin after two weeks of 
administration, the effect gradually increasing over for one month period to reach airs of about once and half 
times over initial values compared with healthy rabbits.
The aqueous extract of vitis vinifera leaves was administered orally 100 mg/mL to each hyperthyroid rabbit to 
assess the affect of the plant extract.
Table (1) showed results of phytochemical screening of aqueous of vitis vinifera leaves.

| Table (1): Result of phytochemical screening of aqueous extract of plant. |
|---------------------------------|-------------------|
| Pytochemical class              | Aqueous extract   |
| Alkaloids                       | +                 |
| Phenolic content                | +                 |
| Carbohydrates                   | +                 |
| Reducing sugar                  | +                 |
| Amino acid + Proteins           | +                 |
| Tannins                         | +                 |
Table(2) shows the immunoglobulin level in the two studied group

Table(2): Illustrate values of IgG, IgA, IgM level in blood rabbit females.

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>L.Thyroxine</th>
<th>L.Thyroxine</th>
<th>L.Thyroxine</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
<td>C'</td>
<td>+ leaf extract 1 month</td>
<td>+ leaf extract 2 month</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mean±S.D</td>
<td>Mean±S.D</td>
<td>Mean±S.D</td>
<td>Mean±S.D</td>
<td>C₁ vs. C'</td>
</tr>
<tr>
<td>IgG mg/dl</td>
<td>2100±50</td>
<td>647±4</td>
<td>2465±10</td>
<td>2566±10</td>
<td>0.05</td>
</tr>
<tr>
<td>IgM mg/dl</td>
<td>21.2±0.5</td>
<td>22.5±0.5</td>
<td>21.0±0.3</td>
<td>21.5±0.3</td>
<td>0.05</td>
</tr>
<tr>
<td>IgA mg/dl</td>
<td>43.3±0.03</td>
<td>44.3±0.2</td>
<td>43.0±0.3</td>
<td>43.5±0.2</td>
<td>0.05</td>
</tr>
</tbody>
</table>

C= control , c*= administrate with L-thyroxin

There was significant decrease in the level of IgG in hyperthyroid rabbits serum when compared with the control group the result agreed with [23] but disagree with [24], while the level of IgM and IgA in rabbit's serum increase when it compared with control group this results agreed with [24].

The intake of vitis vinifera leaves extract (100Mg/ mL) for two month caused significant increase in IgG while reduction IgM and IgA levels to control. NO available literature could be traced concerning the effect of vitis vinifera leaves extract in rabbits on immunoglobulin (IgG, IgA, IgM)

CONCLUSION

It is inferred from the present study that vitis vinifera leaves extract elicited mild and moderate alterations on IgM, IgA and had stimulating effect on IgG.

REFERENCES