Assessment of Antioxidant Changes of Aged Rats Treated With Sumac Extract

M. M. Abbass 1, A. H. Mahmoud 1, Mohamed M. A. Hussein 2 and Salah A. Gabr 1

Biological Applications Dept., Isotopes Applications Division, Nuclear Research Center, Atomic Energy Authority 1, Biochemistry Dept., Faculty of Veterinary Medicine, Zagazig University 2

manalmounir71@yahoo.com

Abstract: It is increasingly being realized that many of today's diseases are due to the "oxidative stress" that results from an imbalance between formation and neutralization of free radicals. Many synthetic drugs are said to protect from oxidative damage but they have adverse side effects. An alternative solution to the problem is to consume natural antioxidants from food supplements and traditional medicine. Sumac extract is a widely used herbal plant that affects the biological activities. The present study has focused on the potential role of sumac extracts on age-related changes in senile rats. Thirty male albino rats were used for this purpose. They were divided into three groups (10 animals in each), 1st group (6 months old) served as adult or control group, 2nd group (24 months old) as senile group, 3rd group (22 months old as senile rats received 200 mg / kg b.w. of sumac extract orally three times a week for two months) as treated group. Blood glutathione (GSH), catalase (CAT), malondialdehyde (MDA), hematomal parameters (Hb, RBCs, WBCs and hematocrit value), some liver enzymes (AST and ALT), total protein, albumin, globulins and hormones (testosterone, triiodothyronine (T3) and thyroxine (T4)) were measured. The sumac extract induced a significant increase in GSH concentration and CAT activity and decrease in the concentration of MDA of senile rats. The plant extract, raised the Hb concentration, RBCs count and hematocrit value. Also, the level of total protein and albumin in treated rats as compared with senile rats was significantly elevated. However, it made no significant change in ALT, AST, T3 and T4. Meanwhile, testosterone level increased significantly in treated rats in comparison with senile rats. These findings demonstrated that sumac extract had a positive effect on antioxidant status through minimize free radical-induced damage which is a main cause of aging.

Keywords: Sumac extract, antioxidant status, hematological parameters, senile rats.

1. Introduction

Medicinal plants can be promising sources of natural products with beneficial activity (Mothana et al., 2009; Krolickewska et al., 2011). Sumac (Rhus coriaria L., family Anacardiaceae) is the common name for genus Rhus that contains over 250 individual species (Rayne and Mazza, 2007). It is widely used as a spice (Chakraborty et al., 2009), also as a herbal remedy in traditional medicine owing to its antifibrogenic, antifungal, anti-inflammatory, antimalarial, antimicrobial, antimutagenic, antioxidant, antithrombin, antitumorogenic, antiviral, cytotoxic hypoglycaemic, leukopaenic and atheroprotective effects (Rayne and Mazza, 2007). Sumac is known to be a rich source of tannins (Zargham and Zargham, 2008). In vitro and in vivo studies have shown that tannins have anticarcinogenic effect (Ram et al., 1997).

Sumac extract was reported to be a source of natural antioxidants (Ozcan, 2003). The methanolic extracts of Rhus showed a remarkable radical scavenging effect even at low concentrations (Mothana et al., 2009). A crude extract of R. coriaria exhibits interesting antioxidant properties, expressed by the capacity to either scavenge superoxide radical or uncompetitively inhibit xanthine oxidase (candan, 2003). One of the main constituents of sumac is gallic acid (Rayne and Mazza, 2007), which possesses potent antioxidant properties (Rozek et al., 2007).

Aging process is accompanied by progressing modifications of most physiological functions and the cells exhibit lower abilities to react with exogenic factors and to stress formation (Jurivich et al., 1997). Numerous studies have reported correlations between age and the accumulation of oxidative damage in cellular macromolecules and this damage could result from a multitude of factors including a decrease in antioxidant defenses (Wolford et al., 1987; stadman, 2001; Posadas et al., 2009).

Physiological aging affects all normal cells, tissues and organ systems as hematological parameters are highly differentiated (Videan et al., 2008). Fibrosis of various organs, including heart, Kidney and Liver is a hallmark of aging (Sum et al., 1998). Moreover, the function of many endocrine glands alters by aging (Correa et al., 2001). In relation to these findings an extensive range of antioxidants, whether synthetic or natural have been presented for the treatment or prophylaxis of disorders attributed to free radical oxidative damages (Maxwell, 1995; Halliwell, 2000).
Restriction on the use of synthetic antioxidants due to their probable side effects has increased the contribution of natural antioxidants (Veliglu et al., 1998). Based on this knowledge, the aim of the present work is to determine the effect of sumac extract (Rhus coriaria) on biochemical and hematological parameters of aged rats.

2. Material and Methods

Sumac extract via supercritical fluid extraction

Supercritical fluid extraction was performed at central laboratories for Radioisotopes Applications Division, Nuclear Research center, Egyptian Atomic Energy Authority on an ISCO supercritical fluid extractor, model SFX 220 equipped with two ISCO 260 D syringe pumps (lincoln, NE, USA). The pressure and temperature were 5000 psi and 80°C respectively. Every 2 gm of plant material was loaded into an extraction cell. The remaining volume was filled with glass wool. The flow rate through the extraction vessel was kept at 1.0 ml/ min during the extraction time (30 minutes). All sumac extracts was collected in 20 ml vials containing 5 ml methanol (Fadel et al., 1999).

Experimental design:

Thirty male (160-220gm) albino rats (bred under specific pathogen- free conditions at the animal Farm of Faculty of Veterinary Medicine, Zagazig University, Egypt) with light/dark cycle:12hrs and transferred to the Animal House of the Nuclear Research Center in Anshas Area. They were kept in metal cages during the whole experiment period under hygienic condition, fed on balanced ration and provided with water ad-libitum through the experiment.

The rats were divided into three equal groups of 10 each; 1st group (6 months old rats) as adult or control group, 2nd group (24 months old rats) as senile group and 3rd group (22 months old) as senescent rats are received 200 mg/kg b. w. of sumac extract according to Mohammadi et al. (2010) three times a week for 2 months through oral gavage, served as treated group.

At the end of the experimental period, the blood was collected and divided into two parts. The first, on EDTA anticoagulant while the second without anticoagulant to separate serum and centrifuged at 4000rpm and kept at -20°C till used for biochemical analysis.

Hematological parameters; Hemoglobin content (Hb), erythrocytic count (RBCs), leucocytic count (WBCs) and hematocrit were determined using Sysmex (KX-21)–cell counter, with a Kit manufactured by (Diamond, Philadelphia, USA). On the other hand, (MDA) was measured spectrophotometrically by the method described by Draper and Hadley (1980). CAT activity and GSH concentration were determined according to Aebi (1984) and Beutler et al. (1963) respectively.

The serum was used for biochemical analysis, the concentration of aspartate aminotransferase (AST), alanine aminotransferase (ALT) were measured according to Reitman and Frankel (1957), total protein, albumin were determined according to Doumas et al. (1981) and Druptr (1974) respectively while plasma globulins were calculated by subtracting the albumin obtained from total protein. All of these parameters were performed on the Hitachi 902 biochemical analyzer.

Testosterone, triiodothyronine (T₃) and thyroxine (T₄) was estimated by radioimmunoassay techniques (RIA) using commercial kits purchased from Siemens Medical Solutions Diagnostics, Los Anglos, USA.

Statistical analysis:

Data were expressed as mean ± standard error of values. Comparison across groups were performed by Anova (one-way classification F- test), following by Duncan's multiple range test according to Lind and Masson (1996). Statistical analyses of data were done using the spss (Statistical package for social sciences) for windows.

3. Results:

The results are presented in table (1) showed significant decrease (p ≤ 0.05) in GSH concentration and CAT activity in senile rats (2nd group) as compared with adult rats (1st control group). Meanwhile, MDA was significantly increased in senile rats.

Treating the senile rats with sumac extract for 2 months lessened the alteration happened in the antioxidant status of aging rats. The results were optimistic since it showed a significant increase (p ≤ 0.05) in GSH concentration and CAT activity and decline in the concentration of MDA in treated group (3rd group) as compared with senile rats (2nd group).

The hematomal parameters of the experimental groups were studied and represented in table (2). In the senile rats (2nd group), hemoglobin content, RBCs count and hematocrit were significantly decreased (p ≤ 0.05) in comparison with adult rats (1st control group). After oral administration of sumac extract to senile rats (treated or 3rd group), Hb concentration, RBCs count and hematocrit were significantly increased (p ≤ 0.05) as compared with senile rats (2nd group).

Biochemical parameters of rats are presented in Table (3). Results showed no changes in AST and ALT activities between senile rats (2nd group) and adult rats (1st control group). Meanwhile, Total protein, Albumin and globulins decreased in senile rats.

After treatment with sumac, statistical evaluation showed significant increase (p ≤ 0.05) in total protein, albumin (3rd group) and restored nearly to their levels in adult rats (1st control group).

http://www.americanscience.org
The pessimistic effect of aging was extended to some endocrine gland functions represented here in T3, T4 and testosterone hormones that demonstrated a significant decrease ($p \leq 0.05$) in senile rats (2nd group) as compared with adult rats (1st or control group) as shown in table (4).

Sumac administration had slight or no effect on T3 and T4 levels. Meanwhile, testosterone level increased significantly ($p \leq 0.05$) in treated rats (3rd group) in comparison with senile rats (2nd group).

Table (1). Blood glutathione (GSH) concentration, catalase activity (CAT) and malondialdehyde (MDA) level in experimental groups (means±SE).

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>GSH (mg/dl)</td>
<td>74.3±7.2a</td>
<td>52.3±6.4b</td>
<td>72.8±6.7a</td>
</tr>
<tr>
<td>CAT (U/ml)</td>
<td>30.7±2.8a</td>
<td>20.2±3.1ab</td>
<td>29.5±2.6a</td>
</tr>
<tr>
<td>MDA (µmol/L)</td>
<td>67.5±7.3a</td>
<td>95.7±8.5a</td>
<td>79.4±7.6a</td>
</tr>
</tbody>
</table>

Means within the same row have different superscripts are significantly different at ($p \leq 0.05$).

Table (2). Hematological parameters in experimental groups (means±SE).

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb (g/dl)</td>
<td>12.9±0.6a</td>
<td>10.3±0.4a</td>
<td>11.8±0.5a</td>
</tr>
<tr>
<td>RBCs (N x 10^6/µl)</td>
<td>7.20±0.3a</td>
<td>5.5±0.2b</td>
<td>6.7±0.2a</td>
</tr>
<tr>
<td>WBCs (N x 10^6/µl)</td>
<td>9.8±0.4a</td>
<td>9.5±0.3a</td>
<td>9.7±0.4a</td>
</tr>
<tr>
<td>Hematocrit (%)</td>
<td>37.6±0.3a</td>
<td>33.4±0.4a</td>
<td>34.8±0.3a</td>
</tr>
</tbody>
</table>

Means within the same row have different superscripts are significantly different at ($p \leq 0.05$).

Table (3). Some Liver enzymes (AST, ALT), total protein, albumin and globulins in experimental groups (means±SE).

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>AST (U/L)</td>
<td>194.3±13.5a</td>
<td>210.6±12.8a</td>
<td>202.5±13.6a</td>
</tr>
<tr>
<td>ALT (U/L)</td>
<td>79.2±6.7a</td>
<td>85.1±5.9a</td>
<td>71.5±5.3a</td>
</tr>
<tr>
<td>Total protein (g/dl)</td>
<td>6.9±0.32a</td>
<td>5.8±2.4b</td>
<td>6.4±0.41a</td>
</tr>
<tr>
<td>Albumin (g/dl)</td>
<td>4.1±0.10a</td>
<td>3.3±0.20b</td>
<td>3.9±0.10a</td>
</tr>
<tr>
<td>Globulins (g/dl)</td>
<td>2.8±0.09a</td>
<td>2.5±0.04b</td>
<td>2.5±0.06b</td>
</tr>
</tbody>
</table>

Means within the same row have different superscripts are significantly different at ($p \leq 0.05$).

4. Discussion

A central tent of green chemistry is the ability to obtain a commercially viable product with desirable properties from a widely renewable feed stock (Rayne and Mazza, 2007). In particular there is significant interest in obtaining extracts with particular biological activities from plants.

Sumac is a well known, popular spice and has been utilized extensively for medicinal and other purposes. Phytochemical studies of this plant reported that its leaves contained phenolic acids (Mavlyanov et al., 1995), anthocyanins, hydrolysable tannis and gallic acid, condensed tannis and several flavonoids (Mavlyanov et al., 1995; Zalacain et al., 2003). These substances have gained interest because may reduce the risk of chronic diseases, reinforcing the defences against free radical species.

Aging is characterized by declining ability of the individual to adapt environmental stress. It is a physiological process involving many biological systems.

The role of antioxidant in aging is of critical importance as normal aging is accompanied by a decline in antioxidant defenses reflected in GSH in blood and different tissues of animals and humans (Muthuswamy et al., 2006). In this study, these findings were confirmed since GSH level and catalase activity were significantly decreased in senile rats (2nd group) as compared to their levels in adult rats (1st group or control) Table (1). This decrease may be due to oxidation of sulfhydryl group and diminished level of GSH (Sarker et al., 1998). Moreover, there is a gradual increase in the level of MDA in senile rats as compared with control group. This increase may be also referred to the failure of antioxidant system to combat oxygen radicals and/or the positive relation between age and oxidative stress. As the increase in age related oxidative damage could result from a multitude of factors including imbalance between antioxidant defenses and oxidative stress (Posadas et al., 2009). Based on the experimental data in this study, the oral administration of sumac extract for two months resulted in a significant increase in the level of...
GSH and CAT activity while MDA level was decreased in treated group as compared with senile rats. This result may be due to the presence of various substances mainly tannin and gallic acids that may be responsible for the antioxidant ability of this plant. It was reported that the antioxidant activity of gallotannins from R. coriaria extract was stronger than that of gallic acid (Beretta et al., 2009). Tannic acid extracted from Rhus chinensis Mill could effectively scavenge the O\(^2\) (Fu, 1991). Glycoprotein isolated from R. verniciflua Stokes fruit improved the antioxidant levels in mice (Oh et al., 2006).

The values of hematological parameters are affected by a number of factors even in apparently healthy populations. These factors include age, sex, nutritional and environmental factors. The present study showed that, the senile rats (2\(^{nd}\) group) had hematological disorders manifested by drop in hemoglobin content, RBCs count and hematocrit value as compared with their values in control group (1\(^{st}\) group). This is consistent with data from Videan et al. (2008) who found a significant age related increase in anemia risk, based on significant decreases in hemoglobin and hematocrit. Also, Wolford et al. (1987) reported that hematological changes with aging. These hematological changes are reflected by a decline in marrow cellularity, increased risk of myeloproliferative diseases and a decline in adaptive immunity (Prabhakar, 2009).

Our results reported that the sumac extract induced an increase in Hb content, RBCs and hematocrit of treated group as compared with the senile rats (Table 2).

The present study demonstrated insignificant increase in some liver enzyme activities (AST, ALT) in senile rats as compared with control (Table 3). Meanwhile, there are decreases in their values after oral administration of sumac extract but they didn’t reach to the normal range.

Albumins represent a very abundant and important circulating antioxidant (Roche et al., 2008). Study of Bourdon et al. (1999) confirmed and extended the idea that serum albumin is an important protein that presents direct protective effects.

Total protein, albumin and globulin levels are performed to evaluate the toxicological nature of various chemicals. Data from the present study, showed a significant decrease in the level of albumin, globulins and total protein in serum of aged rats (2\(^{nd}\) or senile group) as compared with adult rats (1\(^{st}\) or control group) as a result of oxidative stress associated with aging which reduced liver functions and decreased adaptive immunity (Prabhakar, 2009). Several lines of evidence strongly suggest that a reduced serum albumin concentration, although within the normal range, is associated with mortality risk (Bourdon et al., 1999).

Administrating aged rats with sumac extracts caused a significant increase in total protein and albumin levels compared with senile rats. (Table 3). Aging is indication of compromised liver excretory function and impairment of the liver synthetic function, which improved by treating with sumac extracts causing improvement in total protein, albumin and globulin levels.

According to the accumulated data, during aging thyroid function decreases progressively and it is well recognized that the symptoms of aging can easily be confused with those of hypothyroidism (Silvestri et al., 2008). This data matched with our results which found a decrease in the concentrations of T3 and T4 hormones in senile rats as compared with control group (Table 4). In addition, a reduction of testosterone level was observed in senile rats. This is consistent with (Vermeulen and Kaufman 1995; Zirkin and Chen, 2000) who also reported similar finding. This could be due to the changes in the antioxidant environment of aging cells that have been shown to be correlated with the reduced ability of these cells to produce testosterone (Chen et al., 2007). It was also observed an increasing in the level of testosterone of senile rats after their treatment with sumac extract.

The improvement recorded in the studied parameters after oral administration of sumac extract could be attributed to the presence of a relatively high percentage of phenolic compounds with high antioxidant activity (Nokatani, 2000). Epidemiological studies have strongly suggested that consumption of certain plant materials may reduce the risk of chronic diseases related to oxidative stress on account of their antioxidant activity and promote general health benefit (Halliwell, 1997).

5. Conclusion

In conclusion, the importance of this study is to give new information on the application of sumac extract in reducing physiological changes related to aging. Our data show a positive effect of sumac extract on antioxidant status. These findings suggest that sumac extract, reduce the risk of increasing anemia in aged rats and protect the liver from damage associated with aging. However, further studies are required to better understand the full potential of the Rhus genus as part of global green technology based on bioproducts and bioprocesses research program.

Corresponding author

M. M. Abbass

Biological Applications Dept., Isotopes Applications Division, Nuclear Research Center, Atomic Energy Authority

manalmounir71@yahoo.com
References
plant glycoprotein (36kDa) from Rhus verniciflua stokes fruit in Triton WR-1339- induced hyperlipidemic mice. Bioscience, Biotechnology and Biochemistry, 70: 447–456.


