THE ANTIOXIDANT EFFECT OF *ROSA RUGOSA*

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SUMMARY

We examined the effects of *Rosa rugosa* (*Rosa rugosa* Thunb) on lipid peroxidation, alanine transaminase (ALT), aspartate transaminase (AST), glutathione and protein oxidation levels in carbon tetrachloride (CCl_4) treated male Wistar rats. Two control groups and one treatment group of rats were formed. The control groups were fed a standard diet, while the *Rosa rugosa* group was fed a standard diet enriched with 6% by weight dried *Rosa rugosa* fruit powder. After 3 months, a single dose of CCl_4 was injected intraperitoneally in the Control II and *Rosa rugosa* groups (1 ml/kg, as 20% in olive oil) and a similar dose of olive oil was administered i.p. to rats in the Control I group. The rats were sacrificed 2 hours later. Lipid peroxide levels in liver, protein oxidation in liver, glutathione (GSH) levels in liver, and ALT and AST in plasma were measured. The rats in the *Rosa rugosa* group were found to have significantly lower liver peroxide, protein oxidation, glutathione levels and plasma ALT and AST activities compared with the rats in the CCl_4 treated control group. These findings suggest that *Rosa rugosa* possesses antioxidant activity.

KEY WORDS

Rosa rugosa, protein oxidation, ALT, AST, lipid peroxidation, glutathione

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INTRODUCTION

Members of the Rosaceae family have long been eaten because of their constituents. Rosehips are well known to have high vitamin C and flavonoid contents. These compounds are known to possess anti-oxidant, antimutagenic and anticarcinogenic effects. Rose fruits (rosehips), roots and leaves are used in medicine; the fruits are used in food as tea, marmalade, and syrup; and rthe oots are used as tea /1-3/. *Rosa rugosa* (*Rosa rugosa* Thunb) is a member of the Rosaceae family. There is little information about the antioxidant effect of *Rosa rugosa*. We could trace only a few studies on this topic. An *in vivo* study deals with the effect of rose flower extract on antioxidant enzymes and lipid peroxidation /4/. A second article reports the effect of *Rosa rugosa* on lipid peroxidation /5/. Another study was on the *in vitro* 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity of *Rosa rugosa* roots /6/. The aim of the present study was to determine the effects of *Rosa rugosa* fruits on lipid peroxide levels, alanine transaminase (ALT) and aspartate transaminase (AST) activities, protein oxidation and glutathione levels.

MATERIALS AND METHODS

All chemicals were purchased from Merck, Fluka Chemika and Sigma. In order to use the fruits *of Rosa rugosa* in this study, they were collected from Rize, Turkey. Male adult Wistar rats aged 24-25 weeks, weighing 250-300 g, were used in this study. They were accommodated at the Experimental Research and Animal Laboratory Unit, Faculty of Medicine, University of Marmara, Istanbul. Wistar rats were controlled under light conditions (12 hours of dark/light cycle).

Three groups of rats were used: Control I, Control II and *Rosa rugosa* group. Eight rats per group were used. Control I and Control II groups were fed a standard diet, and the *Rosa rugosa* group was fed the standard diet enriched by 6% *Rosa rugosa* powder for 3 months. Rats were fasted 18 hours prior to experiments. A single dose of carbon tetrachloride (CCl_4) was injected intraperitoneally in the Control II and *Rosa rugosa* groups (1 ml/kg, as 20% in olive oil) and a similar dose of olive oil was administered i.p. to the rats in the Control I group. Two hours later, the rats were killed humanely in accordance

with sanctions approved by the Institutional Animal Care and Use Committee (IACUC) appropriate to the species.

Livers of rats were quickly removed and washed in 0.9% NaCl. Liver parts were homogenized in ice-cold 0.15 M KCl (10% w/v) /7/. Plasma AST and ALT activities were measured by auto-analyzer. Lipid peroxide levels in liver were measured by the thiobarbituric acid (TBA) test /8/: 0.2 ml of 10% (w/v) tissue homogenate was added to 0.2 ml of 8.1% sodium dodecyl sulfate (SDS), 1.5 ml of 20% acetic acid solution adjusted to pH 3.5 with NaOH, and 1.5 ml of 0.8% aqueous solution of TBA. Distilled water was used to produce 4.0 ml of mixture. Subsequently, the mixture was heated in a water bath at 95°C for 60 minutes. Once cooled, 1.0 ml of distilled water and 5.0 ml of the mixture of *n*-butanol pyridine (15:1, v/v) were added. Following centrifugation at 4,000 rpm for 10 min, the upper phase was taken and its absorbance measured at 532 nm. 1,1,3,3-Tetraethoxy-propane (TEP) was used as an external standard. Liver glutathione levels were measured by the method of Ellman /9/: 0.5 ml of 10% (w/v) tissue homogenate was added to 1.5 ml of M KCl and 3.0 ml of the mixture of non-proteinization solution. After centrifugation, 0.5 ml of the upper phase was taken and 2 ml of 0.3 M Na_2HPO_4 and 0.5 ml of Ellman reagent were added. Its absorbance at 412 nm was measured. Glutathione (GSH) was used as an external standard. Protein oxidation levels in liver were determined by the methods of Levine *et al.* /10/ and Lowry *et al.* /11/.

Statistical analysis of plasma ALT and AST activities were performed by one way ANOVA and Duncan's test. Statistical analysis of liver lipid peroxide glutathione levels and protein oxidation levels were carried out by Kruskal Wallis one way ANOVA and Mann Whitney U-test.

RESULTS AND DISCUSSION

In the *Rosa rugosa* group, the liver lipid peroxide levels were reduced compared to the Control II group ($p < 0.005$). Moreover, *Rosa rugosa* lowered liver lipid peroxide levels to that of Control I levels (Table 1). Cho *et al.* /5,6/ recently demonstrated that certain polyphenols extracted from *Rosa rugosa* inhibit lipid peroxidation.

When the *Rosa rugosa* group was compared with the Control II group, ALT and AST activities were decreased in the *Rosa rugosa*

TABLE 1

Effect of *Rosa rugosa* on alanine transaminase (ALT) and aspartate transaminase (AST) activities, lipid peroxide, protein oxidation and glutathione levels in male Wistar rats treated with CCl_4

	Control I (n = 8)	**Control II** (n = 8)	***Rosa rugosa*** (n = 8)
Plasma ALT (U/l)	23.1 ± 5.9	44.7 ± 4.6*	30.6 ± 5.7*[a]
Plasma AST (U/l)	124.8 ± 9.6	162.2 ± 8.4*	136.1 ± 10.8*[a]
Liver lipid peroxide (nmol MDA/g wet wt)	121.8 ± 16.2	384.3 ± 70.1*	124.8 ± 26.9[a]
Liver protein carbonyl (nmol carbonyl/mg protein)	4.8 ± 0.6	11.0 ± 2.6*	8.4 + 1.7*[a]
Liver glutathione (μmol GSH/g wet wt)	5.1 ± 1.0	6.5 ± 1.0*	5.1 + 0.8[a]

Results are means ± SD
* $p < 0.05$ in comparison to Control I group.
[a] $p < 0.05$ in comparison to Control II group.

group ($p < 0.005$) (Table 1). These results suggest that *Rosa rugosa* attenuates hepatic injury.

In the *Rosa rugosa* group, the liver glutathione levels were significantly decreased compared to the Control II group ($p < 0.005$) (Table 1). *Rosa rugosa* decreased the liver glutathione levels previously increased by CCl_4 treatment to the Control I levels. For this reason, it is suggested that *Rosa rugosa* reduced the lipid peroxide levels in the liver, and because of this lowering effect the glutathione levels remained low as well. Relevant to this, our previous study revealed that black tea reduced lipid peroxide levels in the liver, and because of this the glutathione levels remained low /12/. *Rosa rugosa* reduced liver protein oxidation levels (Table 1). When the *Rosa rugosa* group was compared with the Control II group, protein oxidation levels were decreased in the *Rosa rugosa* group ($p < 0.005$). According to these results it is suggested that *Rosa rugosa* has antioxidant activity.

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