



## The effects of *Viscum album* ssp. *album* L. extract on biochemical indices in partially hepatectomized liver in rats

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### ABSTRACT

*Viscum* species have been widely used in herbal medicine, such as nervine, hypertensive, cardiac depressant, vasodilator, slowing and steadying of excessive heart rate, relaxant and diuretic. The genus *Viscum* L. (Loranthaceae) comprises semi-parasitic plants which grow on various host tree and shrubs. In Turkey, *Viscum* L. is represented by one species (*Viscum album* L.) and three subspecies; namely ssp. *album*, ssp. *abietis* (Wiesb.) Abromeit and ssp. *austriacum* (Wiesb.) Vollmann and is known with a common name as 'Ökse Otu'. In the present study, the effect of *Viscum album* ssp. *album* L. extract on partially hepatectomized rats was investigated with biochemical assay. The extracts of the plant at the concentrations of 1 and 1.5 ml/100 g body weight/day were administered orally to the two experimental groups including partially hepatectomized rats for 14 days. At the end of the experimental period, animals were sacrificed, blood was collected for the assessment of serum levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST) and blood urea nitrogen (BUN). *Viscum album* ssp. *album* extract given to the hepatectomized rats significantly increased the serum AST and ALT levels when compared to controls at the end of treatment. The increased ALT and AST levels in the serum suggest the possible hepatotoxic effects of extracts. However, AST and ALT levels at the concentration of 1 % were less than those of the levels of 1.5 %. This effect appeared to be dose dependent. On the other hand, we found significant decrease in the level of BUN. This result indicates kidney damage. Thus, it might be due to decreased utilization of urea by damaged liver.

**Key words:** *Viscum album* ssp. *album* L., AST, ALT, Bun, Liver, Hepatectomy

### INTRODUCTION

Liver can rapidly regenerate itself after acute liver injury, chronic hepatic diseases, liver transplantation and partial hepatectomy. Hepatectomy, characterized by increased apoptosis, induces oxidative injury in hepatocytes. Apoptosis (programmed cell death) is an important process during normal development and regeneration of the liver. Due to the significant roles in development, regeneration and tissue homeostasis, it plays a critical role in the balance between regeneration and cell death. Recently, many researches have received a great deal of attention about evaluating apoptotic cell death [1-3]. *Viscum album* L. (VA), also called mistletoe, is a hemi-parasitic plant, originally found in Europe. It (not to be confused with Phoradendron, the American mistletoe) is a semiparasitic shrub that grows on other trees and has been used in cancer treatment for about 80 years. Its extracts contain a variety of biologically active compounds.

*Viscum* species, also known as Mistletoe, are medicinal plants, which have amines (acetylcholine, choline, histamin, tyramin), antioxidant flavonoids (quercetin, chalcone and flavone derivatives) and terpenoids (beta-amyrin, betulinic acid, oleanic acid, beta-sitosterol).

Mistletoes have been used both in traditional and supplementary medicine in the treatment and management of many diseases such as diabetes mellitus, stroke, hypertension, chronic cramps, stomach problems, heart palpitations,

difficulties in breathing and hot flushing in menopause for many years. A number of biological effects such as anticancer, antioxidant, antimicrobial, immunomodulatory and apoptosis inducing activities have been reported [4]. To date most pharmacological studies on mistletoe (*Viscum album* L.) have focused on the therapeutic properties of its polar extracts. In the past decade, there has been a great deal of interest in the use of medicinal plants for the treatment of diseases. Many researches have focused on the therapeutic properties of medicinal plants such as hepatoprotective, antioxidative, antimicrobial and gastric effects [6-7]. Furthermore, many scientific studies have been extensively carried out by using medicinal plants in liver damages [8-9].

The aim of the present study was to investigate the hepatoprotective effect of *Viscum album* ssp. album extracts in the liver of partially hepatectomized rats using biochemical methods.

### EXPERIMENTAL SECTION

Plant material: The host plant, localities, collection time of *Viscum album* L. ssp. album from *Pyrus communis* L. (dried) are Denizli, Tavas in orchard, November 2013, respectively (Figure 1).

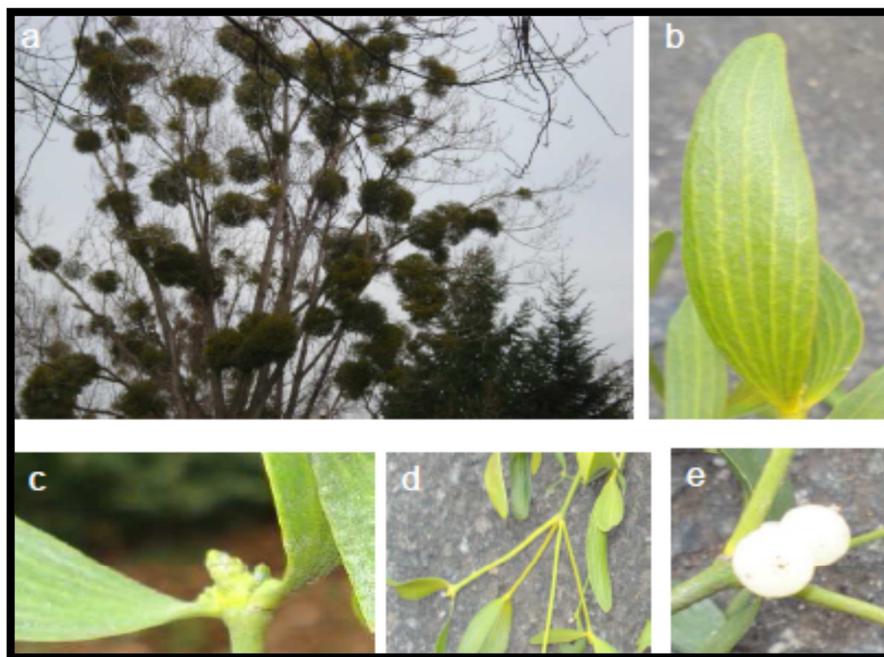


Figure 1. *Viscum album* morphology. a. Image of *Viscum album* growing on poplar; b. Aspect of leaf; c. The flower of mistletoe; d. Stem branched; e. Adult berry containing one seed

**Preparation of Plant Extract:** The plants dried in the shadow for extraction. The air-dried plants were ground to fine powder and then, put in the flask with ethanol for extraction process. The flask mouth was closed with a rubber stopper to protect the extract from contacting with air. The flask was covered with aluminum foil to protect the extract from light. Then, it was placed in a shaker water bath at 55°C for 6 h. The extraction was repeated twice at same condition. These extract were filtered and the solvents were removed in vacuum by a rotary evaporator at 42-49°C. The water in each extract was frozen in freeze-drying machine and then drawn out. Two different concentrations of the extract were prepared: 0.5% and 1%.

**Treatment of Animals:** Male albino rats, weighing approximately 150-200 g, were obtained from the Pamukkale University, Faculty of Medicine, Experimental Research Center, Denizli, Turkey. The animals were allocated into three groups with three rats in each group. Before the experimental period, 50% partial hepatectomy was performed under anesthesia by removing the left lateral lobe from all the groups. After the experimental period, the animals were sacrificed under anesthesia, and blood samples were collected for the biochemical assays

Group I: Control animals received normal rat diet and water, ad libitum (free-feeding).

Group II: The plant extract at concentration of 1 was given orally for 2 weeks.

Group III: The plant extract at concentration of 1.5 was given orally for 2 weeks.

**Biochemical Assays:** Blood samples were taken by cardiac venipuncture at the end of two weeks after the initial treatment. Then, they were centrifuged at 1000 rpm for 10 minutes to collect serum and were stored at -20 °C.

Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were measured for the determination of liver function while blood urea nitrogen (BUN) was measured for the determination of kidney function. The biochemical results of statistical analyses were evaluated using in SPSS 15.0 program.

## RESULTS AND DISCUSSION

Table 1 gives the mean serum AST, ALT and BUN levels for 2 weeks in all groups. *V. album* ssp. album extract given to the hepatectomized rats significantly increased the serum AST and ALT levels when compared to controls at the end of treatment. The increased ALT and AST levels in the serum suggest the possible hepatotoxic effects of extracts. However, AST and ALT levels at the concentration of 1% were less than those of the levels of 1.5%. This effect appeared to be dose dependent (Figure 2). On the other hand, we found significant decrease in the level of BUN. This result indicates kidney damage. Thus, it might be due to decreased utilization of urea by damaged liver. Little information is available in the literature on the effect of *Viscum album* on hepatic injury. Two recent studies done in patients with chronic hepatitis C, treated with a mistletoe preparation as monotherapy for 1 year, reported a significant improvement in elevated transaminases [10]. In a previous study suggests that mistletoe preparations may be a useful therapeutic intervention for patients with chronic liver disease. The mechanism(s) by which *Viscum album* modulates hepatic inflammation remains, however, unclear. The release of aminotransferases into the plasma was increased, indicating a increase in the severity of liver damage. Aminotransferases are sensitive indicators of liver-cell injury and are released into the blood in increasing amounts whenever the liver cell membrane is damaged [11]. In vitro, at very low concentrations (0.17-1ng). Mistletoe extract is highly cytotoxic to many solid and hematological malignancies. The aim of this in-vitro study was the assessment of three commercially available extracts from mistletoe (*Viscum album* L.) grown on ash tree (Abnobaviscum® Fraxini 20 mg), on for (Abnobaviscum® Abietis 20 mg), and on pine (Abnobaviscum® Pini 20 mg) for their potential to interfere with drug metabolism according to current guidelines for in vitro screening of drug–drug interactions [12]. In central Europe, extracts of *Viscum album* [L.] (VAL) are registered for parenteral use and are widely used in adjuvant and palliative cancer therapy, alone or in addition to conventional therapies. The unfavourable side-effects of late-stage pancreatic cancer treatments call for non-toxic and effective therapeutic approaches. They compared the overall survival (OS) of patients receiving an extract of *Viscum album* L. (VAL) or no antineoplastic therapy [13]. The impact on Korean mistletoe lectin II liver cancer cells. In a study that investigated and mechanisms, although not known exactly from the P-53Thelectin II with an independent mechanism to lower the telomerase activity and cell.

It was observed that leads to apoptosis [14-15]. Clastogenicity activity was evaluated by studying micronuclei formation in polychromatic erythrocyte cells in bone marrow. Plasma levels of gamma-glutamyl transferase ( $\gamma$ -GT), Alanine aminotransferase (ALT), Aspartate aminotransferase (AST) and Alanine aminotransferase (ALP) compared to the negative control. The results suggest that pretreatment of rats with either garlic or *Viscum album* extracts reduced the elevated plasma levels of liver enzymes and clastogenicity induced by sodium arsenite in rats [16]. Urea is primarily produced in the liver and secreted by the kidneys. Urea is the major end product of protein catabolism in animals. It is the primary vehicle for removal of toxic ammonia from the body. Urea determination is very useful for the medical clinician to assess kidney function of patients. In general, increased urea levels are associated with nephritis, renal ischemia, urinary tract obstruction, and certain extrarenal diseases, e.g., congestive heart failure, liver diseases and diabetes. Decreased levels indicate acute hepatic insufficiency or may result from over-vigorous parenteral fluid therapy. Increase in BUN level indicates kidney damage but in our study no kidney damage was observed on post-mortem examination. Thus, it might be due to decreased utilization of urea by damaged liver. Our findings pointed out the risk of increased lipid peroxidation, hepatic and renal damage due to long term use of opioids, especially morphine.

Table I. Serum AST, ALT and BUN levels of 2 weeks in partially hepatectomized rats

Groups		AST (IU/l)	ALT (IU/l)	BUN(mg/dL)
Control		124.26±24.04	82.76±7.16	61.26±0.39
Experimentals	1%	125.46±14.05	83.06±0.08	61.08±0.12
	1.5%	129.86±24.21	86.86±0.23	46.03±0.31

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