

The Effect of Hydro Alcoholic Extract of Lettuce on Blood Parameters in Mice

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Abstract: Lettuce (*Lactuca sativa* L.) is one of the most important vegetables in herbal medicine. This plant is belonging to Asteraceae family and has many therapeutic properties in traditional medicine. This study was conducted to investigate the effect of hydro alcoholic extract of lettuce on blood indices, sugar, insulin, cholesterol, triglyceride, HDL and LDL in Balb/C mice. Forty mice with about 25 g weight and the age range of 3-4 months were divided randomly in five groups including control, placebo and three experimental groups. All groups were kept under similar conditions. Hydro alcoholic extract of lettuce were prepared in 50, 100 and 200 mg/kg/day doses and IP injected of treatment groups for 20 days. Normal saline was used to inject to placebo group. The most important studied parameters were number of red and white blood cells, hematocrite, MCH, MCV, MCHC, sugar, insulin, cholesterol, triglyceride, HDL and LDL. At the end of study blood samples were taken and following results were obtained: Red blood cells and hematocrite were decreased significantly in 100 and 200 mg treatments whereas white blood cells were increased significantly ($p < 0.05$) in these treatments. Blood sugar was significantly decreased in all three treatment groups and insulin was reduced only in 200 mg group significantly. Also, cholesterol of 50 mg group reduced significantly whereas other parameters no changed. On the whole, hydro alcoholic extract of lettuce can have positive effects on white blood cells, sugar, insulin and cholesterol dose-dependently.

Key words: Lettuce, blood parameters, sugar, insulin, cholesterol, mice

INTRODUCTION

Scientists have proved in recent years which many plants, such as lettuce have significant effects in the treatment of many diseases. Lettuce (*Lactuca sativa* L.) is from Asteraceae family. This plant is an annual plant with low growing stem which leaves are attached to stem and growing along the short stem. Leaves are smooth and glabrous in dark green, bright green, yellow, red and brown colors. Lettuce seeds have been used in traditional medicine of Iran to treat inflammation and bone pain (Sayyah *et al.*, 2004). Lactucarium which is condensed syrup of this plant and other species of it has less side effects than opium and does not cause anorexia and constipation and is good for painful dysmenorrhea. Lettuce consumption is effective in treating insomnia, heartthrob and for relaxation and also reduces stomachache. Previous studies show the existence of anti oxidants, flavonoids and phenolic compounds in this plant (Lee *et al.*, 2006). Lettuce has aminobutyric acid, sucrose, glucose, fructose and inulin (Sobolev *et al.*, 2007). The root of this plant has organic acids like benzoic, phenyl acetic, cinnamic, lauric, phthalic, vanillic, palmitic and stearic acids (Singh *et al.*, 2008).

Previous studies show that lactucarium extracted from wild lettuce (*Lactuca virasa*) is effective in reducing

acute and chronic pain (Lee *et al.*, 2006). Considering long history of lettuce consumption in traditional medicine as a pharmaceuticals plant and scientific proofs for its medicinal effect, this study was conducted to investigate the effects of hydro alcoholic extract of lettuce on blood parameters, sugar, insulin and blood fat mice.

MATERIALS AND METHODS

The experimental study was done in 2012 in Falavarjan branch of Islamic Azad University. Forty male mice in weight range of 30 ± 5 g were selected and kept in $22-26^\circ\text{C}$ with sufficient food and water. Mice were divided randomly in five experimental groups including control, placebo and treatments groups. To prepare hydro alcoholic extract dried leaves of lettuce were grinded using grinder and 50 g of it was poured in a sterilized erlen. Ethyl alcohol 90% was added to powder and mixture was located on a shaker for 48 h to take the extract. After that extract was filtered using Whatman paper and considering remaining extract in solution, concentration of extract of original solution was calculated and experimental doses were prepared. Hydro alcoholic extract of lettuce was injected to mice of three treatment groups in 50, 100 and 200 mg kg^{-1} every other day and in

peritoneum. In order to achieve basic level of blood indices, control group was not injected and placebo group was injected using physiological serum. At the end of study, blood samples were taken from mice using guillotine method and blood factors including red blood cells, white blood cells, hematocrite, platelets, MCV, MCHC, MCH, sugar, insulin, cholesterol, triglyceride, LDL and HDL were measured by automatic auto analyzer and cell counter machines. Obtained data were analyzed using one way variance analysis of SPSS program and mean comparisons were done using Duncan's multiple range test.

RESULTS AND DISCUSSION

Results of effect of lettuce extract on blood factors are presented in Fig. 1-3. According to results, number of red blood cells was decreased significantly ($p < 0.05$) in all treatment groups. Also, number of white blood cells increased significantly ($p < 0.05$) in second and third treatment groups in proportion to control group.

Hematocrite amount was also reduced significantly ($p < 0.05$) in second and third groups. Blood sugar was decreased significantly ($p < 0.05$) in all three treatment groups.

Insulin concentration showed also significant reduction ($p < 0.05$) in third treatment group. Cholesterol concentration of first treatment group was significantly ($p < 0.05$) less than control. MCH, MCV, MCHC, triglyceride, HDL and LDL have no change in all groups. Considering pharmaceutical properties of lettuce, this study was conducted to investigate the effect of hydro alcoholic extract of lettuce on blood indices, sugar, insulin, cholesterol, triglyceride, HDL and LDL of mice. Results of this study showed that the number of red blood cells was significantly reduced in treatment groups which is probably because of negative effects of lettuce extract on kidneys and reduction in erythropoietin secretion (red blood cells adjuster) in higher doses (Modaresi and Gholchobian, 2012). Also, because hemoglobin and hematocrite are two main parts of red blood cells, it is natural to be affected by globule effecting factors and reduction in red blood cells to be followed by reduction in hemoglobin and hematocrite levels.

About increase in white cells amount we can say that mucosal surface of mice bodies is in contact with external environment and inner secretion of these surfaces are effective in body's defense which increase in oral antigen and maybe lettuce causes entrance and cells migmeiceion

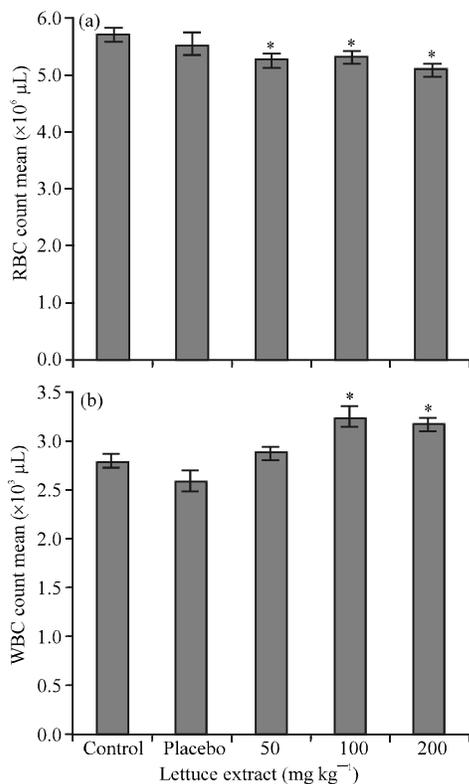


Fig. 1: Number of red and white blood cells

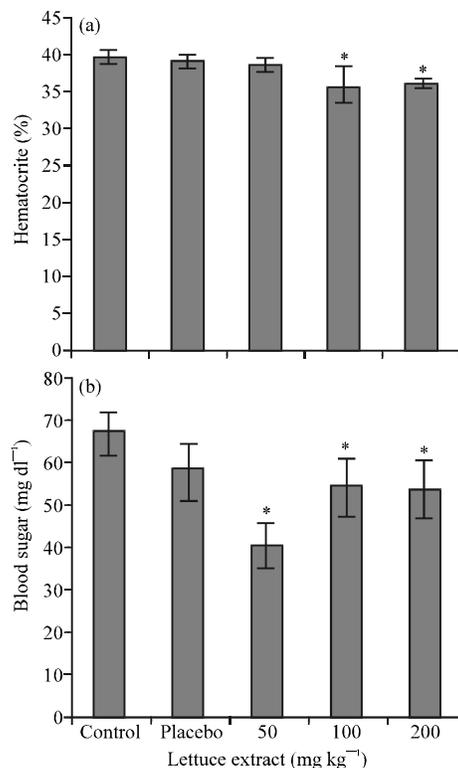


Fig. 2: Percentage of hematocrite and concentration of blood sugar

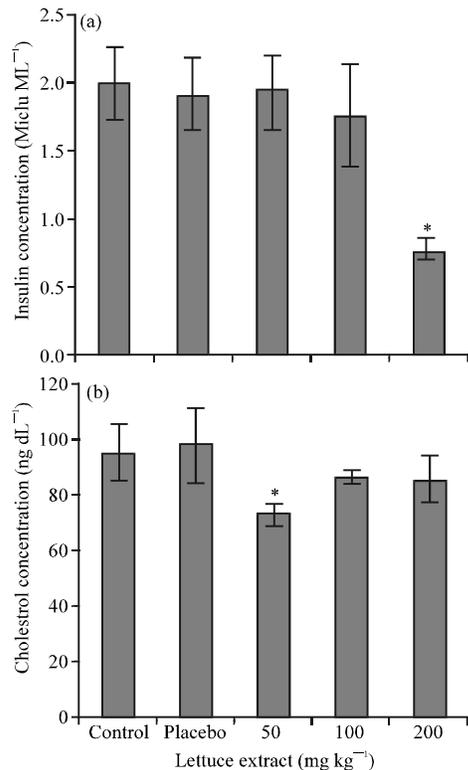


Fig. 3: Insulin and cholesterol concentration

to lymphoreticular tissue and finally IgA construction. In second mechanism, some micro organisms are transferring from digestive system so that they are found in inner tissues like liver, kidney and spleen and can transfer via inter cellular and intra cellular ways. White blood cells are moving units of body's immune system which part of them are made in extravascular spaces of the bone marrow and some in lymph tissue and are carried by blood stream to where they are needed and defend the body against pathogens (Modaresi and Khosravian, 2011).

According to results, there were not seen significant changes in MCHC, MCH and MCV factors which other studies on Astraceae family showed similar results too (Thijssen *et al.*, 2005; Zargari, 2004; Andrade-Cetto and Wiedenfeld, 2001; Heydarian *et al.*, 2011). Blood platelets were reduced non significantly in treatment group one and increased non significantly in second and third groups. Unsaturated fatty acids like linolenic acid of dandelion increase linoleic of platelets and not only reduce thromboxanes but also reduce platelets aggregation which this will be led to reduction in thrombosis possibility (Rajasekaran *et al.*, 2006). Also, extant coumarin in plants of this family like dandelion acts as an anticoagulant matter and prohibits platelets aggregation (Gebhardt, 1998). Blood sugar was decreased in all three treatment groups significantly which is

probably because of extant chlorogenic acid which in other plants of this family like artichoke causes blood sugar reduction (Gong *et al.*, 2009). Blocking feedback effects by lettuce can be second mechanism.

High reduction of insulin in third group can be explained as follows: Probably there is compounds in lettuce extract which affect insulin membrane receptors of skeletal muscles and with activating them increase glucose entrance by these cells. Increase in glucose consumption in mentioned cells causes reduction in blood glucose and then stimulating effect of glucose on β -cells of pancreases is eliminated, therefore insulin amount of serum is reduced (Shahraki *et al.*, 2009). The effect of chlorogenic acid and extant flavonoids in plant in lipid reduction and then decrease in lipoproteins has been proved. These compounds can interfere indirectly in cholesterol synthesis and also can reduce its synthesis via inhibition of hepatic hydroxymethyl glutaryl CoA reductase (Rajabian *et al.*, 2004). But, finally the probable mechanism is potential effects of plant sterols and stanols which while reduce incorporation of cholesterol pathways are able to increase intestinal ABCA1 expression and decrease intestinal absorption of cholesterol and lower cholesterol flows through the liver.

CONCLUSION

According to results of this study, using various levels of lettuce extract had significant effect on blood factors like red blood cells, white cells, hemoglobin and hematocrite. Also, these significant effects were observed in blood sugar, insulin, cholesterol. These significant changes were dose dependent. Increase in white blood cells improves immune system. Also, lettuce could reduce blood sugar and insulin dose dependently and then it can be effective in diabetes prevention. This plant helps fat digestions because of having plenty of fibers and reduces cholesterol, so it can be used as a drug in controlling metabolic syndrome control.

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