



EFFECT OF *OCIMUM SANCTUM* (TULSI) ON HISTOLOGY OF LIVER AND KIDNEY OF ALBINO RAT

Sunanda Dubey* and Sandhya Shah

Department of Zoology, Udai Pratap Autonomous College, Varanasi-221002 (U.P.) India

Abstract

Tulsi has been recognized for thousands of years to be one of greatest health promoting herbs. It has benefits for hundreds of conditions with thousands of years of empirical experience and use. To report the alterations inflicted by Tulsi in Liver and Kidney, the histological studies with these glands were done after administration of a dose of 1 g/ kg body weight for 60 days. The *Ocimum sanctum* leaves seemed to be non toxic on liver and kidney cells as we did not observe any pathological symptom in the histology of these vital organs. Hepatoprotective and nephroprotective effect of *Ocimum sanctum* leaves may be assumed and could be suggested that *Ocimum sanctum* is safe for use, but further studies are needed with further higher doses and duration to complete the safety profile of this plant.

Key words: *Ocimum sanctum*, Tulsi, Liver, Kidney

Introduction

The medicinal plants are rich in secondary metabolites, which are potential sources of drugs and essential oils of therapeutic importance (Wink, 2000). Medicinal plants are widely used in various ailments, because of their safety besides being economical, effective and their easy availability (Siddiqui, 1993; Ahmed *et al.*, 2002; Ammara *et al.*, 2009). According to a survey of World Health Organization (WHO, 1993), the practitioners of traditional system of medicine treat about 80% of patients in India, 85% in Burma and 90% in Bangladesh (Siddiqui, 1993; WHO, 1993). It is stated that Nature has provided a complete store house of remedies to cure ailments of mankind (Rahman *et al.*, 2011). Herbal plants have very important place in our routine life styles, like in food habit, in cosmetics, decoration, worship and most important in medicine. Today a large number of medicinal plants are used as a medicine (Prakash and Gupta, 2005), *Azadirachta indica* (Neem), *Curcuma longa* (Turmeric), like morphine from *Papaver somniferum*, ashwagandha from *Withania somnifera*, *Ephedrine* from *Ephedra vulgaris*, reserpine from *Rouplia serpentine* etc (Singh *et al.*, 2012). The Ayurvedic remedies that are both preventive and therapeutic are primarily made of plants and when

compared with their synthetic counterparts are either nontoxic or less toxic (Baliga and Dsouza, 2011; Kulkarni, 1997). *Ocimum sanctum* are used in Ayurveda and Siddha systems of medicine for prevention and cure of many illnesses and everyday ailments like common cold, headache, cough, influenza, earache, fever, colic pain, sore throat, bronchitis, asthma, hepatic diseases, malarial fever, as an antidote for snake bite and scorpion sting, migraine headaches, fatigue, skin diseases, wound, insomnia, arthritis, digestive disorders, night blindness and diarrhea (Prakash and Gupta, 2005). The leaves are good for nerves and to sharpen memory. Chewing of Tulsi leaves also cures ulcers and infections of mouth (Prajapati *et al.*, 2003).

The liver is the largest internal organ in the body contributing about 2% of total body weight which plays an essential role in the metabolism of foreign substances xenobiotics entering the body. The liver has considerable reserve capacity, can often maintain function in state of significant disease and is one of the few human organs capable of regeneration (Ward and Daly, 1999). Liver plays a major role in detoxification and is generally the major site for intense metabolism (Guyton and Hall, 2004). It is also a site of biotransformation of toxic compounds (Hodgsen, 2004). The liver is the key organ regulating homeostasis in the body. It is involved with almost all the

*Author for correspondence : E-mail :sunandadubey@rediffmail.com

biochemical pathways related to growth, fight against disease, nutrient supply, energy provision and reproduction (Ward and Daly, 1999).

Located at the rear of the abdominal cavity in the retroperitoneal space, the kidneys receive blood from the paired renal arteries, and drain into the paired renal veins. The Kidneys filter the blood in order to make urine, to release and retain water, and to remove waste. They also control the ion concentrations and acid-base balance of the blood. Kidneys also regulate fluid balance, blood pressure and are also responsible for the reabsorption of water, glucose, and amino acids. They also produce hormones calcitriol and erythropoietin and make an important enzyme renin, which affects blood pressure through negative feedback. To observe the effect of *Ocimum sanctum* on these vital organs seems interesting.

Materials and method

A total no. of 60 adult male albino rats weighing between 100-150g were acclimatized to the laboratory conditions for one week. Rats were divided in two groups control (group I, 30 rats) and experimental (group II, 30 rats). They were kept in the laboratory condition with temperature $30\pm 2^{\circ}\text{C}$ and relative humidity 44-56 % with light and dark cycles 14 and 10 h; during the experiments in standard polypropylene rat cages and were provided standard rodent pellet diet and water *ad libitum*. Paste of leaves was suspended in 1 ml water and was administered daily with a dose of 1g/kg body weight/day, orally to each of the experimental group of animals with the help of oral feeding tube. Similarly, each control rat received 1 ml water as a vehicle. 5 rats from control group and 5 rats from experimental group were not provided food and water 24 hours prior to sacrifice of the animals. Animals were sacrificed on 30th and 60th day. Tissues were removed, washed in saline, and fixed in Bouin's fluid for histological examinations. The tissues fixed in Bouin's fluid were processed by the paraffin wax embedding method for tissue sectioning. The 5 micron sections were stained with Haematoxylin and Eosin (H&E) to observe under the microscope, and photographs were taken by the camera attached with microscope (Magnus pro 3.7).

Results and discussion

The histological studies with liver and kidney did not reveal any pathological changes after treatment even with dose of 1 gm/ kg body weight for 60 days. In the present study liver showed a normal arrangement of the hepatocytes, with clearly visible central round nuclei, flat endothelial cells, central vein and sinusoids.

Generally any damage to the parenchymal liver cells results in increase of transaminases in the blood. In our earlier study both alkaline phosphatases and acid phosphatases (Shah and Barai, 2016) decreased as compared with control group while any rise could be taken as first sign of cell damage. Thus no increase in ALP and ACP observed strongly suggests that the *Ocimum sanctum* did not affect the hepatocytes adversely and consequently the metabolism of rats. *Ocimum sanctum* did not show any histological changes in liver and kidney indicating no or protective effect on Liver. *Ocimum sanctum* was found to protect the rats from the hepatotoxic action of paracetamol as evidenced by a significant reduction in the elevated serum enzyme levels, leading to the speculation that the extract treated group was partially protected from hepatic cell damage caused by paracetamol (Chattopadhyay *et al.*, 1992).

The hepatoprotective effect of *Ocimum sanctum* leaves may be due to the antioxidant properties of its constituents (Prakash and Gupta, 2005). *Ocimum sanctum* contains linoleic acid, which is responsible for its anti-inflammatory activity (Singh *et al.*, 1996).

DM (Diabetes Mellitus rats) demonstrated hepatic cell injury and fewer fat vacuoles in hepatocytes with less fat vacuole was shown in DM rats treated with aqueous extract of *Ocimum sanctum* (AQOS). No remarkable lesions were shown in renal tissue of normal control rats. Renal tissues of DM rats showed mild to moderate mesangial cells proliferation and cellular matrix expansion.

AQOS had a free radical scavenging activity which provides organs protection against diabetes. Supporting for this interpretation is that nearly general normal appearances of the liver and renal tissue were shown by histopathological study (Suanarunsawat *et al.*, 2014).

The Aqueous contained a significant amount of phenolic compounds. Several lines of evidences showed that plants with phenolic compounds had anti-lipidemic, anti-oxidative activities to protect risk organ (Fenercioglu *et al.*, 2010). This reflects that phenolic compound in AQOS might be responsible for that action in DM rats, however, it has not yet been known what kinds of phenolic compounds in AQOS that were responsible for those action.

Oxidative stress has been found to be the most important mechanism is hepatotoxicity of antitubercular drugs (Sodhi *et al.*, 1997 and 1996). *Ocimum sanctum* leaf extract has been found to significantly decrease the levels of hepatic lipid peroxidase and increase the levels of superoxide dismutase and catalase (Panda and Kar,

1998). Pretreatment dismutase *Ocimum sanctum* leaf extract has been found to prevent the radiation induced depletion of glutathione, glutathione peroxidase and superoxide dismutase and to prevent increase in lipid peroxidation rate (Ganasoundari *et al.*, 1998). The ursolic acid, which is one of the key constituents of *Ocimum sanctum* leaf extract is responsible for inhibition of lipid peroxidation (Balanehru and Nagarajan, 1991; Liu, 1995). Membrane stabilizing property of *Ocimum sanctum* has been shown to be responsible for hepatoprotective action (Sen *et al.*, 1988).

In the present study sections of Kidney of experimental rats showed normal renal tubules and renal corpuscles. The renal tubules and the glomeruli appeared to be prominent and normal. There were no histological

changes observed in animal, given *Ocimum basilicum* extract.

Ocimum sanctum aqueous leaf extract also known to reduce oxidative stress. The antioxidant activity of *Ocimum sanctum* is recently reviewed (Devi and Ganasoundari, 1999). Phenolic compounds from fresh leaves and stem of *Ocimum sanctum* cirsilineol isothymosin, isothymonin, rosmaric acid, and eugenol have been extracted and found to have antioxidant properties (Gupta *et al.*, 2002). The *Ocimum sanctum* flavonoids orientinn and vicenin also exhibited strong inhibitory effect on the reaction generated hydroxyl radical activity. They have strong antioxidant activity in vitro (Devi *et al.*, 2000; Godhavani *et al.*, 1988; Ganasoundari *et al.*, 1998) and antilipidperoxidative effect *in vivo*, (Devi and

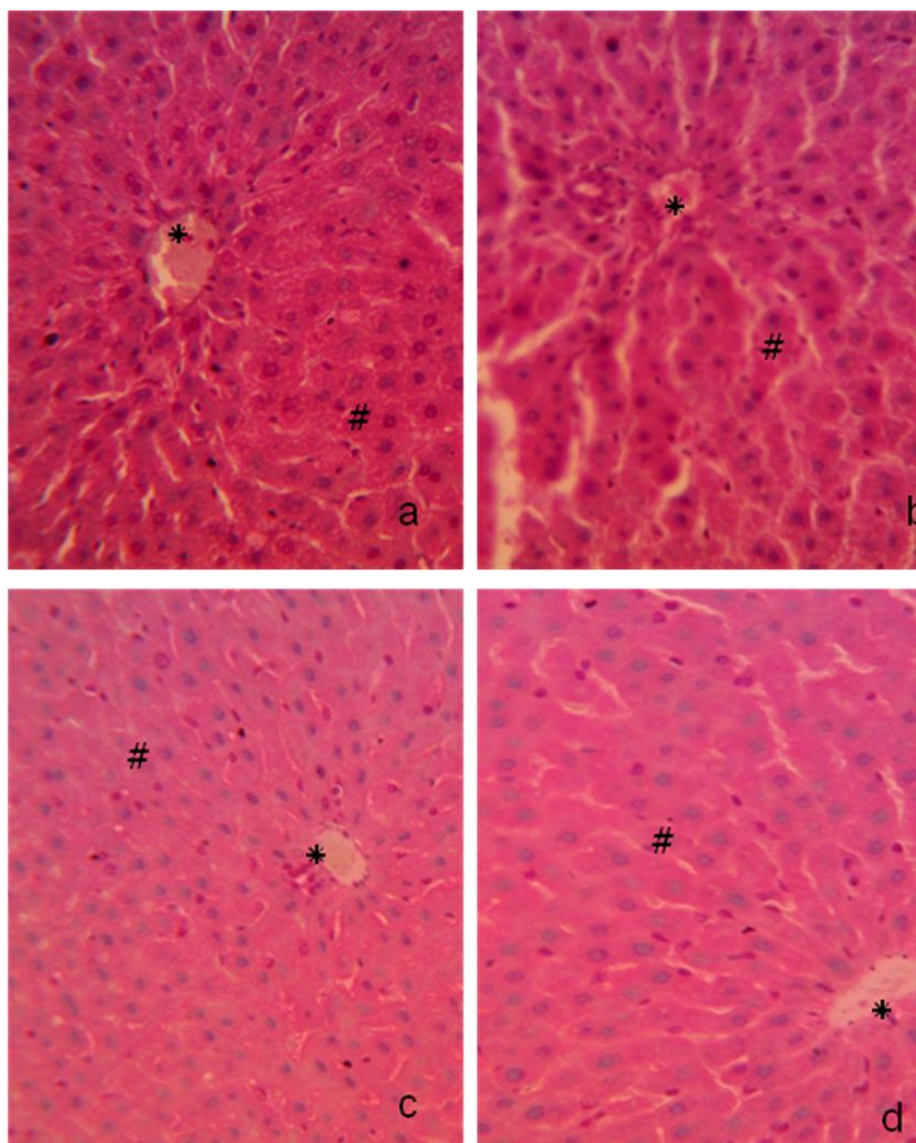


Fig. 1: Liver Showing normal hepatocytes, clearly visible central nuclei, flat endothelial cells and central vein in both control and experimental groups. (a) 30 days control (b) 30 days experimental (c) 60 days control (d) 60 days experimental (#) Hepatocyte and (*) Central vein ($\times 400$).

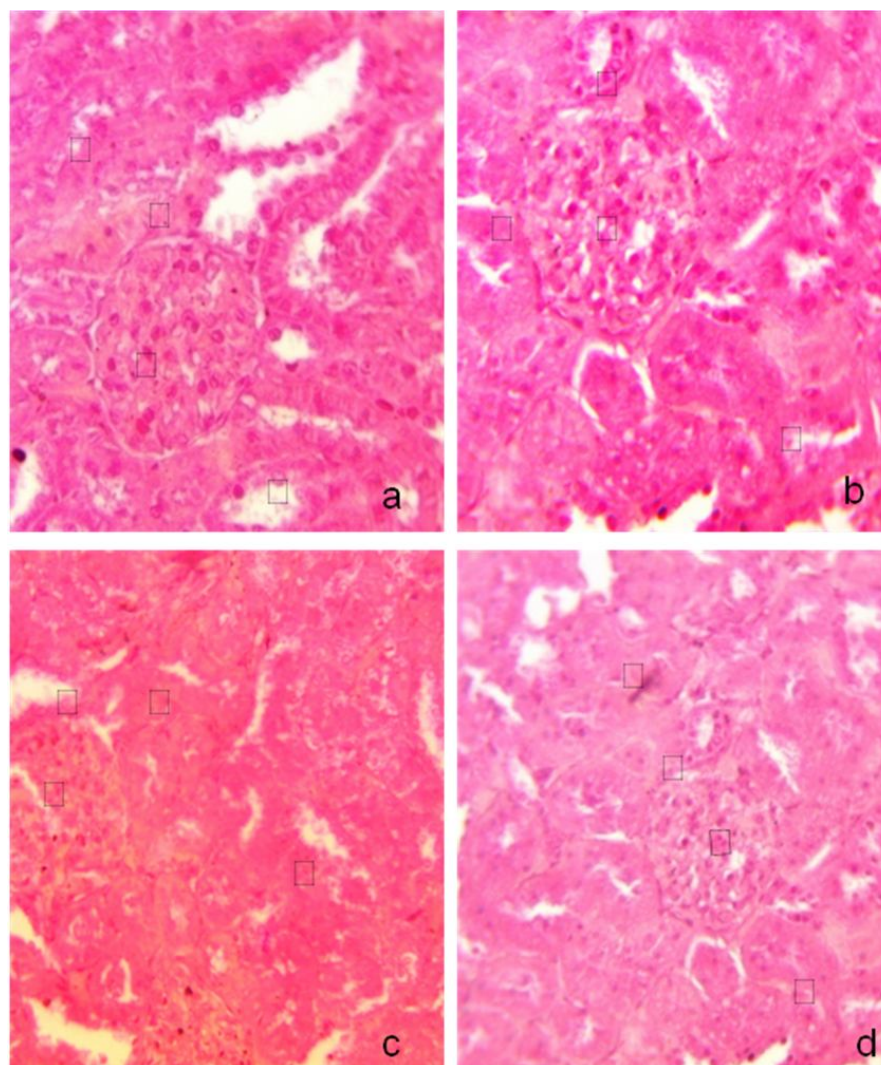


Fig. 2: Kidney Showing renal tubules, renal corpuscles and the glomeruli which appeared to be prominent and normal in both control and experimental groups. **(a)** 30 days control, **(b)** 30 days experimental **(c)** 60 days control **(d)** 60 days experimental. Proximal convoluted tubule (∇), Distal convoluted tubule (\triangle), Bowman's capsule (\ddagger) and Glomerulus (\dagger) ($\times 400$).

Ganasoundari, 1999; Godhavani *et al.*, 1988) strongly suggesting free radical scavenging as a major mechanism by which *Ocimum* products protect against cellular damage.

The antioxidant property and other actions of *Ocimum sanctum* like immunomodulatory properties, anti-inflammatory and some yet unknown properties may have a role in protecting kidneys from toxicity, the exact mechanism remains largely unresolved. Administration of *Ocimum basilicum* improved the histological changes induced in the delta methrin treated kidney. Kidney function was also improved as indicated by significant restoration of serum creatine and urea (Karmala *et al.*, 2011). The significant renal protection shown by long term administration of *Ocimum sanctum* makes it a protective herbal remedy.

Makwana and Rathore (2011) reported that *Ocimum sanctum* leaf extract suppressed histopathological alterations induced by paracetamol in liver and kidney of rats and restore creatinine, urea as well as liver function enzymes to its normal values (Yamamoto *et al.*, 2005) proved that *Ocimum* suppressed hepatic fibrosis and protected liver against parenchymal damage induced by CCl_4 . Our study suggests that no histopathological changes were observed in liver and kidney of albino rat fed with *Ocimum sanctum* leaves, hence, its non toxic role may be suggested.

References

- Ahmed, M., R.N. Ahamed, R.H. Aladakatti and M.G. Ghosesawar (2002). Reversible anti-fertility effect of benzene extract of *Ocimum sanctum* leaves on sperm parameters and fructose content in rats. *J Basic Clin Physiol Pharmacol*, **13**(1):51-9.

- Ammara, H., R. Salma, D. Farah and M. Shahid (2009). Antimicrobial activity of some plant extracts having hepato-protective effects. *Journal of Medicinal Plant Research*, **3(1)**: 020-023.
- Balanehru, S. and B. Nagarajan (1991). Protective effect of oleandolic acid and ursolic acid against lipid Peroxidation. *Biochem Int.*, **24**: 981-990.
- Baliga, M.S. and J.J. Dsouza (2011). Amla (*Embolica officinalis* Gaertn), a wonder berry in the Kulkarni, R.D. (1997) Principles of Pharmacology in Ayurveda. Ram Sangam Graphics, Mumbai, India.
- Chattopadhyay, R.R., K.S. Sarkar, S. Ganguly, C. Medda and K.T. Basu (1992). Hepatoprotective activity of *Ocimum sanctum* leaf extract against paracetamol induced hepatic damage in rats. *Indian Journal of Pharmacology*, **24**: 163-5.
- Devi, P.U. and A. Ganasoundari (1999). Modulation of glutathione and antioxidant enzymes by *Ocimum sanctum* and its role in protection against radiation injury. *Ind. J. Exp. Biol.*, **37**: 262-268.
- Devi, P.U., A. Ganasoundari, B. Vrinda, K.K. Srinivasan and M.K. Unnikrishnan (2000). Radiation protection by the *Ocimum* flavonoids orientin and vicenin: Mechanisms of action. *Radiat Res.*, **154**:455-60.
- Fenercioglu, A.K., T. Sler, E. Gene, H. Sabuncu and Y. Altuntas (2010). The effect of Polyphenol-C containing An-toxidants on Oxidative Stress and Lipid Peroxidation in Type 2 Diabetes Melitus without Compications. *Journal of Endocrinological Investigation*, **33**:118-124.
- Ganasoundari, A. P.U. Devi and B.S.S. Rao (1998). Enhancement of bone marrow radiation protection and reduction in WR-2721 toxicity by *Ocimum sanctum*. *Mutant Res.*, **397**: 303.
- Godhwani, S., J.L. Godhwani and D.S. Vyas (1988). *Ocimum sanctum* a preliminary study evaluating its immunoregulatory profile in albino rats. *J Ethnopharmacol*, **24**:193-8.
- Gupta, S.K., J. Prakash and S. Srivastava (2002). Validation of traditional claim of Tulsi, *Ocimum sanctum* Linn. As a medicinal plant. *Ind. J. Exp. Bio.*, **40**:765-773.
- Guyton, A.C. and J.E. Hall (2004). 'Text book of medical human physiology 10th(ed).
- Hodgsen, E. (2004). "A Textbook of modern toxicology" 3rd edition Jphn Wiley and sons Inc, New Jersey.203-211
- Karmala, K.S., A.Y. Sri latha ch, S.R.T.S. Chandra, V.D. Sreeni and P.P. Amravath (2011). Hematobiochemical changes of lead Poisoning and amelioration with *Ocimum sanctum* in wistar albino rats. *Veterinary World*, **4(6)**: 260-26.
- Kulkarni, R.D. (1997). Principles of Pharmacology in Ayurveda. Ram Sangam Graphics, Mumbai, India.
- Liu, J. (1995). Pharmacology of oleandolic acid and ursolic acid. *J. Ethnopharmacol*, **49**: 57-68.
- Makwana, M. and H.S. Rathore (2011). Prevention of hepatorenal toxicity of acetaminophen with *Ocimum sanctum* in mice. *Int. J. Pharm. Technol*, **3**:1385-96.
- Panda, S. and A. Kar (1998). *Ocimum sanctum* leaf extract in regulation of thyroid function in male mouse. *Pharmacol Res*, **38(2)**:107-110.
- Prajapati, N.D., S.S. Purohit, A.K. Sharma and T.A. Kumar (2003). Hand book of medicinal plant. *Agrobios*, India. 367.
- Prakash, P. and N. Gupta (2005). Therapeutic uses of *Ocimum sanctum* Linn (Tulsi) with a note on engenol and its pharmacological actions: A short review. *Indian Journal of Physiology and Pharmacology*, **49**:125-131.
- Rahman, S., R. Islam, M. Kamruzzaman, A. Khasrul and H.M.A. Jamal (2011). *Ocimum sanctum* L: Review of Phytochemical and Pharmacological Profile. *American Journal of Drug Discovery and Development*. ISSN 215-427X/ DOI: 10.3923.
- Sen, P., V. Dewan, S.K. Bhattacharya, V.S. Gupta, P.C. Maiti and P.K. Mediratta (1988). In brain and Psychophysiology of stress. NewDelhi, ICMR Publication.245.
- Shah, S. and S.R. Barai (2016). Effect of *Ocimum sanctum* (Tulsi) on serum alkaline phosphatase and Acid phosphatase activities on male albino rat. *Shodh Prera (AMQIRRJ)*. Vol. VI. Issue 1. pp. 75-78.
- Siddiqui, H.H. (1993). Safety of herbal drugs-an overview. *Drugs News & Views*, **1(2)**:7-10.
- Singh, S., D.K. Majumdar and M.R. Yadav (1996). Chemical and Pharmacological studies on fixed oil of *Ocimum sanctum*. *Ind. J. Exp. Boil*, **34**:1212-1215.
- Singh, E., S. Sharma, J. Dwivedi and S. Sharma (2012). Diversified potentials of *Ocimum sanctum* Linn (Tulsi). An exhaustive survey. *J. Nat. Prod. Plant Resour.*, **2**:39-48.
- Sodhi, C.P., S. Rana, S. Mehta, K. Vaiphei, R.C. Goel and S.K. Mehta (1997). Study of oxidative stress in rifampicin induced hepatic injury in growing rats with and without protein energy malnutrition. *Hum. Exp. Toxicol.*, **16**:315-321.
- Sodhi, C.P., S.V. Rana, S.K. Mehta, K. Vaiphei, S. Attri and S. Thankur *et al.* (1996). Study of oxidative stress in isonized induced hepatic injury in young rats with and without protein energy malnutrition. *J. Biochem. Toxicol.*, **11**:139-146.
- Suanarunsawat, T., N.D.W. Ayuthaya, S. Thirawarapan and S. Pongshompoo (2014). Anti-Oxidative, Anti-Hyperglycemic and Lipid-Lowering Effect of Aqueous Extract of *Ocimum sanctum* L. Leaves in Diabetic Rats. *Food and Nutritional Sciences*, **5**: 801-811.
- Ward, F.M. and M.J. Daly (1999). Hepatic disease. In Clinical Pharmacy and Therapeutics. R. Walker, C. Edward Eds, Churchill Livingston, Newyork. 195-212.
- WHO survey. (1993). In medicinal plants (Eds. Haq. I.). Karachi: *Hamdard Foundation Press*.
- Wink, M. (2000). Introduction Biochemistry, role and biotechnology of secondary products. In: Biochemistry of Secondary product Metabolism. Florida: CRC press, Bo Raton. 1-16.
- Yamamoto, J., K. Ymada, A. Naemura, T. Yamashita and R. Arai (2005). Testing various herbs for antithrombotic effect. *Nutrition*, **21(5)**:580-587.