Contents lists available at ScienceDirect

Asian Pacific Journal of Tropical Medicine

journal homepage:www.elsevier.com/locate/apjtm

Document heading doi:

Evaluation of anti-inflammatory property of the leaves of *Sansevieria liberica* ger. and labr. (fam: dracaenaceae)

Eze Charity Chinasa*, Inya-Agha Stella Ifeoma, Ezugwu Christopher Obodoike, Ezea Samson Chhukwuemeka

Department of Pharmacognosy and Environmental Medicine, University of Nigeria, Nsukka, Nigeria

ARTICLE INFO

Article history:
Received 20 June 2011
Received in revised form 15 August 2011
Accepted 15 September 2011
Available online 20 October 2011

Keywords:
Anti-Inflammatory activity
Sansevieria liberica
Leaves
Phytochemical screening

ABSTRACT

Objective: To evaluate the anti-inflammatory property of leaves of *Sansevieria liberica* Ger. and Labr. and to ascertain the toxicity and phytochemical profiles of the extract of the leaves. **Methods:** The juice from the fresh leaves was expressed manually and lyophilized. The crude extract (CE) was then fractionated into *n*-hexane fraction (HF), chloroform fraction (CF), ethylacetate fraction (EF) and methanol fraction (MF). The crude extract (CF) and the fractions were screened for anti-inflammatory activity using egg albumen-induced paw (systemic) edema in rats as a measure of acute inflammation. The toxicity test and phytochemical screening were done using standard procedures. **Results:** The CE and the fractions significantly ($P<0.05$) inhibited the development of paw edema induced by egg albumen in rats. The potency/activity of the CE and the fractions increased in the order HF>CE>MF>CF>EF, with the CE and HF at 400 mg/kg exhibiting inhibition comparable to that obtained with 5 mg/kg diclofenac sodium. Acute toxicity test on CE established an oral and intraperitoneal LD_{50} of > 5 000 mg/kg in mice. Phytochemical screening of the CE and the fractions showed the presence of various bioactive substances such as alkaloids, saponins, flavonoids, terpenoids, steroids, glycosides, reducing sugars, tannins, resins, carbohydrates, proteins, acidic compounds, fats and oils. **Conclusions:** The results of the study showed that the leaves of *Sansevieria liberica* Ger and Labr. possess anti-inflammatory effects which may be due to its bioactive constituents. Further purification on these bioactive constituents may result in the development of potent anti-inflammatory agent with low toxicity and better therapeutic index.

1. Introduction

Inflammation is defined as the reactive state of hyperemia and exudation from blood vessels with consequent redness, heat, swelling and pain which a tissue manifests in response to physical and chemical injury or bacterial invasion[1]. Inflammation may also be defined as a defense reaction of vascularised tissues to injury. The causes of inflammation are numerous and include living microorganisms such as pathogenic bacteria (pneumonia, skin infection), animal parasites, physical agents (trauma, thermal and radiant energy), chemical agents comprising exogenous causes (toxins, caustic substances) and endogenous causes including immunological reactions, such as arthritis[2,3]. Each of these precipitating factors causes a particular response but each usually leads to the formation of the five clinical symptoms of inflammation including redness (rubor), swelling (tumor), heat (calor), pain (dolor) and sometimes loss of function[4]. Acute inflammation develops rapidly, usually short lasting, and is a reversible process[5-9] while chronic inflammation may begin with relatively rapid onset or show insidious and even in unnoticed manner and tends to persist for several weeks, months or years and is generally irreversible[4, 10,11].

The overall therapeutic goals in the treatment of inflammation are: the relief of pain which is often the presenting symptom and the major continuing complaint of the patient and the slowing or, in theory, arrest of the tissue damaging process[6].

The cure characteristics and physicomechanical properties of natural rubber vulcanizates filled with fibres of *Sansevieria liberica* (*S. liberica*) and carbon black has

*Corresponding author: Eze Charity Chinasa, Department of Pharmacognosy and Environmental Medicine, University of Nigeria, Nsukka, Nigeria.
E-mail: charity.chinasa@yahoo.com

been reported[12], its amino-acid, mineral and vitamin composition as well as antidiarrhoeal effect have also been reported[13,14]. Due to the numerous ethnomedicinal applications and uses of the leaves of *S. liberica* Ger. and Labr., it is deemed necessary to ascertain the scientific basis for the ethnomedicinal applications and uses of its' leaf extracts in inflammation, determine the safety of the plant and identify the phytochemical constituents responsible for its effect.

2. Materials and methods

All the chemicals used for the extraction and fractionation, phytochemical screening, toxicity test and anti-inflammatory analysis were of analytical grades, and used as procured from their sources.

2.1. Chemicals and drugs

Methanol (BDH, Merck, Germany), *n*-hexane (BDH, Merck, Germany), chloroform (BDH, Merck, Germany), ethylacetate (BDH, Merck, Germany), tween 80 (Janssen, Belgium), diclofenac sodium (Hovid).

2.2. Collection and identification of plant material

Fresh leaves of *S. liberica* Ger and Labr. were collected in August, 2009 from Peoples' Flowering Garden in Nsukka, Nsukka Local Government Area, Enugu State, Nigeria and identified by Mr. A. O. Ozioko of the International Center for Ethnomedicine and Drug Development (Inter-CEDD), Nsukka, Enugu-State, Nigeria.

2.3. Experimental animals

Adult Swiss albino rats (100-300 g) and mice (24-36 g), of either sex were obtained from the animal house, Department of Zoology, University of Nigeria, Nsukka. They were housed in stainless steel cages (rats) and plastic cages (mice) under uniform laboratory conditions. They had free access to standard diet and drinking water and were acclimatized in the laboratory for one week before the start of the experiment.

2.4. Preparation of plant material

The juice from the fresh leave was expressed manually, sieved and stored in containers which was later lyophilized (at NIPRD, Abuja) to powder and stored in air - tight containers in the refrigerator for subsequent use. These samples were brought out and allowed to assume room temperature prior to use for analysis.

2.5. Extraction and fractionation

Sixty six grammes of the crude extract (CE) was adsorbed on silica gel and eluted in succession with *n*-hexane, chloroform, ethylacetate and methanol to yield, *n*-hexane (HF), chloroform (CF), ethylacetate (EF), and methanol (MF) fractions. The fractions HF, CF, EF and MF were screened for anti- inflammatory activity.

2.6. Phytochemical screening

Chemical tests were carried out on the CE and on the fractions using standard procedures[15, 16] and by characteristic colour changes as described by other authors[17,18].

2.7. Acute toxicity test

The acute toxicity profile of the extract was assessed using standard procedures[19].

2.8. Test for anti-inflammatory property

The effect of the extract/fractions on acute inflammation was evaluated by rat paw edema test according to Ratheesh *et al*, 1962[20]. Increase in the right hind paw volume induced by the subplanter injection of fresh egg albumin was used as a measure of acute inflammation[21].

2.9. Egg albumin-induced pedal edema in rats

Adult Swiss albino rats (100-300 g) of both sexes were divided into groups of 5 animals. Each subgroup (n=5) received either of two dose levels 200 or 400 mg/kg of the CE and the fractions in 3% v/v Tween 80, administered orally. Control animals received either diclofenac sodium (5 mg/kg) or equivalent volume of distilled water.

Thirty minutes later, inflammation was induced by injection of 0.1 mL of undiluted fresh egg albumin into the subplanter of the right hind paw of rats. The volume of the paw was measured by water displacement before and at 1, 2, 3, 4 and 5 hours after egg albumin injection. Edema formation was assessed in terms of the differences in the zero time paw volume of the injected paw and its volume at different times after egg albumin injection. For each dose of extract/fractions, the edema of different groups was calculated by subtracting from the paw volume at zero hour, the average of the edema of the group at each hour was obtained, and then the percent inhibition of edema was calculated using the following formula:

$$\text{Inhibition of edema (\%)} = \frac{V_0 - V_1}{V_0} \times 100$$

Where V_o = Average edema of negative control
V_t = Average edema of group test.

2.10. Statistical analysis

Results obtained were analyzed using one - way analysis of variance (ANOVA). Differences between means were accepted as significant at $P< 0.05$. Results were presented as mean±SEM.

3. Results

The expression process yielded 240 g of the CE, and fractionation process yielded 0.16 g (0.24% w/w) of *n* hexane fraction, 1.10 g (1.7% w/w) of chloroform fraction, 0.70 g (1.06% w/w) of ethylacetate fraction and 10.52 g (16.0% w/w) of methanol fraction. Phytochemical Screening showed that the CE tested positive for fats and oils, flavonoids, saponins, proteins, steroids, terpenoids, tannins, reducing sugars, carbohydrates, acidic compounds, alkaloids and glycosides; the MF gave positive reactions for resins, fats and oils, flavonoids, saponins, proteins, steroids, terpenoids, tannins, reducing sugars, carbohydrates, acidic compounds, alkaloids and glycosides; the EF tested positive for resins, fats and oils, flavonoids, steroids, and terpenoids. The CF tested positive for resins, fats and oils, flavonoids, steroids and terpenoids, while the HF gave positive reactions for resins, fats and oils, steroids and terpenoids (Table 1).

Acute toxicity and lethality tests indicated no death in the two phases of the tests and the LD_{50} was thus established to be > 5 000 mg/kg.

The crude extract and the fractions significantly (P<0.05) suppressed paw edema to varying degrees. The CE and the fractions showed significant dose dependent inhibition of paw edema (Figure 1–5). The CE and HF (400 mg/kg) produced inhibition comparable to that of diclofenac sodium (5 mg/kg) (Figure 1 and 2). The potency/activity of the crude extract and the fractions increased in the order: HF>CE>MF>CF>EF (Table 2).

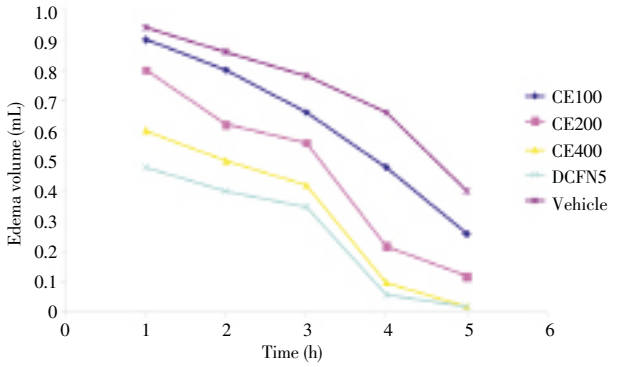

Figure 1. Variation of edema volume with time for the CE.

Table 1
Results of phytochemical analysis.

Phytochemical constituents	Relative abundance				
	CE	MF	EF	CF	HF
Resins	+	+	++	+++	+++
Fats and Oils	+	+	+	++	+++
Flavonoids	++	+	+++	+	–
Saponins	+++	+++	–	–	–
Proteins	+++	+++	–	–	–
Steroids	+	+	++	++	+++
Terpenoids	+	+	++	++	+++
Tannins	+++	+++	–	–	–
Reducing sugars	++	++	–	–	–
Carbohydrates	+++	+++	–	–	–
Acidic compounds	+++	+++	–	–	–
Alkaloids	++	++	–	–	–
Glycosides	++	+++	–	–	–

– = Not present;
+ = Present;
++ = Present in moderately high concentration;
+++ = Present in very high concentration.

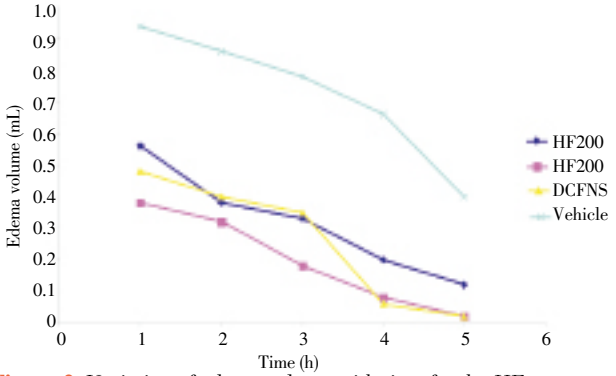

Figure 2. Variation of edema volume with time for the HF.

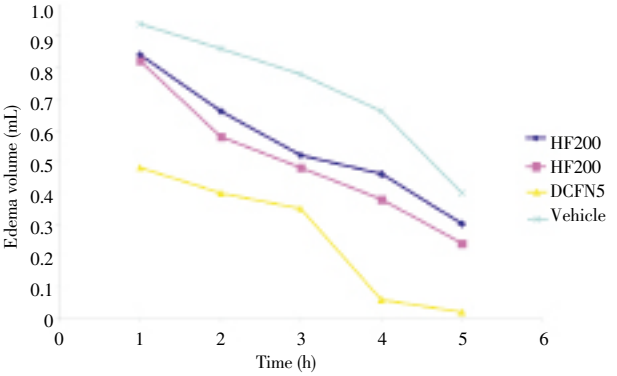

Figure 3. Variation of edema volume with time for the CF.

Table 2
Percentage inhibition of acute inflammation of rat paw.

Extract	Dose (mg/kg)	Inhibition (%)				
		1 h	2 h	3 h	4 h	5 h
CE	100	4.26	6.98	10.81	27.27	35.00
	200	14.89	27.91	24.32	66.67	70.00
	400	36.17	41.86	43.24	78.79	95.00
HF	200	40.43	55.81	56.76	69.70	70.00
	400	59.57	62.79	75.68	90.91	95.00
CF	200	10.64	23.26	32.43	30.30	25.00
	400	12.77	32.56	40.54	42.42	40.00
EF	200	−8.51	−34.88	−54.05	−60.61	−85.00
	400	10.64	−23.26	−16.22	−15.15	−75.00
MF	200	31.92	34.88	37.84	51.52	55.00
	400	38.30	44.19	51.35	57.58	70.00
Diclofenac	5	48.94	53.49	56.76	90.91	95.00

Percent inhibition (%) was calculated relative to the negative control.

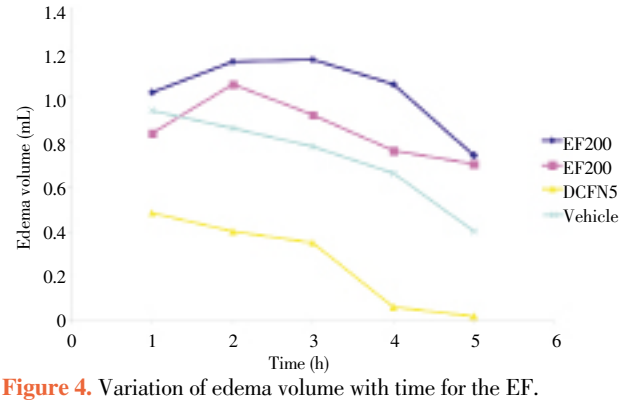

Figure 4. Variation of edema volume with time for the EF.

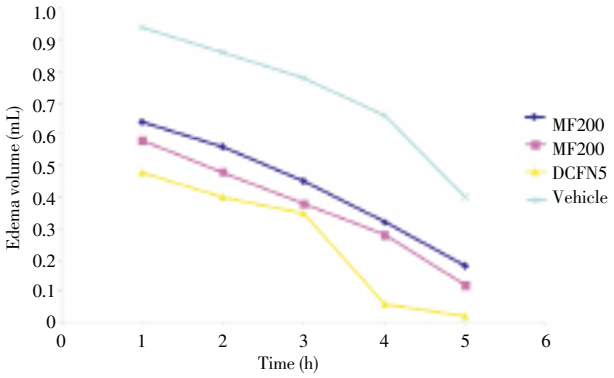

Figure 5. Variation of edema volume with time for the MF.

4. Discussion

The CE and the fractions showed a progressive but significant suppression of acute inflammation induced by egg album in the rat paw and the anti- inflammatory activity obtained for the CE and the fractions has justified the traditional uses of the plant in treating inflammations[22-24]. The toxicological study established the LD_{50} of the CE to be > 5 000 mg/kg and this implies that the leaves of *S. liberica* are safe, hence possibilities are remote for acute intoxication of the leaves in humans. Like the phlogistic agent induced edema, egg albumin-induced edema is probably mediated by histamine, serotonin, kinins, polymorphonuclear leukocytes, prostanoids, nitric oxide, neuropeptide and cytokines[25-29]. The results also suggest that the leaf extract and the fractions of *S. liberica* may be effective in suppressing acute inflammation. The HF consistently produced the highest inhibition of inflammation in the model used. This suggests that the anti-inflammatory constituents of this plant may reside mainly in this fraction.

Phytochemical analysis of the CE and the fractions revealed the presence of biologically active constituents such as alkaloids, carbohydrates, flavonoids, steroids, saponins, terpenoids, tannins etc. The anti- inflammatory activities of most plant extracts can be traced to these bioactive constituents. For instance, various compounds belonging to the terpenoid, and flavonoid groups are known to be biologically active[30-36]. The presence of these phytochemical constituents has been reported for the aqueous root extract of *S. liberica*[14]. The phytochemical constituents found in the leaves of the studied plant are known to possess anti-inflammatory effects. The steroids, terpenoids, alkaloids and flavonoids have been shown to possess anti-inflammatory activity in the adjuvant induced arthritis model[37,38]. Hence, the steroids, terpenoids, alkaloids and flavonoids present in the leaves may possess anti-inflammatory effect and contribute to the anti-inflammatory activity of the leaves. Further purification on these bioactive constituents may result in the development of potent anti-inflammatory agent with low toxicity and better therapeutic index.

Conflict of interest statement

We declare that we have no conflict of interest.

References

[1] Macdonald C. *Butterworth medical dictionary*. 7th ed. Kent: Butterworth and Company Ltd; 2011, p. 115–120.

[2] Cruse JM, Lewis R. *Illustrated dictionary of immunology*. 3rd ed. New York: CRC Press Inc; 2009, p. 166–167.

[3] Greene RJ, Harris ND. *Pathology and therapeutics for pharmacists, a basis for clinical pharmacy practice*. 3rd ed. London: Pharmaceutical Press; 2008, p. 31–38.

[4] Kumar V, Abba AK, Fausto N, Aster J. *Robbins and Cotran pathological basis of disease*. 8th ed. Philadelphia: WB Saunders Company; 2010, p. 50–111.

[5] Gambhire MN, Wankhede SS, Juveka AR. Anti-inflammatory activity of aqueous extract of *Barieria cristata* Leaves. *J Young Pharm* 2009; **1**(3): 220–224.

[6] Katzung BG, Masters SB, Trevor AJ. *Basic and clinical pharmacology*. 11th ed. USA: The McGraw Hill Companies Inc; 2009, p. 578–584.

[7] Botting RM. Inhibitors of cycloxygenases: Mechanisms, selectivity and uses. *J Physiol Pharmacol* 2006; **5**: 113–124.

[8] Guyton CA, Hall JE. *Text book of medical physiology*. 12th ed. Philadelphia: WB Saunders Company; 2011, p. 392–401.

[9] Djukanovic R. *Airway inflammation and repair in asthma*. New York: John Wiley and Sons Inc; 2011, p. 281–294.

[10] Meager A. *Cytokines*. Buckingham: Prentice Hall; 2011, p. 166–172.

[11] Salib RJ, Howarth PH. Transforming growth factor-beta in allergic inflammatory disease of the upper airways: Friend or for? *Clin Exp Allergy* 2009; **1**(1): 65–71.

[12] Osabohien E, Egboh SH. Utilization of bowstring hemp (*Sansevieria liberica*) fibre as a filler in natural rubber compounds. *J Appl Poly Sci* 2007; **107**(1): 210–214.

[13] Ikewuchi CC, Ikewuchi JC. Amino acid, mineral and vitamin composition of *Sansevieria liberica* Ger and Labr. *Pac J Sci Tech* 2009; **10**(1): 477–482.

[14] Adeyemi OO, Akindele AJ, Ogunleye EA. Evaluation of the anti-diarrhoeal effect of *Sansevieria liberica*. *J Ethnopharmacol* 2009; 1**23**(3): 459–463.

[15] Harborne JBC. *Phytochemical methods*. London: Goodreads Inc; 2011, p. 49–51.

[16] Evans WC. *Textbook of pharmacognosy*. 16th ed. London: Elsevier Ltd; 2009, p. 46–47, 832.

[17] Okwu DE, Mbaebie BO. Phytochemical constituents of some Nigerian medicinal plants. *Afric J Biotech* 2005; **4**(7): 685–688.

[18] Obianime AW, Uche FI. The Phytochemical screening and the effects of methanolic extract of *Phyllanthus amarus* leaf on the biochemical parameters of male guinea pigs. *J Appl Sci Environ Manag* 2008; **12**(4): 73–77.

[19] Azra R, Rafeeq A, Shadab A, Syeda A. Assessment of acute toxicity and reproductive capability of herbal combination. *Pac J Pharm Sci* 2010; **23**(3): 291–294.

[20] Ratheesh M, Helen A. Anti-inflammatory activity of *Ruta graveolens* Linn on carrageenan induced paw edema in wistar male Rats. *Afric J Biotech* 2007; **6**(10): 1209–1211.

[21] Okoli CO, Akah PA, Ezugworie U. Anti-inflammatory activity of extracts of root bark of *Securidaca longpedunculata* Fres (Polygalaceae). *Afric J Trad CAM* 2006; **2**(3): 54–63.

[22] Iwu MM. *A handbook of African medicinal plants*. Florida: CRC Press; 2011, p.234 –237.

[23] Jeff N. *Permaculture plants*. Australia: Agaves and Cacti, Jeff Nugent; 2006, p. 29–31.

[24] Joanne B, Habib G, Marie CJ, Michel F, Fernand G, Mansourous M, Joelle QL. Interdisciplinary journal devoted to indigenous drugs. *J Ethnopharmacol* 2009; **122**(3): 439–444.

[25] Shaik Mossadeq WM, Sulaima MR, Tengku Mohamad TA, Chiong HS, Zakaria ZA, Jabit ML, et al. Anti-inflammatory and antinociceptive effects of *Mitragyna speciosa* Korth methanolic extract. *Med Princ Pract* 2009; **18**(5): 378–384.

[26] Thong C, Erika P, Sabah A, Norma G, Robin Hoult J, Susan BD. Neurokinin-1 receptor agonists are involved in mediating neutrophill accumulation in the inflammed, but not normal, cutaneous micro-vasculartur: An *in vivo* study using neurokinin-1 receptor knockout mice. *J Immunol* 2011; **164**: 5424–5429.

[27] Emerson S, Boschi MS, Carlos E, Leite MS, Vasyl C, Saciura MS, et al. Anti-inflammatory effects of low level laser therapy (660 nm) in the early phase in carrageenan-induced pleurisy in rats. *J Lasers Surg Med* 2008; **40**(7): 500–508.

[28] Ninette G, Thierry M, Francois A, Thierry B, Abdel El M, Simon A, et al. T-kinin release from t-kininogen by rat-submaxillary gland endopeptidase K. *Eur J Biochem* 2011; **171**(3): 577–582.

[29] Ialenti A, Ianaro A, Moncada S, Di Rosa M. Modulation of acute inflammation by endogenous nitric oxide, elsevier BV. *Eur J Pharmacol* 2011; **211**(2): 177–182.

[30] Borgi W, Recio MC, Rios JL, Chouchane N. Anti-inflammatory and analgesic activities of flavonoid and saponin fractions from *Zizyphus lotus* Linn Lam. *S Afric J Botany* 2008; **74**(2): 32–34.

[31] Vijayalakshmi A, Ravichandiran V, Velraj M, Hemalatha S, Sudharani G, Jayakumari S. Anti-anaphylactic and anti-inflammatory activities of a bioactive alkaloid from the root bark of *Plumeria acutifolia* Poir. *Asian Pac J Trop Biomed* 2011; **1**(5): 401–405.

[32] Kumar S, Kumar V, Prakash OM. Pharmacognostic study and anti-inflammatory activity of *Callistemon lanceolatus* leaf. *Asian Pac J Trop Biomed* 2011; **1**(3): 177–181.

[33] Mohamed Saleem TK, Azeem AK, Dilip C, Sankar C, Prasanth NV, Duraisami R. Anti-inflammatory activity of the leaf extacts of *Gendarussa vulgaris* Nees. *Asian Pac J Trop Biomed* 2011; **1**(2): 147–149.

[34] Tian YQ, Kou JP, Li LZ, Yu BY. Anti-inflammatory effects of aqueous extract from radix *Liriope muscari* and its major active fraction and component. *Chin J Nat Med* 2011; **9**(3): 222–226.

[35] Wang D, Tang W, Yang GM, Cai BC. Anti-inflammatory, antioxidant and cytotoxic activities of flavonoids from *Oxytropis falcata* Bunge. *Chin J Nat Med* 2010; **8**(6): 461–465.

[36] Yang GM, Wang D, Tang W, Chen X, Fan LQ, Zhang FF, et al. Anti-inflammatory and antioxidant activities of *Oxytropis falcata* fractions and its possible anti-inflammatory mechanism. *Chin J Nat Med* 2010; **8**(6): 285–292.

[37] Gabor M. *The Anti-inflammatory actions of flavonoids*. Budapest, Hungary: Academia Kiado; 2011, p. 6 –15.

[38] Gokani RH, Lahiri SK, Santani DD, Shah MB. Evaluation of anti-inflammatory and antioxidant activity of *Premna integrifolia* Root. *J Comple Integ Med* 2011; **8**(1): 1553–3840.