

A Study on Toxic Effects and Laxative Activity of Senna (*Cassia angustifolia* Vahl.)

R. Sujatha, Jemima Beryl Mohankumar¹ and P. Shanmugasundaram²

Department of Nutrition and Dietetics, Government Arts and Science College for Women,
Bargur, Krishnagiri District, Tamilnadu, India.

¹Department of Nutrition and Dietetics, P.S.G. College of Arts and Science College,
Coimbatore, Tamilnadu, India.

²Department of Chemistry, Thiruvalluvar Government Arts College, Rasipuram,
Namakkal District, Tamilnadu, India.

(Received: 04 June 2011; accepted: 12 July 2011)

Senna is a medicinal plant variety grown predominantly in Tamilnadu and Gujarat. The pods and leaves of the plants are used in confectionary and herbal preparations. The two species used most often for medicinal purposes are Alexandrian senna and Tinnevely senna. The Alexandrian variety is obtained mainly from Egypt and the Sudan. Tinnevely senna is primarily cultivated in India. For the present study the senna plant was collected from Tirunneveli district, Tamilnadu, India and the major active constituents sennoside A and B are identified, microbial analysis of senna leaf powder was carried out and the nutritive value of senna was determined. The toxic effect and laxative activity of senna leaf powder was carried out with animal model.

Key words: Constipation, *Cassia angustifolia*, Senna, Sennosides, Toxicity, Laxative.

In recent times, focus on plant research has increased all over the world and a large body of evidence has been collected to show immense potential of medicinal plants used in various traditional systems (Rao, 2004). Constipation is one of the most common gastrointestinal complaints. It is more common in women than in men and increases with age in the adult population (Walter *et al.*, 2002). The common factors that contribute to the development of constipation in the general

population are diet, altered bowel habits, inadequate fluid intake and lack of exercise (Potluri, 2002). Physiologic factors include inadequate oral intake, dehydration, and inadequate intake of dietary fiber or organ failure. Any or all of these factors can occur because of the disease (Camoiez *et al.*, 2002).

Senna (*Cassia angustifolia* Vahl.) has the powerful cathartic effect. Senna is an important herb in traditional Chinese medicine, Indian Ayurvedic, and Unani medicine. The caution of senna required in hemorrhoids, inflammatory conditions of the gastro intestinal tract, diarrhea and pregnancy. Repeated use of strong purgatives such as senna may aggravate constipation and weaken the tone of the colon. More serious effects include fainting, dehydration, and electrolyte

* To whom all correspondence should be addressed.
E.mail: ramasamysujatha@yahoo.co.in

disorders such as low blood potassium, albuminuria, and hematuria. The leaves and pods of senna contain sennosides A, B, C, D, which are well known for the preparation of laxative and purgative (Kokate, 2004).

Salient Features of Senna

Senna is taken orally. The usual daily dosage delivers 20 to 60 milligrams of its active ingredient. The laxative of senna also known as contact laxatives, act on the intestinal wall. They increase the muscle contractions that move along the stool mass. Senna leaves is used for relief of occasional constipation and bowel irregularity. Senna is a strong purgative that should be taken with care and in proper dosage. It has an irritant effect upon the intestinal membrane, and may cause griping, pain or nausea along with liquid stools or diarrhea (Mitchell *et al.*, 2006). Taking Senna at the same time as drugs or herbals with laxative or diuretic effects may cause potentially dangerous reductions in the amount of potassium in the body.

MATERIAL AND METHODS

In continuance of our work on the development of senna incorporated food products to relieve constipation, the senna plant was collected from Tirunelveli District, Tamilnadu, India. The fresh, dried leaves and extract of senna were analyzed for nutrients like moisture, crude fibre, total ash, acid insoluble ash, iron, calcium, phosphorus, carbohydrate, protein, fat, sodium, potassium, thiamine and riboflavin by standard methods.

Chemical Test

Borntrager Test

- 1) To 25 mg of powdered senna added 50ml of water and 2ml of sulphuric acid. This was heated in a water bath for 15 minutes. Allowed to cool and shake with 40 ml of ether. Dried the ether layer over anhydrous sodium sulphate, evaporated to 5ml. To this added 5 ml of 6% ammonia. The residue develops a yellow (or) orange colour. Heated on a water bath for 2 minutes, a reddish violet colour develops in Tinnevely senna (Christenson and Abdel-Latif 2006).
- 2) A little senna extract is treated with 5N sodium hydroxide and sodium hyposulphite. On heating, red colour

appears indicating the presence of anthraquinones (Harborne, 1983).

Extraction of Sennosides

Sennoside A and B were isolated and identified by the reported procedure (Stool, 1949).

Identification of sennoside A and B by HPLC

Sennosides A and B were identified by HPLC according to the procedure used by Verma *et al.*, (1996).

Heavy Metals like Lead and Cadmium

Cadmium (Cd), Lead (Pb), Arsenic (As), and Mercury (Hg) and other heavy metals are widely dispersed in the environment. These elements have no beneficial effects in humans and there is no known homeostasis mechanism for them and it is well known that chronic exposure to As, Cd, Hg and Pb at relatively low levels can cause adverse effects (Jozef, and Zmudzki 2005). To evaluate the health risks to consumers it is necessary to determine the specific dietary intake of each pollutant for comparison with toxicologically acceptable levels. The analysis of Lead and Cadmium were carried out according to the known procedure (Jozef, 2005).

Pesticides

Pesticides protect the agricultural crops but overuse and incorrect use pose risks to human health and the environment. The increase in the amount and variety of products applied to agriculture makes it necessary to monitor residues in the environment, therefore, the analysis or pesticides has received increasing attention in the last few decades (Acero *et al.*, 2008).

Pesticides are not used for the cultivation of cassia angustifolia vahl in the area from where it was collected but in some places the pesticides like malathion, sumithion and diazinon were used very rarely. HPLC analysis was adopted to determine the pesticides residues viz. organophosphorus pesticides in the selected cassia angustifolia vahl according to the known procedure (Islam et al 2009).

Animal Experimentation for Documentation of Toxic Effects

Animal experimentation is inevitable for obtaining data on toxicity. The investigator submitted the animal study protocol to the Institutional Animal Ethics Committee, which was approved and certified that it was in accordance with the guideline of CPCSEA.

Selection of Animal Model

Either sex of Wistar Albino adult rats weighing between 150 – 250 gm were used. The animals were obtained from animal house in Institute of Road Transport Perundurai Medical College and Hospital, Perundurai, Erode, Tamilnadu, India. On arrival the animals were placed at random and allocated to treatment groups in polypropylene cages with paddy husk as bedding. Animals were housed at a temperature of $24\pm 2^{\circ}\text{C}$ and relative humidity of 30 – 70 %. A 12:12 light: day cycle was followed. All animals were allowed free access to water and fed with standard commercial rat chew pellets obtained from M/s. Hindustan Lever Ltd, Mumbai.

All the experimental procedures and protocols used in this study were reviewed by the Institutional Animal Ethics Committee (Regd No: 688/2/C-CPCSEA) and were in accordance with the guidelines of the CPCSEA.

Acute oral toxicity studies

Acute oral toxicity studies were performed (Vasudevan *et al.*, 2007) according to OECD-423 (Organisation for Economic Co-operation and Development) guidelines (acute toxic class method). Wistar Albino rats ($n=3$) of either sex selected by random sampling technique were employed in this study. The animals were fasted for 4 h with free access to water only. The senna fine powder (suspended with 0.5% w/v Carboxy Methyl Cellulose) was administered orally at a dose of 5 mg/kg initially and mortality was observed for 3 days. If mortality was observed in 2/3 or 3/3 animals, then the dose administered was considered as toxic dose. However, if the mortality was observed in only one rat out of three animals then the same dose was repeated again to confirm the toxic effect. If mortality was not observed, the procedure was then repeated with higher doses such as 50, 300 and 2000 mg/kg.

Screening of Laxative Activity

The use of senna as a laxative is an ancient example of the use of plant products in traditional medicine (Mutwally and Meelad 1997a). In Tibb (Greco-Arabic) system of medicine the use of senna as a laxative first appeared around the late 1000 AD and from there it was adapted into the western medicine. The sennosides have been widely used but the

information relating to their mode of action remains scant. In the last decades, various possible mode of action of sennosides as laxative has been explained including (a) stimulation of colon nerve plexuses thereby leading to defecation (Dobbs *et al.*, 1975); (b) sennosides and their metabolites acting directly on large intestine motility (Leng, 1986a); (c) changes in the colon motility and colonic fluid absorption and finally (d) involvement of prostaglandin E2 in secretagogue action of the sennosides in small intestine has been suggested (Nijs *et al.*, 1991).

The laxative activity was determined according to Capasso *et al.*, (1986). The rats of either sex were fasted for 24 hours before the experiment, but with water provided ad libitum. The animals were divided in to four groups of six rats in each group. The design:

- Group I : Serving as control, received 0.5% w/v carboxy methyl cellulose sodium (CMC).
- Group II : Received orally 100 mg/kg senna fine powder.
- Group III : Received orally 200 mg/kg senna fine powder.
- Group IV : Received orally 400 mg/kg senna fine powder.

Immediately after dosing, the animals were separately placed in metabolic cages suitable for collection of faeces. After 8 hours of drug administration, the faeces were collected and weighted.

Drug-induced Constipation in Rats

Loperamide (5mg/kg, per oral) was used as a drug for inducing constipation (Saito *et al.*, 2002). Wistar rats either sex, fasted for 24 hours before the experiment, and were divided in to five groups of six in each.

- Group I : Serving as control, received 0.5% w/v carboxy methyl cellulose sodium (CMC).
- Group II : Received Loperamide (5mg/kg, p.o) after 1 hour pre-treatment of 0.5% CMC.
- Group III : Received Loperamide (5mg/kg, p.o) after 1 hour pre-treatment of 100 mg/kg, p.o., fine senna powder.
- Group IV : Received Loperamide (5mg/kg, p.o) after 1 hour pre-treatment of 200 mg/kg, p.o., fine senna powder.

Group V : Received Loperamide (5mg/kg, p.o) after 1 hour pre-treatment of 400 mg/kg, p.o., fine senna powder.

Immediately after Loperamide, the animals were separately placed in metabolic cages suitable for collection of faeces. After 8 hours of drug administration, the faeces were collected and weighted.

All the results were expressed as Mean \pm Standard Error (SEM). Data was analyzed using one-way ANOVA followed by Dunnett's t-test and also Tukey-Kramer Multiple Comparison Test. p-values <0.05 were considered as statistically significant.

RESULTS AND DISCUSSION

Identification of Sennosides

In the present study, the major active principles sennosides A and B, which are responsible for the laxative properties were isolated according to the chemical procedure developed by Stoll et al (1949) and the melting points of the solids obtained were compared with the reported values by Stoll et al (1949). It was found that the melting points of the compounds isolated from the leaves matched well with the reported values. Sennoside A melting point 200-240° C (decomposition), reported melting point 200-240° C (decomposition); sennoside B melting point 180-184° C (decomposition), reported melting point 180-186° C (decomposition).

The presence of the sennosides A and B were further confirmed by HPLC analysis of the dried leaf extract. Different compositions of the mobile phase were tested and the desired resolution of the sennosides with symmetrical and reproducible peaks and a stable baseline was achieved by using methanol: water: acetic acid: tetrahydrofuran (60:38:2:2) as mobile phase. Peaks

corresponding to sennoside A and Sennoside B were sharp and well resolved with retention times (Rf) of 9.87 and 7.21 minutes respectively, which is well matched with the standard samples retention times. The resolution factor between the two peaks was 0.52. The desired resolution of the sennosides with symmetrical and reproducible peaks and a stable baseline was achieved by using this mobile phase as given by Verma *et al.*, (1996).

Standard solutions (1mg/10ml) of sennoside A and sennoside B were prepared in methanol. Different amounts of these standards were injected into the HPLC using the chromatographic conditions described and the reproducible retention times were obtained for sennoside A and B. Peaks corresponding to sennoside A (1) and sennoside B (2) were sharp and well resolved with retention times of 9.87 and 7.21 min respectively. The resolution factor between the two peaks was 0.52. The chromatogram obtained with pure standards is given in figure I and for extract of senna is given in figure II.

Heavy Metals like Lead and Cadmium

Atomic absorption spectrophotometry is considered better for time saving, high sensitivity and specificity (Koops and Westerbeek, 1978). The results of our analysis reveals that the lead and cadmium levels were 0.40 and 0.03 mg/kg respectively. These values are lower than the tolerance level of 0.429 mg of lead and 0.055-0.070 mg/kg of cadmium (WHO, 1972).

Pesticides

In the present study, the dried senna leaves (10 g) were powdered and extracted with ethanol (100 ml) and filtered, solvent was removed to get the extract, which was used for HPLC analysis. The analysis was carried out to find the amount of organophosphorus pesticides like diazinon, malathion and sumithion. It was found that these pesticides were not present in the plant

Table 1. Microbial Analysis of *Cassia Angustifolia Vahl.* (Senna) Powder

Storage Packages	Senna Leaf Powder		
	After 45 Days	After 90 Days	After 180 Days
Glass Bottle	Nil	1×10^2 CFU	4×10^2 CFU
Plastic container	Nil	2×10^2 CFU	6×10^2 CFU

CFU – Colony Forming Units

Table 2. Nutrient Analysis of *Cassia Angustifolia Vahl.* (Fresh and Dried Leaves per 100 g)

Nutrient Parameter	Senna leaves	
	Fresh Mean±SD	Dried Mean±SD
Moisture (%)	68.00±1.0	7.7±0.2
Ash (g)	2.88±0.00	8.58±0.14
Crude fibre (g)	6.93±0.39	18.98±0.33
Carbohydrate (g)	11.8±0.3	38.51±0.50
Proteins (g)	2.02±0.008	6.02±0.06
Fat (g)	1.92±0.003	6.96±0.02
Calcium (g)	1.30±0.01	4.56±0.28
Phosphorus (g)	0.90±0.002	3.66±0.10
Sodium (g)	0.19±0.001	0.61±0.00
Potassium (g)	0.55±0.002	1.74±0.01
Iron (mg)	2.50±0.02	7.0±0.17
Thiamine (mg)	0.44±0.001	0.30±0.00
Riboflavin (mg)	0.18±0.003	0.10±0.005

(the limit of detection of diazinon, malathion and sumithion is 0.02 mg/kg, which is lower than the tolerance level, WHO 2005).

Microbial Analysis of *Cassia Angustifolia Vahl.* (Senna) Powder

The foods that we eat are rarely if ever sterile, they carry microbial associations whose composition depends upon which organisms gain access and how they grow, survive and interact in the food over time. The microorganisms present originate from the natural microflora of the raw material and those organisms introduced during harvesting, processing, storage and distribution. The numerical balance in the various types is determined by the organisms themselves and the affects of processing (Adams *et al.*, 2000).

The microbial analysis of senna powder was carried out after being stored at room

Table 3. Effect of Senna Powder on Loperamide - Induced Constipation

Treatment	Dose (mg/kg, p.o)	Faecal Output in 8 hours (mg)
Control	0.5% CMC	85.33±5.05
Loperamide	5	47.67±5.01**
Loperamide & Senna powder	5; 100	58.33±3.37*
	5; 200	103.33±6.16 ^a
	5; 400	166.55±8.34***. ^a

Values are in mean ± SEM. (n=6)* denotes p<0.05 as compared to control group; ** denotes p<0.01 as compared to control group; *** denotes p<0.001 as compared to control group^a denotes p<0.001 as compared to Loperamide group.(One-way ANOVA followed by Tukey-Kramer multiple Comparison Test); p.o – per oral

Table 4. Acute Oral Toxicity Test

Treatment	Dose (mg/kg, p.o)	Fecal Output in 8 hours (mg)
Control	0.5% CMC	85.33±5.05
Senna powder	100	101.83±3.71*
	200	159.17±9.70**
	400	202.17±6.11**

Values are means ± SEM. (n=6) p.o – per oral; * denotes p<0.05 as compared to control group;** denotes p<0.001 as compared to control group. (One-way ANOVA followed by Dunnett’s t-test)

temperature for 180 days (6 months). The results are given in the Table I.

The microbial load of senna powder stored in glass and plastic containers was not detected for a period of 45 days. However, when stored for a period of 90 and 180 days, the microbial load was found to be 1 x 10² and 4 x 10²CFU in glass bottle and 2 x 10² and 6 x 10²CFU in plastic container respectively.

This population of microbes were analysed for 1 g of sample (senna powder). According to the microbial analysis the microbial count was not more than 6 x 10²CFU. The microbes

Staphylococcus albus, *Bacillus species* and *Streptococci species* are air borne microorganism present in our environment, which are not harmful.

The presence of aflatoxins in senna plants was studied by Muller and Basedow (2007) using HPLC method. Only pods of *Cassia Angustifolia* contained aflatoxins; leaves and flowers were free. Since aflatoxins are naturally occurring substances, it is impossible to completely eliminate them from products; however it can be reduced to the minimum possible level. Aflatoxins, especially aflatoxin B1 is highly toxic to mammals and is carcinogenic. Due to the high toxic action of aflatoxins, many countries have established maximum residue levels, generally lying between 4 and 50 µg/kg.

Nutrient Analysis of *Cassia Angustifolia Vahl*

Nutrient analysis of a food is the nutritional composition of that food. It is the estimation of the nutritive value of human food in its chemical form. The nutritive analysis as shown in the Table -II reveals the various nutrient contents of senna.

Screening of Laxative Activity

The higher doses of senna powder (200 & 400 mg/kg, p.o.) showed significant increase of faecal output ($p < 0.05$; $p < 0.001$, respectively) in rats.

In the present study the rats treated with Loperamide (5mg/kg, p.o) to produce constipation, were observed from significant reduction ($p < 0.01$) in faecal output. Faecal out put (mg) in eight hours was used as an indicator of laxative activity. Constipation produced by Loperamide was successfully reversed ($p < 0.001$) by senna powder (200 & 400 mg/kg, p.o.). 200 mg/kg, p.o., showed faecal output like the control animals (Table III).

Acute Oral Toxicity Test

The present study indicates that the senna powder did not produce any mortality even at the highest dose (2000 mg/kg, p.o.) employed. All the doses (5, 50 and 300 mg/kg, p.o.) of senna were thus found to be non-toxic. Three doses (100, 200 and 400 mg/kg, p.o.) of senna powder were selected for further pharmacological studies in humans (Table IV).

Results of the present study indicated that the senna (*Cassia Angustifolia Vahl.*) used for the development of incorporated food products, contains the major active constituents sennoside A and B, which are responsible for its laxative

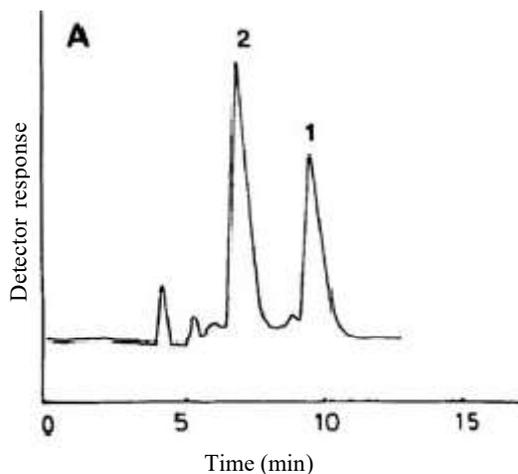


Fig. 1. HPLC Separation of Sennoside A(1) and Sennoside B(2)-Chromatogram obtained with Pure Standards

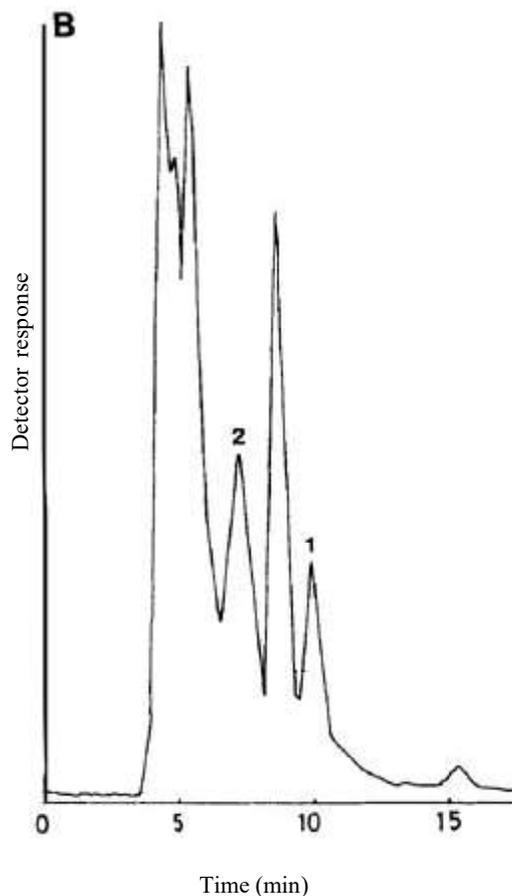


Fig. 2. Chromatogram of Extract of Senna

property. The results of our analysis revealed that the lead and cadmium levels were 0.40 and 0.03 mg/kg respectively. These values are lower than the tolerance level of 0.429 mg of lead and 0.055-0.070 mg/kg of cadmium. The analysis also was carried out to find the amount of pesticides like diazinon, malathion and sumithion. The pesticide residue analysis showed that the pesticides diazinon, malathion and sumithion were not present in the plant (the limit of detection of diazinon, malathion and sumithion is 0.02 mg/kg).

The nutritive analysis shows the nutrient content of senna. The microbial load of senna powder stored in glass and plastic containers was not detected for a period of 45 days. However, when stored for a period of 90 and 180 days, the microbial load was found to be 1×10^2 and 4×10^2 CFU in glass bottle and 2×10^2 and 6×10^2 CFU in plastic container respectively.

Toxic effect study of senna showed that the higher doses of senna powder (100, 200 & 400 mg/kg, p.o.) were found to be non-toxic and showed significant increase of faecal output ($p < 0.05$; $p < 0.001$, respectively) in rats. The highest administered dose (2000mg/kg) does not cause any effect in general behaviours and it does not produce any mortality which gives an indication for the safety of senna. Constipation produced by Loperamide was successfully reversed ($p < 0.001$) by senna powder (200 & 400 mg/kg, p.o.). 200 mg/kg, p.o., showed faecal output like the control animals.

REFERENCES

- Rao T. "Phytochemical study for the content of cassia angustifolia (senna)." *J Agri Food Chem*, 2004; **52**: 260:6673-6842.
- Kokate, C.K. Pharmacognancy, Irali Prakashan Publishers, Pune, 2004; 57-59.
- Potluri H.M.A. "Laxative effect of cassia senna." *Sau J Biol Sci*, 2002; **9**:12-22.
- Camoiez L, Nyman M and Yossuck P. "Measurement of low dietary fibre intake as a risk factor for chronic constipation in adult." *J Gastro Nutr*, 2002; **29**(2):132-135.
- Walter S, Hallbook O, Gotthard R, Bergmark M and Sjobahl R. "A population-based study on bowel habits in a Swedish community: prevalence of aecal incontinence and constipation." *Scand J Gastroenterol*, 2002; **37**: 911-916.
- Mitchell, J.M, Mengers U and Mc Pherson S. "An oral Carcinogenicity and toxicity study of Senna (Tinnevelly Senna fruits) in the rat." *Arch Toxi*, 2006; **80**(1): 34-44.
- Christenson B.V and Abdel-Latif I.A. "Colorimetric and Fluorometric Studies on the Borntrager reaction for anthraquinone drugs." *J Am Pharmal Ass*, 2006; **38**(9): 487-489.
- Mukherjee A, Mukhopadhyay M. J, Saha A. Dutta A and De B. "Genotoxicity of sennosides on the bone marrow cells of mice." *Food Chem Toxi*, 1998; **36**:937-940.
- Jozef S and Zmudzki J. "Determination of lead and cadmium in biological material by graphite furnace atomic absorption spectrometry method." *Bull Vet Inst Pulawy*, 2005; **49**:89-92.
- Acero J.L, Benitez F.J, Real F.J and Gonzalez M. "Oxidation of chlorfenvinphos in ultrapure and natural waters by ozonisation and photochemical method." *J Hazard Mater*, 2008; **153**: 320.
- Islam S, Hossain M.S, Nahar M, Mosihuzzaman and Mamun M.I.R. "Application of HPLC to the Analysis of Pesticide Residues in Eggplants." *J App Sci*, 2009; **9**(5): 973-977.
- Mutwally H.M.A and Meelad M.M.H. "A series in medical effect of wild plants of saudiArabia:2a- Effect of senna mutant Cassia senna on the spontaneous activity of locust foregut and hindgut muscles." 1997a; **67**.
- Dobbs H.E, Lane A.C and Macfarlane I.R. "Transport and mode of action of sennosides." *Pharma Sci*, 1975; **30**(2):147-158.
- Leng P.E "Acceleration of large intestine transit time in rats by sennosides and related compounds." *J Phar Pharm*, 1986a; **38**(5): 369-373.
- Nijs G, de-Witte P, van-Hoestenbergh A, Geboes K and Lemli J. "Direct and indirect evidence for the involvement of prostaglandins in the secretagogue action of rhein anthrone in the small intestine." *Acta Gastroenteroll Belgica* 1991; **54**(2): 184-190.
- Capasso F, Mascolo N, Autore G and Romano V. "Laxatives and the production of antacoids by rat colon." *J Pharm Pharmacol*, 1986; **38**: 627-629.
- Saito T, Mizutani F, Iwanaga Y, Morikawa K and Kato H. "Laxative and anti-diarreal activity of polycarbophil in mice and rats." *Jap J Pharma*, 2002 **89**: 133-141.
- Stool A, Becker B, and Kussamaul W. "The isolation of anthraglycosides from senna drugs." *Helvetica Chimica Acta*, 1949; **32**:1892-1903.
- Verma R.K, Uniyal G.C and Singh S.P. "Reverse-phase High Performance Liquid
J. Pure & Appl. Microbiol., **6**(1), March 2012.

- Chromotography of Sennosides in *Cassia angustifolia*.” *Phyto Analysis*, 1996; 7: 73-75.
20. Narres H.D, Carola M and Markus S. “Metal analysis in difficult materials with platform furnace Zeeman-Atomic Absorbtion Spectroscopy”. *Z Labnsm Unters Forsch*, 1985; **181**: 111-116.
 21. Koops J and Westerbeek D. “Determination of lead and cadmium in pasteurised liquid milk by flameless atomic absorption spectrophotometry”. *Neth Milk Dairy*, 1978; **32**: 149-169.
 22. Alda M.L, Kuster M and Barcelo D, “Mass spectrum Rev”. 2006; **25**: 900.
 23. WHO, WHO monographs on selected medicinal plants, volume 1, First Indian edition, A.I.T.B.S. Publishers and Distributors, Delhi, 2005; 256.
 24. Adams E, Shyu S and Brandle R. “Quality Evaluation”. *Indian J Nut Diet*, 2000; **39**(4): 194-195.
 25. Muller P and Basedow T. “Aflatoxin contamination of pods of Indian *Cassia senna* L. (*Caesal pinacea*) before harvest, during drying and in storage: Reasons and possible methods of reduction”. *J Stor Prod Res*, 2007; **43**: 323-329.