Journal of Stored Products and Postharvest Research Vol. 2(5) pp. 97 – 103, May 2011
Available online http://www.academicjournals.org/JSPPR
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Full Length Research Paper

Sennoside contents in Senna (*Cassia angustifolia Vahl.*) as influenced by date of leaf picking, packaging material and storage period

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Accepted 31 January 2011

A field experiment was conducted in the Senna (*Cassia angustifolia Vahl.*) during the Rabi season of 2008 to 2009 in the Dusty Area of the Department of Crop and Herbal Physiology, Jawaharlal Nehru Krishi Vishwa Vidyalaya. The experiment was carried out in a complete randomized design asymmetrical factorial with three treatments and three replications. *Cassia angustifolia* leaves were picked at three different stages of growth period viz. 90, 110 and 130 DAS (days after sowing) with different type of packaging material and storage period. The Sennoside A and B were determined by High Performance Thin Layer Chromatography. The Sennoside A and B content were found to be highest in leaf picking stage of 90 DAS followed by 130 and 110. Maximum contents of Sennoside A and B were estimated in black polythene followed by aluminum foil bag and transparent polythene.

Key words: Senna, *Cassia angustifolia,* Sennoside A and B, leaf picking stage, HPTLC.

INTRODUCTION

Cassia angustifolia (family Caesalpiniacea) popularly known as Senna, is a valuable plant drug in Ayurveda and modern system of medicine for the treatment of constipation (Atal and Kapoor, 1982; Das et al., 2003; Martindale, 1977; Sharma, 2004). Cultivation of Senna does not require much expenditure on irrigation, manuring, pesticides, protection and other pre- and post harvest care. This makes the plant ideal crop for acid regions where water provision, wasteland development, desertification control and sand dune stabilization are the major challenges (Tripathi, 1999). Senna is a sun-loving crop and requires bright sunshine for its successful growth. The crop is raised from seed and has a hard and tough seed-coat, ascertain amount of abrading of its surface is necessary to induce quick germination. The Sennosides had been extracted from Senna leaves, stems, pods, buds and flowers but no Sennosides were found in the seeds.

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Sennoside levels in native Senna plants were similar to those found growing in other countries (Babash et al., 1985). The laxative property of Senna is based on two glycosides viz. Sennoside A and sennoside B whereas Sennosides C and D have also been reported in the plant. These Sennosides are the aloe-emodin diantraone diglucosides. Apart from Sennosides, the pod and leaf also contain glycosides of anthraquinones, rhein and chrysophenic acid. Recently, two naphthalene glycosides have also been isolated from leaves and pods (Gupta, 2008). The flavanoids that are reported from this plant are Kaenferol, Kaempterin and isorhamnetin. It also contains beta-sitosterol (0.33%). From the germplasm collected in Tamil Nadu, Andhra Pradesh and Gujarat, India the superior accession 'Sona' was derived, following evaluation and selection for leaf and pod yield. Cultivar Sona (released in 1997) is 115 to 120 cm tall, leafy (mean leaf area 11.28 cm^2), highly branched, flowering after 100 to 115 days, large number of pods $plant^{-1}$ (5 cm long and 1.25 to 1.75 cm wide). Dry leaf and pod yield is 11 and 4 quintals ha^{-1}, respectively, while sennoside content average 3.5% (Singh et al., 1997). The deflowering

Table 1. Effect of storage material and duration on the content of Sennoside A and B in Senna leaves picked at 90 DAS.

Storage period	Packaging materials					
	Sennoside A (%)			Sennoside B (%)		
Month	P_1	P_2	P_3	P_1	P_2	P_3
S_0 (0) Initial content		0.126			0.069	
S_1 (1)	0.123	0.117	0.113	0.067	0.065	0.064
S_2 (2)	0.113	0.110	0.107	0.063	0.061	0.060
S_3 (3)	0.100	0.095	0.088	0.057	0.054	0.055
S_4 (4)	0.088	0.077	0.081	0.048	0.046	0.047
S_5 (5)	0.065	0.060	0.058	0.043	0.041	0.042
S_6 (6)	0.054	0.045	0.045	0.036	0.034	0.035

P1- Black polythene bags, P2- Aluminum foil bags, P3- Transparent polythene bags.

increased Sennoside A and B concentration (percent dry weight) in leaves by 25%, total leaf dry mass by 63% and harvest index by 22% with the result that the Sennoside A and B yield (grams) per plant doubled in response to flowering. During the day time, net photosynthesis remained consistently lower in the deflowered plants. Youngest leaves had the greatest Sennoside A and B concentration. A clone raised from cuttings of one seedling had lower Sennoside A: B ratio than the plants raised from the seedlings (Ratnayaka et al., 2002). In the traditional medicine, *C. angustifolia* is used for its purgative properties. A new anthraquinone glycoside (emodin 8-O-sophoroside), and seven known glycosides, were isolated from the leaves of *C. angustifolia* and their structures were elucidated by spectral analysis (Kinjo et al., 1994). The *Cassia-angustifolia* is used in traditional medicine for its cathartic properties and in the treatment of tumours. Kaempferol 3-O- beta -glucoside and isorhamnetin 3-O-beta -glucosides were newly isolated in its leaves (Singh et al., 1995). No study was carried out about the growth stages on which the crop should be harvested to obtain the maximum sennoside content and how long its storage durability. Therefore, the said study was undertaken to evaluate the effect of different growth stages on sennoside content of Senna and after that its post-harvest technology. The main objectives were to observe the sennoside content in Senna leaves at different dates of leaf picking and the impact of packaging materials and storage periods.

MATERIALS AND METHODS

The experiment was conducted in the Department of Crop and Herbal Physiology at the dusty area, Jawaharlal Nehru Krishi Vishwa Vidyalaya Jabalpur, Madhya Pradesh during the year 2008 to 2009. The experiment was carried out in a complete randomized design (CRD) asymmetrical factorial with three treatments and three replications. The sowing date was 3rd July, 2008. The Leaf - picking stages (day after sowing) were 90 days (G_1), 110 days (G_2) and 130 days (G_3). The packaging materials (storage containers) were black polythene (P_1), aluminum bag (P_2) and transparent polythene (P_3). The storage time (duration after packaging) were having one month intervals up to six month (S_1 to S_6). The Sennoside A and B (%) were estimated by High Performance Thin Layer Chromatography (Rajpal, 2002). The statistical analysis of data taken on different variables was carried out through CRD asymmetrical factorial design to know the degree of variation among the treatments.

RESULTS AND DISCUSSION

Initial observation before packaging

The initial content of Sennoside A and B in Senna leaves were estimated before packaging to be 0.126 and 0.069%, 0.112 and 0.064%, 0.114 and 0.066% at 90%, 110% and 130% DAS respectively.

Storage and leaf picking effect on Sennoside A and B

The analysis of Sennoside was carried out in different types of packaging materials and storage periods. The results are presented in Tables 1 to 4 shows there were significant differences in the Sennoside levels in the different containers. The highest content of Sennoside A and B were 0.123 and 0.067%, 0.110 and 0.063%, 0.113 and 0.064% in leaves picked at 90%, 110% and 130% DAS respectively in black polythene bag (P_1). During the storage minimum content of Sennoside A and B that is, 0.113 and 0.064%, 0.100 and 0.060%, 0.108 and 0.062% were recorded in transparent polythene bag (P_3) at 90%, 110% and 130% DAS respectively. Hence the Sennosides content were highest in black polythene bag (P_1), followed by aluminium foil bag (P_2) and the least in transparent polythene bag (P_3). After one month of storage, Sennoside A and B

Table 2. Effect of storage material and duration on the content of Sennoside A and B in Senna leaves picked at 110 DAS.

Storage period (Month)	Packaging materials					
	Sennoside A (%)			Sennoside B (%)		
	P_1	P_2	P_3	P_1	P_2	P_3
S_0 (0) Initial content		0.112			0.064	
S_1 (1)	0.110	0.105	0.100	0.063	0.061	0.060
S_2 (2)	0.101	0.095	0.090	0.057	0.054	0.053
S_3 (3)	0.095	0.091	0.087	0.054	0.052	0.050
S_4 (4)	0.080	0.074	0.068	0.045	0.043	0.042
S_5 (5)	0.061	0.056	0.054	0.041	0.040	0.039
S_6 (6)	0.050	0.044	0.042	0.034	0.033	0.031

P_1- Black polythene bags, P_2- Aluminum foil bags, P_3- Transparent polythene bags.

Table 3. Effect of storage material and duration on the content of Sennoside A and B in Senna leaves picked at 130 DAS.

Storage period (Month)	Packaging materials					
	Sennoside A (%)			Sennoside B (%)		
	P_1	P_2	P_3	P_1	P_2	P_3
S_0 (0) Initial content		0.114			0.066	
S_1 (1)	0.113	0.110	0.108	0.064	0.062	0.062
S_2 (2)	0.102	0.100	0.097	0.060	0.057	0.055
S_3 (3)	0.095	0.089	0.087	0.055	0.053	0.051
S_4 (4)	0.086	0.084	0.070	0.047	0.044	0.043
S_5 (5)	0.064	0.061	0.052	0.042	0.041	0.040
S_6 (6)	0.052	0.041	0.040	0.035	0.033	0.032

P_1- Black polythene bags, P_2- Aluminum foil bags, P_3- Transparent polythene bags.

Table 4. Interaction of packaging material and storage period on content of Sennoside A and B.

Treatments	Sennoside A (%)			Sennoside B (%)		
	SEd	SEm	CD 5%	SEd	SEm	CD 5%
G	0.0002	0.0002	0.0004	0.0002	0.0001	0.0004
P	0.0002	0.0002	0.0004	0.0002	0.0001	0.0004
GXP	0.0004	0.0003	0.0007	0.0003	0.0002	0.0007
S	0.0003	0.0002	0.0006	0.0003	0.0002	0.0005
GXS	0.0005	0.0004	0.0011	0.0005	0.0003	0.0009
PXS	0.0005	0.0004	0.0011	0.0005	0.0003	0.0009
GXPXS	0.0009	0.0007	0.0018	0.0008	0.0006	0.0016

G - Growth stages, P - Packaging materials, S - Storage period.

contents were maximum in leaves picked at 90, 110 and 130 DAS respectively in P_1 and were found at par with P_2 followed by P_3. At two months, of storage Sennoside A and B contents were found maximum with (0.113 and 0.063%, 0.101 and 0.057%, 0.102 and 0.060%) in leaves picked at 90, 110 and 130 DAS respectively in P_1 and were statistically at par with P_2 and P_3. Minimum Sennoside A and B were found observed in transparent polythene bags with values of 0.107 and 0.060%, 0.090 and 0.053%, 0.097 and 0.055%) for leaves picked at 90, 110 and 130 DAS respectively.

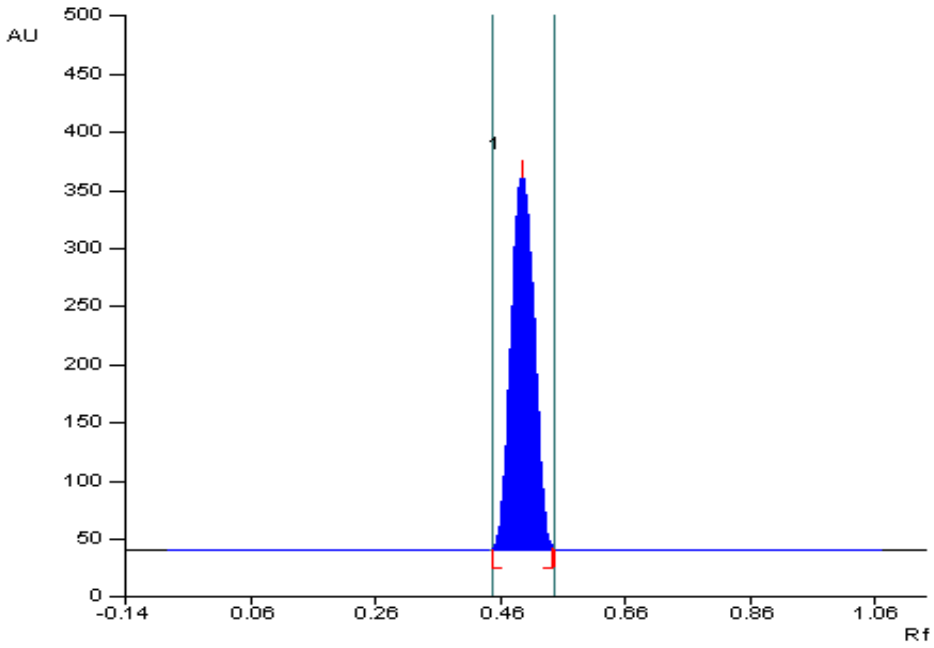

Figure 1. Chromatogram of Sennoside A standard.

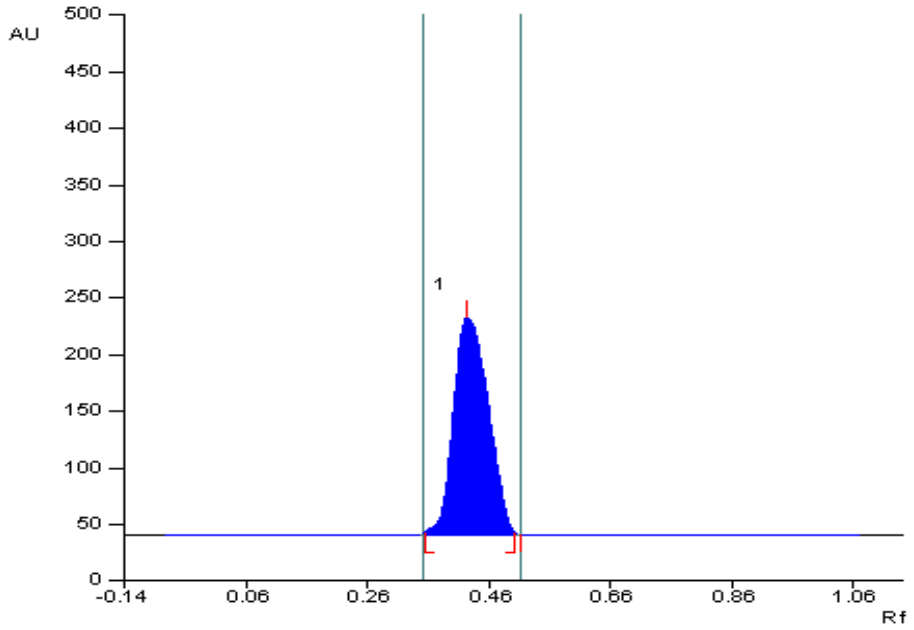

Figure 2. Chromatogram of Sennoside A content in leaf picked at 90 DAS and stored in black polyethylene bag at the 1st month.

At three month of storage, the significantly highest Sennoside A and B contents that is, 0.100 and 0.057%, 0.095 and 0.054%, 0.095 and 0.055% at 90, 110 and 130 DAS respectively were estimated in black polythene bags and least were (0.088 and 0.055%, 0.087 and 0.050%, 0.087 and 0.051%) at 90, 110 and 130 DAS respectively noted in transparent polythene bags P_3 (Figures 1 to 6).

At four month of storage, the maximum contents of Sennoside A and B were recorded to be that is, 0.088 and 0.048%, 0.080 and 0.045%, 0.086 and 0.047% at 90, 110 and 130 DAS respectively in

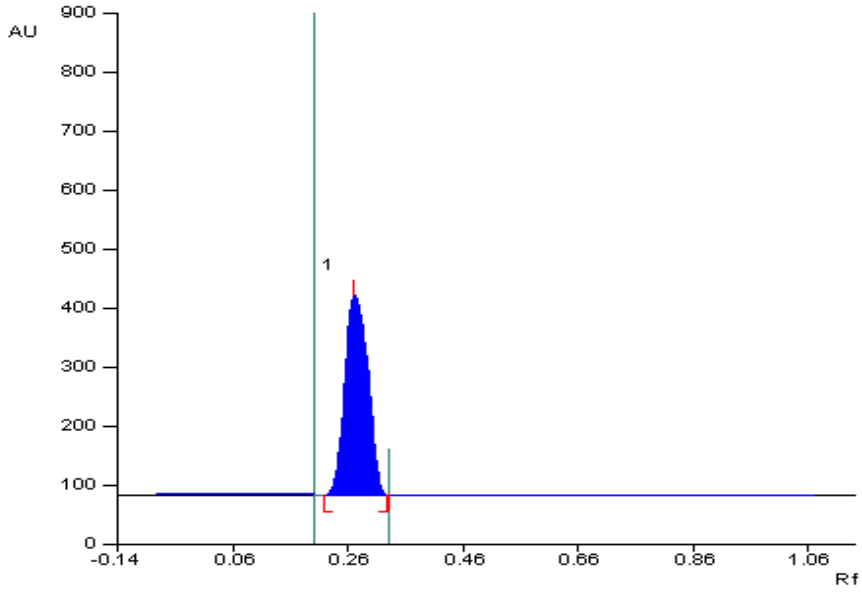

Figure 3. Chromatogram of Sennoside A content in leaf picked at 90 DAS and stored in a black polyethylene bag at the 6th month.

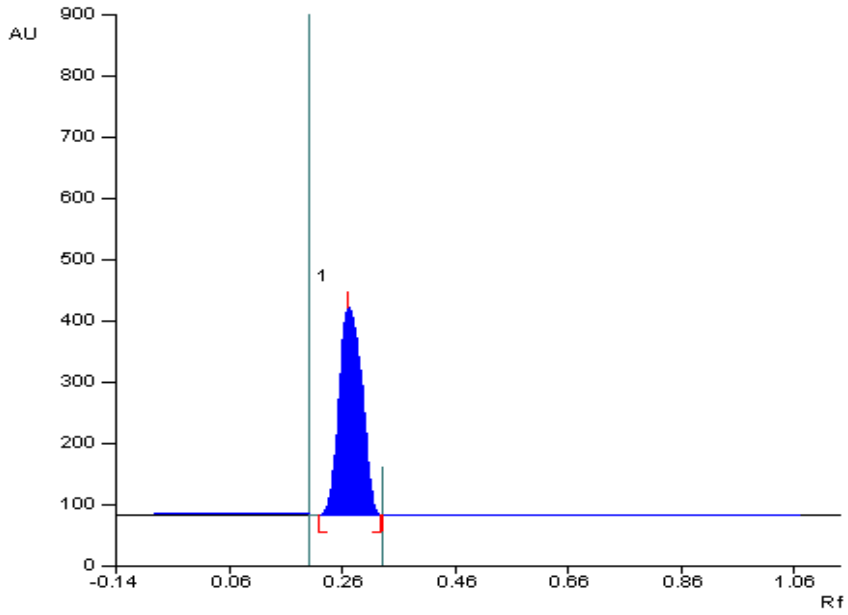

Figure 4. Chromatogram of Sennoside B standard.

black polythene P_1. The least contents were observed in transparent polythene bags (0.081 and 0.047%, 0.068 and 0.042%, 0.070 and 0.043%) in leaves picked at 90, 110 and 130 DAS.

At the fifth and sixth months, of storage period, the trend continued with leaves stored in black polythene bags having the maximum Sennoside A and B levels whereas the lowest was found in the transparent polythene bags. Though these values were lower than what was obtained in the previous months.

The results revealed that Sennoside A and B contents were significantly reduced in different storage containers that is, P_1, P_2 and P_3 (6.17 and 10.46%). Furthermore, it was found that the Sennoside A and B were significantly reduced with

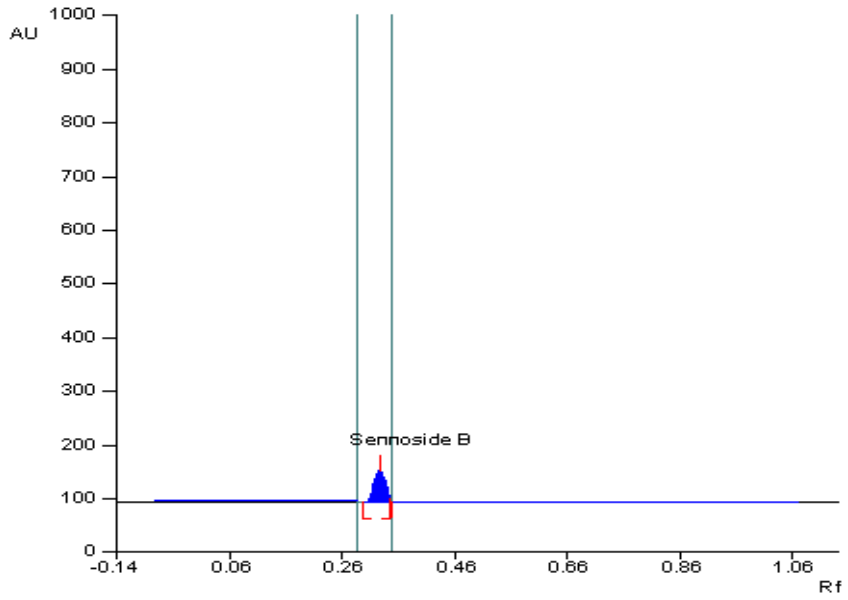

Figure 5. Chromatogram of Sennoside B content in leaf picked at 90 DAS and stored in a black polythene bag at the 1st month.

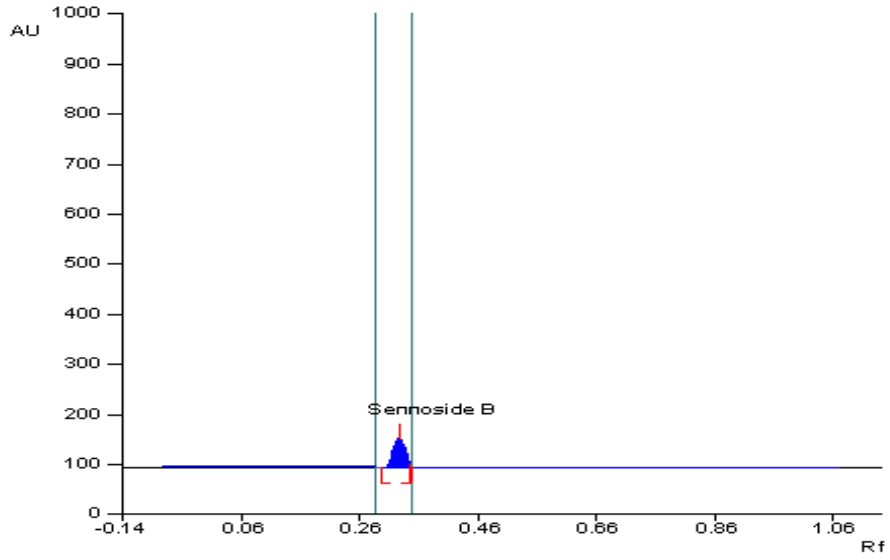

Figure 6. Chromatogram of Sennoside B content in leaf picked at 90 DAS and stored in a black polythene bag at the 6th month.

increasing storage period from the first month till the sixth month. The Sennoside A and B in all three leaves picking (90 DAS, 110 DAS and 130 DAS) were packed in different three packaging materials that is, black polythene bags (P_1), aluminum foil bags (P_2) and transparent polythene bags (P_3) and stored for six month. Maximum content of Sennosides A and B was obtained in freshly harvested Senna leaves but was found to decline significantly for rest of the storage period up to six months. The losses of Sennosides A and B in storage were found to be lowest in black polythene bags as compared to aluminum foil and transparent polythene bags. The storage of leaf should be protected from light and moisture (European Pharmacopoeia, 2005). Srivastava, et al. (1980) have reported that the glycosides is more in the leaves than in pods and it decreases with the age during the growth of

plant, thus harvesting of the leaf crop between 40 and 70 days improved biological activity to the maximum in the crude drug. It is also found that prolonged exposure of growing Senna plants to cycle light and darkness affects the biosynthesis of anthracene derivatives in their leaves. They also found that in the absence of light (after 48 h), the conversion of non-rhein to rhein is suppressed as indicated by high proportion of non-rhein glycosides to total glycosides. Pareek, et al. (1983) has reported that total Sennosides content in stored produce fall after one year. Similar decrease was reported by Anonymous (2006) of total alkaloid content during storage in medicinal roots. Storage methods have also been reported to have a significant effect on saponin content. Decrease in saponin contents was observed at the later period of storage. Significantly higher saponin contents were retained due to storage in polythene bags. Similarly, Adom et al. (1996) reported that the powdered dry okra kept in polythene package survived storage best. So the package material and storage time significantly affected the quality parameters.

CONCLUSIONS

1) The Sennoside content in leaves was found highest in those picked at 90 DAS and was lowest at 110 days after sowing. So it is suggested that the senna leaves should be harvested at 90 days after sowing to get maximum Sennosides contents.
2) The best packaging material for storage of Senna leaves was found to be black polyethylene bags, then aluminum foil bags and transparent polyethylene bags. The reduction of Sennoside was found to be lowest in black polyethylene bags. Further, the storage of the leaves should be done in black polythene or in any dark container so that the deterioration of Sennoside should be minimum.
3) In conclusion, it is suggested that the Senna leaves should be harvested at 90 days after sowing to get maximum Sennosides contents. Secondly, dark-coloured packaging material should be used to store in order to minimize the degradation of Sennosides.

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