

Deterioration of chemical constituents in roots of drug *Stereospermum chelonoides* DC. under storage

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ABSTRACT

*In the present study fresh and market roots of drug *Stereospermum chelonoides* DC. were analyzed for study in changes of chemical constituents under storage. Root samples were stored under different 30, 50, 75, 96 and 100 % relative humidity and different incubation days 15, 30, 45 and 60 days. Quantitative estimation of carbohydrates, proteins and phenols in fresh and market roots was done. The results indicated that biodeterioration of selected chemical constituents were observed under high relative humidities 75, 96 and 100% RH and with increased incubation days (45 and 60). More deterioration of chemical constituents recorded in case of market samples as compared to fresh samples. Analysis of variance also showed that the effect of relative humidity and incubation days on biodeterioration of chemical constituents amount were significant.*

Key words: deterioration, relative humidity, incubation days, *Stereospermum chelonoides*

INTRODUCTION

Stereospermum chelonoides (Syn: *S. suaveolens*) is a large sized tree, deciduous, branches and usually 9 to 10 m tall and distributed in sub Himalayan tract, central parts of India. It is commonly called as "Patla and "Padri" and belongs to the "Bignoniaceae" family. This drug is famous for medicinal uses, such as decoction of the root is antipyretic and it is useful in asthma, cough and excessive thirst. The bark and all parts contain a naphthaquinone and lepachol. The wood also contains lepachonone. All the parts are used in medicine. Flowers are used in bleeding disease, sore throat and diarrhoea; fruits are useful in blood diseases. The root-bark is an ingredient of Dashmoola and it is regarded as cooling, astringent cardio tonic, bitter, diuretic and tonic and generally used in combination with other medicine the ashes of this plant are used in the preparation of alkaline water and caustic pastes. Fruits are useful in hic cough and blood diseases (Negi, 2000). If storage of medicinal plant is not properly there may be possibility of contamination of different organism and these microorganisms and their growth are responsible for the deterioration and changes of chemical constituents. Therefore, it is necessary to study the changes in chemical constituents in roots of this drug. So that, fresh and market roots of this plant stored at various relative humidity 30, 50, 75, 96 and 100% RH for different incubation periods 15, 30, 45, 60, 75 and 90 days. The effect of various relative humidity and incubation days on changes in chemical constituents studied.

MATERIALS AND METHODS

The fresh roots of drug *Stereospermum chelonoides* DC. were collected in healthy, flowering and fruiting conditions from different localities. Besides the genuine drugs, market survey has also been carried out for the collection of the market (bazaar) drug samples. These drug samples were collected from various Kashthausadhi shopkeepers. The

collected samples were brought to the laboratory, maintained and stored in separate polyethylene bags to avoid further aerial contamination. For study to deterioration of chemical constituents, organ form of roots cut to small pieces and were stored in small muslin clothes bags under different level of RH i.e. 30, 50, 75, 96 and 100 % RH for 90 days and at $28 \pm 3^\circ\text{C}$ temperature (Wink and Sears, 1950). At an interval of 15 days, root samples were taken out and thoroughly washed with distilled water and were dried in oven for chemical analysis. Chemical analysis was estimated by the procedure described by Lowry *et al.* (1951) for total protein, Singh *et al.* (1978) for total phenols. Anthrone methods for Total sugars (TS) and Dinitrosalicylic acid (DNSA) method for Reducing sugar (RS) amount were also followed (Sadasivam and Manickam, 1992). Simple correlation were run between selected parameters using Statistical Package for Social Science (SPSS) software in which statistical significance was determined at 0.05 % probability levels.

RESULTS

Fresh roots of *S. chelonoides* stored at various relative humidity in different incubation days 15, 30, 45, 60, 75 and 90 days. The control roots contained 51.30% TS and 8.23% RS. After 15 days of incubation period value of TS under 30 and 50 % RH didn't change but value of RS observed 8.19 %, at same RH after 90 days the value of TS deteriorated to 49.57 % and value of RS reached to 8.09 in both of RH. In cases of 75, 96 and 100 % RH after 15 days value of TS and RS observed 51.13, 8.14%; 51.09, 8.09% and 50.96, 8.05% respectively. These contents of TS and RS deteriorated to 49.53, 7.69 %; 48.73, 7.28 % and 48.35, 7.28 % at 75, 96 and 100 % RH (Table1). Market sample of this drug after 15 days of incubation showed minimum deterioration under all tested RH 30, 50, 75, 96 and 100% RH, 40.05, 6.38; 32.40, 6.24%; 30.93, 5.88; 29.99, 5.47% and 29.41, 5.38% respectively. Maximum deterioration of total sugars and reducing sugars showed after 90 days of incubation, they include: 27.77, 4.07%; 27.52, 3.25%; 26.70, 2.35%. More reduction of sugar contents observed at 96 and 100 % RH after 90 days, 11.36, 1.85% and 7.82, 1.44% (Table 2).

Table 1: Deterioration of Total sugars (TS) and Reducing sugars (RS) content (mg/100mg) in root of *Stereospermum chelonoides* (Fresh sample) at different relative humidities

Incubation days	Control		30%		50%		75%		96%		100%	
	TS	RS	TS	RS	TS	RS	TS	RS	TS	RS	TS	RS
1 day	51.30 ±3.78	8.23 ±0.16	51.30 ±3.78	8.23 ±0.16	51.30 ±3.78	8.23 ±0.16	51.30 ±3.78	8.23 ±0.16	51.30 ±3.78	8.23 ±0.16	51.30 ±3.78	8.23 ±0.16
15 days	51.30 ±3.78 ^c	8.23 ±0.16 ^c	51.30 ±3.78 ^c	8.19 ±0.19 ^c	51.30 ±3.78 ^c	8.19 ±0.17 ^c	51.13 ±3.77 ^b	8.14 ±0.14 ^b	51.09±3.8 ^a	8.09 ±0.10 ^a	50.96 ±3.7 ^a	8.05 ±0.20 ^a
30 days	51.30 ±3.78 ^c	8.23 ±0.16 ^c	48.19 ±8.94 ^c	8.19 ±0.19 ^c	51.17 ±3.81 ^c	8.14 ±0.21 ^c	50.75 ±3.50 ^b	8.09 ±0.21 ^b	50.54 ±3.66 ^a	7.87 ±0.22 ^a	50.21 ±3.79 ^a	7.69 ±0.069 ^a
45 days	51.30 ±3.78 ^c	8.23 ±0.16 ^c	50.96 ±3.94 ^c	8.14 ±0.17 ^c	50.75 ±3.50 ^c	8.14 ±0.22 ^c	50.29 ±3.11 ^b	7.91 ±0.18 ^b	49.91 ±3.89 ^a	7.69 ±0.090 ^a	49.70 ±3.67 ^a	7.64 ±0.17 ^a
60 days	51.30 ±3.78 ^c	8.23 ±0.16 ^c	49.57 ±3.71 ^c	8.09 ±0.22 ^c	49.57 ±3.55 ^c	7.91 ±0.27 ^c	49.53 ±3.42 ^b	7.69 ±0.069 ^b	48.73 ±3.17 ^a	7.28 ±0.24 ^a	48.35 ±3.39 ^a	7.28 ±0.045 ^a
75 days	51.30 ±3.78 ^c	8.23 ±0.16 ^c	48.73 ±3.17 ^c	7.78 ±0.27 ^c	48.44 ±3.36 ^c	7.69 ±0.30 ^c	48.75 ±3.20 ^b	7.23 ±0.11 ^b	47.03 ±2.86 ^a	6.92 ±0.23 ^a	45.05 ±1.94 ^a	6.78 ±0.026 ^a
90 days	51.30 ±3.78 ^c	8.23 ±0.16 ^c	47.60 ±2.95 ^c	7.69 ±0.052 ^c	47.18 ±3.17 ^{bc}	7.23 ±0.25 ^{bc}	43.81 ±0.76 ^b	6.78 ±0.19 ^b	39.98 ±0.84 ^a	6.33 ±0.069 ^a	39.07 ±1.08 ^a	5.79 ±0.23 ^a

Data are the mean of three replicates ± standard deviation. P- Value denoted the significance of differences between the mean by univariate comparison statistics. The value followed by different letters differ significantly by Duncan's multiple rang test at $P = \text{Sig} = 0.05$

Fresh and market roots of *S. chelonoides* contained 13.75 and 13.25% total proteins at the first day (Table 3). Under 30, 50 % RH showed minimum loss in protein contents as compared to 75, 96 and 100 % RH showed maximum deterioration in protein values after different incubation days. In case of 30 and 50 % RH after 15 days showed 13.08, 11.75 %; 12.85, 12.7 % in fresh and market sample respectively. These values reduced to 10.41, 9.16%; 7.09, 5.55 % after 90 days of incubation in both of kind of samples, respectively. In cases of 96 and 100% RH after 15 days of storage the value of total proteins showed 10.86, 9.66%; 7.63, 6.26% but after 90 days of storage these amounts deteriorated to 6.80, 5.16% and 3.18, 3.05 % in fresh and market sample, respectively.

Table 2: Deterioration of Total sugars (TS) and Reducing sugars (RS) content (mg/100mg) in root of *Stereospermum chelonoides* (Market sample) at different relative humidities

Incubation days	Control		30%		50%		75%		96%		100%	
	TS	RS	TS	RS	TS	RS	TS	RS	TS	RS	TS	RS
1 day	40.44 ±0.19	6.47 ±0.14	40.44 ±0.19	6.47 ±0.14	40.44 ±0.19	6.47 ±0.14	40.44 ±0.19	6.47 ±0.14	40.44 ±0.19	6.47 ±0.14	40.44 ±0.19	6.47 ±0.14
15 days	40.44 ±0.19 ^c	6.47 ±0.14 ^c	40.05 ±1.33 ^c	6.38 ±0.11 ^c	32.40 ±0.51 ^c	6.24 ±0.18 ^c	30.93 ±0.45 ^b	5.88 ±0.069 ^b	29.99 ±0.072 ^{ab}	5.47 ±0.22 ^{ab}	29.41 ±0.12 ^a	5.38 ±0.13 ^a
30 days	40.44 ±0.19 ^d	6.47 ±0.14 ^d	36.61 ±0.62 ^c	5.83 ±0.30 ^c	31.94 ±0.38 ^c	5.52 ±0.069 ^c	30.68±0.21 ^b	5.33 ±0.069 ^b	29.04 ±4.04 ^{ab}	5.20 ±0.11 ^{ab}	19.19 ±0.91 ^a	5.02 ±0.17 ^a
45 days	40.44 ±0.19 ^c	6.47 ±0.14 ^c	35.60 ±1.07 ^c	5.92 ±0.40 ^c	35.22 ±0.73 ^c	5.33 ±0.11 ^c	28.91±0.69 ^b	5.20 ±0.18 ^b	25 ±0.52 ^{ab}	5.02 ±0.13 ^{ab}	15.15 ±0.78 ^a	4.43 ±0.21 ^a
60 days	40.44 ±0.19 ^d	6.47 ±0.14 ^d	33.83 ±1.90 ^c	5.42 ±0.29 ^c	33.08 ±1.8 ^c	5.02 ±0.069 ^c	20.04±1.20 ^b	4.88 ±0.23 ^b	15.02 ±0.33 ^{ab}	4.52±0.069 ^{ab}	12.62 ±0.69 ^a	4.07 ±0.045 ^a
75 days	40.44 ±0.19 ^c	6.47 ±0.14 ^c	30.68 ±0.64 ^c	4.57 ±0.43 ^c	29.29 ±0.52 ^{bc}	4.43 ±0.21 ^b	26.51 ±0.76 ^b	3.71 ±0.069 ^b	12.75 ±0.072 ^a	3.16 ±0.23 ^{ab}	11.61 ±0.52 ^a	2.66 ±0.24 ^a
90 days	40.44 ±0.19 ^c	6.47 ±0.14 ^c	27.77 ±1.75 ^c	4.07 ±0.36 ^c	27.52 ±1.90 ^{bc}	3.25 ±0.25 ^{bc}	26.50 ±1.90 ^{ab}	2.35 ±0.23 ^{ab}	11.36 ±0.12 ^a	1.85 ±0.045 ^a	7.82 ±0.66 ^a	1.44 ±0.23 ^a

Data are the mean of three replicates ± standard deviation. P- Value denoted the significance of differences between the mean by univariate comparison statistics. The value followed by different letters differ significantly by Duncan's multiple range test at P=Sig= 0.05

Table3: Deterioration of phenols content (mg/100mg) in root of *Stereospermum chelonoides* (Fresh and market samples) at different relative humidities

Incubation days	Control	30%	50%	75%	96%	100%
1 day	9.58±0.039	9.58±0.039	9.58±0.039	9.58±0.039	9.58±0.039	9.58±0.039
15days	9.58±0.030 ^a	9.58±0.11 ^b	9.58±0.019 ^a	9.58±0.022 ^a	9.58±0.040 ^a	9.62±0.55 ^a
30days	9.58±0.019 ^c	9.58±0.030 ^b	9.54±0.019 ^a	9.46±0.030 ^a	9.44±0.030 ^a	9.38±0.040 ^a
45 days	9.58±0.019 ^d	9.56±0.011 ^d	9.52±0.019 ^d	9.42±0.030 ^c	9.31±0.034 ^b	9.23±0.085 ^a
60 days	9.53±0.034 ^e	9.44±0.060 ^c	9.27±0.085 ^c	9.29±0.030 ^c	9.09±0.030 ^b	8.91±0.078 ^a
75 days	9.54±0.052 ^e	9.27±0.10 ^d	9.055±0.15 ^d	8.81±0.10 ^e	8.20±0.19 ^b	7.89±0.097 ^a
90 days	9.52±0.069 ^e	9.055±0.04 ^d	8.68±0.12 ^d	8.20±0.20 ^c	7.81±0.101 ^b	7.48±0.097 ^a

Incubation days	Control	30%	50%	75%	96%	100%
1 day	4.48±0.39	4.48±0.39	4.48±0.39	4.48±0.39	4.48±0.39	4.48±0.39
15days	4.47±0.032 ^b	4.13±0.30 ^b	3.74±0.24 ^a	3.52±0.13 ^a	3.48±0.19 ^a	3.50±0.37 ^a
30days	4.47±0.16 ^d	3.75±0.20 ^c	3.75±0.41 ^c	3.34±0.12 ^b	3.16±0.54 ^b	2.17±0.013 ^a
45 days	4.45±0.21 ^d	2.18±0.12 ^d	3.56±0.11 ^c	3.18±0.11 ^c	3.12±0.50 ^b	1.99±0.041 ^a
60 days	4.45±0.35 ^d	3.32±0.108 ^c	3.07±0.099 ^c	3.7±0.152 ^c	2.55±0.34 ^b	1.80±0.041 ^a
75 days	4.45±0.068 ^b	3.01±0.10 ^{ab}	2.55±0.131 ^{ab}	2.34±0.187 ^{ab}	1.96±0.115 ^{ab}	1.79±0.67 ^a
90 days	4.45±0.69 ^d	2.79±0.022 ^c	2.18±0.11 ^c	1.96±0.22 ^{bc}	1.77±0.09 ^{ab}	1.59±0.027 ^a

Data are the mean of three replicates ± standard deviation. P- Value denoted the significance of differences between the mean by univariate comparison statistics. The value followed by different letters differ significantly by Duncan's multiple range test at P=Sig= 0.05

Table 4: Deterioration of proteins content (mg/100mg) in root of *Stereospermum chelonoides* (Fresh and market samples) at different relative humidities

Incubation days	control	30%	50%	75%	96%	100%
1 day	13.75±0.74	13.75±0.74	13.75±0.74	13.75±0.74	13.7 ±0.74	13.7 ±0.74
15days	13.75±0.85 ^c	13.08±0.38 ^c	11.75±0.52 ^b	11.38±0.37 ^b	10.86±0.24 ^b	9.66±0.044 ^a
30days	13.70±0.36 ^d	12.79±0.35 ^d	11.11±0.86 ^{bc}	11.11±0.16 ^c	9.84±0.36 ^{ab}	9.45±0.076 ^a
45 days	13.78±0.69 ^d	11.15±0.78 ^{cd}	9.73±0.22 ^c	9.59±0.12 ^c	8.33±0.081 ^b	5.94±0.068 ^a
60 days	13.75±0.89 ^d	10.09±0.70 ^d	9.58±0.35 ^d	8.34±0.080 ^c	7.50±0.15 ^b	5.56±0.013 ^a
75 days	13.74±0.36 ^d	10±0.59 ^d	9.30±0.50 ^e	7.66±0.48 ^b	7.22±0.16 ^b	5.55±0.089 ^a
90 days	13.70±0.65 ^d	10.41±0.37 ^c	9.16±0.63 ^c	7.52±0.63 ^b	6.80±0.068 ^b	5.16±0.089 ^a

Incubation days	control	30%	50%	75%	96%	100%
1 day	13.25±0.45	13.25±0.45	13.25±0.45	13.25±0.45	13.25±0.45	13.25±0.45
15days	13.25±0.40 ^e	12.85±0.40 ^c	12.7±0.043 ^c	11.75±2.38 ^c	7.63±0.34 ^{ab}	6.26±0.019 ^a
30days	13.25±0.58 ^d	12.91±0.45 ^c	11.43±0.46 ^c	11.5±0.13 ^c	7.65±1.20 ^a	5.56±0.16 ^a
45 days	13.25±0.96 ^d	8.34±0.75 ^{cd}	7.37±0.32 ^{bc}	5.97±0.52 ^c	5.13±1.32 ^{ab}	4.79±0.88 ^a
60 days	13.25±0.36 ^d	7.36±0.125 ^d	6.09±0.575 ^c	5.58±0.156 ^b	4.18±0.15 ^b	4.01±0.14 ^a
75 days	13.25±0.68 ^d	7.22±0.089 ^d	5.68±0.75 ^c	4.27±0.74 ^{bc}	4.13±0.073 ^b	3.47±0.20 ^a
90 days	13.25±0.79 ^d	7.09±0.076 ^c	5.55±0.72 ^c	4.16±0.65 ^c	3.18±0.12 ^a	3.05±0.17 ^a

Data are the mean of three replicates ± standard deviation. P- Value denoted the significance of differences between the mean by univariate comparison statistics. The value followed by different letters differ significantly by Duncan's multiple range test at P=Sig= 0.05

Fresh roots of *S. chelonoides* stored at various relative humidity in different incubation days 15, 30, 45, 60, 75 and 90 days. The control roots contained 9.58% total phenols. After 15 days of incubation period value of total phenols under 30 and 50 % RH observed 9.58%, but this amount increased to 9.62 % under 100%RH but after that gradually decreased while after 90 days the value of total phenols deteriorated to 9.055 and 8.68 %. In cases of 75, 96 and 100% RH after 15 days value of phenols amount observed 9.58, 9.58 and 9.62 %, respectively. These contents deteriorated to 8.20, 7.81, 7.48%, at 75, 96 and 100% RH. Market sample of this drug after 15 days of incubation showed minimum deterioration under all tested RH 30, 50, 75, 96 % RH, 4.13, 3.74, 3.52 and 3.48 %, respectively but in case of 100%RH increased to 3.50%. Maximum deterioration of phenols showed after 90 days of incubation, they include: 2.79, 2.18, 1.96, 1.77 and 1.59% (Table 4).

Data analysis of carbohydrates, proteins and phenol amounts indicated that there is a significant correlation between incubation days and relative humidity with deterioration of sugar amount at 5% levels of significance (P value <0.05).

DISCUSSION

Each herb contains large number of diverse compounds and it is not possible to analyze for presence or absence for all compounds. Medicinal plants may be associated with a broad variety of microbial contaminants, represented by bacteria, fungi and viruses. Inevitably, this microbiological background depends on several environmental factors and exerts an important impact on the overall quality of herbal products and preparations. During storage, the fungal organisms thrive on drug plants by utilizing various components including the active ingredients and changes in therapeutically active content such as carbohydrates, alkaloids, phenols, glycosides and proteins of plant parts like roots, barks, stems, fruits and seeds are influenced both by fungi as well as physical factors (Roy, 2003 and Wallace *et al.* 1976). Other factors such as use of fresh or dried plants, season, light exposure, water availability, nutrients period, time of collection, storage, transportation of raw material, age and part of the plant collected, and methods of collecting, drying and packaging can greatly affect the quality and consequently the therapeutic value of herbal medicines deteriorates. These factors also account for variability of individual constituents in herbal preparations. Unscientific drying, storage methods and good environmental conditions favor association of various microbes with stored products.

CONCLUSION

The result of this investigation indicate that relative humidities 75, 96 and 100% RH show the significant reduction in total sugars, proteins and phenols amounts with increased storage period. More reduction in market samples of this drug as compared to fresh samples are showed. This may be due to unscientific methods of harvesting, collecting, handling and storage in unsuitable places, transporting and drying. Increased storage period (45 and 60 days of incubation) also is effective on biodeterioration of these chemical constituents.

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