*Indian Journal of Ecology (2024) 51(1): 135-139*
*DOI: https://doi.org/10.55362/IJE/2024/4206*

**Manuscript Number: 4206**
**NAAS Rating: 5.38**

# Effect of Size Reduction and Packaging Materials on Storage Stability of Dried Stevia under Ambient Conditions

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***Abstract*:** Experiments were conducted to study the effect of particle size of dried stevia leaves and packaging material on storage stability under ambient conditions. Three forms of dried stevia - Fraction A (<180 µm) in powder, Fraction B (>180 µm and <350 µm) in powder and dried leaves were packed in different packaging materials viz. glass jars, PET jars and punnets, stored under ambient conditions and evaluated for quality attributes at regular intervals throughout the storage period. The quality assessment of various forms of dried stevia suggested that type of packaging material and form of dried stevia had significant effect on the quality and shelf life of dried product. The glass jars were the best packaging material followed by PET jars. Stevia stored in dried leaves retained better quality characteristics in comparison to stevia stored in powdered form during nine months of storage. Out of the two fractions of powder, stevia powder-Fraction B retained better quality characteristics throughout the storage period.

***Keywords*:** Stevia, *Stevia rebaudiana.* Packaging materials, PET, Stevioside

Stevia, botanically known as *Stevia rebaudiana* Bertoni (Family- Asteraceae) is a sweet herb and leaves of stevia are the source of sweet glycosides. The chemical compound obtained from stevia is the best alternative source of sugar especially for diabetes patients. More importantly, stevia contains a high percentage of phenols, flavonoids, and antioxidants. There are eight groups of active compounds that are a source of sweetness in stevia that can be divided into: stevioside, steviobioside, ducloside and rebaudiosides A, B, C, D and E. The two main glycosides are stevioside (St) traditionally being 5-10% of the dry weight of the leaves and rebaudioside A (R-A) being 2-4% (Hossain et al 2017). Fresh stevia leaves are perishable in nature which leads to their easy spoilage due to climatic factors, handling, microorganisms, and structural and chemical changes during storage. Drying is an important step in post-harvest technology of stevia because every agricultural product in its natural state contains a certain amount of water called the moisture. To preserve the quality of the product and its storage life, this moisture content needs to be reduced to a level that is safe for storage. Drying of stevia needs to be done as soon as the crop is harvested to protect it from any kind of infestation. Shelf life of the dried product is very much dependent on the packaging materials and storage conditions under which the product is stored. Besides these, the form of the dried product is also an important parameter that determines the quality of the dried product during the storage as it relates to the surface area available for various losses to occur. The type of package, form of dried product and storage also affects the final quality of the product over time. The present study was planned to study the effect of size reduction and packaging materials on storage stability of dried stevia under ambient conditions.

## MATERIAL AND METHODS

The leaves of stevia were dried in solar assisted mechanical tray dryer at temperature of 55°C. Once dried, half of the stevia leaves were grounded to powder in a grinder. The powder thus obtained, was then subjected to sieve analysis and two fractions of powder were obtained. Therefore, dried stevia was kept in three forms, viz, stevia powder- Fraction A (<180 µm), stevia powder-Fraction B (>180 µm and <350 µm) and dried leaves. The dried stevia leaves and powder were then packed in three containers namely, glass jars, punnets and PET jars and stored under ambient conditions over a period of nine months. The samples were tested for various physio-chemical parameters at regular intervals. The weights of dried stevia leaves and both fractions of stevia powder stored in all three containers were evaluated for gain in weight. The percentage gain in weight of various forms of dried stevia during storage was calculated by using the formula:

$$\text{Gain in weight (\%)} = \left( \frac{W_2 - W_1}{W_1} \right) * 100$$

Color was determined using Hunter Lab Miniscan XE-Plus Colorimeter. The instrument gives readings of three

parameters namely, L*, a* and b*. Values of a, b closer to zero indicates grey color. L indicates the intensity of color i.e., lightness which varies from L=100 for perfect white to L=0 for black. The stevioside content from the stevia extract was estimated by method described by Kaur (2009). Steviol glycoside extract was hydrolyzed with 5N HCL at 70°C for 1 hour. The glucose units liberated from the stevioside upon hydrolysis took part in the reaction with 5% phenol and 95% sulphuric acid ($H_2SO_4$). The intensity of orange, brown color was read at 490 nm.

The flavonoid content was determined using method given by Balabaa et al (1974). Methanolic extract (3 ml) mixed with stevia extract was evaporated to dryness. The residue left was dissolved in 10 ml of 0.1 M methanolic solution of aluminium chloride. Intensity of yellow color so developed was read at 420 nm against blank. Protein content was determined by Lowry *et al* method (1951). The crude fibre method was determined using method given in (AOAC, 1980).

**Statistical analysis:** The data was analyzed for the effect of storage period, form of dried stevia and type of packaging material on the quality of dried stevia during storage using general linear model using Statistical Package for Social Sciences (SPSS).

## RESULTS AND DISCUSSION

**Gain in weight (%)**: Storage period and packaging material had significant effect on the gain in weight of dried stevia irrespective of its form during storage of nine months. The minimum gain in weight was for samples packed in glass jars while maximum for punnets (Fig. 1). This could be because of higher permeability of punnets as compared to glass and PET jars. Among the various forms of dried stevia, stevia powder-Fraction A showed the highest percentage gain in weight and dried stevia leaves had the lowest percentage gain in weight. This could be attributed to the fact that in dried leaves, lesser surface area was exposed and thus gain in moisture was comparatively less. The gain in weight of dried stevia was significantly affected by storage period, form of dried stevia and type of packaging materials.

**Color:** The L value, taken as a measurement of brightness, varied from 21.35 to 28.07 with storage period irrespective of the type of packaging material. Minimum L value was for stevia powder-Fraction A stored in punnets. The maximum 'a' value recorded for different forms of dried stevia i.e stevia powder-Fraction A, stevia powder-Fraction B and dried stevia leaves were -1.09, -1.18 and -1.23, respectively, when packed in punnets at storage of nine months. The increase in b value was also observed with storage period irrespective of the packaging material and storage period. The highest 'b' value for all the three forms of dried stevia viz. stevia powder-Fraction A, stevia powder-Fraction B and dried stevia leaves was 3.39, 3.42 and 3.61 respectively, for punnets. Moreover, the color change ($\Delta E$) of dried stevia powder as well as leaves witnessed increasing trend with storage period throughout the storage period of nine months. The increase in color change was significantly affected by storage time and packaging materials and could be due to the absorption of moisture by all the forms of dried stevia caused by an inadequate barrier provided by the packaging materials resulting in higher color variation with storage. The maximum '$\Delta E$' value was 26.19, 27.91 and 27.83 of all three forms viz. stevia powder-Fraction A, stevia powder-Fraction B and dried stevia leaves, respectively. However, the least color change was observed for dried stevia stored in glass jars. This change in color might be due to the reduction of

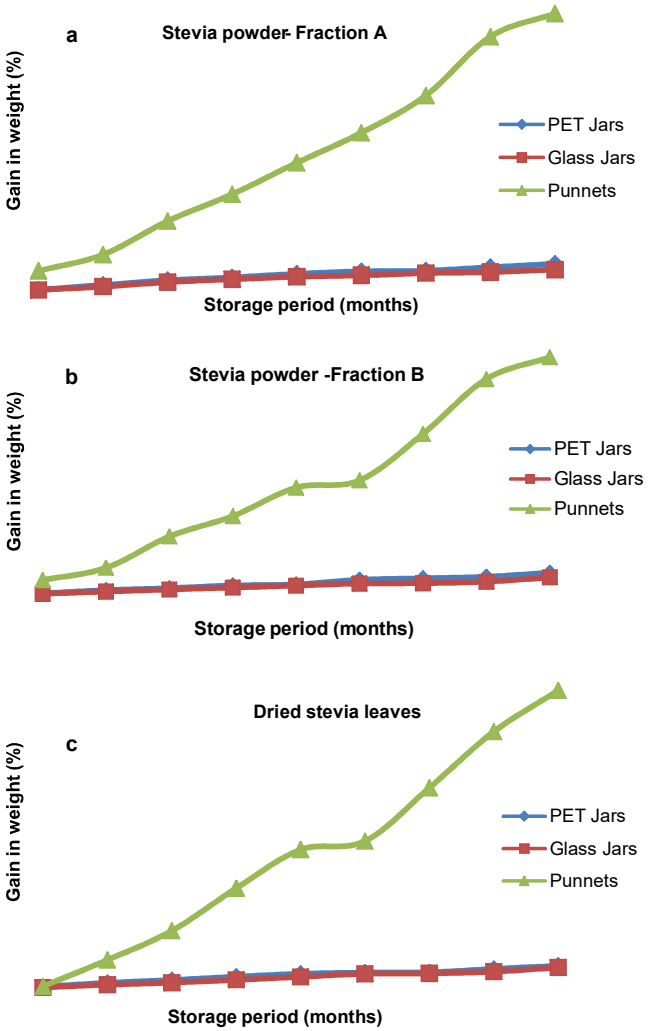


**Fig. 1.** Effect of packaging material and storage period on gain in weight of various forms of dried stevia. Stevia powder-Fraction A, (b) Stevia powder-Fraction B and (c) Dried stevia leaves

chlorophyll pigments as a result of photo oxidation reaction in the cells (Maskan et al 2002). The form of dried stevia and the type of packaging materials had significant effect on the color of dried stevia.

**Stevioside content:** The stevioside content varied from 5.15 to 13.61 % with the storage period irrespective of the type of packaging material (Fig. 3). The least stevioside content was observed in stevia powder-Fraction A when stored in punnets. The highest stevioside content at the end of storage period was in dried stevia leaves stored in glass jars. The decrease was significant and may be attributed to the moisture gain by all forms of dried stevia during the storage period that led to the deterioration of the stevioside content. Dried stevia leaves were observed to retain more stevioside content as compared to the two fractions of powder. This could be because lesser the surface area exposed; less are the losses from the surface. Since dried leaves had less exposed surface area as compared to the two fractions of powder, thus the losses in dried leaves were comparatively less. It was observed that punnets retained the least stevioside content out of all three packaging materials at the end of storage. It could be because of higher permeability of moisture and air in case of punnets as compared to PET and glass jars. The stevioside content of dried stevia was effected significantly by storage period, form of dried stevia and type of packaging materials.

**Flavonoid content:** The least flavonoid content was in stevia powder-Fraction A stored in punnets and maximum value of flavonoid content was observed in dried stevia leaves stored in glass jars after a storage period of nine months. The flavonoid content decreased with storage period (Fig. 4). This could be due to conversion of the natural constituents into other compounds as a result of chemical reactions with enzymes, oxygen and light during storage.

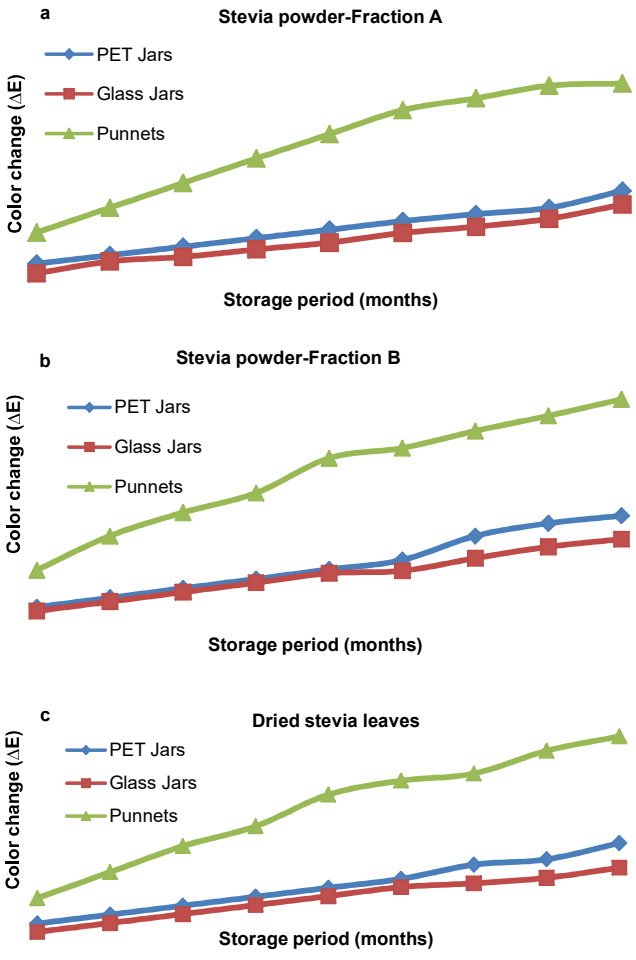


**Fig. 2.** Effect of packaging material and storage period on color change of various forms of dried stevia (a) Stevia powder-Fraction A, (b) Stevia powder-Fraction B and (c) Dried stevia leaves

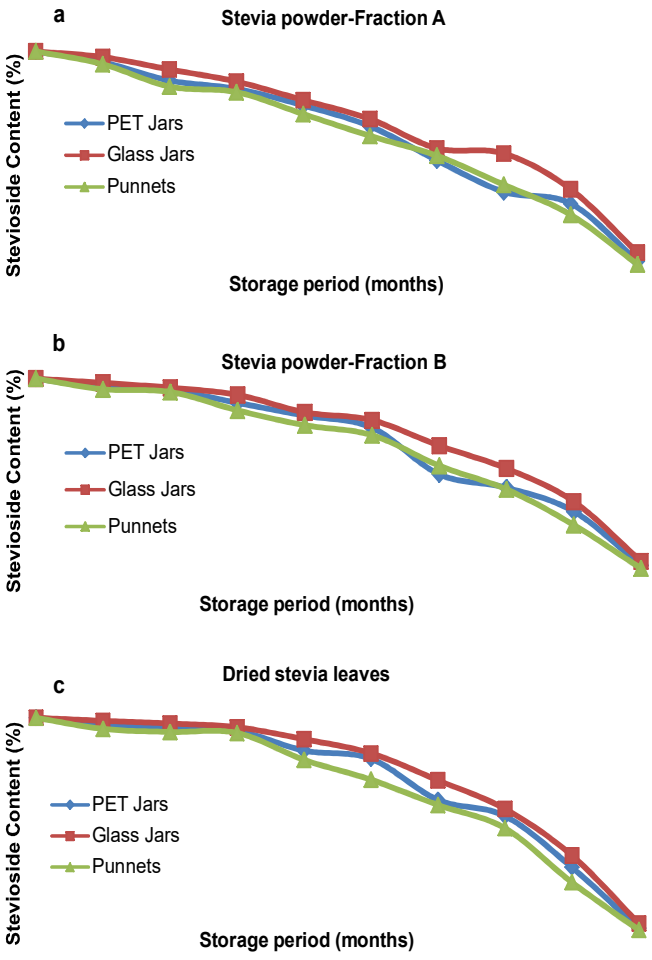


**Fig. 3.** Effect of packaging material and storage period on stevioside content of various forms of dried stevia (a) Stevia powder-Fraction A, (b) Stevia powder-Fraction B and (c) Dried stevia leaves

Some volatile compounds tend to escape away from the ground powder and leaves during storage time. Similar results were reported by Meghwal and Goswami (2014). The glass jars retained the highest flavonoid content irrespective of the form of dried stevia and punnets retain the minimum flavonoid content at the end of storage period. The dried leaves retained the flavonoid content better than both fractions of stevia powder. There was significant effect of storage period, form of dried stevia and type of packaging materials on the flavonoid content of the dried stevia.

**Protein content:** The least protein content was observed in stevia powder-Fraction A when stored in punnets and the maximum protein content was recorded in dried stevia leaves stored in glass jars (Fig. 5) for storage over nine months. The protein content of dried stevia leaves and both fractions of stevia powder followed a decreasing trend during the storage period i.e. the protein content for dried stevia decreased with an increase in the storage period, irrespective of the type of packaging materials. The decrease in protein content of dried stevia leaves and powder during storage period could be due to the fact that after drying, the process of protein synthesis stopped and denaturation of protein took place and thus the protein content either remained constant or decreased with storage period. The decrease was not much because during this time, denaturation of protein took place, thus the protein changed its form but was not lost completely (Zhang et al 2013). The maximum protein content was retained in dried stevia leaves in glass jars whereas, the least protein content was found in stevia powder-Fraction A in punnets at the end of nine months. The protein content of dried stevia was significantly affected by the storage period, form of stevia as well as the packaging material used.

**Crude fibre content:** The least crude fibre content was recorded in stevia powder-Fraction A during the storage period of nine months (Fig. 6). The crude fibre content decreased with increase in storage period in all three

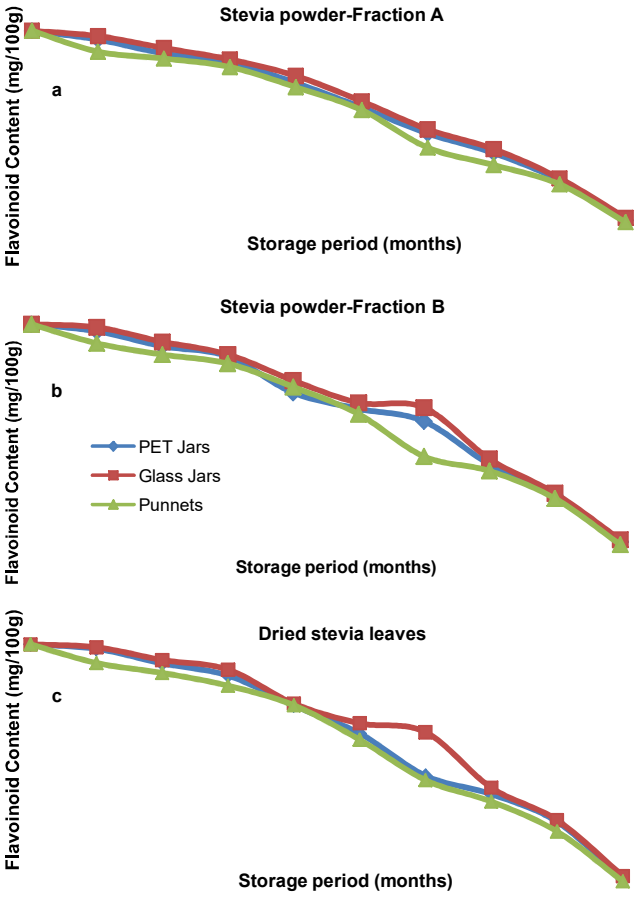


**Fig. 4.** Effect of packaging material and storage period on flavonoid content of various forms of dried stevia (a) Stevia powder-Fraction A, (b) Stevia powder-Fraction B and (c) Dried stevia leaves

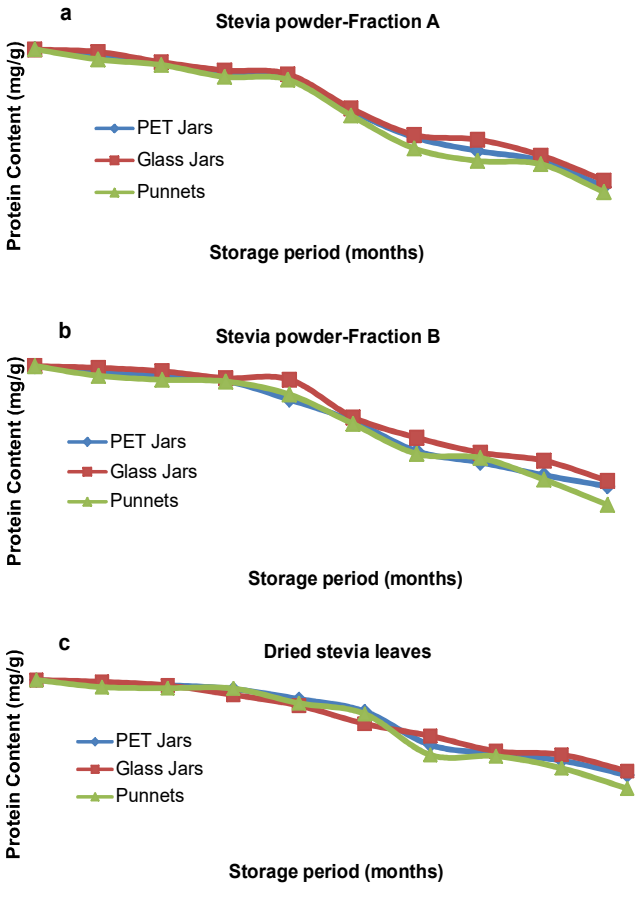


**Fig. 5.** Effect of packaging material and storage period on protein content of various forms of dried stevia (a) Stevia powder-Fraction A, (b) Stevia powder-Fraction B and (c) Dried stevia leaves

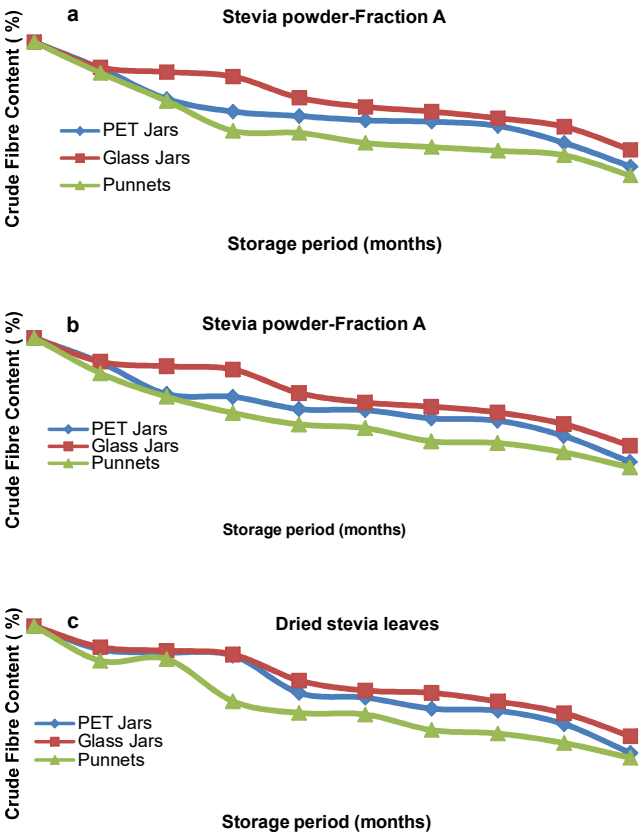


**Fig. 6.** Effect of packaging material and storage period on crude fibre content of various forms of dried stevia (a) Stevia powder-Fraction A, (b) Stevia powder-Fraction B and (c) Dried stevia leaves

packaging materials viz. PET jars, glass jars and punnets during storage. This might be because of the activity of lipase enzyme which increased during ambient conditions leading to more loss of fibrous content during storage (Kalim et al 2016). The least amount of crude fibre content was retained in stevia powder-Fraction A while the maximum was in dried stevia leaves. This could be attributed to the fact that the powder fractions were subjected to physical and chemical deterioration at the time of grinding. The storage period had significant effect on the quality of dried stevia irrespective of the packaging materials and form of stevia.

*Received 18 September, 2023; Accepted 12 January, 2024*

## CONCLUSIONS

The quality assessment of various forms of dried stevia suggested that storage period, type of packaging material and the form of dried stevia significantly affected the keeping quality of dried stevia. In order to store the dried stevia, the glass jars were the best packaging materials followed by PET jars. The stevia stored in form of dried leaves retained better quality characteristics throughout the storage in comparison to stevia in powdered form. Stevia, in powder (>180 µm and <350 µm)form retained better quality characteristics in comparison to powder with <180 µm size throughout the storage of 9 months.

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