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Long-term Storage of Turmeric (Curcuma longa) Rhizomes at Ambient Conditions in Jamaica:- *t*-Test for Matched Pairs and PCA Analysis

Collin SCANTLEBURY[1]*, Gillian ROWE[1], Oreane COLLINS[1], Danielle WILLIAMS[1] and Ryan FRANCIS[2]

[1]*Biotechnology Department, Scientific Research Council, P.O. Box 350, Hope Gardens Complex, Kingston 6 Jamaica, W.I.*

[2]*Product Research and Development, Biotechnology Department, Scientific Research Council, P.O. Box 350, Hope Gardens Complex, Kingston 6 Jamaica, W.I.*

***Correspondence:**

Dr. Collin SCANTLEBURY, Biotechnology Department, Scientific Research Council, P.O. Box 350, Hope Gardens Complex, Kingston 6 Jamaica, W.I, Tel: 1 (876) 322 7526.

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ABSTRACT

The research generated benchmark data from turmeric rhizomes, which may be specific to that variety and its growth environment. Ambient laboratory storage conditions allowed storage in excess of seven months while maintaining shoot germination viability. The most common weigh class of rhizomes in the sample was 10.5 – 15.0 g. After seven months, rhizomes lost on average 4.1 ± 3.3 g per rhizome or 32.6 ± 6.2 % of fresh weight. The t-test for matched pairs showed that the difference in rhizome mass pre and post storage was significant, with t = 4.83 (14 df) exceeding the tabled critical value of 2.15 (P = 0.05). IBM SPSS version 30.0.0.0 (172) PCA for factor analysis with data reduction showed that 94 % of the variance in the turmeric rhizome data was explained by three principal components (PC). These facilitate better identification of rhizome traits that may be used to enhance optimum rhizome storage. PC1 identified factors that promoted side branching. PC2 described factors that produced higher branching and larger rhizomes that increase weight loss. PC3 indicated that there were factors enhancing high shoot length and number of nodal rings which countered loss of weight during storage.

Keywords

Ginger, Turmeric, Rhizomes, Long–term storage, Nodal rings, Principal component analysis.

Introduction

Among the range of local spices in common use in Jamaican cuisine, the Biotechnology Department Departmentof the Jamaica Scientific Research Council (SRC) has been propagating ginger (*Zingiber officinale*), turmeric (*Curcuma longa*) and some vanilla (*Vanilla planifolia)* by *in vitro* plant tissue culture. Jamaican pimento (*Pimenta dioica*), cinnamon (*Cinnamomum verum*) and nutmeg (*Myristica fragrans*) have not gotten much tissue culture interest because these are traditionally propagated by zygotic seeds, *ex vitro* rooting of stem cuttings and air layering. These activities are however not outside of the purview of the Biotechnology department at SRC. Apart from tissue culture to eliminate fungus and viruses there is scope for mutagenesis breeding solutions regarding inducing disease resistance, improved yield and alteration of selected traits.

The ginger production at SRC fulfils specific client funded contracts as well as various initiatives funded by FAO/National Tissue Culture Technical Working Group, JaSPice, and International Atomic Energy Agency (IAEA) mutagenesis breeding. SRC also produces turmeric for client contracts exclusively. While much is known of its medicinal value [1,2], in Jamaica it is used mostly as a spice, coloring and flavoring for food. In the Biotechnology department, turmeric has stimulated research attention due to its apparent enhanced viability and capability for long-term storage relative to ginger. Understanding turmeric storage may also be useful in management of ginger rhizomes.

As a member of the Zingiberaceae or ginger family, there is anecdotal observations that turmeric store longer than ginger post harvest. There are observations that when ginger and turmeric are stored under similar conditions, within a month ginger has

germinated, while turmeric has showed no change (Figure 1). Ginger and turmeric cannot be grown all year round in natural field environment with diminishing day length. Storage of rhizomes facilitates breaking of any dormancy and allows farmers preparation for the optimum planting time.

Across the globe, traditional conservation of ginger rhizomes postharvest involves storage in a cool place, mitigation of potential rot from fungal spores and protection from moisture absorption [3,4]. Elimination of light is an important requirement that is taken for granted, but these are not novel, following long standing and well-researched recommendations [5]. The role of temperature in reducing sprouting is also well known [6] along with the use of plant growth regulators. The more recent development has been the use of radiation treatments to inhibit sprouting [7].

Figure 1: Ginger sprouted after four to six weeks at SRC Biotechnology facility.

The management of ginger and turmeric in the SRC shade-house involves competencies in handling weaned tissue cultured ginger plants that senesce prematurely and form mini rhizomes. There is a need to maximize germination and recovery of senesced plants especially from mini or undersized rhizomes. A large corpus of knowledge is available on storage of these rhizomes dating back to 1960s research [5,8]. Among the current research there are recommendations of optimum storage of turmeric for up to 20 weeks at 12-14 °C [4]. The purpose of the investigation is an evaluation of data of turmeric rhizomes which remained viable following storage in excess of seven months to identify some determinants of storage ability. It will include use of principal component analysis (PCA) to identify the main factors accounting for differences between stored turmeric rhizomes and their capacity to remain viable and germinate.

Methodology

Turmeric was procured locally from a supermarket in Liguanea, Kingston Jamaica. The fresh weight (FW) of turmeric rhizomes (30) was recorded, 7 November 2023. Mean fresh weight (FW), maximum and minimum rhizome mass was determined. Rhizomes were surface sterilized (15% bleach, 10 minutes; rinsed with water). Surface sterilized rhizomes were placed in a tray (28 cm x 54 cm x 5 cm) lined with brown wrapping paper and overlain with moist tissue paper (Figure 2). To conserve moisture a similar inverted tray was used as a lid to cover the rhizomes and create the humidity chamber. Rhizomes were incubated for seven months at the SRC lab on Hope Road, Kingston (Google map reference 19.057375, -77.126622).

Figure 2: Turmeric at 4-6 weeks in tray lined with brown wrapping paper and moist tissue paper.

Rhizomes were incubated at ambient lab temperature (24 to 27 ° C) and post storage data recorded June 2024. Frequency graphs were prepared to identify the most common weight class for the turmeric from a sample of 30 rhizomes pre storage. Data was collected from a smaller sample of 15 rhizomes June 2024 and comparisons made with data collected November 2023. For each of 15 rhizomes monitored from pre and post storage, 27 data fields populated in an XL spreadsheet comprising mass, length and related count data.

The *t*-test for matched pairs was also applied to verify that the difference in masses of rhizomes pre and post storage was significant, following Fowler et al. [9] where *d* is the difference in mass and *n* is the number of rhizomes:

$$t = \frac{\Sigma d}{\sqrt{\frac{n(\Sigma d^2)-(\Sigma d)^2}{n-1}}}$$

Statistical analysis also included pairwise correlations and PCA using IBM SPSS version 30.0.0.0 (172) to identify the main factors accounting for differences between the rhizomes.

Observations and Results

The most common mass class for the sample of turmeric pre storage where n = 30, was 10.5 – 15.0 g. This comprised almost 40 % of the sample (Table 1, Figure 3). While the masses of individual rhizomes ranged from less than 5.0 to 30.0 g, there were a few 40.5 – 45.0 g, less than 5%. The mean mass for the sample of 30 rhizomes was 14.3 ± 8.7 g per rhizome.

Table 2 summarizes descriptive data comprising 27 rhizome parameters (mean ± St. dev) for the sample of 15 rhizomes held over seven months. With this smaller sample size tracked over seven months in storage, the pre storage mean mass was 13.2 ±

10.7 g and 6.2 ± 7.5 g post storage. The mass classes changed and there was loss of some mass classes after storage. The most common mass class pre storage was 10.5 – 15.0 g (30 %) and post storage it was 5.5 – 10.0 g (40 %), Figure 4 and 5.

Table 1: Frequency histogram for masses of turmeric rhizomes (n=30).

Mass classes (g)	Frequency (%)
0.0 - 5.0	10.0
5.5 -10.0	23.3
10.5 - 15.0	36.7
15.5- 20.0	13.3
20.5 -25.0	3.3
25.5 - 30.0	10.0
30.5 - 35.0	0.0
35.5 -40.0	0.0
40.5 -45.0	3.3

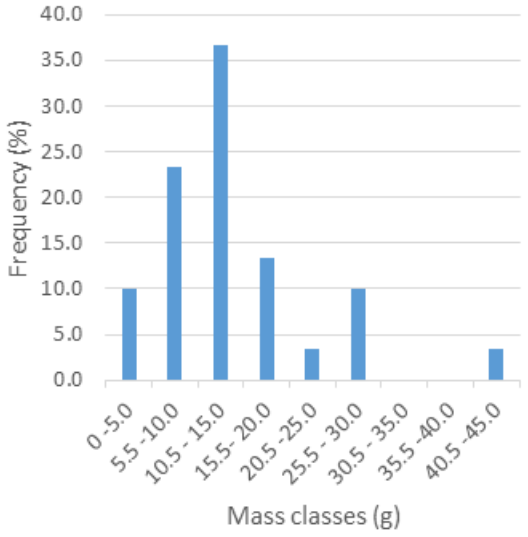

Figure 3: Frequency histogram of turmeric rhizomes mass classes (n=30).

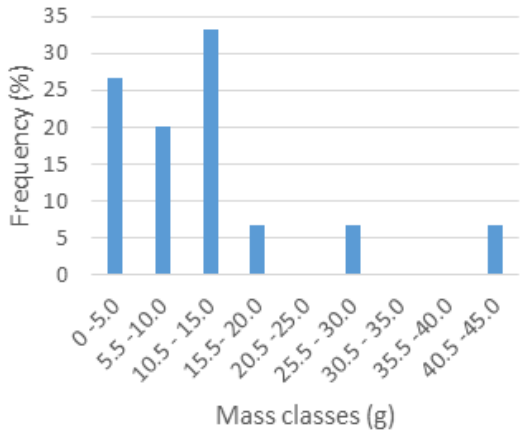

Figure 4: Frequency histogram of turmeric rhizome mass classes pre storage (n=15).

The *t*-tests for matched pairs showed that the differences in rhizome mass pre and post storage (or loss of mass) 4.1 ± 3.3 g or 32.6 ± 6.2 % was significant. The statistic $t = 4.83$ (14 *df*) exceeded the tabled critical value of 2.15 (P = 0.05).

Table 2: Descriptive Data and data fields used in the PCA (n=15).

#	Parameter (trait)	Mean ± t. dev
1	Post storage rhizome mass (g)	**6.2 ± 7.5**
2	Post storage rhizome volume (cm^3)	**9.1 ± 7.5**
3	Post storage rhizome density gcm^{-3}	**4.0 ± 6.7**
4	Original pre storage rhizome mass (g)	**13.2 ± 10.7**
5	Rhizome weight loss during storage (g)	**4.1 ± 3.3**
6	Percentage weight loss during storage (%)	**32.6 ± 6.2**
7	Number of branches (or bud extensions)	**0.9 ± 1.4**
8	Length of rhizome main or central branch (mm)	**80.6 ± 30.9**
9	Length of rhizome branch 1 (mm)	**6.93 ± 10.8**
10	Length of rhizome branch 2 (mm)	**5.7 ± 9.3**
11	Length of rhizome branch 3 (mm)	**3.7 ± 8.0**
12	Length of rhizome branch 4 (mm)	**0.7 ± 2.6**
13	Total length of rhizome main + branches (mm)	**97.5 ± 52.1**
14	Total length of rhizome ide branches (mm)	**16.9 ± 29.3**
15	Average length of side branches or buds (mm)	**5.7 ± 8.7**
16	Mass of rhizome main or central branch (g)	**7.5 ± 5.7**
17	Mass of rhizome side branch 1 (g)	**0.7 ± 1.5**
18	Mass of rhizome side branch 2 (g)	**0.3 ± 0.7**
19	Mass of rhizome side branch 3 (g)	**0.2 ± 0.6**
20	Mass of rhizome side branch 4 (g)	**0.0 ± 0.1**
21	Total mass of rhizome side branches (g)	**1.3 ± 2.6**
22	Average mass of rhizome side branches (g)	**0.4 ± 0.8**
23	Total mass of rhizome branches as fraction of total rhizome mass (%)	**8.9 ± 18.4**
24	Number of un-germinated buds (eyes)	**4.9 ± 1.5**
25	Number of nodal rings	**12.7 ± 4.0**
26	Number of shoots	**0.9 ± 0.3**
27	Shoot length (mm)	**36.8 ± 15.5**

Sampling half of the rhizomes, even if randomly, would have changed the frequency distribution of the rhizome mass classes. A comparison of Figure 3,4 and 5 showed that the smaller sample had a similar most common or modal weight class 10.5 -15 g to the total population; similar ca. 20 % between 5.5- 10.0 g but the weight class 0 – 5.0 g increased from 10% to 26%. Post storage the most common weight class shifted to 5.5 -10.0g (40 %). There were increases in the 0-5.0 g weight class and reduction in the 10.5 – 15.0 g class.

Principal Components

The extraction value for the variable "number of shoots on sprouted turmeric" was 0.48. Removal of this variable along with "number of buds" and "density" resulted in extraction values all closer to 0.9 in the output of communalities for the traits in Table 2. This indicated that the extracted components represented the variables well. The scree plot identified three principal components, those on the steep slope with eigenvalues greater than 1.0 (Figure 6).

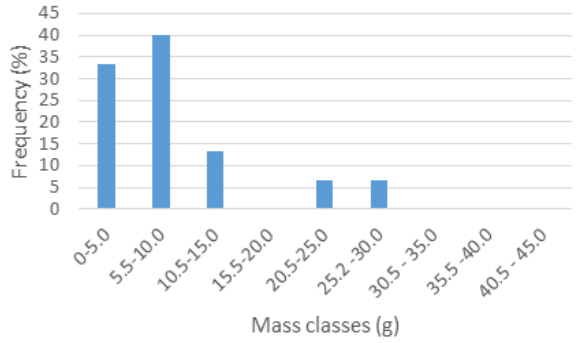

Figure 5: Frequency histogram of turmeric rhizome mass classes post storage (n=15).

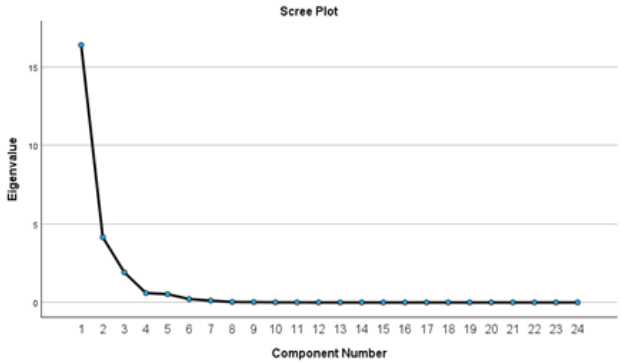

Figure 6: Scree plot highlighting principal components on steep slope.

The application of the rotation matrix maintained the cumulative percentage of variation explained by the extracted components but the variation was spread more evenly over the components. For easier viewing, in the rotated component matrix (Table 4) only correlations equal to or greater than 0.5 are shown. The first component, PC1 comprises the factors that promote side branches, account for their mass, length and ratios of the total rhizome mass. These appear to be limited to smaller numbers of branches (up to 3), the earlier formed branches that are larger more so than late forming, slower growing or smaller branches.

The correlations indicate that a different set of factors PC2 determine the length and mass of branches where there are larger numbers of branches. PC2 is highly correlated with larger rhizomes (high volume, weight and length). PC2 is also correlated with high weight loss.

PC3 describes a common determinant that influences high shoot length. It indicates that the number of nodal rings on rhizomes are correlated to shoot length. The analysis shows that these factors while enhancing shoot length and number of nodal rings, they effectively reduced percentage weight loss in turmeric rhizomes.

Rotated Component Matrix[a]

	Component		
	1	2	3
SB fraction of total (%)	.948		
av.mass SB(g)	.947		
mass branch 1 (g)	.932		
average branch length (mm)	.906		
length b1 (mm)	.906		
length b2 (mm)	.901		
total mass SB (g)	.892		
# branching	.881		
total branch length (mm)	.865		
mass branch 2 (g)	.804	.590	
length b3 (mm)	.778	.559	
length b4 (mm)		.906	
mass branch 4 (g)		.906	
volume (cm3)		.884	
weight loss (g)		.820	
original mass (g)		.774	.509
length main branch (mm)		.764	
mass (g)		.751	.532
mass main branch (g)		.740	.647
mass branch 3 (g)	.685	.715	
Total length (mm)	.553	.715	
shoot length (mm)			.905
# nodal rings			.844
% weight loss			-.769

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.
a. Rotation converged in 8 iterations.

Table 4: Rotated Component Matrix.

Discussion

The PCA analysis is a simple yet powerful tool that allows a better interpretation of the turmeric data to identify correlations and associations that would not be possible by looking at a vast array of raw data. It is intuitive that more experience and a review of more data and replications are required to validate inferences. The PCA

Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	16.409	68.369	68.369	16.409	68.369	68.369	10.343	43.096	43.096
2	4.155	17.314	85.683	4.155	17.314	85.683	8.197	34.153	77.249
3	1.917	7.990	93.673	1.917	7.990	93.673	3.942	16.423	93.673

Table 3: Total variance explained, extracted components.

however remains a useful predictive tool. PCA has identified key traits that influence long-term storage of turmeric rhizomes. Based on trait data we may have better understanding of the expectations and take the appropriate actions to enhance storage. PCA has been used in turmeric for analysis of multivariate trait data [10], genetic variability studies in accessions [11,12], and predicting yield from weather data [13] to name a few practical applications.

It is speculative that comparing data pre and post storage for the 27 parameters would have furnished more information than just the post storage data. Secondly it is also debatable a bigger sample of 30 rhizomes would have yield more reliable results than the smaller set of 15 for rhizome masses. instructive to verify . Benchmark data has been provided as an estimate of selected rhizome parameters for a random sample of turmeric rhizomes. There is expected to be variance with varieties, growing conditions, climate etc. The preliminary investigation demonstrated how the SPSS PCA tool originally designed for social science applications can be useful in Agriculture and related applied earth sciences.

Turmeric varieties are classified by the length of primary rhizomes as either short <50 mm, medium 50 – 100 mm or long > 100 mm and straight or curved [14,15]. Our data and inferences were made from a sample of turmeric rhizomes that were medium with straight fingers (80.6 ± 30.9 mm). These were comparable to medium varieties in Jan et al. with length (79 ± 11 mm). Branching as a descriptor may have been used loosely in the article, but linear non-branching fingers are mostly depicted in literature where photos are available [16]. Raut et al. [17] depicts pictures of more branched long types.

Among issues arising from the preliminary survey is that research is now required to identify the determinants of linear rhizomes and branching in turmeric. In root crops this may be genetic but is influenced by soil texture and physical compaction. Irregularity of nutrient and water supply, microbial factors and interaction with various chemical stimuli may also be important [18-20]. There was no initial focus on addressing whether straight or branched rhizomes were more suited for long-term storage. However, it is felt that long-term rhizome storage may be enhanced if the rhizomes are optimum shape and size. For this, the appropriate environment must be available.

It is interesting that PC2 also correlates with side branching and not the branching described in PC1. PC2 only describes where there are as many as four side branches. It is intuitive and easy to visualize that a large or a long main branch is more likely to have a larger number of branches than a shorter branch. Also high mass, volume, long length are expected to be correlated in rhizomes. It is the water content that contributes to the mass hence the correlation with weight loss.

It was surprising that the extraction communalities or the variance accounting for "number of sprouted shoots on rhizomes" was low. So this was not a variable that was well represented in any extracted component. However, shoot length had a high extraction value. The analysis showed that emergence of buds, and shoots was not influenced by rhizome mass. This is interesting since a reasonable expectation of growers is for heavier rhizome setts to produce higher yields.

Similarly, nodal rings were not influenced by rhizome length. This may only be a reasonable expectation if nodes perform specific independent functions and are not mere artifacts of rhizome elongation. The correlation between shoot length and number of nodal rings is significant because the longer the shoot length and the more nodal rings present, the less weight loss is observed during storage. The nodal rings may therefore be predictive of the relative shoot length and relative weight loss during storage.

How PCA helps us to better manage turmeric rhizomes is intuitive. It is a fair expectation that there will be variability in the storage potential of rhizomes. There is likely to be differences due to the degree of branching. Differences are expected between those with one to three branches and those with more branches. These will be related to length and mass differences. The significance of the nodal rings are yet to be determined but the more nodal rings and the more shoots present on the rhizome, the less weight reduction during storage.

Conclusion

While the research was on turmeric, the findings bear some relevance to ginger. These species both belong to the Zingerberaceae family. They share similar requirements for storage. Whether sprouting to provide *in vitro* explant material or single bud techniques (SBT) for *ex vitro* propagation the handling and processing is identical. Varietal effects are expected with highly branching rhizomes showing higher mass. The mass is due largely to higher water content and potential for associated higher water loss. Certain traits are predictive with the high presence of nodal rings and high bud or shoot formation indicative of reduced weight or water loss. Overall selection of these traits is expected to enhance storage time and quality of ginger and turmeric rhizomes.

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