

**FINAL TECHNICAL REPORT**

**PHYSIOLOGICAL BASIS FOR STORAGE LIFE EXTENSION IN FRESH  
SWEET POTATO**

**R NUMBER: R6314 (A0422)**

**RNRRS PROGRAMME: Crop Post-Harvest Programme**

**PROGRAMME MANAGER: Natural Resources Institute**

**SUB-CONTRACTOR: none**

**RNRRS PROGRAMME PURPOSE: 4: Storage and Processing Losses Reduced.**

**RNRRS PRODUCTION SYSTEM: Forest/Agriculture Interface and Hillsides**

**COMMODITY BASE: Non-grain Starch Staples (Sweet potatoes)**

**AUTHORS OF REPORT: D. Rees, R. Bancroft, D. Matters and A. Pollard**

## **EXECUTIVE SUMMARY**

The overall objective of this project was to work towards the identification of the major physiological and environmental factors affecting perishability of the fresh sweet potato root, and thereby to facilitate the identification of strategies for increasing shelf-life through cultivar selection and improved handling practice.

Specific objectives were:

- (i) To provide technical assistance to the Tanzanian National Root and Tuber Crops Programme on trials to assess the post-harvest characteristics of local sweet potato varieties.
- (ii) To carry out laboratory studies of the post-harvest physiology of sweet potato roots under conditions which they would typically experience in East Africa.
- (iii) To survey existing handling practice for sweet potatoes in selected areas of Tanzania, and thereby to develop recommendations for improved practice and future work on handling.

Technical assistance was provided to the Tanzanian National Root and Tuber Crops Programme to plan, carry out and analyze trials carried out at five research institutes to assess the post-harvest characteristics of 5-6 sweet potato cultivars at each institute. (The results of the trials are reported under R6204). Several methods of post-harvest assessment were tried out, and have led to the production of a Methods Manual to be used for post-harvest assessment of cultivars in future.

A rapid method for estimating sugar content of sweet potato roots (an important quality characteristic) in the field, by measurement of the soluble solids in root "sap" by refractive index, has been tested in the laboratory. Comparison of this method with extraction and measurement of sugars by HPLC (High Performance Liquid Chromatography) has indicated that it provides a good estimation of sugar changes during storage for two out of the four cultivars tested. For the other two cultivars there appeared to be interference due either to the presence of non-sugar soluble compounds or textural characteristics preventing efficient "sap" extraction. Given that this method is being used by a number of

sweet potato researchers in several countries of the world, it will be important to resolve this problem.

An understanding of the physiological factors associated with perishability of sweet potato roots is important for efficient selection of improved cultivars within breeding programmes, especially if indirect selection criteria for longer-keeping varieties can be identified. This is important for breeding programmes carried out by the National Programmes, but probably even more for longer term breeding programmes as carried out by the International Potato Centre. As most of the physiological work reported in the literature has been carried out on North American and Japanese cultivars, it was necessary to start by collecting baseline data on which future work can be based. A study on the post-harvest metabolism of roots of four Kenyan cultivars (grown in Nairobi by the International Potato Centre) maintained under storage conditions typical of East Africa was therefore conducted at NRI to compare perishability and relate this to physiological characteristics. Two of the cultivars were reported by Kenyan farmers and traders to be perishable, while the other two were reported to have long shelf-life. The roots used in this study were free of insect infestation and were pre-treated with fungicide so that only perishability due to physiological deterioration was assessed. To facilitate this, and future studies a rapid method for analysing sugar composition by room temperature water extraction of freeze-dried samples, followed by HPLC analysis was developed. Data were obtained on changes in fresh weight, total dry matter, sugar composition and respiration rates. Other observations included shrivelling, sprouting, internal tissue breakdown and tissue browning. Of the four cultivars, one, KSP20, (reported to have long shelf-life) was distinguished by a lower dry matter content, distinct sugar composition (lower sucrose and higher reducing sugar content), low rates of respiration and low rates of sprouting. It is postulated that this cultivar may be particularly suitable for long-term storage and that low sucrose levels may be a valuable selection criterion for long storing cultivars. Further investigation to compare KSP20 with other cultivars on the basis of storability, and how this is associated with the above characteristics, should be carried out to confirm these findings and seek additional selection criteria for longer shelf-life.

The advances made to date on characterisation of Tanzanian germplasm, and on the physiological behaviour of roots under storage conditions will be further exploited within a subsequent project funded by the RNRRS Crop Post-Harvest Programme which includes work to define more precisely the consumer criteria of quality for sweet potato roots in Tanzania and further post-harvest evaluation of Tanzanian germplasm. Also

included is a PhD programme to continue and expand the work to establish the physiological characteristics associated with long shelf-life. A second PhD programme is planned during which field work will be conducted in Tanzania to investigate pre-harvest factors (production practice and environment) on post-harvest characteristics.

Surveys of the marketing systems for fresh sweet potato were conducted in Tanzania, in the urban districts of Mwanza (Lake Zone), Morogoro and Dar es Salaam (Coast Zone). The target groups investigated included producer and producer-traders, traders, wholesalers, retailers and snack food vendors etc. Informal semi-structured interviews were undertaken with over 80 respondents in more than 20 markets and other locations. Trading and handling practices, and production and post-harvest problems were characterised. Preliminary findings indicate that the growing demand for fresh sweet potato in urban areas is leading to a shift from predominantly small-scale production to cash cropping, usually of a narrow selection of favoured varieties. From the perspective of producers and producer/traders, sweet potato is considered one of the most profitable of food crops, the market potential of which is only limited by its seasonality and restricted storage life. Seasonality is exacerbated by the fact that in-ground storage of roots is limited by weevil infestation and a decline in culinary quality. Once harvested, fresh sweet potato roots were considered to have a maximum shelf-life of seven days. Post-harvest losses of 30-50% were occasionally cited, but were usually considered to be of the order of 10-15%. There is, however, no hard data to confirm this. These surveys have provided the necessary first step to planning the more detailed studies needed to identify appropriate measures for reducing losses. The results have been used to develop a programme to be carried out as a collaboration between NRI, the Tanzanian National Root and Tuber Crops Programme and the Tanzania Food and Nutrition Centre, funded within the ODA RNRRS Crop Post-Harvest Programme.

## **BACKGROUND**

1. Several surveys conducted in East Africa have indicated that short shelf-life is an important constraint for sweet potato and have led to the recommendations that less perishable sweet potato varieties should be developed and that handling techniques be devised to minimise post-harvest losses. These include surveys conducted both by NRI, and by the Tanzanian National Programme. (e.g. Fowler and Stabrawa (1993), Kapinga *et al.* (1995) ). Precise economic losses have not been quantified, although it has been estimated that losses of sweet potato can range from 35-95% in developing countries (Anon 1978). Although such high figures require confirmation, it is clear that the pattern of consumption and marketing is controlled by the short shelf-life of the commodity, and that a longer shelf-life would increase opportunities for consumption and marketing .

2. The Tanzanian Ministry of Agriculture is giving increasingly high priority to post-harvest issues such that the National Agricultural and Livestock Research Masterplan gives first priority to post-harvest research in several zones of the country. This is illustrated by the fact that the Ministry of Agriculture has agreed to fund the establishment of a post-harvest unit within the National Root and Tuber Crops Research Programme based in Lake Zone.

3. Improvements in the shelf-life and quality of sweet potato roots could be brought about in a number of ways: by identifying improved methods of handling, harvesting and production, and through selection of higher quality and less perishable cultivars. In the case of cultivar selection, cultivars could be selected from existing germplasm, or improved cultivars could be developed as part of a longer-term breeding programme. To make selection more efficient an understanding of those characteristics associated with good post-harvest behaviour is very important. This would lead to the development of selection techniques to be used by national programmes and in longer-term breeding programmes, such as those undertaken by the International Potato Centre (CIP). Although in the long-term it is envisaged that the main benefits will be obtained through the breeding of improved varieties, the diversity of the existing East African Germplasm (see below) provides great potential for short-term improvements through selection and promotion of the cultivars most appropriate for each set of conditions and use.

## **Diversity of East African Germplasm**

4. The diversity of the sweet potato germplasm in East Africa is indicated by the germplasm collection initiated by the Tanzanian National Root and Tuber Crops Programme in 1992, which already includes several hundred landraces. Characterisation of this germplasm has started over the last 2-3 years, but cross-referencing between the separate germplasm collections in Tanzania and between countries, has not taken place.

5. As will be described below, there have been very few scientific studies on the post-harvest behaviour of East African germplasm. However, evidence obtained during informal interviews of farmers and traders has indicated that Tanzanian cultivars are perceived as differing significantly in their perishability (Fowler and Stabrawa, 1993, Kapinga *et al.* 1995).

### **Root deterioration through post-harvest physiological changes.**

6. Deterioration of a sweet potato root can occur through physiological changes, rotting and insect infestation. The storage root is primarily a starch storage organ, with some sugars (mainly sucrose, glucose and fructose) and low levels of protein. The main post-harvest physiological changes are associated with water loss, and carbohydrate (starch and sugar) metabolism.

7. The extent of water loss depends primarily on the intactness of the periderm. If this is damaged, as will often happen during harvesting and handling, the site of damage becomes an area of rapid water loss, as well as providing a site for infection by pathogens (see below). The root, however, has a natural defence mechanism to heal such wounds by forming a "wound" periderm.

8. The process of wound healing involves the deposition of a suberin-like polymer in the surface cell layers which subsequently die to form a dessicated layer. This is followed by the formation, by cell division, of an underlying wound periderm with a composition very similar to normal periderm (Walter and Schadel 1982, 1983). Work carried out on the control of wound healing in sweet potato at the molecular level has indicated that ethylene, which is known to stimulate many other plant responses, such as fruit ripening, is involved in the control of the wound response (e.g. Imaseki *et al.* 1968, St.-Amand and Randle 1991).

9. In the practice of curing, sweet potatoes are exposed to conditions to promote wound healing. This is used routinely in the storage of sweet potatoes in temperate regions. For example, in the United States where sweet potatoes are stored at 13-15°C, this is preceded by 5-7 days at a warm temperature of about 30-33°C and a high humidity of 85-95%. There are a number of reports in which different curing conditions for sweet potato are compared (e.g. Gull and Duarte 1974, Lawrence 1985, Gooding and Campbell 1964, Delate and Brecht 1985). A high relative humidity is recommended since low relative humidity causes surface cells to die by desiccation and thus inhibits wound periderm formation. The optimum temperature is determined by the temperature dependence of the enzymes involved. Thompson (1972) postulates that roots grown under tropical conditions may need higher temperatures to cure than those grown under temperate conditions. There is evidence, mostly with respect to North American varieties that the efficiency of wound healing may vary between varieties (Gull and Duarte 1974, Clark 1992, Clark *et al.* 1989). If this is the case among East African germplasm, it could be a very useful characteristic to select for.

10. As the storage root is a living organ, low levels of metabolism are necessary to maintain the integrity of the cells. High rates of metabolism, however, can be detrimental to quality, by changing the carbohydrate composition, or in the extreme case, by metabolising so much starch that air spaces form, and the texture of the root becomes spongy. Most work on carbohydrate metabolism and respiration has been carried out on North American or Japanese cultivars under the temperature regimes used in refrigerated stores (typically 13-15°C) (e.g. Woolfe 1992 and references therein, Takahata *et al.* 1995). These have generally shown that sugar levels increase during storage (Woolfe 1992). However, the metabolic rate is temperature dependent, and the cultivars can vary significantly in their metabolic characteristics (e.g. Ahn *et al.* 1980), so that there is a need to determine how the major East African cultivars behave under tropical conditions. A better understanding of the control of the metabolic rate would help us to select cultivars and conditions for slow metabolism.

11. During longer term storage, sprouting of the root can cause loss. Although sprouting is not considered to be a major problem in East Africa under the present handling conditions, it could become so if longer term storage was feasible (Devereau 1996). Sprouting can be controlled by sprout suppressants, but this is not economically

feasible under most situations in East Africa. Thus the identification of cultivars that sprout less easily would be advantageous for long-term storage.

#### **Root loss through post-harvest rots.**

12. Evidence that resistance to rotting organisms exists within the American germplasm (e.g. Clark *et al.* 1989 and references therein) strongly suggests that such resistance would exist within the African germplasm, and observations from surveys support this (Fowler and Strabawa 1993, Kapinga *et al.* 1995). Mechanisms of resistance include the formation of physical barriers, as in the formation of wound periderm and hypersensitive cell death, and the production of chemicals toxic to invading pathogens such as phytoalexins, proteinase inhibitors and phenols. A greater understanding of the role of these different mechanisms among the African germplasm could lead to the development of indirect selection techniques to facilitate selection from existing germplasm, and in future breeding programmes.

#### **Root loss through insect infestation**

13. Infestation by sweet potato weevils (*Cylas* spp.) is a major constraint both pre- and post-harvest worldwide. Variation in susceptibility to infestation among sweet potato cultivars has been reported (Rajamma and Pillai 1987, Macfarlane 1987, Mullen *et al.* 1985, Oboh *et al.* 1989). Breeding programmes have led to the release of cultivars in the United States with a degree of resistance to *Cylas formicarius*. The basis of this resistance may be related to deep rooting, in which case it would not confer post-harvest resistance. There is some evidence of resistance based on chemical composition of the root surface (e.g. Son *et al.* 1991, J. Bohac pers. comm.), although this is not as clear as the evidence for resistance against rots.

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## PROJECT PURPOSE

14. The overall objective of this project was to work towards the identification of the major physiological and environmental factors affecting perishability of the fresh sweet potato root, and thereby to facilitate the identification of strategies for increasing shelf-life through cultivar selection and improved handling practice.

15. Specific objectives were:

**To provide technical assistance to the Tanzanian National Root and Tuber Crops Programme on trials to assess the post-harvest characteristics of local sweet potato varieties.**

The aims of these trials were: to obtain in-country data on the deterioration of sweet potato roots to support a programme of research on post-harvest physiology of the sweet potato root (see below), to assess the shelf-life and quality of major Tanzanian sweet potato cultivars so that those with most appropriate post-harvest characteristics can be promoted, and to develop a standardised methodology of post-harvest assessment that can be used by the National Programme in future. As well as providing technical support during the trials themselves and analysis of results, an additional objective was to develop analytical methods suitable for use in the field.

*(The trials are reported in detail elsewhere under R6204).*

**To carry out laboratory studies of the post-harvest physiology of sweet potato roots under conditions which they would typically experience in East Africa.**

Given that most of the work reported in the scientific literature has been carried out on North American and Japanese cultivars under conditions typical of chilled stores, it was necessary to collect baseline data for East African cultivars at higher temperatures, one of the aims being to determine to what extent existing information could be extrapolated to East African cultivars and conditions.

**To survey existing handling practices for sweet potatoes in selected areas of Tanzania, and thereby to develop recommendations for improved practice and future work on handling.**

## **RESEARCH ACTIVITIES**

### **Technical assistance to the Tanzanian national programme on trials to assess the post-harvest characteristics of local sweet potato varieties.**

16. Post-harvest evaluation trials were conducted by the National Root and Tuber Crops Programme at five agricultural research institutes in Tanzania: Kibaha in Coast Zone, Ukiriguru in Lake Zone, Uyole in the Western Highlands (Two sites at Uyole and at Mitalula were used), Tengeru, Arusha, and at Chollima-Dakawa, Morogoro.

17. Prior to the initiation of the trials a meeting was held at Ukiriguru Agricultural Research Institute, supported with funds from this project and R6204 (X0287), the International Potato Centre (CIP) and the Southern Africa Root Crops Research Network (SARRNET) specifically to standardise methods for pre- and post-harvest assessment of sweet potato and cassava cultivars within breeding programmes. Methodologies for the post-harvest evaluation of sweet potato were discussed at this meeting.

18. A staff member from NRI assisted in the initiation of trials at Ukiriguru, and at Kibaha, and also in the analysis of results.

19. At each station the varieties were assessed at the time of harvest for yield, visual and physical characteristics of the roots and consumer acceptability. This was followed by a storage trial to assess rates of deterioration due to physiological changes and rotting. The storage environment during the trial (roots stored in a fertiliser sack which was closed for 2 days and then opened) was chosen to simulate the conditions to which the roots would normally be subjected during marketing and household storage. As previous observations indicate that resistance to mechanical damage is important for good shelf-life, a second storage treatment in which the roots were damaged before storage was also used. Changes in consumer acceptability during storage was assessed at one station.

20. Changes in physical characteristics of the roots were measured in an attempt to relate these physical characteristics to shelf-life and quality.

21. In-country costs for the trials were funded through an extra-mural contract. The trials are therefore reported in more detail in the Final Technical Report of R6204. A

manual of methods to be used in future post-harvest evaluations has been produced for approval by the National Programme.

### **Development of methodologies appropriate to field work.**

22. To support the trials described above, work was conducted at NRI to develop and validate methodologies appropriate for use in the field. Root sugar content can be an important factor with respect to quality. As well as affecting root taste and therefore consumer acceptability, changes in sugar composition are an important indication of post-harvest metabolism of the sweet potato root. Facilities are not available at most agricultural research institutes in Tanzania to carry out sugar analyses by standard laboratory methods such as chemical assay, or high performance liquid chromatography (HPLC). As an alternative, during the field trials, the use of hand-held refractometers was tested. It therefore became important to check the validity of this method.

23. For the measurement a small amount of root tissue was grated, and the juice squeezed out using a garlic press. The refractive index of the juice measured by the refractometer depends on the soluble solid content. This can be used to estimate sugar content in cases where most of the soluble solids are sugars and is a standard method for measuring sugar content of fruits. There are reports of this method being used in sweet potato research programmes (e.g. USA, Japan, South Africa). However, to our knowledge, there are only two reports of work conducted to validate the use of refractive index measurements, one was carried out on cooked sweet potato, and the other involved extraction of juice using an electric blender (Report in 1992, Annual Report of Kyushu National Agricultural Experiment Station, MAFF, Japan. Walter, WM (1992) HortiScience, 27; 333-335 Use of Refractive Index to Monitor Changes in Sugar Content of Sweetpotatoes). A study was therefore carried out at NRI to compare sugar levels measured using a hand-held refractometer with those measured by HPLC.

*Methods used are given in more detail in Appendix 1.*

**Laboratory studies of post-harvest physiology of sweet potato roots under tropical conditions.**

24. As part of the overall objective to identify physiological factors associated with perishability, a study was conducted to follow the post-harvest physiological changes of the roots of four Kenyan sweet potato varieties maintained under conditions typical of the tropics. Of the four varieties considered; Kemb10, Kemb36, SPK004 and KSP20, two (Kemb10 and SPK004) have a reputation in Kenya for being perishable, while two (Kemb36 and KSP20) are reputed to keep well. The roots were grown in trials near Nairobi by the International Potato Centre, and were air-freighted to the UK. Immediately following harvest the roots were treated with fungicide to prevent rotting.

25. The roots were stored for eight weeks at 27°C, relative humidity 90-95% and a normal atmospheric gas composition. The temperature and humidity were chosen with reference to long-term pit and clamp storage trials that were previously conducted in Uganda. No information was available, however, as to the gas composition within the stores. A normal atmospheric composition was therefore chosen for these experiments in order to be relevant to more ventilated storage. (An experiment was conducted during which CO<sub>2</sub> was allowed to rise and O<sub>2</sub> to fall, but the results are too preliminary to report here).

26. Throughout the period of storage, observations of root quality characteristics, including fresh weight, sprouting and shrivelling were made. At regular intervals roots were sampled to determine internal appearance, hardness, latex production, flesh browning. Samples of tissue were taken for analysis of sugar and starch composition and dry matter content.

27. The respiration rates of selected roots were also measured at regular intervals as an indication of metabolic rate.

*Methods used are given in more detail in Appendix 1.*

## **Development of Analytical Methods**

28. In order to carry out the study outlined above, some development of analytical methods was necessary. For example, given that a large number of samples were to be analysed for sugar composition, more rapid methods of analysis of sugar and starch composition were developed and validated.

29. Standard methods for sugar analysis of root crops involve hot extraction with 80% ethanol are time consuming and expensive. An alternative method of extraction by water, which is simpler, cheaper and considerably faster, was tested and validated for sweet potatoes. As an alternative to time consuming biochemical sugar assays, an HPLC system for sugar analysis, capable of fructose, glucose, sucrose and maltose was set up, and compared to analysis by biochemical assay.

### **Surveys of existing handling practice for sweet potatoes in Tanzania.**

30. In collaboration with the Tanzania Food and Nutrition Centre (TFNC) and the Tanzanian National Root and Tuber Crops Programme, surveys of the marketing systems for fresh sweet potato were conducted in Tanzania, in the urban districts of Mwanza (Lake Zone), Morogoro and Dar es Salaam (Coast Zone). The target groups investigated included producer and producer-traders, traders, wholesalers, retailers and snack food vendors etc. In the Lake Zone the survey was combined with the collection of information on processed sweet potato and cassava. Informal semi-structured interviews were undertaken with over 80 respondents in more than 20 markets and other locations. Trading and handling practices, and production and post-harvest problems were documented.

## OUTPUTS

### **Technical assistance to the Tanzanian national programme on trials to assess the post-harvest characteristics of local sweet potato varieties.**

31. The results obtained from the post-harvest evaluation trials conducted by the Tanzanian National Root and Tuber Crops Programme are reported in full in the technical report for R6204. The results confirmed that varietal differences with respect to perishability do exist, although from this years data no firm conclusions could be drawn as to the physiological characteristics underlying perishability.

32. Several methods of post-harvest assessment were tried out, and have led to the production of a Methods Manual to be used for future trials.

33. Some problems in interpretation of data were experienced due to the fact the methods were being assessed, and were therefore adapted and refined throughout the trials. Additional problems were encountered due to the low level of resources in the Tanzanian National Root and Tuber Crops Programme. For example dry matter determinations could not be carried out reliably in two of the participating institutes.

34. In some cases, a need for additional collection of information was identified. For example little data exists on the the quality criteria used by urban consumers for fresh sweet potato roots, or on the most important types of damage to which fresh roots are exposed during marketing. Both these issues are to be addressed in a subsequent project funded by the Crop Post-Harvest Programme, R6507 (A0499).

### **Development of methodologies appropriate to field work.**

35. In order to check the use of refractive index measurements for estimation of storage root sugar content, parallel measurements of refractive index and HPLC analysis of extracted sugars were made for four Kenyan sweet potato cultivars during storage trials under tropical conditions conducted in the laboratory at NRI. The results obtained are shown in Figure 1; which indicates a very poor correlation between sugar levels measured by the two methods.

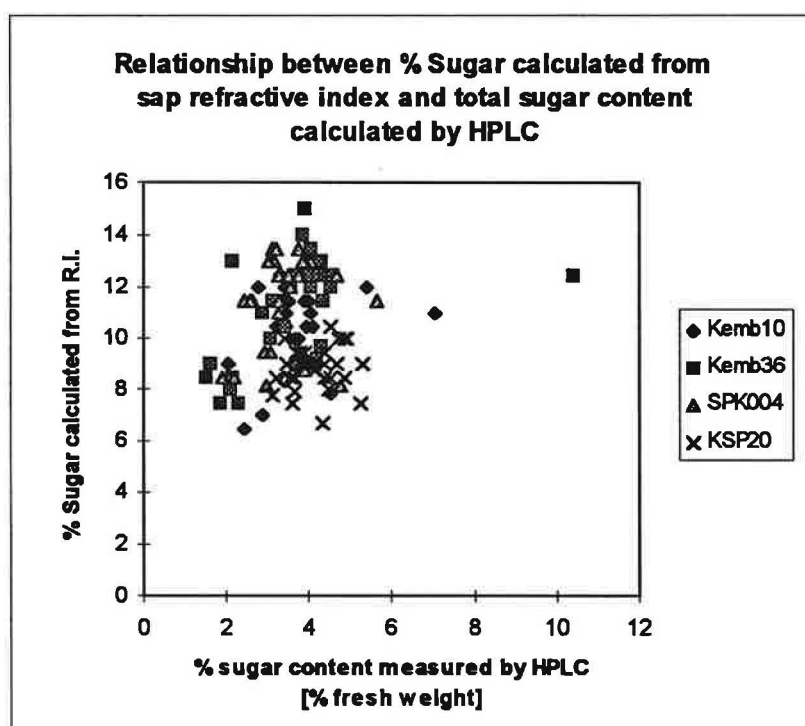


Given that refractive index is considered a reliable method for measuring sugar content of fruits, further experiments were conducted to investigate why there was such a poor correlation between the two methods in this case. It was hoped that by identifying the source of the problem, the method could be adapted.

36. It was considered that the two most likely problems were either:

- a) roots contain high levels of soluble solids other than sugars, or
- b) the method for extracting root sap does not give a representative sample of the root tissue.

Figure 1.



37. To investigate these two possibilities, the following three measurements were compared.

- a) Refractive index measurements of root sap extracted by grating and pressing (i.e. the original refractive index method).
- b) Refractive index measurements of water extracts of ground, freeze dried root tissue, results expressed with respect to fresh weight of tissue.

c) HPLC analysis of water extracts of ground, freeze dried root tissue, results expressed with respect to fresh weight of tissue (i.e. the original HPLC method).

38. If the main problem with the refractive index measurement were due to the fact that soluble solids other than sugars were present in the sweet potatoes, then this would be indicated by differences between results obtained by methods b) and c) but not between a) and b). On the other hand, if the main problem were due to the fact that the method for extracting root sap does not give a representative sample of the root tissue, then this would be indicated by differences between results obtained by methods a) and b), but not between b) and c).

**Table 1: Correlation between methods a), b) and c)\*.**

\* Methods as described in text.

a) correlation between method a) and method c)

Cultivar	Correlation	R2	slope	intercept
Kemb10	0.37	13%	1.49	5.01
Kemb36	0.75	55%	2.09	3.65
SPK004	0.61	37%	1.41	6.39
KSP20	0.36	13%	0.74	5.89

a) correlation between method b) and method c)

Cultivar	Correlation	R2	slope	intercept
Kemb10	0.30	14%	0.47	5.21
Kemb36	0.93	86%	1.13	2.56
SPK004	0.86	73%	1.13	3.77
KSP20	0.46	21%	0.54	3.89

c) correlation between method b) and method a)

Cultivar	Correlation	R2	slope	intercept
Kemb10	0.60	36%	0.19	4.90
Kemb36	0.89	79%	0.39	2.24
SPK004	0.64*	41%	0.37	3.54
KSP20	0.42	17%	0.24	3.94

\* Data in brackets is calculated omitting one outlying data point.

39. The results given in Table 1 (a-c) are disappointing in that they do not clearly indicate that either of the postulated sources of error is alone responsible for the variation observed, but rather that both may contribute. Furthermore, they indicate that the accuracy of the method is cultivar dependent, as the correlations obtained in all cases is higher for Kemb36 and SPK004, than the other two cultivars. A comparison of replicate measurements indicated that methods b) and c) had small errors, with a 95% confidence limit of 0.7 for method b) and 0.6 for method c). (No duplicate measurements had been made for method a).)

40. Some further study of this problem is justified, given the relative widespread use of this method. Nevertheless, the present conclusion of this study must be that an alternative field method for measuring sugar content should be sought.

#### **Laboratory studies of post-harvest physiology of sweet potato roots under tropical conditions.**

##### **Development of a rapid method for the analysis of sugars in sweet potato.**

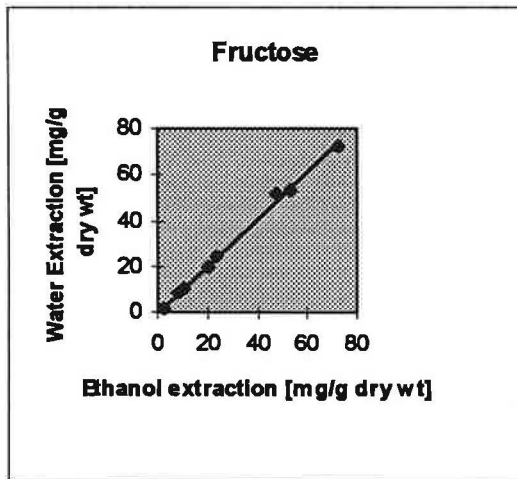
41. Standard methods of analysis of sugars from root crops involve extraction of freeze-dried tissue samples in 80% ethanol by refluxing on a Soxhlet apparatus for 4 hours, followed by the use of chemical assays of reducing and non-reducing sugars. A more appropriate method involving water extraction followed by HPLC analysis has been developed.

42. The use of HPLC (Hypersil APS2 column, 80% acetonitrile mobile phase) allows the separation of the major sugars found in sweet potato; fructose, glucose, sucrose and maltose. The standard ethanol extraction method (see Appendix 1 for details) has the disadvantages that it is time consuming, gives sugar concentrations which are often close to or below the detection limit of RI (Refractive index) detectors usually used for HPLC, and even with careful removal of ethanol, produces a large solvent peak when injected onto the HPLC. As an alternative, therefore, a method of water extraction was developed, which involved shaking 1 g of freeze-dried sample in 20 ml of water for one hour at room temperature.

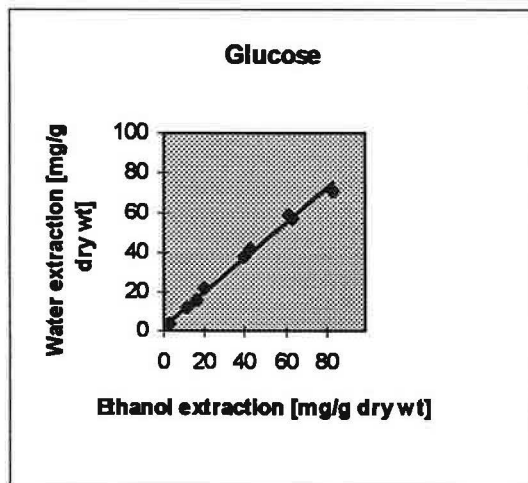
43. In order to check the efficiency of water extraction, the levels of fructose, glucose and sucrose obtained from 10 freeze-dried sweet potato samples by the two methods were compared. The results are shown in Figure 3 (a-c). Correlation coefficients of 0.998, 0.997 and 0.979 were obtained between the two methods for fructose, glucose and sucrose respectively. Water extraction was therefore used for all subsequent analyses.

**Figure 2:** A comparison of ethanol extraction and water extraction of sugars from freeze-dried sweet potato samples.

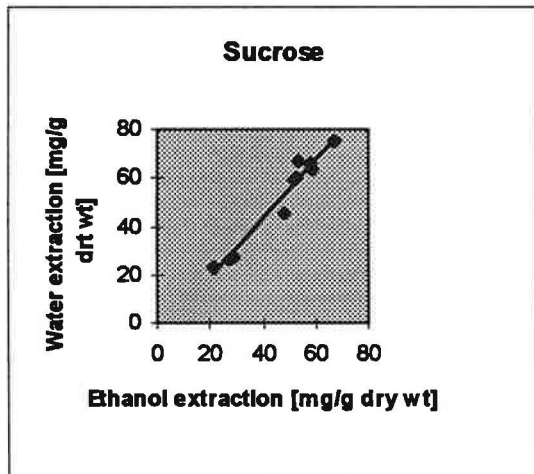
(a)



(b)



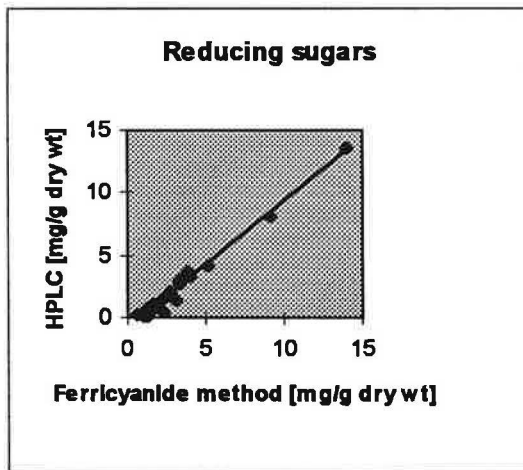
(c)



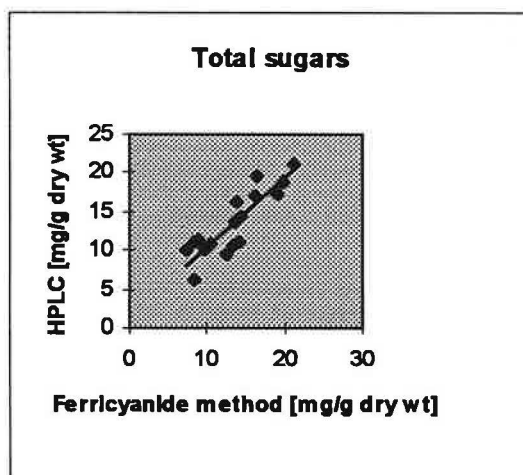
44. The analysis of sugars by HPLC was compared to the measurement of reducing sugars and total sugars by chemical assay (For detailed methods see Appendix 1). Highly significant correlations were obtained between the two sets of measurements (0.992 and 0.875 for reducing sugars and total sugars respectively), indicating the validity of the HPLC measurement. The results obtained are illustrated in Figure 3 (a,b)

**Figure 3:** A comparison of sugar concentrations measured by HPLC and chemical analysis.

(a)



(b)



### **Post-harvest characteristics of four Kenyan cultivars of sweet potato.**

45. The results obtained from a study to compare the post-harvest physiological characteristics of four Kenyan sweet potato cultivars are summarised in Tables 2-6 and Figures 4 and 5. Two of the cultivars; Kemb36 and KSP20 are perceived by Kenyan traders to have long shelf-life, while the other two; Kemb10 and SPK004 are perceived to be perishable (Carey T. *pers. comm.*). A comparison of the post-harvest physiological characteristics of these cultivars was carried out with the objective of determining which of these characteristics may be associated with long shelf-life. For ease of interpretation, as an indication of storability, in subsequent tables Kemb36 and KSP20 are marked with a +, and Kemb10 and SPK004 with a - .

46. In addition to cultivar effects, pre-harvest factors can affect post-harvest characteristics. This is demonstrated in Table 2, showing initial root composition, where results from a preliminary study using roots harvested during an earlier season are also given. Despite seasonal differences, the results indicate distinct cultivar differences, with KSP20 having a lower dry matter content than the other three cultivars. Consistent with previous observations on sweet potato storage roots (Woolfe 1992), the major sugars detected were sucrose, glucose, fructose, with low amounts of maltose. KSP20 was again distinct from the other three cultivars in having lower sucrose content and higher levels of glucose and fructose.

47. During storage for almost seven weeks, loss of fresh weight was near 10% for all cultivars (Table 3). No consistent increase in dry matter content was observed, suggesting that the weight loss was due to the metabolism of carbohydrate, rather than water loss. This is supported by the fact that the rates of root respiration observed (Figures 5 a,b , see below) were close to the theoretical levels for the rate of loss of dry matter (125 mg CO<sub>2</sub>/Kg dry weight would lead to approximately 10% loss of dry matter over 7 weeks). Towards the end of the storage period, pithiness, thought to be a result of cell shrinkage due to metabolism of starch reserves, was observed in a small number of roots.

48. It has been stated that the main cause of loss during storage of sweet potato roots is physiological deterioration. The results shown here indicate that even at tropical temperatures depletion of starch reserves is unlikely to be a major cause of loss unless very long-term storage is undertaken. A greater cause of loss during long-term storage is likely to be due to sprouting (Devereau 1996). KSP20 was found to have a much lower

rate of sprouting than the other three cultivars. (Table 4 (a,b)), suggesting that it may be a particularly suitable cultivar for longterm storage. Sprouting has previously been associated with an increase in sucrose levels (Collins *et al.* 1990). The results shown here are consistent with this observation. Figure 6 indicates that all the cultivars show increasing levels of sucrose during storage, whereas KSP20 starts with the lowest sucrose levels of all the cultivars, and shows a slower rate of increase.

49. In agreement with studies on North American cultivars (Kays 1985) rates of respiration (Figure 5) tend to decrease over the first three weeks of storage, although this is less marked for SPK004 and Kemb36. Although KSP20 starts with a high rate of respiration, in the longer term, KSP20 and Kemb36, (the two cultivars classified as less perishable), have lower respiration rates than the other two cultivars. These results are contrary to observations on North American cultivars (Kays 1985) that cultivars with lower dry matter have higher rates of respiration and do not store well.

50. In summary, due to the conditions chosen for this study the information obtained is a better indication of the behaviour of the cultivars during long-term storage, than the short-term storage that they would experience during immediate transport to market. Contrary to observations in the literature, even at tropical temperatures, physiological losses due to the rate of starch degradation is likely to be a problem only for storage for more than two months. In this case, the selection of cultivars with lower respiration rates, such as KSP20 or Kemb36, would be advantageous. Losses due to sprouting can also be reduced by selection of low sprouting cultivars such as KSP20. Contrary to the conclusions from work on North American cultivars, cultivars with low dry matter are not necessarily bad for storage. These conflicting observations could be associated with the fact that the East African cultivars tend to have higher dry matter contents than North American cultivars. Low levels of sucrose may turn out to be a good selection criterion for storable cultivars.

51. It should be noted that the classification of cultivars as perishable and non-perishable in this study was based on reputation alone. More information is needed on the conditions and causes of loss associated with these cultivars. Water loss under conditions of low humidity, and susceptibility to rots, both of which are likely to be important, have not yet been considered. Nevertheless the identification of KSP20 as a cultivar which stores well and has distinct physiological characteristics provides us with an opportunity to learn more about the nature of perishability through comparative studies.



**Table 2 :** Characteristics of four Kenyan varieties of sweet potato at the start of the study.

		<b>Kemb10</b>	<b>Kemb36</b>	<b>SPK004</b>	<b>KSP20</b>
		-	+	-	+
<b>Fresh weight:</b> [g]	<b>Expt 1</b>	203 +/- 30	312 +/- 47	248 +/- 31	260 +/- 21
	<b>Expt 2</b>	<b>247 +/- 8</b>	<b>289 +/- 23</b>	<b>270 +/- 20</b>	<b>195 +/- 8</b>
<b>Dry matter content:</b> %	<b>Expt 1</b>	36.0 +/- 1.3	36.3 +/- 1.0	36.1 +/- 0.1	28.4 +/- 0.2
	<b>Expt 2</b>	<b>32.2 +/- 1.2</b>	<b>33.0 +/- 1.0</b>	<b>32.3 +/- 0.4</b>	<b>27.1 +/- 1.0</b>
<b>Sugar [% dry matter]</b>					
<b>Sucrose</b>	<b>Expt 1</b>	13.1 +/- 1.1	11.0 +/- 1.0	9.8 +/- 1.5	9.3 +/- 1.5
	<b>Expt 2</b>	<b>5.5 +/- 0.3</b>	<b>3.7 +/- 0.5</b>	<b>5.5 +/- 0.4</b>	<b>2.8 +/- 0.2</b>
<b>Glucose</b>	<b>Expt 1</b>	0.4 +/- 0.2	0.8 +/- 0.2	0.6 +/- 0.3	4.8 +/- 1.0
	<b>Expt 2</b>	<b>1.1 +/- 0.4</b>	<b>0.5 +/- 0.1</b>	<b>0.4 +/- 0.1</b>	<b>6.6 +/- 0.7</b>
<b>Fructose</b>	<b>Expt 1</b>	1.0 +/- 0.5	1.1 +/- 0.3	1.0 +/- 0.5	5.0 +/- 1.1
	<b>Expt 2</b>	<b>1.0 +/- 0.4</b>	<b>0.5 +/- 0.1</b>	<b>0.2 +/- 0.0</b>	<b>6.3 +/- 1.0</b>
<b>Maltose</b>	<b>Expt 1</b>				
	<b>Expt 2</b>	<b>0.8 +/- 0.2</b>	<b>1.0 +/- 0.1</b>	<b>0.8 +/- 0.2</b>	<b>1.0 +/- 0.4</b>

*Results are expressed as mean +/- s.e. (n = 5 for all measurements except for fresh weight in which case n= 23, 24, 25, 23 for expt 1 and 66,60,86,120 for expt 2 for Kemb10, Kemb36, SPK004 and KSP20 respectively.)*

**Table 3:** Fresh weight of roots expressed as a percentage of the weight on the first day of storage.

<b>Days of storage</b>	<b>Kemb10</b>	<b>Kemb36</b>	<b>SPK004</b>	<b>KSP20</b>
	-	+	-	+
<b>4</b>	97.1	96.8	97.5	97.5
<b>11</b>	94.6	94.3	95.1	94.9
<b>18</b>	93.6	93.3	93.5	94.1
<b>26</b>	92.2	92.4	92.1	93.2
<b>33</b>	90.7	91.8	91.1	92.6
<b>40</b>	88.7	91.0	90.2	91.4
<b>47</b>	88.1	90.3	88.9	90.6

*Percentage of initial weight was calculated by weighing each individual root on each of the days, and comparing it with the weight at the start of the study. The number of roots weighed decreased as roots were selected for destructive sampling. Thus on day 4 n=56,50,56,89 and on day 47 n=22,17,13,25 for Kemb10, Kemb36, SPK004 and KSP20 respectively.*

**Table 4: Dry matter content [%]**

Days of storage	Kemb10	Kemb36	SPK004	KSP20
	-	+	-	+
0	32.2 +/- 1.2	33.0 +/- 1.0	32.3 +/- 0.4	27.1 +/- 1.0
4	32.8 +/- 1.4	32.7 +/- 0.5	31.4 +/- 0.5	26.4 +/- 0.8
11	35.9 +/- 0.9	33.7 +/- 0.6	32.7 +/- 0.3	26.4 +/- 0.7
19	35.9 +/- 1.0	34.3 +/- 1.0	31.7 +/- 1.7	25.1 +/- 0.4
26	31.7 +/- 1.0	33.3 +/- 0.9	29.9 +/- 0.2	27.7 +/- 0.1
33	35.9 +/- 1.2	33.9 +/- 0.6	32.4 +/- 1.9	26.9 +/- 0.8
40	32.3 +/- 2.4	33.7 +/- 1.0	33.4 +/- 1.5	28.0 +/- 0.8

*Values are expressed as the mean +/- s.e. (n=4 except for day 0 where n=5)*

**Table 5: Sprouting during storage****(a) number of sprouting areas**

Days	Kemb10	Kemb36	SPK004	KSP20
	-	+	-	+
4	2.07 (2.11)	4.90 (3.12)	8.04 (2.23)	0.09 (0.39)
11	2.91 (2.44)	7.79 (3.18)	9.94 (3.77)	0.16 (0.50)
18	3.67 (2.70)	7.68 (2.85)	10.07 (4.23)	0.35 (0.80)
26	3.59 (2.39)	8.69 (3.31)	10.60 (3.25)	0.67 (1.20)
33	3.55 (2.47)	8.23 (2.23)	12.65 (5.30)	1.14 (1.67)
40	3.81 (2.62)	7.95 (1.96)	12.24 (3.38)	1.33 (1.87)
47	4.17 (2.73)	8.12 (1.80)	13.46 (3.99)	1.48 (2.00)
54	4.39 (2.83)	6.94 (1.71)	10.85 (2.67)	2.28 (2.40)

**(b) length of sprouts**

Days	Kemb10	Kemb36	SPK004	KSP20
	-	+	-	+
4	2.11 (1.93)	2.96 (2.37)	7.44 (2.33)	0.19 (1.12)
11	4.96 (4.44)	7.00 (2.34)	15.92 (6.61)	0.52 (2.20)
18	7.81 (6.44)	9.56 (2.98)	21.79 (9.01)	1.00 (2.68)
26	10.70 (7.13)	12.41 (4.05)	29.00 (9.79)	2.67 (5.56)
33	11.16 (7.79)	14.38 (5.55)	27.00 (8.65)	4.14 (7.83)
40	20.58 (16.50)	15.38 (9.41)	28.82 (8.93)	3.91 (7.08)
47	30.50 (27.86)	19.12 (7.75)	38.85 (20.43)	6.87 (10.61)
54	95.17 (79.00)	32.65 (17.33)	75.38 (30.99)	14.33 (20.64)

*Results are expressed as the mean of observations. Standard deviation is given in brackets.*

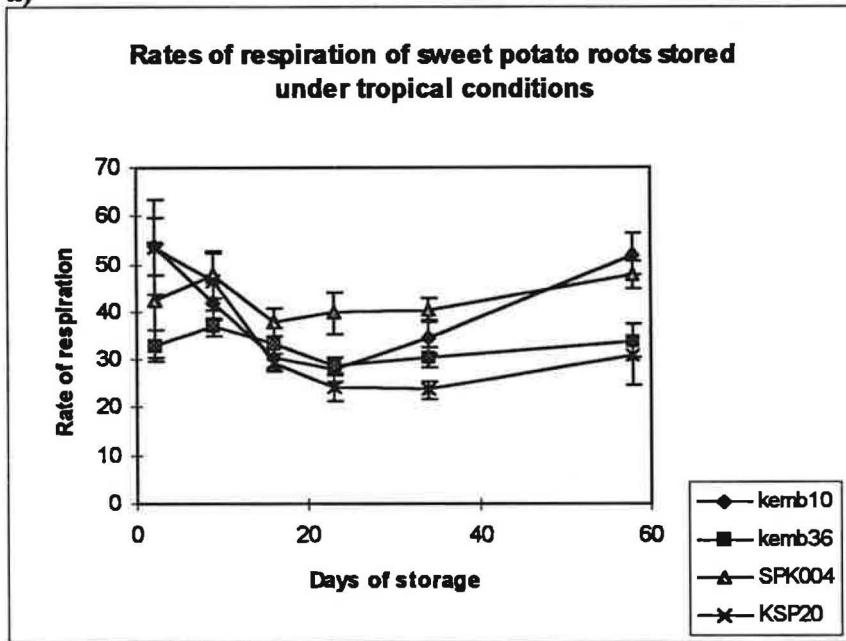
**Table 6:** Rates of respiration measured during storage.

Days of storage	Rates of respiration [ mg CO <sub>2</sub> /Kg fresh weight/hour]			
	Kemb10	Kemb36	SPK004	KSP20
	-	+	-	+
2	53.78 +/- 5.94	32.84 +/- 3.37	42.23 +/- 11.96	53.49 +/- 9.88
9	42.15 +/- 3.93	36.96 +/- 1.86	47.82 +/- 5.02	46.33 +/- 6.05
16	30.47 +/- 1.89	33.49 +/- 3.33	37.80 +/- 2.88	29.37 +/- 1.74
23	27.83 +/- 2.48	28.84 +/- 1.78	39.75 +/- 4.33	24.24 +/- 2.70
34	34.58 +/- 3.29	30.44 +/- 2.06	40.50 +/- 2.40	23.69 +/- 1.98
58	51.91 +/- 4.30	33.86 +/- 3.44	47.73 +/- 2.89	31.05 +/- 6.23

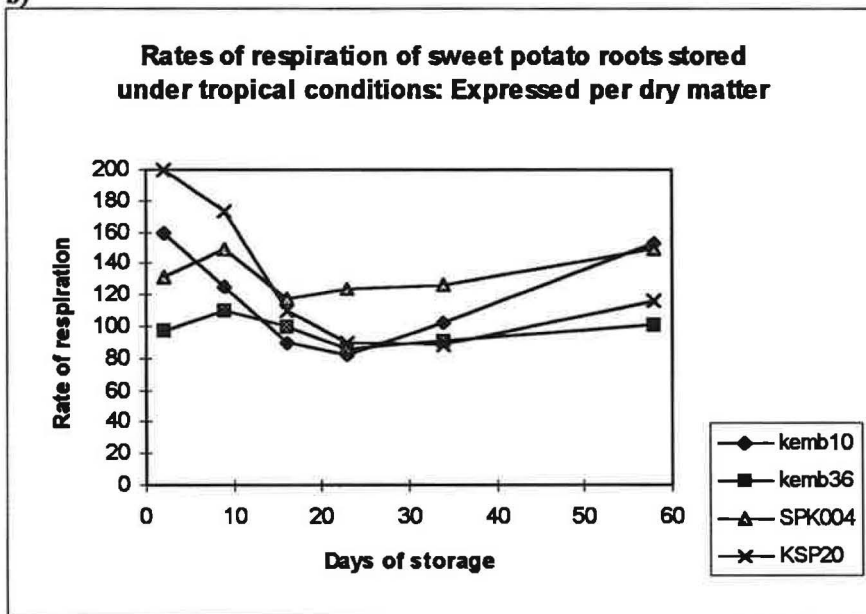
*Results are expressed as the mean +/- standard error (n=8, except for day 2 where n=4)*

**Figure 5:**

a)



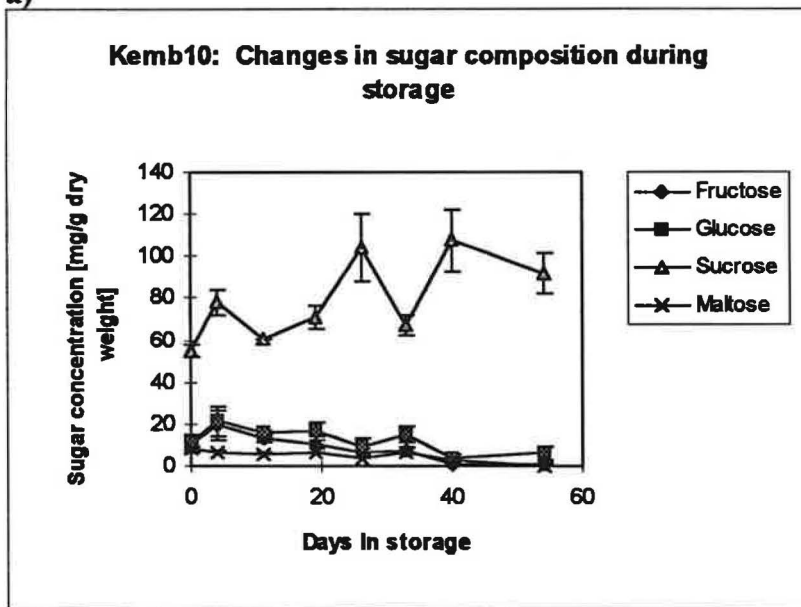
b)



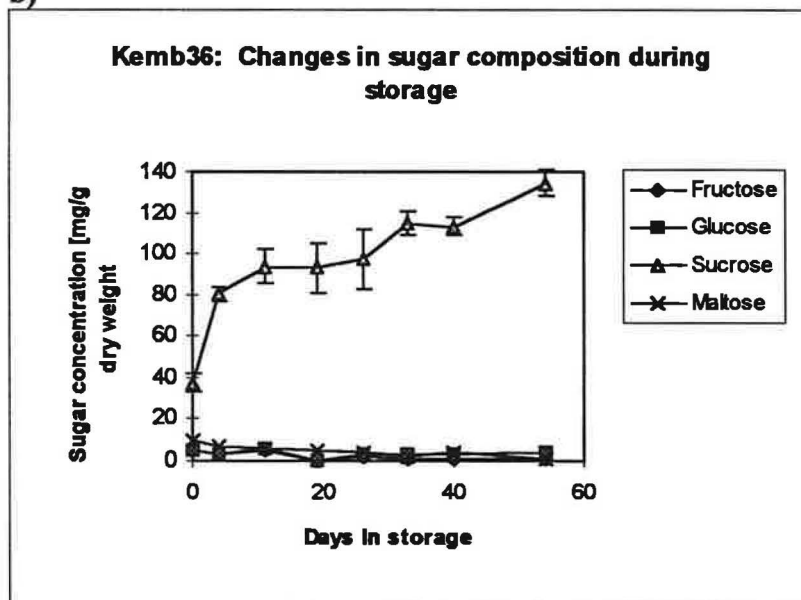
Rate of respiration was measured over 5-7 hours in a closed system, using gas chromatographic measurements of  $CO_2$ . Data is expressed as mean  $\pm$  s.e. of 8 roots. For a) rate of respiration is expressed as  $mg\ CO_2/Kg\ fresh\ weight/h$ , for b) rate of respiration is expressed as  $mg\ CO_2/Kg\ dry\ matter/h$  and was calculated using average dry matter contents measured during the whole storage experiment, as no consistent changes were seen (Kemb10 33.8%, Kemb36 33.5% SPK004 32.0% KSP20 26.8%)

**Figure 6: Changes in sugar composition during storage.**

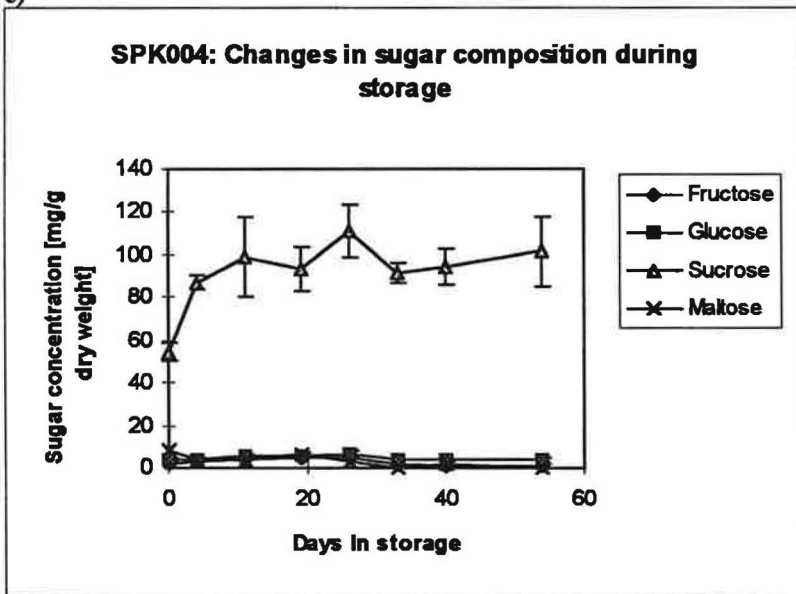
a)



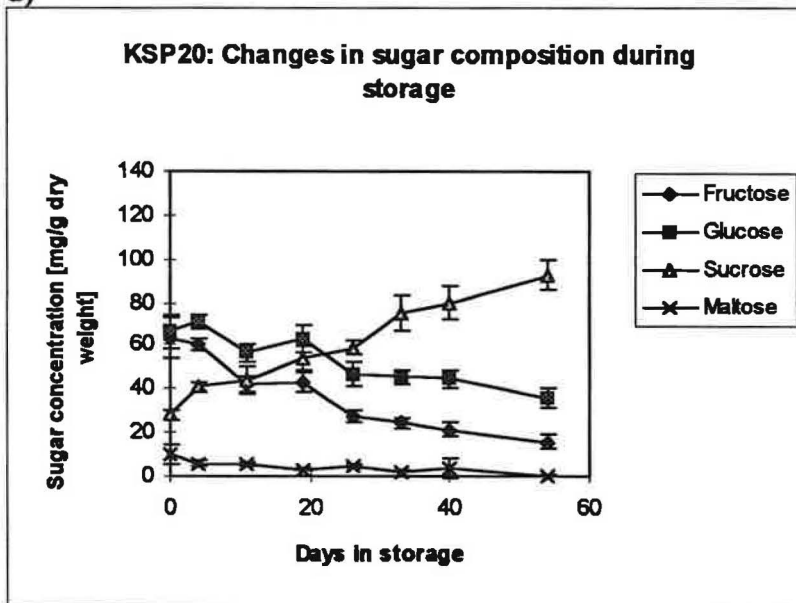
b)



c)



d)



*Sugar levels were measured by HPLC analysis of freeze-dried samples following water extraction. Data is expressed as the mean +/- s.e. of four roots.*

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## Survey of existing handling practices for sweet potatoes in selected areas of Tanzania.

52. Inferences drawn from primary data collected by the 'Transfer of Needs Assessment Methodologies and Post-Harvest Technologies for Non-Grain Starch Staple Food Crops in Sub-Saharan Africa Project' and some secondary sources have suggested that a variety of constraints may impact on the trading of fresh sweet potato in Tanzania. Although previous work has gathered some information on these issues, data specific to the urban and peri-urban marketing of sweet potato in the Lake Zone and areas supplying Dar es Salaam has been lacking. To rectify this deficiency, and as a contribution to the Crop Post-Harvest Programme (CPHP) funded from the Natural Resources Department [NRD], a survey was conducted in Tanzania during the period mid October to early December 1995 with the objectives of characterising the marketing systems for sweet potato in these two areas, documenting the perceived constraints identified by traders engaged in the transport and marketing of fresh sweet potatoes and recording trader and consumer varietal preferences.

53. Various staff drawn from the Natural Resources Institute (NRI), the Tanzania Food and Nutrition Centre (TFNC) and the Agricultural Research Institute, Ukiriguru and Chollima Agro-Scientific Research Station, Dakawa contributed to gathering market intelligence in the urban districts of Mwanza (Lake Zone), Morogoro and Dar es Salaam [DSM] (Coast Zone). The target groups investigated included producers and producer-traders, traders, wholesalers, retailers and snack food vendors and customers etc. Informal semi-structured interviews were undertaken with over 100 respondents at in excess of 20 markets and other locations.

54. A major trend recorded was the ever growing importance of fresh sweet potato as both a domestic and convenience food in the expanding urban centres. Demand for produce had increased over recent years to the extent that in, particular areas such as Ukerewe (Lake Zone) and Morogoro (Coast Zone) producers were changing from predominantly small-scale or subsistence farming of a variety of crops to cash cropping of sweet potato. In response to the demands of urban consumers particular emphasis was now placed on the production of a narrow selection of favoured varieties. Predominant amongst these is the variety grown in the area of 'Gairo' in the vicinity of Morogoro. Over the last decade this area had developed into the major sweet potato supplier for the markets of Dar es Salaam.

55. From the perspective of producers and producer/traders, sweet potato was considered one of the most profitable of food crops, the market potential of which was only limited by its seasonality and restricted storage life. Constraints to production were often identified as lack of planting material either in absolute terms (lack of vegetative 'slips' carried over from the previous growing season) or on account of the prevailing cost of purchasing slips at the beginning of the growing season. The availability of such planting material was invariably aggravated by extended dry seasons and the infestation of nursery plots by a variety of insect pests. Once established, the major constraints to the quality and extended in-ground storage of roots were weevil infestation and the decline in culinary quality associated with the delayed harvesting of the crop. Both factors which limited the potential length of the cropping season irrespective of the prevailing rainfall patterns. Despite the enhanced importance of sweet potato crop, producers do not apply fertilisers to their plots and, on account of lack of finance, the use of insecticides to control pests was not considered an option.

56. Once harvested, fresh sweet potatoes were considered to have a maximum shelf-life of 7 days and marketing systems had developed to accommodate this major constraint. In-ground or pit storage of harvested sweet potatoes, although known to be of some benefit, was not practised by the majority of producers neither were any attempts made to 'cure' the sweet potato roots after harvest to extend their potential storage-life. Traders in particular complained of storage rots causing losses in consignments either on account of poor grading at the point of harvest and/or extended periods in transit. Losses of 30 to 50% were cited occasionally but it would appear that post-harvest losses throughout the marketing chain were usually considerable less than this and more of the order of 10-15%. There is, however, no hard data to confirm this impression.

57. The traditional drying of sweet potato chips although still practised in rural areas in the Lake Zone was not observed in the urban area around Mwanza and respondents suggest that there is little demand for this product in the urban populations. In the Coast Zone the prevailing level of relative humidity precludes the successful drying of sweet potato chips in the sun neither are dried chips a traditional component of the diet.

58. These observations suggest that there is scope for improving the versatility of the sweet potato crop by judicious selection of indigenous landraces and/or the introduction of germplasm from abroad in line with consumer preferences and production constraints.

**Improvements in the handling practices and the introduction of on-farm storage techniques could also help to extend the shelf-life of harvested roots and extend the seasonal availability of particular varieties of sweet potatoes.**

**59. The information obtained about the structure of the marketing systems has been used to plan more detailed surveys in the same areas, to be carried out within the Crop Post-Harvest Programme.**

## **CONTRIBUTION OF OUTPUTS**

60. This project is focused towards Purpose 4 of the RNRRS Crop Post-Harvest Programme, which seeks to reduce storage and processing losses. The technology to store sweet potato roots for several months, usually by refrigeration, exists and is used in developed countries. However, these technologies are expensive, and inappropriate for resource poor countries such as in East Africa, the region of focus for this work.

61. One route to reducing losses in developing countries, is by appropriate low-cost improvements to handling practice. Surveys of the marketing system carried out within this project have provided the necessary first step to planning the more detailed studies needed to identify appropriate measures. The results have been used to develop a programme to be carried out as a collaboration between NRI, the Tanzanian National Root and Tuber Crops Programme and the Tanzania Food and Nutrition Centre, funded within the RNRRS Crop Post-Harvest Programme.

62. The main emphasis of this project, however, is towards the selection and promotion of more storable cultivars. Improvement of germplasm is a means to improve the product with little or no cost to farmers and traders. Technical support provided to the Tanzanian National Root and Tuber Crops Programme has helped them to initiate the establishment of a methodology for assessing varieties for such characteristics. The advances made to date will be further exploited within a subsequent project funded by the RNRRS Crop Post-Harvest Programme which includes post-harvest evaluation of Tanzanian germplasm. In order to assess a sufficiently wide range of varieties, a procedure that is less labour intensive and uses less plant material will need to be developed.

63. To help the National Programmes of Tanzania and other developing countries to carry out assessments and research more effectively, the development of appropriate field methods is very important. In addition the identification of problems with techniques, such as reported here for the use of refractive index for sugar determination is necessary to allow improvement of methodologies, as well as to prevent the collection of false information. Work on this method and others will continue at NRI.

64. An understanding of the physiological factors associated with perishability of sweet potato roots is important for efficient selection of improved cultivars within

breeding programmes, especially if indirect selection criteria for longer-keeping varieties can be identified. This is important for breeding programmes carried out by the National Programmes, but probably even more for longer term breeding programmes as carried out by the International Potato Centre.

65. As most of the physiological work reported in the literature has been carried out on North American and Japanese cultivars, it was necessary to start by collecting a certain amount of baseline data on which future work can be based. Through this work, one variety, KSP20 has been identified which stores well and has distinct physiological characteristics. Comparative studies of this variety with more perishable varieties will provide us with an exciting opportunity to learn more about the nature of perishability. The preliminary observation that low sucrose levels may provide a selection criteria for cultivars suitable for long-term storage requires confirmation.

### **Publications**

Two publications in preparation:

POLLARD, A and REES, D. Rapid Method for Analysis of Sugars in Sweet Potato Roots.

REES, D., POLLARD, A, CAREY, E and MATTERS, D. Post-harvest Metabolism of Storage Roots of Four Kenyan Sweet Potato Cultivars.

### **Internal Reports**

BANCROFT, R. (1995) Visit to Tanzania to continue cassava loss reduction sub-project and initiate commodity systems market characterisation with specific reference to Sweet Potato. 10 Oct - 29 Nov 1995

REES, D. (1995). Report on a Visit to Tanzania to Participate in a Training Course on the Standardisation of Research Methodologies for Root and Tuber Crops. 25 April - 12 May

REES, D. (1995) Back to Office Report. Visit to East Africa to Discuss On-going and Completed Trials on the Post-Harvest Evaluation of Sweet Potato Cultivars in Tanzania, and to Develop a Proposal for Submission to the EU (DGXII). 5-25 August.

REES, D. (1995) Back to Office Report. Visit to Mwanza, Tanzania to Attend the National Co-ordinating Meeting for Root and Tuber Crops Research, and to Discuss On-going and Future Collaborative Projects. 10-25 October 1995

REES, D. (1996) Physiological basis for storage life extension in fresh sweet potato. A0422 Final Technical Report.

REES, D, RWIZA, E., NDONDI, T., KILIMA, M.S., CHILOSA, N.N.V., MAYONA, C.M., and KAPINGA, R., (1996) Post-Harvest Evaluation of Local Sweet Potato Cultivars in Tanzania. R6204 Final Technical Report.

#### **Other Dissemination of Results.**

CHILOSA, N.N.V., MAYONA, C.M., RWIZA, E., NDONDI, T., KILIMA, M.S., MBILINYI, L.B., KAPINGA, R., and REES, D (1995) Collaborative research between the Root and Tuber Research Programme and NRI: Post-harvest evaluation of sweet potato varieties. Paper presented at the National Root and Tuber Co-ordinating Committee Meeting held at Mwanza on 16th-17th October 1995.

REES, D, RWIZA, E., NDONDI, T., KILIMA, M.S., CHILOSA, N.N.V., MAYONA, C.M., and KAPINGA, R., (1996) Sweet potato post-harvest evaluation trial: Method Manual.

REES, D., POLLARD, A, MATTERS, D and CAREY, E. Post-Harvest Physiology of Sweet Potato Storage Roots Related to Storability.

Poster accepted for Second International Symposium on the Biology of Root Formation and Development. Jerusalem, Israel, June 23-28 1996.

## APPENDIX 1

### LABORATORY METHODS

#### **Measurement of refractive index of root sap.**

For measurement of refractive index of root sap, the root was first cut in half longitudinally. A layer of root tissue was removed along the whole surface of one half of the root using a cheese grater. The grated material was mixed, and a sample of this was then pressed using a garlic press. Two drops were placed on the viewing surface of a hand-held refractometer (David Bishop 0-30% Brix) for measurement. The brix value (expressed as % sucrose) was measured directly from the scale.

#### **Measurement of sugars by chemical assay and by HPLC**

##### **Extraction of sugars by ethanol**

Samples of sweet potato root tissue were freeze-dried and ground to a fine flour. 1 g of dry flour was extracted in 200 ml 85% v/v ethanol for 2 hours on a soxhlet apparatus. The extract was made aqueous and made up to 50 ml.

##### **Extraction of sugars by water.**

Samples of sweet potato root tissue were freeze-dried and ground to a fine flour. 1 g of dry flour was extracted in 20 ml distilled water by shaking for one hour at room temperature.

##### **Measurement of sugar by chemical assay**

Sugars were measured as described in "Methods Manual for the Assessment of the Quality Characteristics of Non-Grain Starch Staples" NRI publication. Reducing sugars were determined spectrophotometrically at 380 nm by reaction with ferricyanide. Total sugars were determined after acid hydrolysis using hydrochloric acid for 24 hours at room temperature.

##### **Measurement of sugars by HPLC**

Prior to analysis samples were diluted with acetonitrile to 80% acetonitrile, and were filtered through a 0.45 µm syringe filter. Samples were injected onto an amino-bonded HPLC column (Hypersil APS-2, 20 cm) maintained at 30°C, using 80% acetonitrile running at 0.5 ml/min as the mobile phase. Sugars were detected using a refractive index detector (Varian RI-4), and peak sizes were calculated using a Perkin Elmer LCI-100 Integrator. Peak heights were used to calculate sugar concentrations.

### **Sweet potato roots:**

Four cultivars: KSP20, SPK004, KEMB10 and KEMB36 were grown in field trials outside Nairobi, Kenya by the International Potato Centre, were harvested during the period 20-24 March, and imported to the UK on 24th March. Roots were chosen that were free from insect infestation. They were dipped in fungicide following harvest to prevent fungal rotting.

On arrival at NRI 30-40 roots of similar size were chosen for each cultivar. Each of these was labelled with variety and number towards one end with a permanent marker.

### **Storage conditions:**

Approximately 7.5 Kg of roots were stored in each of 8 black plastic bins (2 for each variety) (diameter 50 cm, height 100 cm), on a rack supported at a height of 40cm above the base of the bin which contained water to a depth of 8 cm. Air was bubbled through the water to maintain a high relative humidity and prevent the build up of CO<sub>2</sub>. The bins were maintained in a constant environment room at a temperature of 27°C. The temperature and relative humidity to which the roots were exposed were monitored frequently by means of thermistors and hair hygrometers. The temperature within the bins was 27°C and the relative humidity was maintained between 90 and 100% throughout the experiment.

### **Atmospheric composition**

The composition of the air surrounding the roots was monitored by gas chromatography. This was sampled by means of fine nylon tubing placed so as to allow sampling from the centre of the pile of roots, and also from the boundary layer immediately above the roots (the boundary layer was assumed to extend up to 1 cm from the surface of the top layer of roots.) Composition was found to be close to ambient throughout the experiment.

### **Analysis of roots:**

At the start of the experiment, weight, length and maximum diameter was measured for each root.

At weekly intervals, the roots were carefully removed from each bin to be weighed individually, and assessed for sprouting and rotting. Sprouting was recorded on the basis of the number of sprout clusters, and the average length of sprouts. Rotting roots were removed from the experiment.

Sample roots (5 of each cultivar at the start of the experiment, and 4 of each cultivar on later occasions) were removed for the following analyses.



**Visual assessment of external appearance:**

Shrivelling (1-5 subjective scale)

Moulds visible at surface (1-5 scale: 1 0%, 2 1-10%, 3 10-25%, 4 25-50%, 5 >50% . (Any roots scoring 4 or 5 for moulds were omitted from further analysis.)

**Penetrometer readings:**

The roots were peeled. An electronic gauge with 6mm flat ended probe, mounted on a mechanical drill stand was then used to measured penetrating force [Kgf]. 4 measurements were made on each root; 2 halfway along the root at opposite sides, and 2 toward the ends of the root.

N.B. towards the end of the experiment, a handheld penetrometer was used instead of the electronic gauge, and was found to be more reliable.

**Visual assessment of internal appearance:**

Each root was cut in half widthways and left for 5 mins to assess browning (1-3) and exudation of latex (+/-).

The roots were then cut lengthways and assessed for:

Pithiness (+/-)

Internal rots (1-5) [scoring as above]. Any roots scoring 4 or 5 were omitted from further analysis.

**Refractive Index of sap:**

Material was taken from the longitudinal surface of the root by grating. A sample of this was then pressed in a handheld press and the refractive index of the sap obtained was measured using a hand-held refractometer

**Dry weight:**

One longitudinal half of the root was cut into cubes. A sample of this was used for dry weight determination. About 10 g was weighed, and then dried in a vacuum oven at 60oC for 24 hours followed by 2 hours at 90oC in a conventional oven.

**Analysis of sugar and starch content.**

The second longitudinal half of the root was cut into cubes. A weighed portion (about 20 g ) was frozen and freeze-dried for later analysis for sugar and starch content.

**Measurement of rates of respiration**

On 6 dates during the storage period (27/3, 3/4, 10/4, 17/4, 28/4, 22/5) (days 2, 9, 16, 23, 34, 58) the rates of respiration of selected roots were measured. 8 roots were selected for each variety (4 from each bin, but otherwise at random). (N.B. only 2 roots from each bin was selected on day 2). Each root was weighed and placed in a 3L glass jar, equilibrated to 27°C, containing a beaker with 30ml water to maintain a high humidity, and was placed in an incubator maintained at 27°C. Each jar was sealed for between 5 and 7 hours. At the end of this time a gas sample was removed from the jar through a teflon port using a gas tight syringe and was analysed for CO<sub>2</sub> and O<sub>2</sub> concentration by gas chromatography.

For calculation of the volume of gas in the jar, the volume of the root was calculated from its weight assuming a specific gravity of 1.05.