INFLUENCE OF STORAGE CONDITIONS ON DEVELOPMENT OF HARD TO COOK DEFECT IN COMMON BEANS AND THE SUBSEQUENT NUTRITIONAL CHANGES

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Abstract
Common beans form one of the main sources of proteins in poor sub-Saharan Africa. However, common beans are succumbed to the Hard-to-cook (HTC) defect which is suggested to develop due to storage at temperatures higher than 25ºC and relative humidity of more than 70%. This results in increased cooking time, fuel consumption and water use thus lowering the rate of utilization of beans at domestic level. This study was aimed at understanding the development and the effects of hard to cook on an easy and hard to cook bean varieties. Samples were collected from KARI (Kenya Agricultural Research Institute), Thika. The hard (Pinto) and easy (Red Haricot) to cook beans were stored at varying temperatures (25, 35, and 45°C) and humidity (75 and 85%) levels, followed by pre-treatment (soaking in deionized water, sodium carbonate and calcium chloride) and thermal treatment. The samples were assessed for physical (Seed density, seed porosity, characteristic dimension of beans, hydration coefficient, and swelling coefficient) changes. Beans stored at 45°C at 75% RH were significantly harder than those stored at 25ºC/75% and 35ºC/85% after 2 months. The hydration coefficient and swelling coeffient decreased with increasing storage time while the geometric properties did not change. Bean hardness, solutes and electrolytes leaching after 16hr soaking substantially increased with increased temperature. It was concluded that the longer the beans are stored at higher temperatures the more they develop the hard to cook defect.

Key words: *Phaseolus vulgaris L.*, Hard to cook defect, Physico-chemical properties, Storage conditions

1.0 Introduction

Legumes generally, and the common bean (*Phaseolus vulgaris* L.) in particular, act as important sources of protein and other essential nutrients for most of the world's population. Food legumes have significant nutritional and health advantages for consumers. They provide high levels of plant protein and are particularly important in plant-based diets where cereals are predominantly used to compliment amino acids thereby enhancing protein quality. The world leader in production of dry bean is Brazil, followed by India and then China. In Europe, the most important producer is Germany. The regions of highest bean consumption in the world include Latin America, where legume consumption ranges from 1 kg/capita per year (Argentina) to 25 kg/capita per year (Nicaragua). Common beans dominate and account for 87 % of the total legume product consumption. Bean consumption in Eastern and Southern Africa exceeds 50 kilograms per person per year. Cooking time is one of the main criteria used in evaluating pulse cooking quality (Moscoso *et al*., 1984). Long cooking times are a major constraint to wider acceptance and use of pulses. Furthermore, overcooking of pulses has been reported to result in a reduction in the nutritive value of the protein (Walker and Kochhar, 1982). HTC development is associated with storage of beans under conditions of high temperature (≥ 25°C) and high relative humidity (≥ 65%) (Shiga *et al*., 2004).Beans with this defect are characterized by extended cooking times for cotyledon softening, are less acceptable to the consumer, and are of lower nutritive value. Mechanisms involved in the HTC defect have not been elucidated satisfactorily. The most important hypotheses that have been proposed to explain the cause of the hardening are lipid oxidation and/or polymerization, formation of insoluble pectates, lignifications of the middle lamella and multiple mechanisms (Reyes-Moreno and Peredes-Lopez, 1993). According to Stambolie *et al*., (1995), HTC development in common beans is influenced mainly by two environmental conditions during growth, namely the soil mineral composition and the amount of rainfall in a particular area. Presence of high levels of Ca and Mg contribute to the formation of insoluble pectates in the intercellular spaces of the cotyledon which hinder rapid movement of water in the seed during coo cooking or soaking. This leads to long cooking time (Stambolie *et al*., 1995). On the other hand heavy rainfall results in the formation of a thin seed coat which allows faster movement of water across the seed coat during cooking hence short cooking time (Stambolie *et al*., 1995). Differences in bean varieties in HTC defect development, were however not dependent on differences in Ca and Mg content. The varietal differences seemed to be more related to the size of the bean grain, with the variety with small seeds exhibiting higher HTC development. According to the work done by Stanley *et al*., (Stanley *et al*., 1990) this could be explained by the fact that the small seeded beans tend to lose moisture faster than bigger ones resulting in concentration of phytases and poly-methyl esterases which accelerates liginification of the beans. Proneness to HTC is a function of variety and storage conditions (Giselle *et al*., 2004).

High relative humidity is known to favor bean hardening. Moisture plays a very big role in preserving the quality of beans and the quality of beans can be maintained by reducing the moisture content or the water activity (Paredes *et al*., 1989). High moisture content and relative humidity also stimulate phytase and pectin methyl esterase activity (PME) (Jones and Boulter, 1983). The phytase liberates Mg from phytic acid and PME hydrolyses pectin to pectinic acid and methanol. The pectinic acid combines with Mg forming magnesium pectate which cements cells together resulting in extended cooking time.

In order to prevent the development of the hard to cook (HTC) defect, several procedures have been proposed including appropriate storage, controlled atmospheres, pre-treatments and development of materials less prone to HTC defect. Research in HTC would best be aimed at decreasing cooking times, increasing nutritive value, improving sensory properties of the seeds and understanding the mechanisms leading the phenomenon which would aid in appropriate methods to prevent it. This study was aimed at understanding the development and the effects of hard to cook on an easy and hard to cook bean varieties stored at varying temperatures (25, 35, and 45°C) and humidity (75 and 85%) levels, followed by pre-treatment (soaking in deionized water, sodium carbonate and calcium chloride) and thermal treatment.

2.0 Materials and Methods

2.1 Plant Material

Common beans (*Phaseolus vulgaris L*) were collected from KARI (Kenya Agricultural Research Institute), Thika. The hard to cook Pinto and the easy to cook Red haricot were divided for storage at various conditions.

2.2 Conditions and Duration of Storage

The hard (Pinto) and easy (Red Haricot) to cook beans were kept in baskets and stored at varying temperatures (25, 35, and 45°C) and humidity (75 and 85%) levels for eight months to see how the hard to cook defect develops.

2.3 Physical Properties

2.3.1 Hydration Coefficient (Imbibition Value)

Hydration coefficient was determined by soaking 5 g of bean seeds at room temperature in 25 ml deionized water (ratio of 1:5). The beans were removed after 18 h from the soaked water, cut into two halves along the fissure and the testa separated from the cotyledon followed by free water removal using a blotting paper and re-weighing. Gain in weight was taken as the amount of water absorbed and expressed as the hydration coefficient (El-Refai *et al*., 1988):

$$\text{Hydration coefficient} = \frac{\text{Weight of bean seeds after soaking}}{\text{Weight of bean seeds before soaking}} \times 100$$

2.3.2 Swelling Coefficient

The volume of raw bean seeds before and after soaking in deionised water for 18 h at 25^{0}C was determined by water volume displaced in a graduated cylinder and expressed as the swelling coefficient (El-Refai *et al*., 1988):

$$\text{Swelling coefficient} = \frac{\text{Volume of bean seeds after soaking}}{\text{Volume of bean seeds before soaking}} \times 100$$

2.3.3 Electrolytes and Solutes Leaching

After 18 h soaking in deionized water the soak water was collected and leached electrolytes were quantified by assessing conductivity with a digital conductivity meter (Sisabata model SC-17A) in mmh/cm at 25^{0}C (Hentges, Weaver, & Nielsen, 1991). To measure solutes leached from beans during soaking, soak water was evaporate, dried in a hot air oven (105^{0}C), cooled in a dessicator, weighed and expressed as mg/g dry weight of beans.

2.3.4 Bulk and Seed Density

A measuring cylinder was filled with beans and the contents weighed in triplicate. Bulk density was calculated as the ratio of the bulk weight and the volume of the container (g/ml). Seed/ true density was obtained by liquid displacement using a top loading balance. Seeds were immersed in distilled water in a beaker. The mass of the displaced water was the balance reading with the seed submerged minus the mass of the beaker plus water. The seed volume was estimated by dividing the mass of displaced water (g) by the density of water (g/cm^3). Seed density was determined by dividing the seed mass by the measured seed volume.

2.3.5 Seed Porosity

This is the property of the grain which depends on its bulk and kernel densities. It was calculated as: $P = 1 - Pb/Pt \times 100$

Where Pb is bulk density (kg/m^3) and Pt is seed density (kg/m^3)

2.3.6 Characteristic dimension of beans

Seeds were selected randomly and their width and length determined using a vernier caliper to an accuracy of 0.001mm, in triplicate. The bulk seed sample for each variety was classified into small (≤9.50), medium (9.50-10.49) and large (≥ 10.50).

2.3.7 Testa-cotyledon test

10 seeds of each variety were weighed in triplicate on weighing balance. The testa was separated from the cotyledon and each weighed separately.

2.3.8 Colour

Seeds packed in transparent polythene bags for the purpose of analysis were selected for their colour determination using colour L.A.B equipment (Minolta Chroma Metre CR-200). The instrument was first calibrated using standard black and white plates (with transparent papers placed on the side plates). After calibration, colour measurements were randomly taken in triplicates. The hue angle of difference (HD) which describes the visual sensation according to an area which appears to be similar to one or proportions of two of the perceived colors, red, yellow, green and blue was calculated according to the following formula:

Hue angle of difference (HD)= $[\tan^{-1}(b^*_x/a^*_x) - \tan{-1}(b^*_0/a^*_0)]^{0.5}$

Where L^*_0, a^*_0, *and* b^*_0 *are values at time zero (time for calibration) and* L^*x, a^*_x, *and* b^*_x *are at time X of object analysis.*

2.4 Data Analysis

Differences between varieties and various treatments was determined using two way analysis of variance. The least significant difference (LSD) at 5% probability level was used for mean separation. Correlation between the cooking time and other parameters was also carried out to determine their relationship.

3.0 Results and Discussion

3.1 Effects of Storage Temperature and Humidity on Physical Properties

Substantial differences occurred in physical characteristics of common beans stored at different conditions (Table 1 and 2) for 6 months. Beans stored at 45°C at 75% RH were significantly harder than those stored at 25ºC/75% and 35ºC/85% after 2, 4, and 6 months. After 16 h soaking followed by 2 h cooking, the puncture force required for seeds stored at 45°C at 75% was higher, whereas seeds stored at 25ºC/75% required a much lower puncture force (Table 1). Hydration and swelling coefficients that reflect the capacity to imbibe water in a reasonable length of soaking time was substantially affected by storage temperature. After 12 h soaking at 25 ºC, the hydration and swelling coeffient was significantly ($p \leq 0.05$) lower in samples stored at higher temperatures, especially those stored at 45°C/75%, compared to samples stored at lower temperature 25ºC/75% and 35ºC/85%. Low hydration and swelling coefficients following storage at high temperature can be due to structural and chemical changes in the testa making it harder and less permeable to water so that it acts as a barrier, preventing water reaching the cotyledons (Liu *et al.,* 1992). Bean hardness, solutes and electrolytes leaching after 16hr soaking substantially increased with increased temperature. Beans stored at 45°C/75% for 2, 4 and 6 months exhibited a higher solute leakage than seeds stored 25ºC/75% and 35ºC/85% for the same period of time.

Table 1: Physical properties of different bean varieties stored at 45ºC 75% for 0-6 months

		PINTO	RHC	PINTO	RHC	PINTO	RHC	PINTO	RHC	PINTO	RHC	PINTO	RHC	PINTO	RHC
	Time (months	Hydration coefficient (%)		Swelling coefficient (%)		Conductivity (mmh/cm)		Leached solutes (%)		100 seed weight		Length		Width	
45/75	0	174.7 ± 5.8	180.6 ± 2.6	207.1 ± 10.1	247.2 ± 10.3	4.7 ± 0.7	4.5 ± 0.3	0.7 ± 0.1	0.9 ± 0.1	39.4 ± 0.4	22.2 ± 0.6	11.4 ± 1.0	11.2 ± 0.6	8.7 ± 0.9	7.1 ± 0.3
	2	172.6 ± 5.1	175.6 ± 9.4	204.2 ± 7.2	200.0 ± 0.0	5.1 ± 0.1	5.5 ± 0.5	1.7 ± 0.2	1.4 ± 0.3	39.7 ± 0.8	25.8 ± 0.8	11.8 ± 0.8	10.9 ± 1.4	8.9 ± 0.8	6.6 ± 0.5
	4	166.1 ± 2.0	170.9 ± 3.9	200.0 ± 16.7	192.9 ± 10.1	6.4 ± 0.1	6.2 ± 0.6	2.1 ± 0.6	2.4 ± 0.7	37.7 ± 2.8	24.3 ± 0.3	11.8 ± 0.7	10.6 ± 0.5	8.7 ± 0.4	6.6 ± 0.5
	6	164.1 ± 1.4	169.7 ± 3.9	197.6 ± 4.1	188.4 ± 11.1	9.4 ± 0.2	9.1 ± 0.1	2.8 ± 2.6	2.6 ± 1.2	37.6 ± 0.4	24.6 ± 0.7	11.9 ± 0.8	10.9 ± 0.7	8.1 ± 0.6	6.8 ± 0.2
		True seed density		Bulk seed density		Porosity		Colour (L)		A		B			
	0	1.2 ± 0.0	1.3 ± 0.0	0.7 ± 0.0	0.8 ± 0.0	40.2 ± 1.0	38.6 ± 0.7	42.1 ± 2.1	53.9 ± 2.8	7.9 ± 2.7	6.6 ± 0.3	0.4 ± 0.7	9.7 ± 0.8		
	2	1.2 ± 0.0	1.2 ± 0.0	1.0 ± 0.0	1.0 ± 0.0	18.4 ± 0.9	17.8 ± 1.1	52.0 ± 2.1	43.7 ± 1.9	7.5 ± 1.0	7.7 ± 1.4	11.6 ± 1.4	0.3 ± 0.1		
	4	1.2 ± 0.0	1.3 ± 0.0	1.0 ± 0.0	1.1 ± 0.0	17.2 ± 1.5	18.5 ± 2.5	47.6 ± 2.5	38.6 ± 1.1	7.8 ± 0.7	9.1 ± 1.4	8.1 ± 0.6	0.8 ± 0.6		
	6	1.3 ± 0.1	1.3 ± 0.1	1.0 ± 0.0	1.1 ± 0.0	22.9 ± 5.8	15.7 ± 3.9	35.8 ± 3.7	21.8 ± 4.9	13.4 ± 0.3	12.8 ± 2.9	17.6 ± 1.7	2.2 ± 1.3		

Values = Mean ± SD. Each value is a mean of 3 replicates. S.D (standard deviation) (p≤0.05)

Table 2: Physical properties of different bean varieties stored at 35ºC 85% for 0-6 months

		PINTO	RHC	PINTO	RHC	PINTO	RHC	PINTO	RHC	PINTO	RHC	PINTO	RHC	PINTO	RHC
	Time (months	Hydration coefficient (%)		Swelling coefficient (%)		Conductivity (mmh/cm)		Leached solutes (%)		100 seed weight		Length		Width	
35/85	0	174.6 ± 5.8	180.6 ± 2.6	207.1 ± 10.1	247.2 ± 10.3	4.7 ± 0.7	4.5 ± 0.3	0.7 ± 0.1	0.9 ± 0.1	39.4 ± 0.4	22.2 ± 0.6	11.4 ± 1.0	11.2 ± 0.6	8.7 ± 0.9	7.1 ± 0.3
	2	169.7 ± 6.8	177.6 ± 5.3	205.6 ± 9.6	245.8 ± 7.2	4.8 ± 0.6	5.4 ± 0.2	1.8 ± 0.7	1.7 ± 0.7	38.1 ± 1.0	25.3 ± 1.9	11.6 ± 0.1	10.0 ± 0.7	8.5 ± 0.5	6.3 ± 0.5
	4	168.3 ± 2.5	175.0 ± 7.3	204.2 ± 5.9	215.7 ± 13.7	4.9 ± 0.1	5.7 ± 0.2	2.1 ± 0.4	1.9 ± 0.7	37.4 ± 0.8	24.2 ± 0.8	10.8 ± 0.5	10.9 ± 0.8	8.1 ± 0.3	7.2 ± 0.7
	6	166.4 ± 3.5	167.0 ± 1.8	202.4 ± 8.3	200.0 ± 0.0	5.6 ± 0.2	6.1 ± 0.9	2.4 ± 1.3	2.5 ± 0.9	37.4 ± 0.6	24.5 ± 0.0	11.4 ± 0.9	10.2 ± 0.3	8.7 ± 0.5	6.5 ± 0.3
		True seed density		Bulk seed density		Porosity		Colour (L)		A		B			
	0	1.2 ± 0.0	1.3 ± 0.0	0.7 ± 0.0	0.8 ± 0.0	40.2 ± 1	38.6 ± 0.7	42.1 ± 2.1	53.9 ± 2.8	7.9 ± 2.7	6.6 ± 0.3	0.4 ± 0.7	9.7 ± 0.8		
	2	1.2 ± 0.0	1.2 ± 0.0	1.0 ± 0.0	1.0 ± 0.0	13.1 ± 2.2	10.9 ± 1.5	54.9 ± 2.0	41.9 ± 4.0	6.1 ± 0.4	9.8 ± 0.9	12.4 ± 4.5	0.7 ± 0.4		
	4	1.2 ± 0.0	1.3 ± 0.1	1.0 ± 0.0	1.0 ± 0.0	16.3 ± 7.1	17.9 ± 1.7	49.5 ± 4.1	44.8 ± 3.0	6.6 ± 0.8	5.8 ± 1.0	8.6 ± 0.8	0.9 ± 0.5		
	6	1.3 ± 0.0	1.4 ± 0.0	1.0 ± 0.0	1.0 ± 0.0	24.7 ± 1.4	27.9 ± 2.2	40.1 ± 5.2	28.6 ± 4.0	9.9 ± 1.1	10.6 ± 3.1	15.9 ± 1.7	4.3 ± 4.0		

Values = Mean ± SD. Each value is a mean of 3 replicates. S.D (standard deviation) (p≤0.05)

3.2 Effects of Storage Temperature and Humidity on Moisture Uptake during Cooking

Storage of beans under unfavorable conditions affects their quality by reducing water uptake during cooking. The moisture uptake of beans stored at low temperature and low relative humidity was recorded and compared to beans stored at high temperature and high relative humidity for the same period of time. Figure 1 and 2 shows the differences in moisture uptake of Easy to cook and hard to cook beans stored at different temperature and humidity for the same period of time. After cooking for 3 hours, beans stored at low temperature and humidity had a much higher water uptake during cooking than for beans stored at high temperature and relative humidity. This shows that beans stored at elevated temperature (45ºC) and low relative humidity (75%) exhibit the greatest loss in moisture during storage and increased bean hardness. Hard to cook beans are characterized by prolonged cooking times than for easy to cook beans, this is because the cell wall of hard to cook beans is more compact, having smaller intercellular spaces (Garcia et al., 1993), so more time is needed for water penetration.

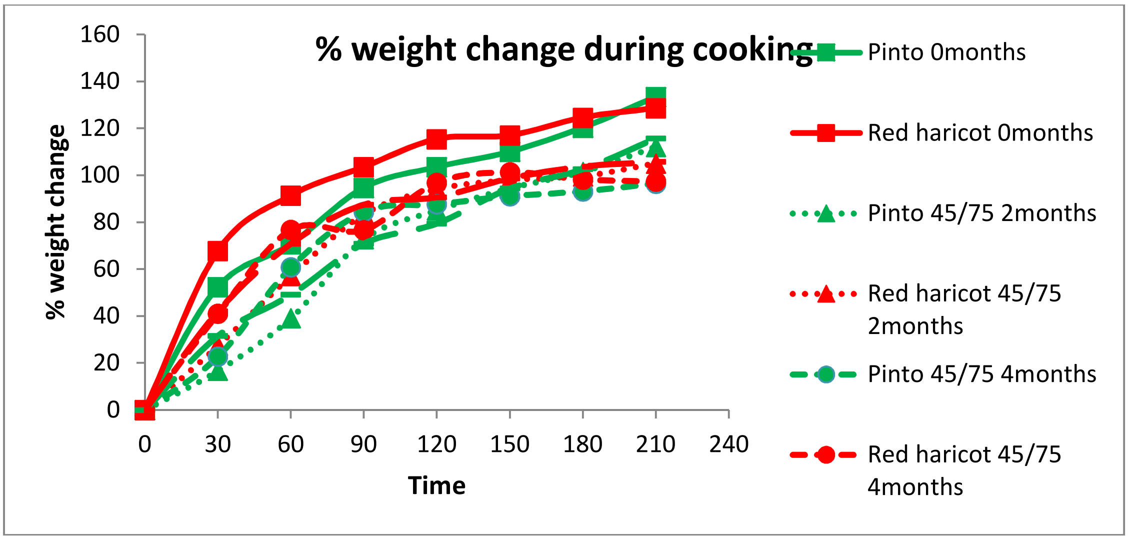

Figure 1: Percentage moisture uptake during cooking for beans stored at 45^{o}C

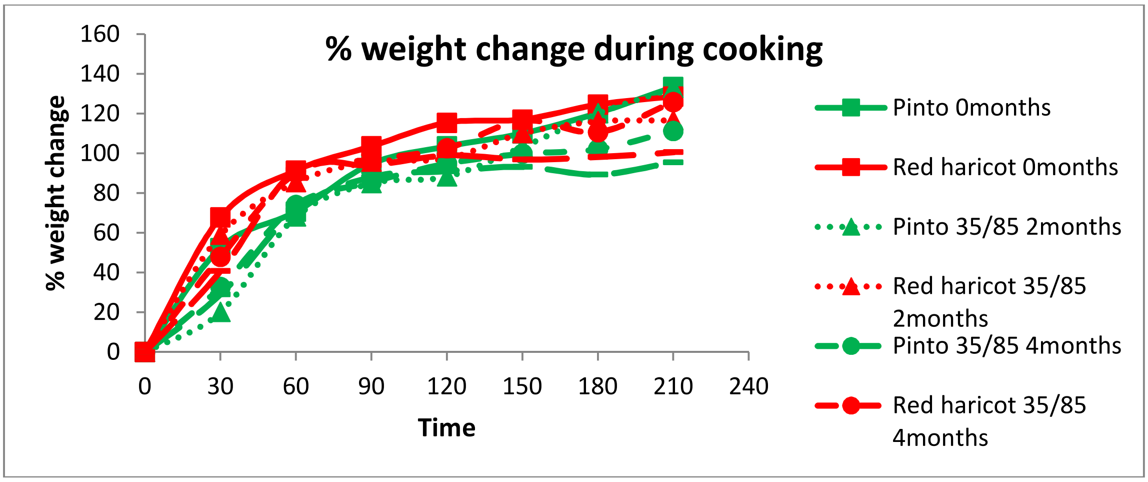

Figure 2: Percentage moisture uptake during cooking for beans stored at 35^{o}C 85% humidity

3.3 Effects of Storage Temperature and Humidity on Cooking Rates

The cooking quality of beans is related to post harvest handling and storage conditions. Beans stored at high temperature hydrate unevenly during cooking resulting in uneven bean softening and to the presence of hard to cook beans. Hard to cook beans fail to soften enough to be eaten after cooking for a reasonable time. Figure 3 and 4 shows the differences in cooking rates of an easy to cook and a hard to cook bean variety stored at different temperature and humidity for the same period of time. After 3 hours of cooking, beans stored at low temperature and humidity cooked at a faster rate than for beans stored at high temperature and relative humidity. This slow cooking rate of beans stored at higher storage conditions is attributed to the development of bean hardness during storage.

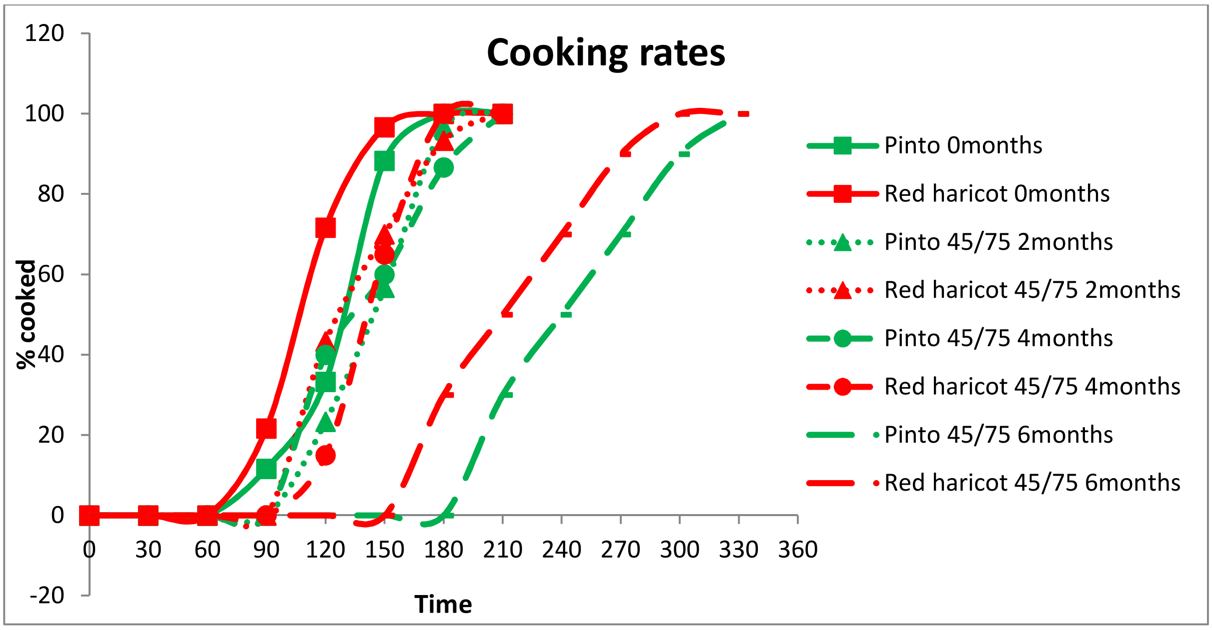

Figure 3: Cooking rates of beans stored at 45ºC 75% for 0-6 months

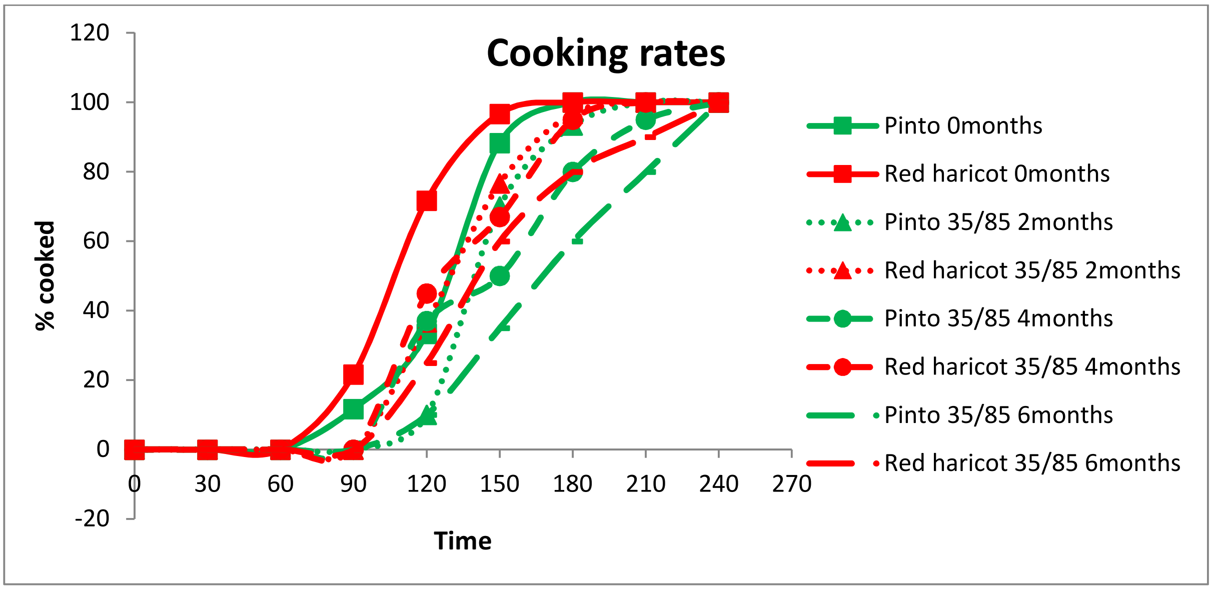

Figure 4: Cooking rates of beans stored at 35ºC 85% for 0-6 months

4.0 Conclusion

Storage of bean varieties under adverse conditions of high temperature and relative humidity rendered them susceptible to hardening. Several changes in the quality of beans were affected by storage under these conditions: decrease in the rate of water absorption, loss of colour lightness, development of browning and darkening. It is important to store beans at cool temperatures and in the absence of light to maintain colour and cooking quality.

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