PHYSIOLOGICAL AND BIOCHEMICAL STUDIES ON THE QUALITY OF KENAF (*Hibiscus cannabinus* L.) SEED AS AFFECTED BY STORAGE PERIOD, TYPE OF CONTAINER AND GENOTYPE

BY

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CERTIFICATE

This is to certify that the thesis entitled, "Physiological and biochemical studies on the quality of kenaf (*Hibiscus cannabinus* L.) seed as affected by storage period, type of container and genotype" submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE in AGRONOMY embodies the result of a piece of *bona fide* research work carried out by Shamima Akther, Registration No. 23992/00219 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that such help or source of information, as has been availed of during the course of this investigation has been duly acknowledged by her.

Dated:
Place: Dhaka, Bangladesh

02/02/08

(Dr. Selina Begum)
Supervisor


Dedicated to my
Beloved Parents

LIST OF ABBREVIATIONS OF SYMBOLS AND TERMS

Full Word	Abbreviation
And others (at elli)	*et al.*
Centimeter	cm
Coefficient of Variation	CV
Degree Celsius (Centigrade)	^{0}C
Etcetera	etc.
Example	e.g.
Gram	g
Hour	hr
Least significant difference	LSD
Liter	l
Meter	m
Micron	µ
Milligram	mg
Milligram per gram	mg/g
Microgram per gram	µg/g
Milliliter	ml
Namely	viz.
Percent	%
Square meter	m^2

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By

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ABSTRACT

This study aimed to determine the storage performance of kenaf seeds and type of container used during storage. Study also aimed to find out the probable causes of seed deterioration. The experiments were conducted from December 2004 to June 2005. Results revealed that tin container was found less permeable to moisture transmission compared with jute sack and tin container also retained higher seed quality attributes throughout the storage period. Results from the experiments on biochemical basis of seed deterioration showed that no appreciable difference of sugar content was detected in fresh seeds and seeds those were stored in airtight tin container. But significant increase of sugar content in all genotype was recorded when seeds were stored in jute sack. Again after storage in jute sack, significant decrease of protein content was observed in all genotype but no appreciable change was recorded in seeds stored in tin container.

CHAPTER I

Introduction

CHAPTER I

INTRODUCTION

Kenaf (*Hibiscus cannabinus* L.) is one of the most promising bast fibre crop. The species belongs to the section Furcaria of the family Malvaceae. It occupies the most important position next to jute. Kenaf produces more biomass in poor soil where jute can not be grown even.

The kenaf plant has a wider range of adaptation to climate and soils than any other fibre plant grown for commercial use. Within the past few years, research has been carried out on kenaf stems as a raw material for pulp and paper, and the leaves as a high protein animal feed (White *et al.*, 1970). At present, kenaf is considered as the main renewable source of cheap raw materials for paper pulp production. Kenaf twigs are also fed to milch cattle and its dry stem is used for match sticks and as climbing stick of betel leaf and some vegetable crops. Kenaf is also cultivated for the production of edible oil from seed.

Kenaf is rapidly replacing jute because the crop has less intensive labour requirements, is cheaper to produce, may be grown on a wide range of soils under varied climatic conditions and is not necessarily competitive with food crops. While kenaf is somewhat coarser than jute, it has greater tensile strength, lighter in color, and has greater resistance to moisture.

To the farmers, a high quality seed is not only desirable but is also a statutory requirement in developed countries. Maintenance of seed quality in storage is important not

only for crop production in the following years but also for the maintenance of genetic integrity of the seeds because of constant threat of genetic erosion.

Seed storage and the retention of seed viability have always been an important consideration in agricultural practice. Poor storage conditions give rise to deterioration of seed quality and the resultant loss of viability. Deteriorated seeds when sown are also more susceptible to the attack of micro-organisms and insects and by resulting in poor seedling establishment, lead to reduced competitiveness against weeds so that crop yield may be affected.

As jute, kenaf seeds are very delicate and can be hydrated and dehydrated with ambient moisture. It is very deteriorous to use seed containers, which are permeable to moisture and oxygen. The maintenance of good germinability of carry-over kenaf seed is of great importance to seed producers. The viability of kenaf seed in the warm humid climate of Bangladesh is a major problem to growers. Due to lack of proper storage condition farmers do not generally store seed for more than some weeks. The growers, therefore, throw away surplus seed if any is left from the last year's stock. The wastage may be reduced if a proper method of storing for a few years could be adopted (Islam and Ali, 1981).

Storage condition plays a significant role in seed preservation. Storage containers of semi-permeable status may be of noteworthy for short term as well as long term seed preservation. Seed growers at farm level use varieties of container (Hossain *et al.*, 1994C; Khandakar, 1982) although most of these are not conducive to seed health because they are permeable in nature. Under high humid condition, permeable containers allow moisture penetration, which in turn increases humidity surrounding the seeds with the presence of

excess moisture and with the rise of ambient temperature during summer months tend to germinate at storage even with the absence of other conditions required for seed germination. In the process, seeds gradually lose vigour and eventuate complete destruction of viability. Once the tendency of germination grows in seeds at storage, these seeds deceive germination second time in the field (Hossain *et al.*, 1994C).

The farmers of Bangladesh usually use four types of container, which are metal cointainer, clay pot, polythene bag and jute sacks (Hossain *et al.*, 1994C).The efficiency of clay pot and jute sack, as storage containers have been proved worst (Razzaque, 1980), and that of polyethene bag yet to confirm. Only the airtight metal containers have been found to restrict moisture penetration to an acceptable range.

Knowledge is still inadequate as to how seeds survive in storage as well as how seeds die in storage. Many theories have so far been evolved on the mechanism of seed deterioration. Christensen (1972) considered that the loss of seed viability was due to storage fungi and the extent of deterioration was related to seed moisture content, storage temperature and the availability of oxygen. Khandakar and Bradbeer (1983) have reported that seed quality mainly depends on preharvest environment, post harvest processing and storage; and the sowing environment of the seeds in the following season.

A very little information is available relating to storability of kenaf seed. So it is very important to evaluate the proper storage condition for kenaf seed.

Therefore, the present study was undertaken with the following objectives:

1. To see the effects of storage period, type of container and genotype on seed moisture content and retention of seed quality.

3. To find out the biochemical causes of quick deterioration of seeds.

CHAPTER II

Review of Literatures

CHAPTER II

REVIEW OF LITERATURE

Research on kenaf, mainly on variety development have been carried out, but very little attention was paid to the researches on its seed storage technologies in the past. Seed moisture content being an important consideration for retention of seed quality during storage, its interaction with type of storage container may help to devise a suitable device of seed preservation for the use of seed growers and seedman. The information is essentially needed for the management of seed quality during seed storage. The present research programme has been undertaken to generate information in this regard and so a review of the research problem has been presented here with the available literature pertinent to this research programme.

2.1 Factors affecting seed quality at storage

Bhattacharyya and Dutta (1972) conducted storage experiment with storing jute seeds in glass bottle, double plastic bag and cotton bag. A big fluctuation in moisture content was observed at storage in all seed samples. Double plastic bag with silica gel was found to be the most effective storage practice.

Khare *et al.* (1974) reported that the lowest viability loss was observed in wheat seed 1.5 % in metal dram and the highest of 4.3 % in gunny sack, where moisture absorption was also the highest.

Prodhan and Mukherjee (1975) also emphasized significant importance on the airtight container that must be useful for minimizing storage loss.

Roberts and Roberts (1972) worked out homographs of a number of seeds (rice, barely, wheat, peas, and broad bean) which can be used to predict the viability of seeds during storage at any given combination of temperature and moisture content.

Christensen (1972) considered that the loss of seed viability was due to storage fungi and the extent of deterioration was related to seed moisture content, storage temperature and the availability of oxygen.

Khatun (1988) reported that the optimum method for long term storage of jute, kenaf and mesta seeds was in laminated aluminium foil packets at - 20°C and for medium term storage in aluminium foil packets at 4°C. Seed moisture content should be 6–7 percent at the time of packaging for storage.

Khandakar and Bradbeer (1983) recommended that both *Corchorus capsularis* and *Corchorus olitorius* seeds could be stored safely for one year with moisture content of 10%.

Khatun and Sobhan (1986) also reported that jute seeds with moisture content of 4-7% maintained 85% viability up to 12 months at room temperature.

Sobhan and Khatun (1986) in another experiment reported that kenaf and mesta seeds stored with moisture content of 14.3-24.5% had a sharp decline in viability and vigour with the increase of storage period. However, jute seeds with 4-7 % moisture content maintained

80 % germination, while kenaf seeds dropped viability to 48-58 % with moisture content of 5.5-7.4 %in 6 months.

Harrington (1973) expressed that starchy seeds above 12 % moisture and oily seeds above 9 % moisture should be packed in moisture resistant containers.

Sijbring (1973) reported that moisture content of seeds stored in jute sacks would eventually reach a value which is in equilibrium with the atmospheric humidity of the store.

Sangakkara and Somarathe (1988) stored *Vigna radiata* seeds in paper, jute, transparent polyethylene and cloth bags, thus, reported that seed moisture content increased and percentage germination decreased over 30 weeks of storage irrespective of the container used, but markedly the greatest effect with paper bag and the least effect with polyethylene bag.

Idem (1987) in successive trials during 1978-83 at Mokwa, Nigeria stored jute and kenaf at room temperature (minimum-maximum temperature, 13.7-24.5 °C and 23.4-40.2 °C, respectively) in a baft bag with one or two polyethylene layers or in a refrigerator (6.6-15 °C) in a baft bag with or without two polyethylene liners. Kenaf seeds germination decreased with increasing storage time and the highest germination percentage after 36 months was 83.3% when stored in a baft bag with 2 liners at room temperature. Germination of jute seed increased with storage time up to 43 months and decreased thereafter.

Walton (1977) reported that polyethylene storage improved lupin seed germination.

Srivasta (1978) also reported that polyethylene bags had minimum loss of viability of soybean seeds.

Jalote and Vanish (1978) revealed that reduction of viability in rice seed was less with high moisture content stored in polyethylene. Rao (1978) reported that metal container was better than polyethylene lined gunny bag for preservation of sunflower seeds.

Majid and Nahar (1981) observed both metal container and polyethylene bag were suitable for short duration storage (4 months) for soybean seeds.

Khandaker (1982) reported that lamofoil porches proved to be the best container. After three years of storage, the low land species of jute (*Corchorus capsularis*) still maintain above 90% germination when stored at 9 % seed moisture content. The high land species (*Corchorus olitorius*) gave a similar result when stored at 5-7 % moisture content.

Hossain *et al.* (1994 c) also reported that seed moisture content was perhaps, the most important factor that regulated longevity of seeds at storage.

Harrington (1973) stated that at higher temperature, polyethylene was more permeable to moisture vapour transmission than at lower temperature.

Jain and Saha (1971) stored jute seed in glass Stoppard bottles and observed *Corchorus capsularis* seeds maintained viability better than *Corchorus olitorius* varieties; they also observed variations within species in maintenance of viability. After 38 months, the mean viability was 79.9 % for *C. capsularis* and 68.4 % for *C. olitorius.*

Razzaque (1980) reported that bamboo made dully and gunny bag individually proved to be the worst, but tin can, drum, polyethylene bag covered with gunny bag and earthen pot coated with coal tar outside appeared to be effective.

Hossain (2003) stated that plastic container and tin can were found less permeable to moisture transmission compared with polyethylene bag in earthen pot and polyethylene bag in jute sack. Plastic container and tin can also retained higher seed quality attributes throughout the storage period.

Boyd *et al.*, (1960) stated that seed moisture range for safe storage should be 12-14 % or less depending on kinds of seed and storage condition.

Dhesi (1963) reported that increase of temperature in combination with high seed moisture content increased the life activities of seeds. Combining the temperature and seed moisture content, Harrington (1963) developed two Thumb rules that were easily understood and reasonably approximate the effect of moisture and temperature on seed longevity, which were as follows:

1. For each 1 % decrease in seed moisture content, the life of the seed is doubled (between 4 and 14 % moisture).

2. For each 5 ^{o}C decrease in seed temperature, the life of the seed is doubled (at least between 0 ^{0}C and 50 ^{0}C).

Harrington and Douglas (1970) estimated the storage life of cereal seeds in relation to seed moisture ranges at start of storage practice. The estimated storage life can be seen in the following chart.

Seed moisture content	Storage life
11-13%	1-2 year
10-12%	1 year
9-11%	2 years
8-10%	4 years

If the seeds are kept in high moisture content mentioned in the above chart, the loss could be very rapid due to mould growth in the seed (12-14 % moisture) or due to heating (18-20 % moisture). Within the normal range, biological activity of seeds, the insects and moulds further increase as the temperature increases. The higher the moisture content of seeds, the more they are adversely affected by both upper and lower limit of temperature.

Hossain *et al.* (1994c) stated that jute seeds with high moisture content had germination tendency with the rise of temperature at storage even with the absence of other conditions required for seed germination. This tendency of germination started physiological activities in seed, which affected seed vigour and eventually its viability fell.

Heydecker (1969) reported that poor storage conditions give rise to quality deterioration, greatly affects seed vigor and resulted loss of viability.

Seeds stored in ordinary condition, absorb moisture and reduce germination percentage (Razzaque, 1980; Rahman *et al.*, 1985).

Metal cans when properly sealed provided an absolute barrier to moisture penetration (Bass *et al.*, 1961, Grable and Isley, 1969, Harrington, 1973) and was found to be a completely satisfactory container for maintaining seed viability.

2.2 Biochemical manifestation of seed deterioration

According to the reports of pioneer workers, poor storage condition greatly affects seedling vigour (Heydecker, 1969), disrupts protein synthesis and glucose utilization at an early stage of germination (Abdul-Baki, 1969, Wood stock, 1969).

Koostra (1973) found that seed deterioration was associated with disintegration of the plasma- lemma and other cellular membranes during aging.

Jones *et al.* (1942) investigated changes in proteins of wheat seeds stored for 24 months under conditions which induced different levels of deterioration. Ching and Schoolcraft (1968) reported reductions in seed proteins of crimson clover and perennial rye-grass with concomitant increases in amino acids of deteriorated seeds

Roberts and Osborne (1973) attributed the loss of viability to the deterioration of DNA molecules. This was considered by many workers to be the most acceptable explanation for the mechanism of seed deterioration.

Abdul-Baki and Anderson (1972) reported that the activities of enzymes such as alcohol dehydrogenase, amylase, catalase, cellulase, cytochrome oxidase, glutamate, decarboxylase, malate dehydrogenase, peroxides and phenolase are degraded during seed aging.

Varier and Agarwal (1982) observed that soluble protein content increased with increasing storage period. Nautiyal *et al.* (1985) also observed that soluble proteins were present in the non viable seeds.

All the literatures reviewed in relation to type of storage container on seed deterioration indicate that moisture has tremendous influence on the longevity of seeds at storage. Thus it is of paramount importance to determine the safe moisture level and also to identify cheap and handy storage container for preservation of kenaf seeds for the use of seedmen & seed growers at farm level.

CHAPTER III

Materials and Methods

CHAPTER III

MATERIALS AND METHODS

3.1 Experimental site:

The experiment was conducted at the Physiology laboratory, Agronomy division, Central station, Bangladesh Jute Research Institute (BJRI), Manikmia Avenue, Dhaka-1207, during December 2004 to June 2005.

3.2 Seed collection:

Fresh seeds were collected from Central station of Bangladesh Jute Research Institute (BJRI), Dhaka. Seeds were harvested on the 18th December 2004, dried in sun for 7 days and then stored in air tight tin container and jute sack.

3.3 Characteristics of the study materials:

The two kenaf (*Hibiscus cannabinus* L.) varieties (HC-2 and HC-95) and an advance line (CPI-72126/1) were selected for the experiment. Kenaf seeds are produced in an ovoid capsule like fruit. The fruits are pointed villous, half the length of the calyx with 20 to 26 seeds in each fruit. Seeds are triangular, angles are more or less acute, and color is ash gray with pointed light yellowish warty spots. Hilum brown and relatively small.

3.4 Treatments of the study:

Storage period: Kenaf seeds were stored for seven months. Storage period commenced on 25 December, 2004 and moisture and germination percentage were recorded at an interval of one month till June, 2005. However, estimation of protein and sugar of seed were done only on December, 2004 and June, 2005.

Types of storage container

i. Tin container

ii. Jute sack

Genotype (variety /line)

i. HC-2

ii. HC-95

iii.CPI-72126/1

The experiment was conducted following Completely Randomized Design (CRD) with three replications.

3.5 Initial seed physiology study:

3.5.1 Calculation of vigour value:

Vigour value was employed to qualitate of the viable seeds which was assessed by using the results of the above mentioned germination test according to Khandakar (1982), using the following formula:

$$\text{Vigour value (\%)} = \frac{(a/1 + b/2 + c/3 + d/4) \times 100}{S}$$

Where a, b, c and d are seed germinated after 1, 2, 3 and 4 days from the start of the germination test and S is the total numbers of seeds germinated.

3.5.2 Measurement of seedling growth and development:

For growth measurement, seeds were allowed to germinate in an incubator set at 30 °C. Seeds of kenaf were kept to grow in petridish on top of two layers of Whatman No.1 filter paper. The root and shoot length of 10 seedlings were recorded at an interval of 24 hr.starting from 24 hr.after placement. The experiment was terminated after 72 hours.

3.6 Seed quality study:

3.6.1 Determination of germination (%):

One hundred seeds were collected randomly from each containers and set for germination test in four glass petridishes having equal number of seed. Germination tests were carried out in an incubator set at 30 °C. Seeds were evenly placed on the top of 9 cm Whatman No.1 filter paper in each petridish. Filter papers were then kept moist by adding 5 ml distilled water. From 2nd days after setting germinated seeds were counted and recorded daily for four days. Seeds with radicle extended up to 1 cm in length or more were considered to be germinated.

3.6.2 Determination of seed moisture (%):

Seed moisture content was determined by the air-oven method developed by Roberts and Roberts (1966). Approximately 1 g of seeds was accurately weighed in a small pre-weighed porcelain crucible with lid. After 16 hr. in the oven at 105° C, the crucible was allowed to cool in a desiccator over silica gel. The weight was recorded again and the percent moisture content of seeds was determined as follows:

$$\text{Moisture content (\%)} = \frac{(m_2-m_3) \times 100}{m_2-m_1}$$

m_1 = weight of crucible + lid.

m_2 = weight of crucible + lid +fresh seeds.

m_3 = weight of crucible + lid +dried seeds

Determination of moisture content of seeds of each sample was replicated three times.

3.7 Studies on biochemical basis of seed deterioration:

3.7.1 Soluble sugar content in seeds:

Soluble sugar content in the seeds was quantified following anthrone-sulphuric acid method as was described by Shirlaw and Gilchrist (1967). One g seeds of each genotype of HC-2 and HC-95 and CPI-72126/1 were allowed to soak in distilled water for 4 hr. and then crushed. After crushing more distilled water was added up to a volume of 100 ml. Crushed sample was then centrifuged and filtered through whatman No.1 filter paper. The filtrate was boiled for 20 minutes, as a result of which the protein materials coagulated. The samples were then centrifuged again for 20 minutes. The supernatant were washed twice with diethyl ether and 0.2 ml of elute obtained was pipetted in a test tube containing 5 ml of ice-cold anthron ($C_6H_4.COC_6H_4CH_2$) solution (0.2% in sulphuric acid). The elute and anthrone-

sulphuric acid solution were mixed rapidly in a test tube and the sample was placed in a water bath for 10 minute. It was then cooled by running tap water. The optical density was measured at 625 nm. Glucose was used to plot a standard curve for the estimation of sugar content in the solution (AnnexureII). Each sample was replicated three times.

3.7.2 Soluble protein contents in seeds

a) Preparation of reagents :

i) 0.1M NaOH + 1% anhydrous Na_2CO_3 were made up to 1 liter

ii) 1% $CuSO_4.5H_2O$

iii) 2% Na-K-Tartrate.

Reagents: 49 ml of i) 0.5 ml of ii) and 0.5 ml of iii) were mixed.

b) Folin-Ciocalteau phenol reagent :

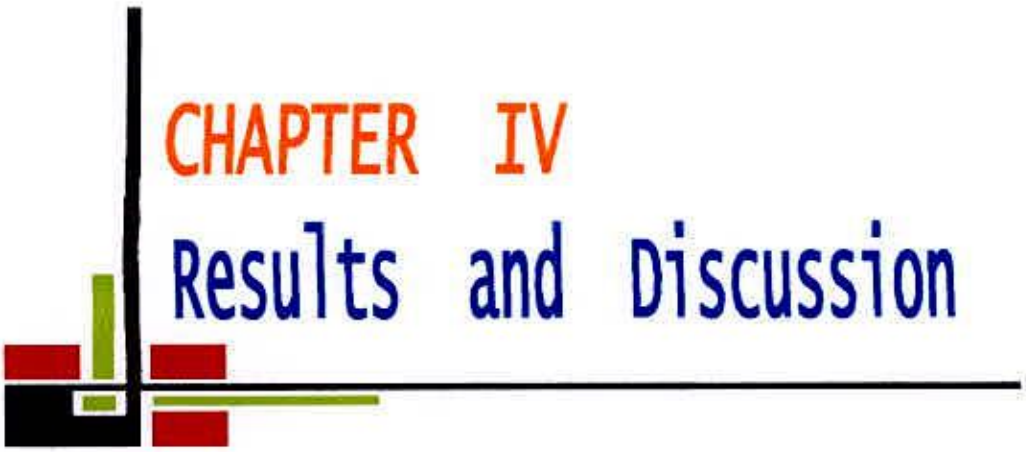
Folin-Ciocalteau phenol reagent was diluted with water in 1:1 ratio.

Protein content of seeds was estimated following the method described by Lowry *et al.* (1951) .One g of each seed sample were soaked with distilled water for 4 hours and then crushed. After crushing, more distilled water was added up to a volume of 100 ml. Then crushed sample was centrifuged and then filtered through whatman No.1 filter paper. The filtrate was defatted by adding diethyl ether. Then 0.4 ml of the supernatant was pipetted in a test tube containing freshly prepared 5 ml of reagent (1), and 0.8 ml of distilled water. After that 0.5 ml of freshly prepared Folin-Ciocalteau Phenol reagent was added. The mixture of these three solutions were then shaken and allowed to stand for 30 minute. The optical density was measured in a spectrophotometer at 750 nm. Bovin Serum albumin was used to

plot a standard curve for estimation of protein content in the solution (Annexure II). Each sample was replicated three times.

3.8 Analysis of data:

The recorded data under the present study were statistically analyzed using IRRI STAT programme. The level of significance and analysis of variance along with the Least Significance Difference (LSD) Test were done following Gomez and Gomez (1984).



CHAPTER IV

Results and Discussion

CHAPTER IV

RESULTS AND DISCUSSION

4.1 Physiological profile of fresh kenaf seeds

4.1.1 The rate of germination and vigour value of fresh kenaf seeds

Results presented in table 1 shows that more than 95 % of the seeds of all genotype germinated at 24 hr. after placement. The germination percentages were 96, 97 and 95 % in HC-2, HC-95 and CPI-72126/1, respectively. After 48 hr. only 1-2 % seeds germinated. No germination was observed after 72 and 96 hr. This finding is in accordance with that of Jain and Saha (1971) and Khandaker (1982). They observed that 90 % of the fresh seeds in both species of jute germinated during the first 24 hr. at 30-33 °C, with most of the remaining seeds sprouted on the second day. Results also showed that vigour values of fresh seeds were higher and those were 98.98, 99.49 and 99.48 % in HC-2, HC-95 and CPI-72126/1, respectively.

Table 1 Germination and vigour value of fresh seeds of kenaf

Time of germination recorded / Genotype	Germination (%)				
	24 hr	48 hr	72 hr	96 hr	Vigour value (%)
HC-2	96	2	0	0	98.98
HC-95	97	1	0	0	99.49
CPI-72126/1	95	1	0	0	99.48

4.1.2 Root and shoot growth of seedlings raised from fresh seed of kenaf

One hundred fresh seeds (25 in each of 4 petridishes) were allowed to germinate on filter paper at 30 °C in an incubator. Roots and shoots elongation was measured every 24 hr for three days. Results presented in Figure 1 shows that at every sampling time the highest root length observed in HC-2 (4.86 cm) followed by line CPI-72126/1 (4.18cm) and HC-95 (3.75 cm).

Irrespective of genotype, shoot length increase with the increase in time. Results presented in Figure 2 shows that at every sampling time the highest shoot length observed in HC-2 (6.90 cm) followed by line CPI-72126/1 (6.65 cm) and HC-95 (5.95 cm).It is interesting to note that higher root and shoot length was attained by HC-2 in all stages of growth. Thus, the findings of the present investigation are in accordance with those of khandakar (1982). He reported that at 30 °C root and shoot length of seedlings of *Corchorus* spp increased during the first three days.

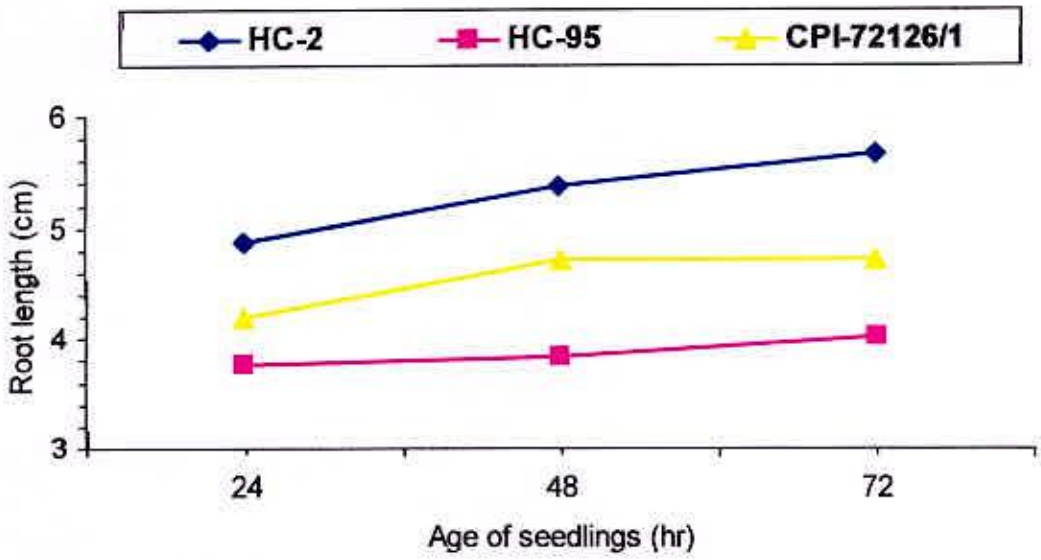


Fig 1 Root length of seedlings raised from fresh seeds of kenaf genotype

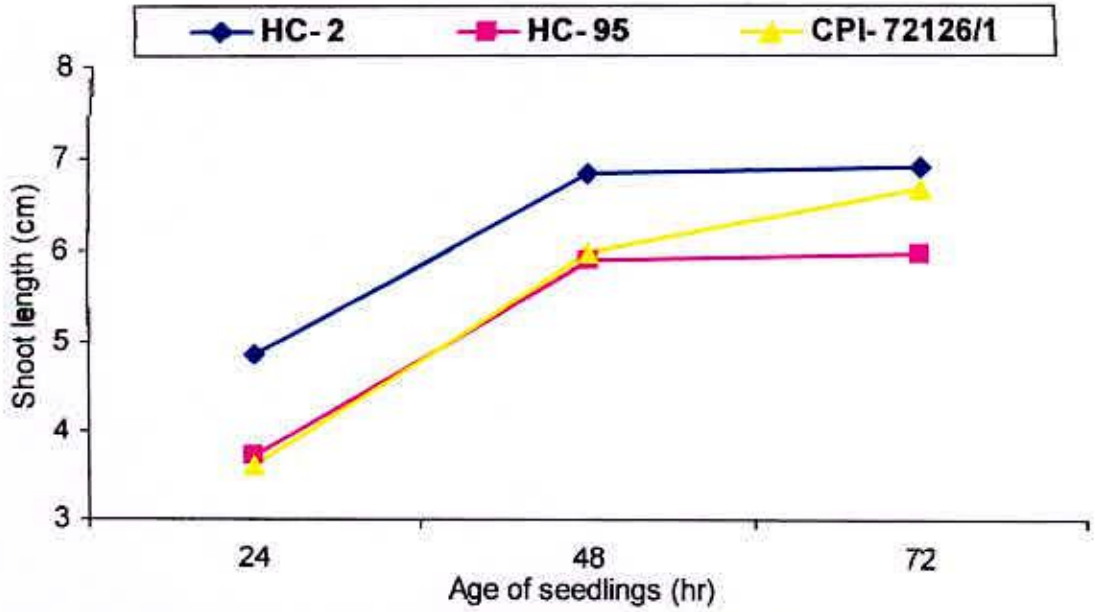


Fig 2 Shoot length of seedlings raised from fresh seeds of kenaf genotype

4.2 Effect of storage period, container and variety on quality of kenaf seeds

4.2.1 Effect of storage period on germination percentage of kenaf seeds

Results presented in Figure 3 shows that there was significant effect of storage period on kenaf seed germination (Annexure I). The highest germination percentage was recorded at December (96.06 %) and the lowest germination percentage was recorded in the month of June (74.44 %).

Results also showed that germination percentage reduced gradually with the extension of storage duration after storage. Significant difference was found in between each month observation up to May. No significant difference was found in May and June observation.

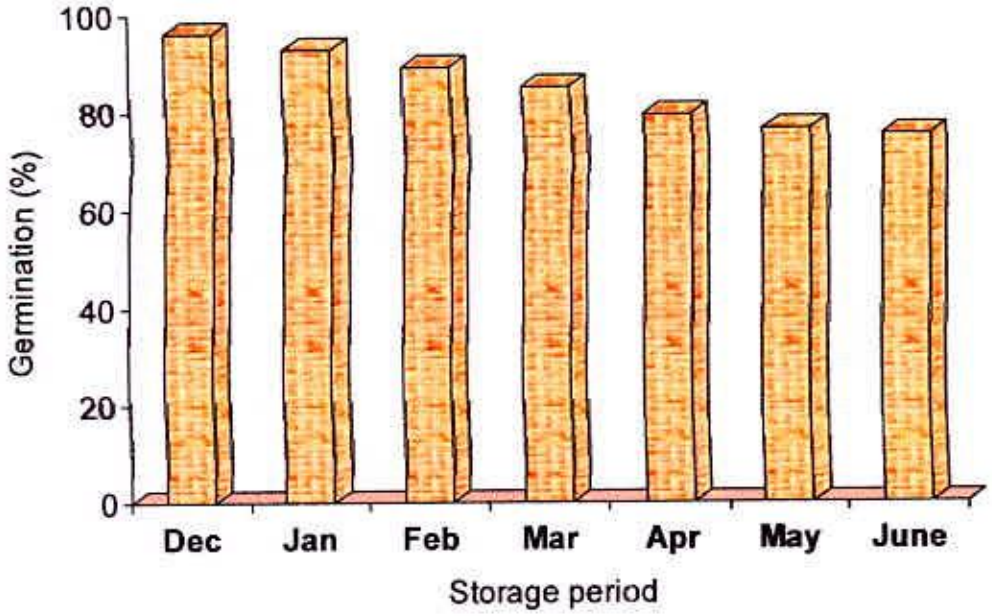


Figure 3 Month wise germination (%) in kenaf seeds

These results agree with Idem (1987) who reported that kenaf seeds germination decreased with the increase in storage time. Sangakkara and Somarathe (1988) found similar results on *Vigna radiata* seeds.

4.2.2 Effect of storage container on germination percentage of kenaf seeds

After 7 month of storage in tin container germination percentage was found 91.49 % and in jute sack it was 78.65 %. Significant effect was observed in storage container on seed germination. Tin container showed better performance than jute sack as shown in Table 2. Thus, the findings of the present investigation are in accordance with Prodhan and Mukherjee (1975) who stated that airtight container minimizes storage loss.

Table 2 Container wise germination percentage of kenaf seeds

Container	Germination (%)
Tin	91.49
Jute sack	78.65
LSD (1% level)	22.34

4.2.3 Effect of genotype on germination percentage of kenaf seeds

Significant effect of germination was found on genotype. The highest germination percentage was recorded in CPI- 72126/1 (85.74%) and it was significantly higher than those in other varieties at 1 % level (Table 3).

Table 3 Genotype wise germination percentage of kenaf seeds

Genotype	Germination (%)
HC-2	85.17
HC-95	84.31
CPI-72126/1	85.74
LSD (1% level)	0.55

4.2.4 Effect of storage period and container on germination percentage of kenaf seeds

Before storage germination percentage of seed was observed around 96. After seven months of storage in air tight tin container germination percentage decreased slightly and it was around 88 % (Table 4).

Table 4 Interaction effect of storage period and container on germination percentage of kenaf seeds

Storage period	Storage container	
	Tin	Jute sack
December	96.00	96.00
January	94.33	91.67
February	92.11	86.33
March	90.78	80.00
April	89.67	69.67
May	89.56	63.89
June	88.00	62.89
LSD (1 % level)	3.44	

Germination percentage declination in jute sack container storage was higher than tin container where the highest germination percentage was observed before storing in December (96 %) and the lowest germination percentage was recorded in the month of June (62.89 %). Results also showed that significant effect (at 1 % level) of storage period and container was observed on germination percentage of seeds.

4.2.5 Effect of storage period and genotype on germination percentage of kenaf seeds

Results presented in table 5 shows that initial germination percentage was observed and those were 96.17, 97.17 and 94.83 for HC-2, HC-95 and CPI 72126/1, respectively. After 7 months of storage germination percentage decreased significantly to 75.68, 73.50 and 77.17 in HC-2, HC-95 and CPI-72126/1, respectively. Results also showed that significant effect of storage period and genotype on germination percentage of seeds.

Table 5 Interaction effect of storage period and genotype on germination percentage of kenaf seeds

Storage period	Genotype		
	HC-2	HC-95	CPI-72126/1
December	96.17	97.17	94.83
January	94.00	93.17	91.83
February	91.67	89.00	87.00
March	84.67	85.17	86.33
April	77.17	78.00	83.83
May	76.83	74.17	79.17
June	75.67	73.50	77.17
LSD (at 1% level)	3.47		

4.2.6 Effect of storage container and genotype on germination percentage of kenaf seeds

Interaction effect of storage container x genotype has significant effect on germination as shown in Table 6. Tin container performed better than jute sack for each genotype. For HC-2, germination percentages were observed 91.67 and 78.67 %, for HC-95, germination percentages were 90.29 and 78.33 % and for CPI-72126/1 germination percentage were 92.52 % and 78.96 % in tin container and jute sack, respectively. Results also showed that the effect of storage container and genotype on germination percentage was significant at 1 % level.

Table 6 Interaction effect of genotype and container on germination (%) of kenaf seeds.

Genotype	Storage container	
	Tin	Jute sack
HC-2	91.67	78.67
HC-95	90.29	78.33
CPI-72126/1	92.52	78.96
LSD (at 1% level)	6.03	

4.2.7 Effect of storage period, container and genotype on germination percentage of kenaf seeds

For variety HC-2:

Results presented in Figure 4 shows that before storage germination percentage of the seeds was 96 %. After 7 month of storage slight decrease in germination percentage was observed in tin container and it was 87.67 %. But in jute sack drastic decrease in germination percentage was observed and it was 63.67 % and the decrease in germination was statistically significant.

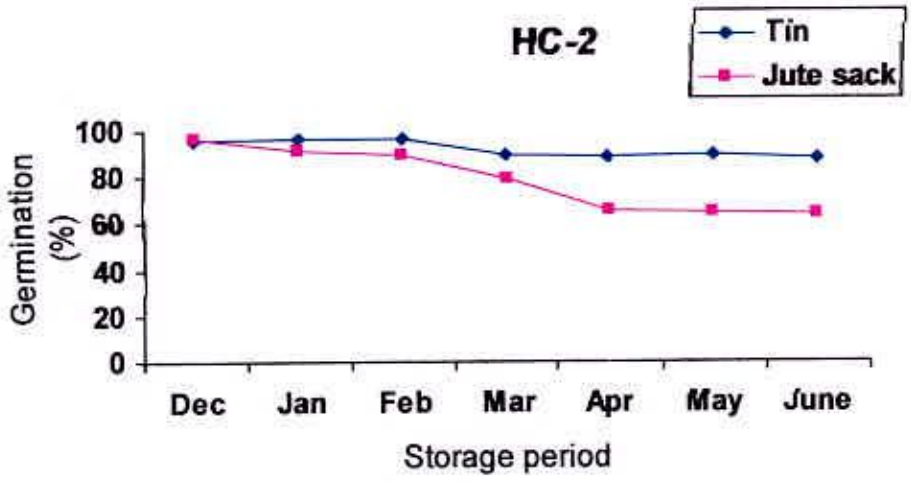


Figure 4 Effect of storage period and container on germination percentage of HC-2

For variety HC-95:

Results presented in Figure 5. shows that germination percentage reduced gradually in tin but in jute sack drastic reduction of germination percentage was observed. In tin container germination percentage decreased from 97.0 % to 86.67 % after 4 months of storage and then remains unchanged.

In jute sack container, germination percentage decreased significantly up to May. No significant difference was found between May and June observation.

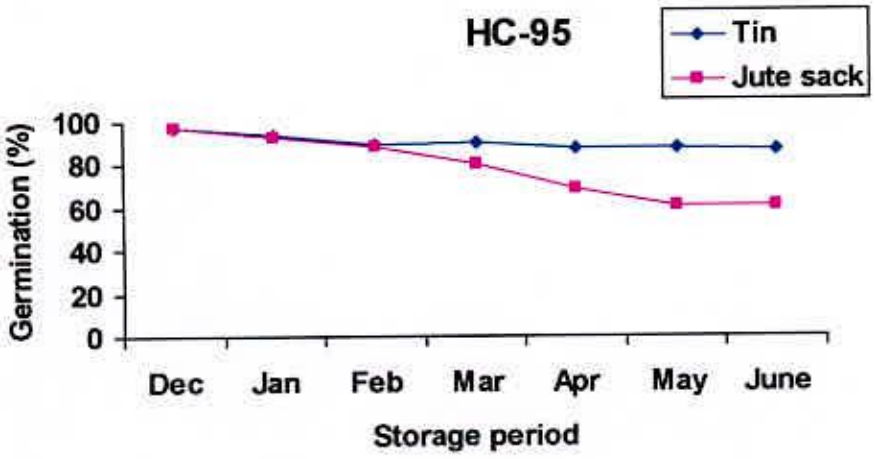


Figure 5 Effect of storage period and container on germination percentage of HC-95

For line CPI 72126 /1:

Germination percentage decreased very slowly in tin container storage but in jute sack container germination percentage decreased rapidly (Figure 6) and there was significant different (at 1 % level) which was observed in between December (94.67 %) and June (77.17 %) observation.

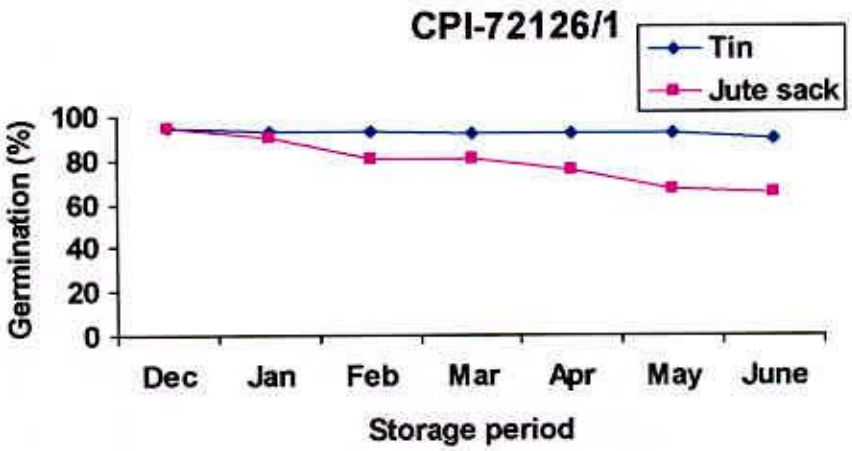


Figure 6 Effect of storage period and container on germination percentage of CPI-72126/1

4.2.8 Effect of storage period on moisture percentage of kenaf seeds

Moisture content of kenaf seed significantly increased due to storage. The highest moisture content (10.99 %) was observed in the month of June (Figure 7). In the first two observations December (8.7 %) and January (8.7 %) no significant difference in moisture content was observed. But significant increase in moisture content was observed from January (8.7 %) to June observation (10.99 %).

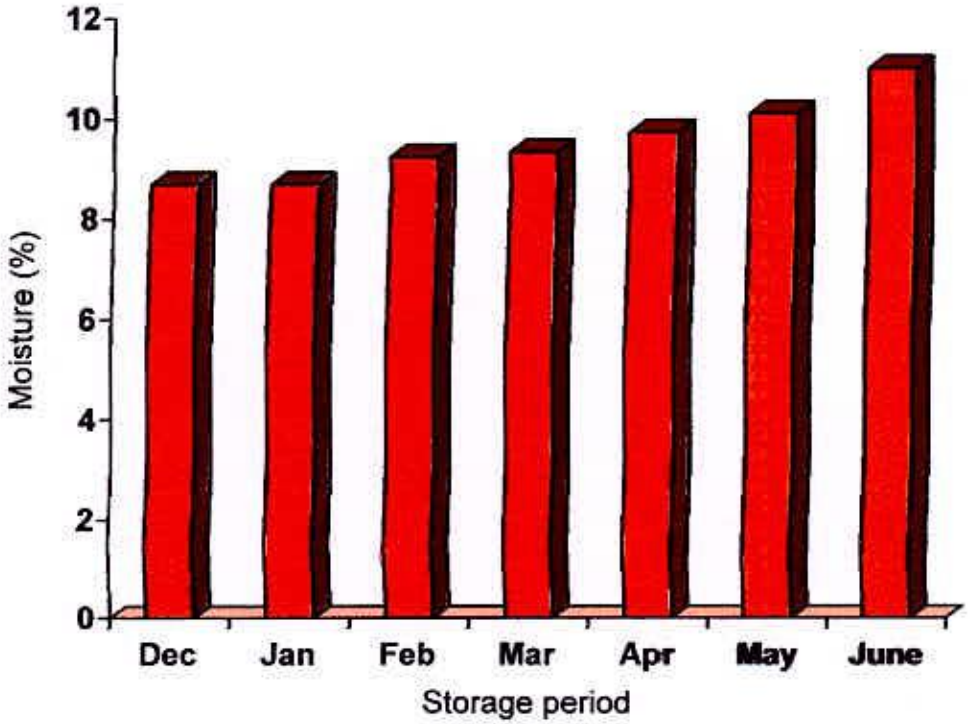


Figure 7 Month wise moisture (%) in kenaf seeds

4.2.9 Effect of storage container on moisture percentage of kenaf seeds

Storage container has significant effect on moisture content of kenaf seeds (Table 7). After storage in jute sack container moisture content was observed 10.50 % which was statistically higher than tin container (8.54 %) at 1% level of significance. Seed stored in jute sack container absorbed moisture from atmosphere. Results obtained during the investigation are in agreement with Sangakkara (1988) who reported that seeds stored in jute bag increased moisture over 30 weeks of storage. Khare *et al.* (1974) also stated that in jute sack moisture absorption was the highest.

Table 7 Container wise moisture percentage of kenaf seeds

Container	Moisture (%)
Tin	8.54
Jute sack	10.50
LSD (1% level)	1.10

4.2.10 Effect of genotype on moisture percentage of kenaf seeds

Variety HC-95 shows significantly higher moisture percentage (9.63 %) than other genotype at 1 % level of significance (Table 8). Variety HC-2 and line CPI- 72126/1 had obtained 9.48 and 9.48 % moisture, respectively.

Table 8 Genotype wise moisture percentage of kenaf seeds

Genotype	Moisture (%)
HC-2	9.48
HC-95	9.63
CPI-72126/1	9.48
LSD (1% level)	0.21

4.2.11 Effect of storage period and container on moisture percentage of kenaf seed

Results presented in Table 9 shows that before storage seed contained 8.71 % moisture. After 7 months of storage slight change in moisture content was observed in seeds of tin container but in jute sack container, moisture percentage gradually increased to 13.46 % in June and the increase was statistically significant in 1 % level. Results are in agreement with that of Hossain (2003) who observed that plastic container and tin can were found less permeable to moisture transmission compared with polyethylene bag in earthen pot and polyethylene bag in jute sack.

Table 9 Interaction effect of storage period and container on moisture percentage of kenaf seeds

Storage period	Container	
	Tin	Jute sack
December	8.71	8.71
January	8.62	8.78
February	8.58	9.90
March	8.55	10.11
April	8.48	10.96
May	8.44	11.74
June	8.37	13.46
LSD (1% level)	0.17	

4.2.12 Effect of storage period and genotype on moisture percentage of kenaf seeds

Results presented in Table 10 shows that before storage (December) variety HC-2, HC-95 and CPI-72126/1 had obtained 8.60, 8.77 and 8.73 % seed moisture, respectively. After storage in the month of June, seeds of all genotype had gained moisture and those were 11.26, 10.78 and 10.71 % for HC-2, HC-95 and CPI 72126/1, respectively. The increase in moisture percentage for the genotype was statistically significant at 1% level.

Table 10 Interaction effect of storage period and genotype on moisture percentage of kenaf seeds

Storage period	**Genotype**		
	HC-2	HC-95	CPI-72126/1
December	8.60	8.77	8.73
January	8.60	8.78	8.71
February	9.25	9.43	9.05
March	9.34	9.53	9.13
April	9.60	9.82	9.75
May	9.70	10.31	10.26
June	11.26	10.78	10.71
LSD (at 1% level)	0.17		

4.2.13 Effect of storage container and genotype on moisture percentage of kenaf seeds

Interaction of storage container × genotype had significant effect (at 1 % level) on moisture percentage. Seeds of jute sack container showed higher moisture percentage than tin container for each genotype (Table 11). Variety HC-2 had 10.57, 8.39 moisture; HC-95 had 10.60, 8.67 % and CPI-72126/1 had 10.40, 8.56 % moisture in jute sack and tin container.

Table 11 Interaction effect of genotype and container on moisture percentage of kenaf seeds

Genotype	Container	
	Tin	Jute sack
HC-2	8.39	10.57
HC-95	8.67	10.60
CPI-72126/1	8.56	10.40
LSD (at 1% level)	0.29	

4.2.14 Effect of storage period, container and genotype on moisture percentage of kenaf seeds

For variety HC-2:

Results obtained in Table 8 shows that 7 months after storage the highest moisture percentage 14.36 % (jute sack container) and the lowest moisture percentage 8.16 % (tin container) was recorded. Moisture percentage trend was almost static for tin container but in case of jute sack container after two months of storage seeds had gained moisture very quickly and in the month of June it was maximum (14.36 %).

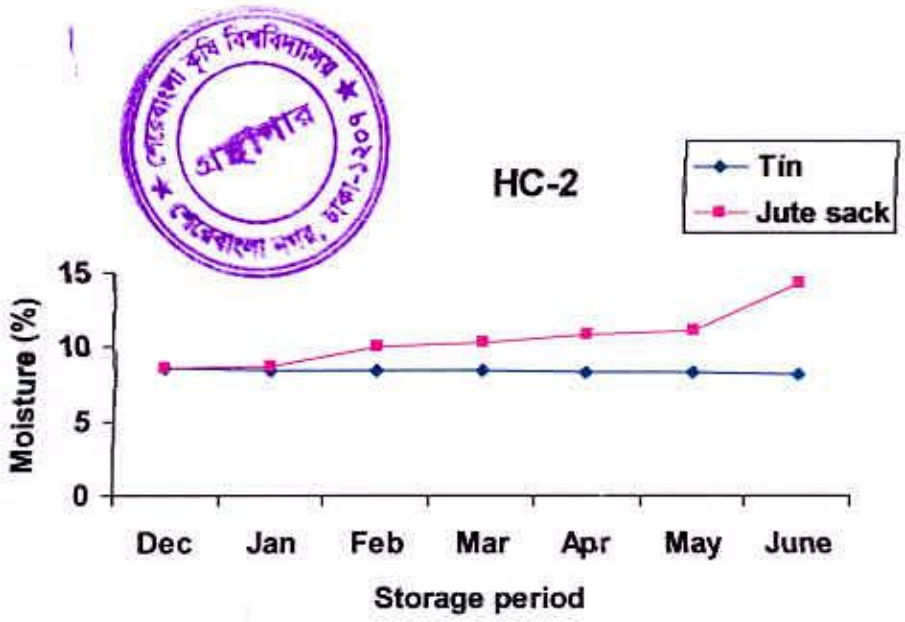


Figure 8 Effect of storage period and container on moisture percentage of HC-2

For variety HC-95:

Figure 9 shows that after 7 months of storage the highest moisture percentage recorded was 13.00 % (jute sack container) and the lowest moisture percentage recorded was 8.567 % (tin container). Moisture percentage trend was almost static for tin container but in case of jute sack container after two months of storage seeds gained moisture gradually and it was maximum at June (13.00 %).

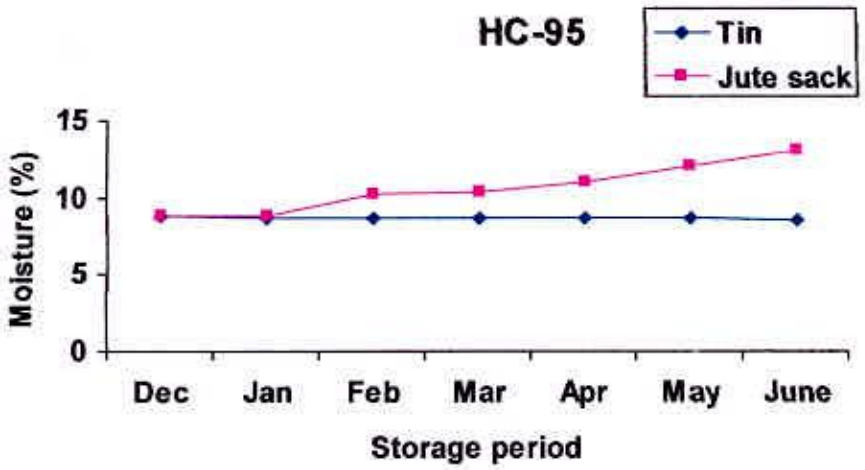


Figure 9 Effect of storage period and container on moisture percentage of HC-95

For CPI 72126 /1:

Results presented in Table 10 shows that during storage moisture percentage trend was found similar as HC-2 and HC-95. After 7 months storage in June, the highest moisture percentage recorded was 13.03 % (jute sack container) and the lowest moisture percentage recorded was 8.4 % (tin container).

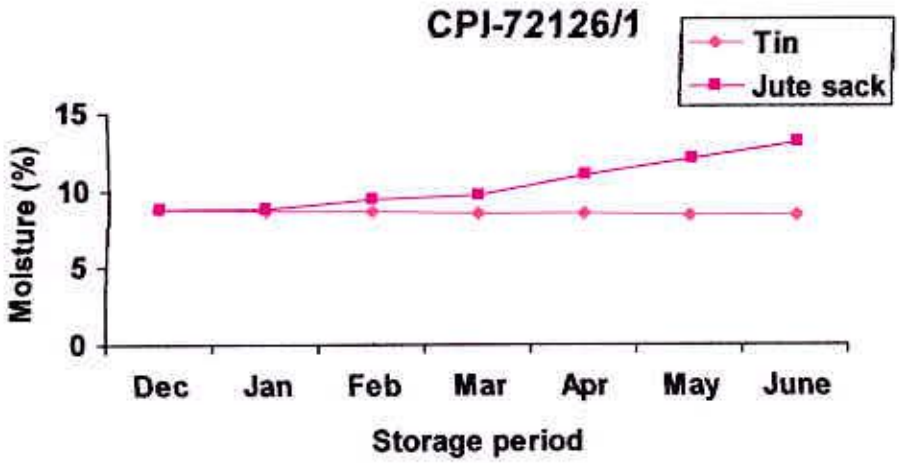


Figure 10 Effect of storage period and container on moisture percentage of CPI-72126/1

4.3 Studies on the biochemical basis of deterioration of kenaf seeds

To understand the causes of seed deterioration some biochemical investigations of fresh and stored seed were done. Biochemical parameters were soluble sugar and protein content.

4.3.1 Effect of storage period on sugar and protein content of kenaf seeds

Kenaf seeds are stored in different container (tin and jute sack) for 7 months. Results presented in Table 12 shows that at initial stage of storage sugar content of seeds recorded was 246.75 mg/g of seed but after 7 months of storage it increased to 332.83 mg /g and the increase was statistically significant at 1 % level. The results agree with the findings of Khandaker (1982) who reported that in jute seed loss of vigour during storage correlated closely with the increase in sugar exudation. Results obtained here are in agreement with those of Baki and Anderson (1972) who reported that when seeds deteriorated, synthesis of carbohydrate occurs.

Table 12 Month wise sugar and protein content of kenaf seeds

Storage period	Amount of sugar (mg/g)	Amount of protein (µg/g)
December	246.75	182.92
June	332.83	97.12
LSD (1% level)	62.89	7.44

Results also shows that initially seed contained 182.92 µg protein /g of seed but after 7 months of storage it decreased to 97.12 µg /g of seed and the decrease was statistically significant at 1% level.

Protein content in seed indicates quality of seed. Results revealed that after storage protein content of seeds decreased and at that time seed quality deteriorates. Results obtained during the investigation are in agreement with those of Abdul Baki and Anderson (1972) who reported degradation of protein occurred when seeds deteriorates.

4.3.2 Effect of storage container on sugar and protein content of kenaf seeds

Kenaf seeds are stored in different container (tin and jute sack) for 7 months. Results presented in Table 13 shows that seeds stored in jute sack and in tin container contained 330.70 and 248.87 mg sugar per g of seed, respectively. Amount of sugar of seeds stored in jute sack container was statistically higher than that of seed stored in tin container and the difference was significance at 1 % level of significance.

Results also shows that seeds stored in tin and jute sack container contained 179.48 and 100.59 µg protein/g of seed, respectively and the difference was statistically significant at 1 % level of significance.

Table 13 Container wise sugar and protein content of kenaf seeds

Container	Amount of sugar (mg/g)	Amount of protein (µg/g)
Tin	248.87	179.48
Jute sack	330.70	100.59
LSD (1% level)	62.89	7.44

4.3.3 Effect of genotype on sugar and protein content of kenaf seeds

Seeds of kenaf genotype (HC-2, HC-95 and advance line CPI-72126 /1) were stored in different storage container. Variety HC-95 achieved the highest sugar content (318.39 mg/g) and advance breeding line CPI-72126/1 showed the lowest sugar content (273.11 mg/g) but statistically HC-2 and CPI-72126/1 were identical and lower than variety HC-95 (Table 14).

Advance breeding line CPI-72126 /1 achieved the highest protein content (147.29 µg/g) which is higher than those of HC-95 (143.07 µg/g) and HC-2 (129.7 µg/g) and the difference in amount of protein content was significant at 1 % level of significance.

Table 14 genotype wise sugar and protein content of kenaf seeds

Genotype	Amount of sugar (mg/g)	Amount of protein (µg/g)
HC-2	277.86	129.71
HC-95	318.39	143.07
CPI-72126/1	273.11	147.29
LSD (1% level)	12.08	1.42

4.3.4 Effect of storage period and container on sugar and protein content of kenaf seeds

Results depicted in figure 11 and 12 shows that storage period and container has significant effect on protein and sugar content of seeds. In case of tin container no significant difference in sugar content was observed between initial stage of storage (246.65 mg / g) and 7 months after storage (251.09 mg/g). In jute sack container, sugar content in seed increased significantly (at 1% level) due to storage (246.84 mg/g to 414.55 mg /g).

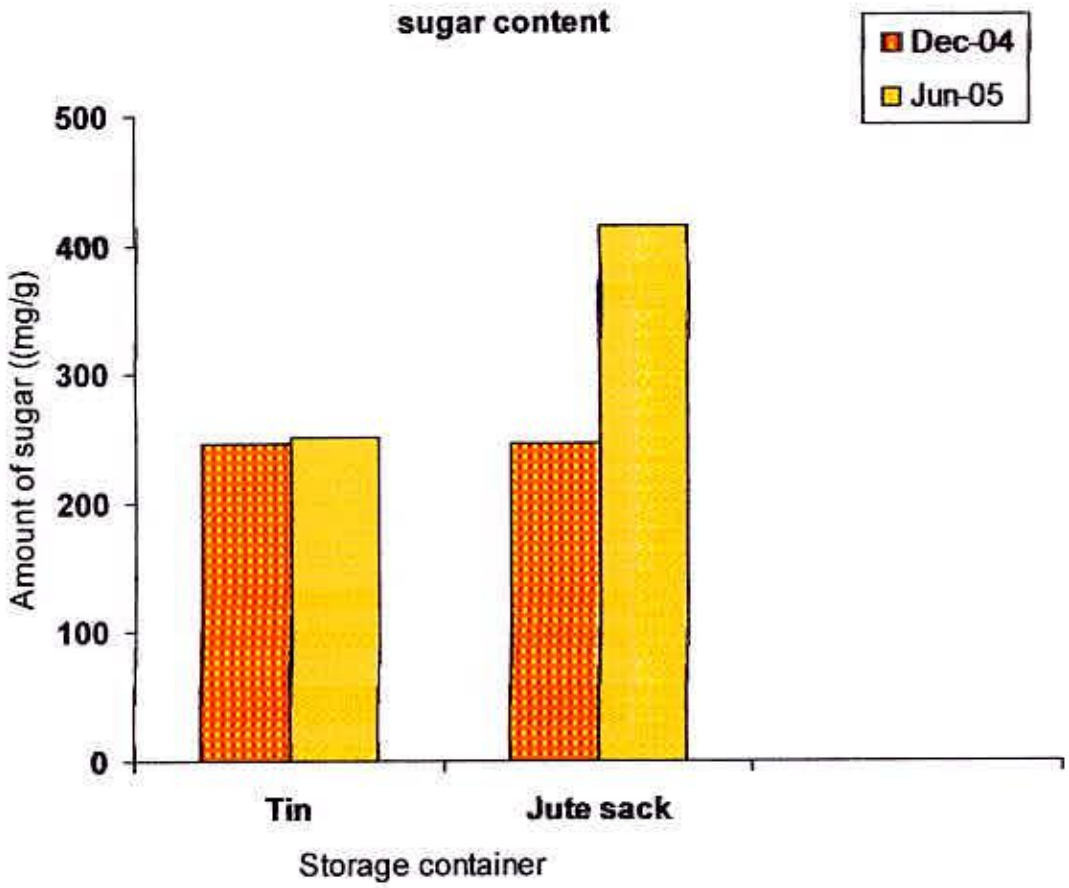


Figure 11 Interaction effect of storage period and container on sugar content of kenaf seeds

Slight decrease in protein content was observed in seeds of tin container and it decreased from 182.98 µg/g to 175.96 µg/g but in jute sack container protein content decreased drastically (from 182.85 µg/g to 18.27 µg/g) due to storage (Figure 12).

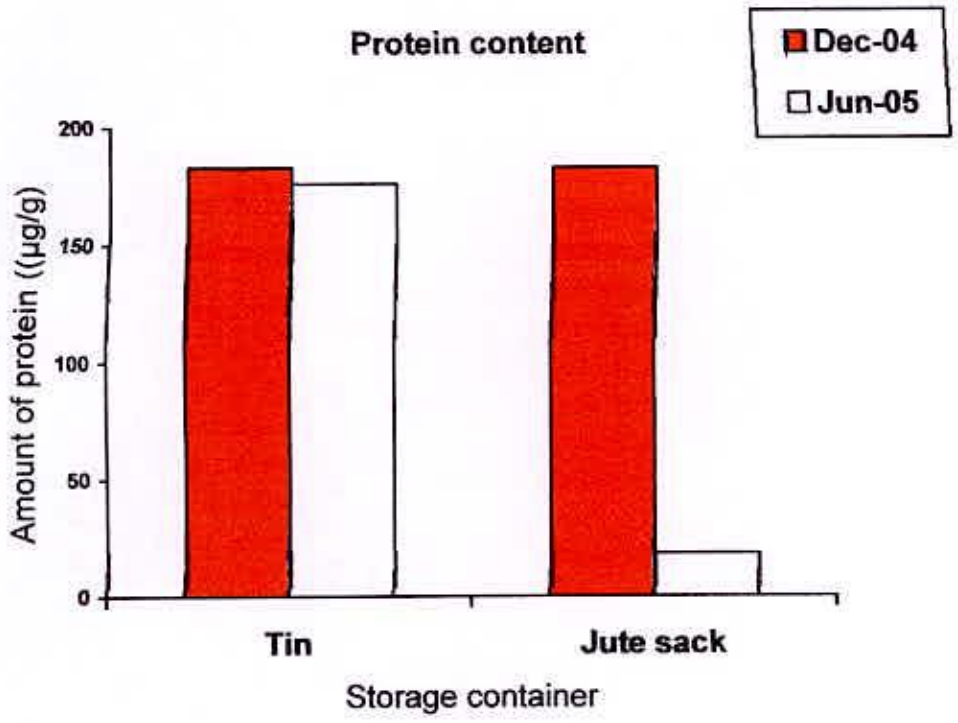


Figure 12 Interaction effect of storage period and container on protein content of kenaf seeds

Results revealed that tin container hold protein content for longer time but jute sack cannot retain protein content in seed and it indicates that jute sack cannot hold quality for longer time. This result agrees with that of Hossain (2003) who stated that tin container retained higher seed quality attributes through out the storage period.

4.3.5 Effect of storage period and genotype on sugar and protein content of kenaf seeds

Results depicted in Table 15 shows that seeds of variety HC-2 initially contained comparatively highest sugar content (263.95 mg/g) than HC-95 (240.32 mg /g) and CPI-72126 /1 (235.96 mg /g), but after 7 months of storage the highest sugar content was recorded in HC-95 (396.46 mg /g) and HC-2, CPI-72126/1 had obtained 291.76 and 310.25 mg/g of seeds, respectively. This increase of sugar content for the genotype was significant at 1 % level of significance.

Protein content was higher in CPI-72126 / 1 (194.86 µg/g) at before storage than HC-95 (184.60 µg/g) and HC-2 (169.30 µg/g) and after storage highest protein content was recorded in HC-95 (101.53 µg/g) and lowest protein content was recorded 90.11 µg/g for HC-2 variety. Protein content declination was higher in CPI 72126 / 1 (194.86 -99.71) = 95.15 µg/g than HC-95 (184.60 -101.53) = 83.07 and HC-2 (169.30 -90.11) = 79.19 µg/g. The decrease of protein content was significant at 1 % level of significance.

Table 15 Interaction effect of storage period and genotype on sugar and protein content of kenaf seeds.

Genotype	Amount of sugar (mg/g)		Amount of protein (µg/g)	
	December 2004	June 2005	December 2004	June 2005
HC-2	263.95	291.76	169.30	90.11
HC-95	240.32	396.46	184.60	101.53
CPI-72126/1	235.96	310.25	194.86	99.71
LSD (1% level)	5.76		2.00	

4.3.6 Effect of storage container and genotype on sugar and protein content of kenaf seeds

Kenaf seeds were stored in tin and jute sack container for 7 months. The highest sugar content was found in HC-95 variety with jute sack container (394.89 mg/g) and the lowest in CPI 72126 / 1 with tin container (237.60 mg/g). Sugar content in HC-95 seed with jute sack container was statistically higher than all other combination.

The highest protein content was recorded in CPI 72126 / 1 (189.83 µg/g) with tin container which was significantly higher than all other combinations and the lowest protein content was recorded in HC-2 (93.04 µg/g) in jute sack container. For each genotype tin container showed the higher protein content than jute sack container that shown in Table 16.

Table 16 Interaction effect of genotype and container on sugar and protein content of kenaf seeds

Genotype	Amount of sugar (mg/g)		Amount of protein (µg/g)	
	Tin	Jute sack	Tin	Jute sack
HC-2	267.13	288.58	166.36	93.04
HC-95	241.88	394.89	182.22	103.90
CPI-72126/1	237.60	308.63	189.83	104.75
LSD (1% level)	5.76		2.00	

4.3.7 Effect of storage period, storage container and genotype on sugar and protein content of kenaf seeds

Sugar and protein contents of kenaf seeds stored in air tight tin and jute sack containers were determined twice, first in December 2004 and second in June 2005.

Sugar content

Tin container

Results presented in Table 17 shows that before storage sugar content of seeds of HC-2, HC-95 and CPI-72126 were 263.86, 240.32 and 235.76 mg/g of seed, respectively. Sugar content increased due to storage in all genotype of seeds and it increased to (270.40 - 263.86) = 6.54 mg/g, (243.44 - 240.32) = 3.12 mg/g and (239.43 - 235.76) = 3.67 mg/g for HC-2, HC-95 and CPI-72126, respectively. Storage duration did not exert any significant effect on sugar content in kenaf seeds when stored in airtight tin container.

Jute sack container

Sugar content in seeds rapidly increased in jute sack container than in tin container. Sugar content increased (313.12-264.03) = 49.09 mg / g, (549.47-240.32) = 309.15 and (381.08-236.16) =144.92 mg / g for HC-2, HC-95 and CPI 72126/1, respectively (Table 17). There is significant difference at 1% level between December observation and May observation for all genotype i.e, sugar content in kenaf seeds significantly increased with the increase in storage duration when seeds were stored in jute sack container.

Protein content

Tin container

Protein content decreased due to storage in all genotype of seed. Protein content decreased (169.43-163.29) = 6.14 µg/g, (184.70-179.75) = 4.95 µg/g and (194.83-184.83)

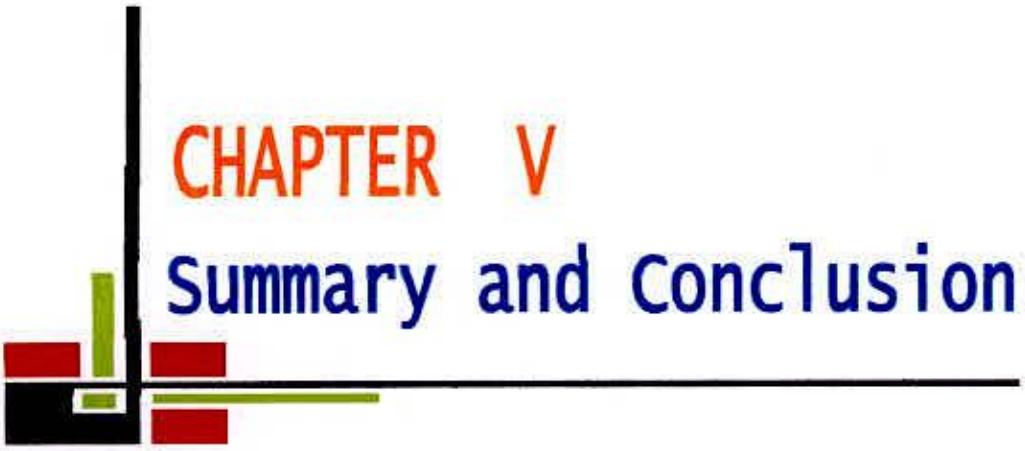
=10.00 µg/g for HC-2, HC-95 and CPI 72126 /1 respectively. There is no significant difference between December observation and May observation for all genotypes .This means that storage duration had little or no effect on protein content in kenaf seeds when stored in tin container.

Jute sack container

Protein content in seed drastically decreased in jute sack container than in tin container. Protein content decreased to (169.16-16.93) = 152.05 µg/g, (184.50-23.30) =161.2 µg/g and (194.90-14.60) = 180.3 µg/g for HC-2, HC-95 and CPI 72126/1, respectively. Significant differences were found between December and May observation at 1% level in all genotypes i.e,protein content in kenaf seeds significantly decreased with the increase in storage duration when seeds were stored in jute sack container.

Table 17 Interaction effect of storage period, genotype and container on sugar and protein content of kenaf seeds

Storage period	Genotype	Amount of sugar (mg/g)		Amount of protein (µg/gm)	
		Tin	Jute sack	Tin	Jute sack
December 2004	HC-2	263.86	264.03	169.43	169.16
	HC-95	240.32	240.32	184.70	184.50
	CPI-72126/1	235.76	236.16	194.83	194.90
June 2005	HC-2	270.40	313.12	163.297	16.93
	HC-95	243.44	549.47	179.75	23.30
	CPI-72126/1	239.43	381.08	184.83	14.60
	LSD (1% level)	16.81			



CHAPTER V

Summary and Conclusion

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SUMMARY AND CONCLUSION

The study was conducted at Bangladesh Jute Research Institute during the period from December 2004 to June 2005. The study included 1) storing of seeds and 2) physiological and biochemical records of fresh and stored seeds.

Seeds of two kenaf varieties (HC-2 and HC-95) and an advance line CPI-72126/1 were stored in jute sack and tin container. After 7 months of storing in air tight tin container germination percentage and moisture content remained unchanged but deterioration of viability was observed in seeds stored in jute sack. At that time moisture content increased significantly.

The type of storage container influenced seed moisture content throughout the storage period. The jute sack resisted little moisture vapour penetration and seed of all genotype stored in this container gained moisture to higher levels. Tin container on the other hand, resisted seed moisture absorption to a higher extent and maintained it below or close to the critical at storage. Rise in seed moisture content of all genotype showed inverse relationship with the magnitude of seed quality attributes. In jute sack, seed moisture rose sharply and it provided adverse effect upon seed quality and thus seed quality retarded sharply. However, tin container maintained seed viability and vigour much higher compared to those of jute sack and thus, tin container expected to come into effect as satisfactory containers for storage of kenaf seeds over the season.

Experiments on biochemical basis of seed deterioration, soluble sugar and protein content of seeds (before and after storing) was measured. In case of sugar no appreciable difference was observed in seeds of all genotype stored in tin container but significant

increase in sugar content was observed in all genotype when seeds were stored in jute sack. Again higher amount of protein was observed in fresh seeds of kenaf varieties. After 7 months of storage in jute sack significant decrease in protein content was observed in all genotype but no appreciable change was observed in seeds stored in tin container.

During investigation on the viability of stored kenaf seeds some biochemical changes have been detected as they deteriorate. These include increasing soluble sugar and decreasing protein level. Seeds stored in jute sack gained moisture very quickly and seed quality retarded sharply and it was assumed that the extent of deterioration was related directly to seed moisture content.

Our knowledge of biochemical deterioration of seeds is still limited, and therefore, it does not permit making firm conclusions. Therefore, in discussing some of the major biochemical changes that are observed in seeds as they deteriorate, allowance will be made for the possibility that most of the changes are results than causes.

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ANNEXURE

ANNEXURE I: Analysis of variance for different parameters

01. Germination percentage

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F- Value
Treatments	41	15921.69048	388.33391	300.18**
Time (T)	6	7063.96825	1177.32804	910.08**
Container (C)	1	5194.29365	5194.29365	4015.22**
Variety (V)	2	43.42857	21.71429	16.79**
TXC	6	3112.76190	518.79365	401.03**
TXV	12	336.12698	28.01058	21.65**
CXV	2	14.15873	7.07937	5.47**
TXCXV	12	156.95238	13.07937	10.11**
Error	84	108.66667	1.29365	
Total	125	16030.35714		

CV= 1.38 %

** = Significant at 1% level

02. Moisture percentage

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F- Value
Treatments	41	288.0146992	7.0247488	2220.57**
Time (T)	6	68.0064492	11.3344082	3582.88**
Container (C)	1	123.8294294	123.8294294	39143.27**
Variety (V)	2	0.6624063	0.3312032	104.70**
TXC	6	88.1613540	14.6935590	4644.73**
TXV	12	3.0936603	0.2578050	81.49**
CXV	2	0.6797397	0.3398698	107.44**
TXCXV	12	3.5816603	0.2984717	94.35**
Error	84	0.2657333	0.0031635	
Total	125	288.2804325		

CV= 0.68 %

** = Significant at 1% level

03. Protein content

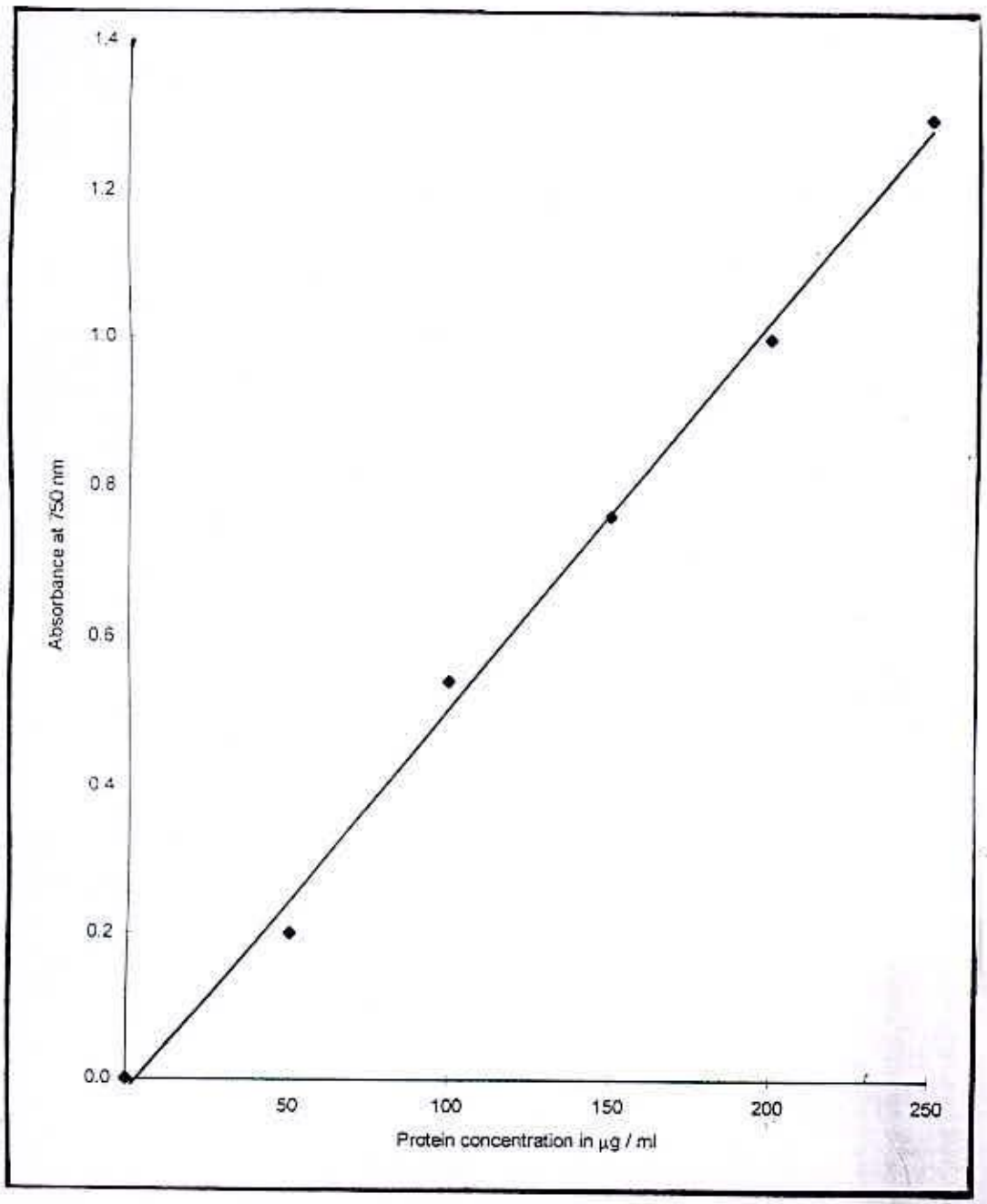
Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F- Value
Treatments	11	181016.7197	16456.0654	133795.19**
Time (T)	1	66258.1920	66258.1920	538708.82**
Container (C)	1	56039.5147	56039.5147	455626.39**
Variety (V)	2	2022.1680	1011.0840	8220.57**
TXC	1	55850.2934	55850.2934	454087.94**
TXV	2	415.8347	207.9173	1690.46**
CXV	2	209.2071	104.6035	850.47**
TXCXV	2	221.5098	110.7549	900.49**
Error	24	2.9519	0.1230	
Total	35	181019.6716		

CV= 0.38%
** = Significant at 1% level

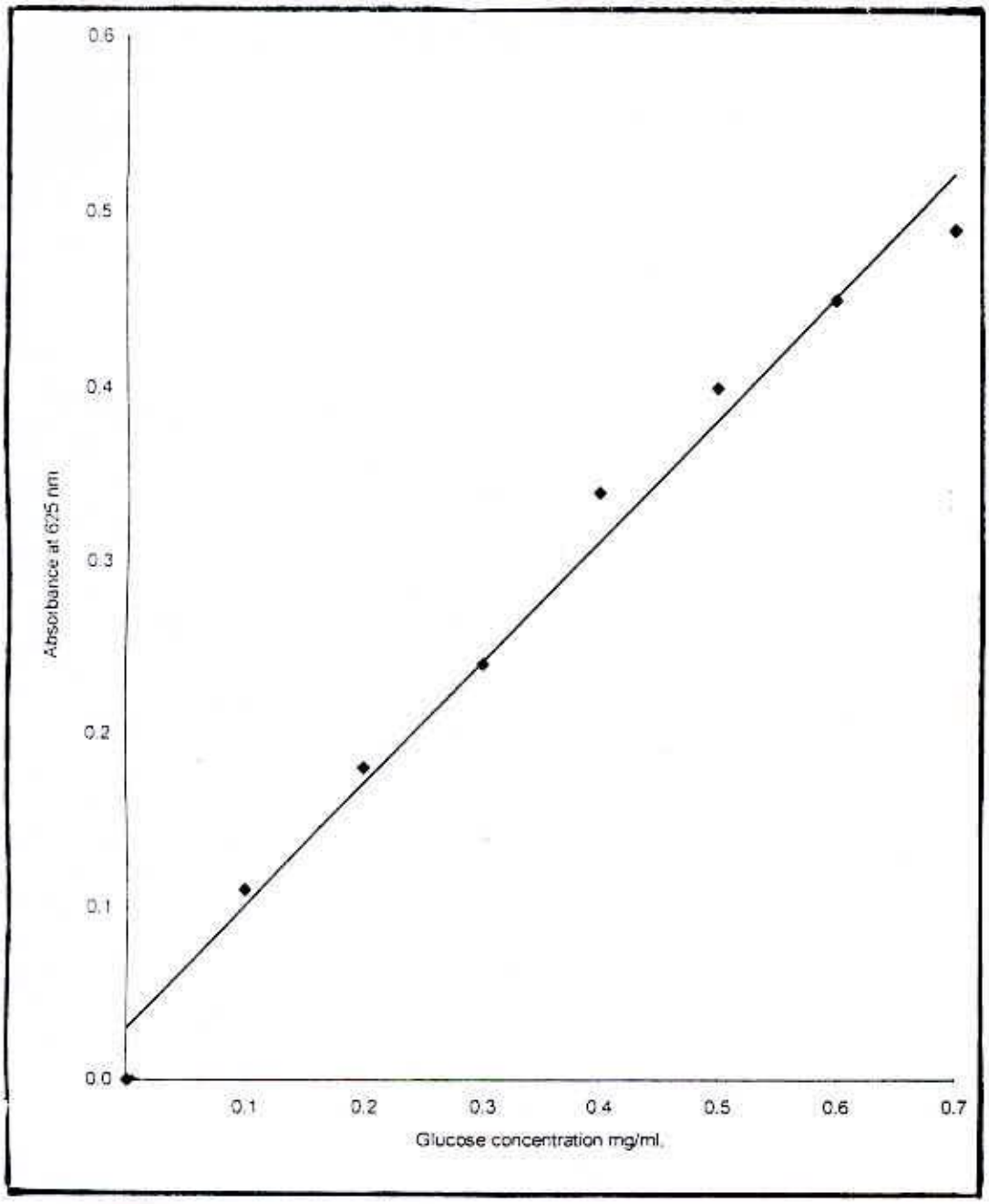
04. Sugar content

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F- Value
Treatments	11	280179.4936	25470.8631	2864.39**
Time (T)	1	66685.3152	66685.3152	7499.26**
Container (C)	1	60261.2487	60261.2487	6776.83**
Variety (V)	2	14863.1851	7431.5926	835.74**
TXC	1	59983.3572	59983.3572	6745.58**
TXV	2	25322.9152	12661.4576	1423.88**
CXV	2	26488.4620	13244.2310	1489.41**
TXCXV	2	26575.0102	13287.5051	1494.28**
Error	24	213.4139	8.8922	
Total	35	280392.9076		

CV= 1.00 %
** = Significant at 1% level



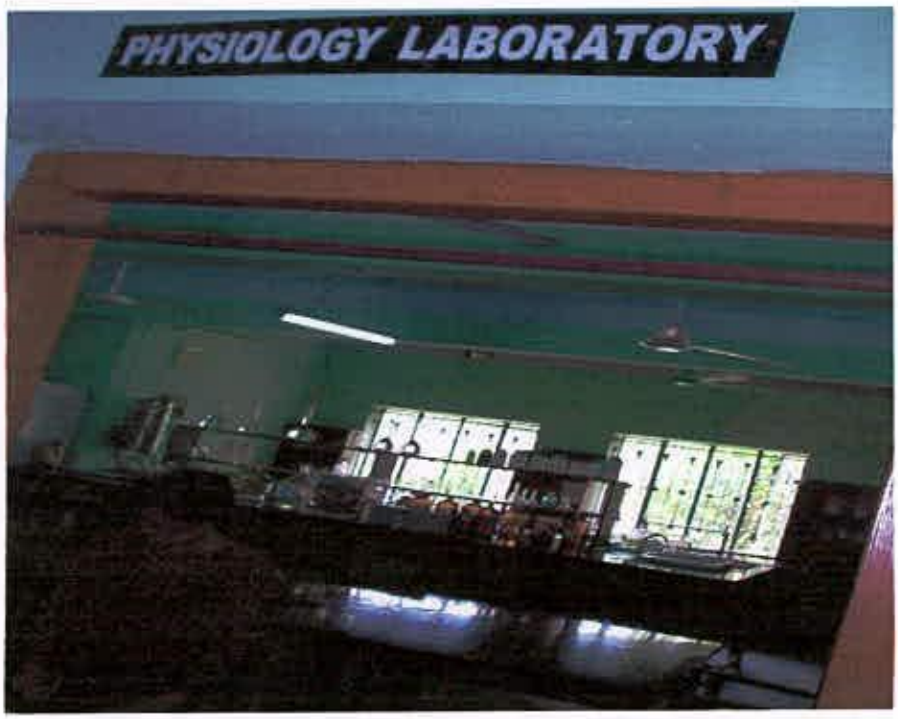
Annexure II (a): Standard calibration curve for protein estimation by Folin-Ciocalteau Phenol reagent



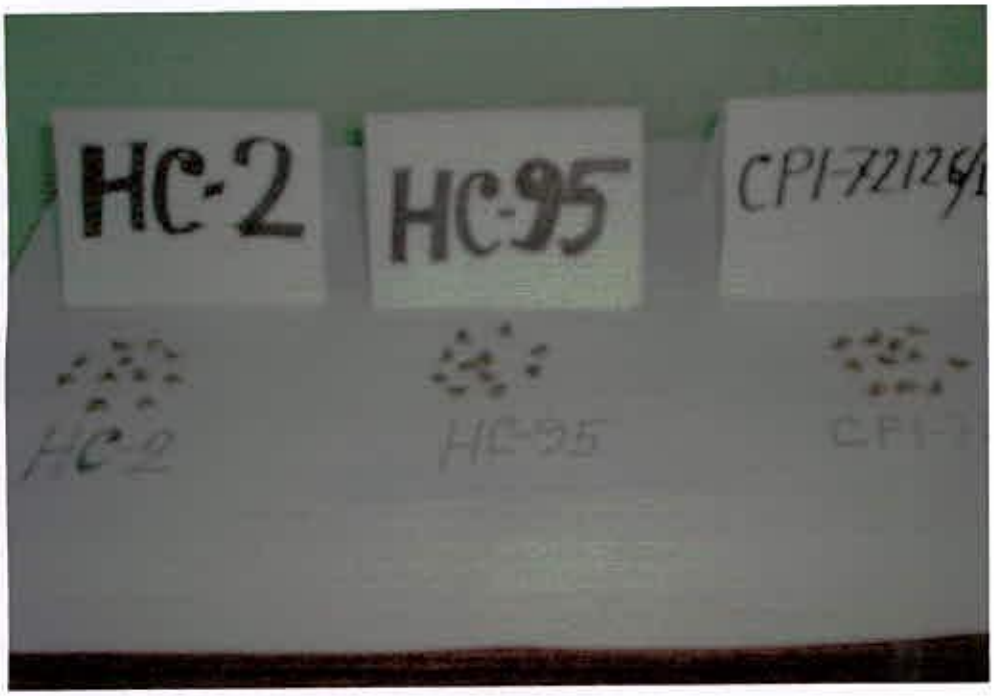
Annexure II (b). Standard calibration curve for sugar estimation

ANNEXURE III : Photograph of the experiment

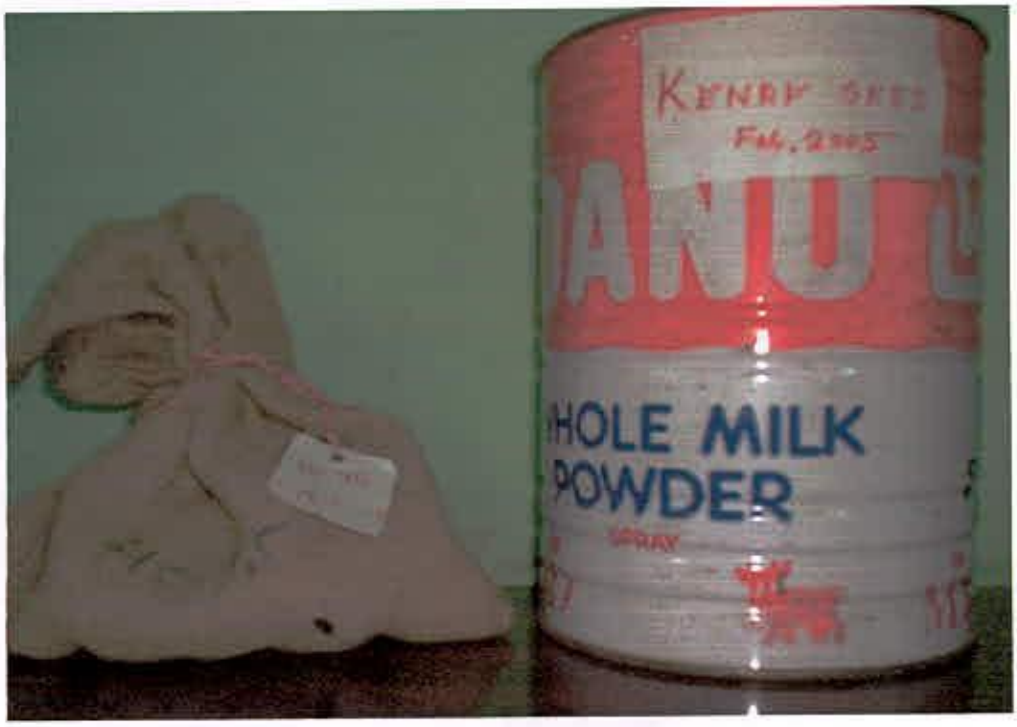
Picture 1 A view of three kenaf varieties/line



Picture 2 A view of Physiology Laboratory of BJRI, Dhaka.



Picture 3 Seeds of kenaf varieties/lines



Picture 4 Photograph of storage container (Jute sack and Tin container)