

Radiation Preservation of Black Velvet Tamarind (*Dialium guineense* Wild.)

This thesis is submitted to the University of Ghana, Legon in partial fulfilment of the requirement for the award of **MPhil Radiation Processing degree**

BY

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DECLARATION

This thesis is the result of research work by Daniel Osei Ofosu in the Department of Nuclear Agriculture and Radiation Processing, University of Ghana, under the supervision of Prof. G.T. Odamtten. Except for references of other peoples' work which I have duly cited, this thesis has not been presented either in whole or in part for another degree elsewhere.

Sign 

Daniel Osei Ofosu

1st Dec 2010

Date

Sign 

Prof. G.T. Odamtten

18th Dec 2010

Date



DEDICATION

This project is dedicated to the Almighty God who has brought me this far. To my mother, Miss Comfort Ama Tawiah Thompson, siblings, Francis Martinson and Francisca Martinson, relatives, friends and anyone who prayed for me to complete this course, this work is for you.

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TABLE OF CONTENTS

	Page
DECLARATION	ii
DEDICATION	iii
ACKNOWLEDGEMENTS	iv
TABLE OF CONTENTS	v
LIST OF TABLES	viii
LIST OF PLATES	x
LIST OF MAPS	xi
LIST OF FIGURES	xii
ABSTRACT	xiv
CHAPTER ONE: INTRODUCTION	
1.1 Background information	1
1.2 Research problem	4
1.3 Objectives and scope of the study	5
1.4 Relevance of the project	6
CHAPTER TWO: LITERATURE REVIEW	
2.1 <i>Dialium guineense</i>	7
2.2 Distribution of <i>Dialium guineense</i>	10
2.2.1 Seasonality and economic value	10
2.2.2 Current knowledge of the nutritive value of <i>Dialium guineense</i>	14
2.3 Importance of food preservation	15
2.4 Irradiation processing of foods to extend shelf-life	16

2.5 Food Packaging	18
CHAPTER THREE: MATERIALS AND METHODS	
3.1 Questionnaire Administration	20
3.2 Sample collection	20
3.3 Physical Characteristics, Packaging, Irradiation and Storage of Black Velvet Tamarind fruits	21
3.4.1 Determination of percentage moisture content of the pulp	21
3.4.2 Determination of water sorption isotherm of the pulp and the whole fruit under varying Equilibrium Relative Humidities (ERH's) at 30 °C	22
3.4.3 Determination of pH of pulp	25
3.4.4 Determination of total titratable acidity of pulp	25
3.4.5 Determination of ash content of pulp	25
3.4.6 Determination of reducing sugar content of pulp	26
3.4.7 Determination of crude fat content of pulp	27
3.4.8 Determination of crude protein content of pulp	27
3.4.9 Mycoflora of whole and dehulled fruit before and after irradiation and storage in different packaging materials	28
3.4.10 Determination of elemental composition of <i>Dialium guineense</i>	28
3.5 Hidden insect infestation of fruits	31
3.6 Sensory evaluation	31
3.7 Statistical analysis	33

CHAPTER FOUR: RESULTS

4.1 Rapid appraisal method for assessing and determining sources of <i>Dialium guineense</i> on the local markets in Ghana	34
4.2 Physical characteristics of the fruit	40
4.3 Moisture sorption isotherms of whole and de-hulled fruit (pulp) of <i>D. guineense</i> at varying equilibrium relative humidities (ERH's)	43
4.4 Mycoflora resident in the fruit of black velvet tamarind (<i>D. guineense</i>)	46
4.5 Hidden insect infestation of fruits	52
4.6 Influence of gamma irradiation and packaging on some mycological quality of the pulp of <i>D. guineense</i> stored at $29\pm 1^{\circ}\text{C}$ for 3 months	54
4.7 Influence of gamma irradiation and packaging on the physico-chemical properties of the pulp of <i>D. guineense</i> stored at $29\pm 1^{\circ}\text{C}$ for 3 months	64
4.8 Elemental composition of the fruit coat, pulp and seed of <i>D. guineense</i>	74
4.9 Sensory evaluation of the fruit of <i>D. guineense</i> treated with gamma irradiation and stored in different packaging materials for 3 months	76

CHAPTER FIVE: DISCUSSION

5.0 General Discussion	83
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CHAPTER SIX: CONCLUSIONS AND RECOMMENDATIONS

6.0 Conclusions and Recommendations	94
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REFERENCES	96
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APPENDICES	107
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LIST OF TABLES

Table		Page
1	Herbarium records of <i>Dialium guineense</i> Willd in Ghana	11
2	Specific markets in which questionnaire was administered	20
3	Formulation of glycerol : water mixtures used to establish Equilibrium Relative Humidity in the desiccators	23
4	Nuclear Data (IAEA TECDOC-564)	30
5	Local names of <i>D. guineense</i>	36
6	Mean values for physical characteristics of <i>D. guineense</i> fruit	42
7	Total list of resident fungi isolated from pulp of <i>D. guineense</i> at 28±1°C	49
8	Total list of resident fungi isolated from the outer covering of <i>D. guineense</i> at 28±1°C	50
9	pH of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGy of gamma irradiation	67
10	Total titratable acidity of the fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGy of gamma irradiation	68
11	Ash content of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGy of gamma irradiation	69
12	Moisture content of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGy of gamma irradiation	70

Table	LIST OF TABLES (CONT'D)	Page
13	Crude fat content of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGy of gamma irradiation	71
14	Reducing sugar content of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGy of gamma irradiation	72
15	Crude protein of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGy of gamma irradiation	73
16	Elemental composition of the fruit coat, pulp and seed of <i>D. guineense</i>	75
17	Descriptive sensory evaluation of samples stored in polyethylene bags	80
18	Descriptive sensory evaluation of samples stored in polypropylene bags	80
19	Descriptive sensory evaluation of samples stored in jute sack	81
20	Preference sensory evaluation of samples stored in polyethylene bags	81
21	Preference sensory evaluation of samples stored in polypropylene Bags	82
22	Preference sensory evaluation of samples stored in jute sack	82

LIST OF PLATES

Plate		Page
1	Habit of <i>D. guineense</i> in the field	9
2	A bunch of <i>D. guineense</i> fruits showing the black velvety exterior of the pod	9
3	Photograph showing the arrangement of the black velvet tamarind inside glass desiccators (Note: The glycerol: water mixtures of simulating the prescribed ERH's were poured into the bottom of the container)	24
4	Adult (A) and larva (B) of <i>E. cautella</i> (Walker)	53

LIST OF MAPS

Map		Page
1	Flow of sale of <i>D. guineense</i> fruits obtained from markets in the indicated regions of Ghana	35

LIST OF FIGURES

Figure		Page
1	People who regularly purchase <i>D. guineense</i> fruits from the market	37
2	Choice of transporting and storage material for the <i>D. guineense</i> fruits	39
3	Major sources of post harvest loss in <i>D. guineense</i> fruits	39
4	Class-size distribution of the pod weight of <i>D. guineense</i>	41
5	Class-size distribution of the mean diameter of the pod of <i>D. guineense</i>	41
6	Moisture sorption isotherm of the whole fruit of <i>Dialium guineense</i> at different indicated equilibrium relative humidities (ERH's) at $29\pm 1^{\circ}\text{C}$ for 30 days	44
7	Moisture sorption isotherm of the pulp of <i>Dialium guineense</i> at different indicated equilibrium relative humidities (ERH's) at $29\pm 1^{\circ}\text{C}$ for 30 days	45
8	Percentage occurrence of selected fungi in the pulp of the fruits cultured on either DG 18 or OGYE at $28\pm 1^{\circ}\text{C}$ for 7 days	47
9	Percentage occurrence of selected fungi in the outer coat of the fruits cultured on either DG 18 or OGYE at $28\pm 1^{\circ}\text{C}$ for 7 days	48
10	Changes in the total number of resident fungi in the pulp immediately after gamma irradiation	56
11	Changes in the total number of resident fungi in the pulp three months after gamma irradiation	57

12	Percent occurrence of mycoflora on the pulp of <i>D. guineense</i> stored in polyethylene bags immediately after irradiation with indicated doses	58
13	Percent occurrence of mycoflora on the pulp of <i>D. guineense</i> stored in polyethylene bags for three months after irradiation with indicated doses	59
14	Percent occurrence of mycoflora on the pulp of <i>D. guineense</i> stored in polypropylene bags immediately after irradiation with indicated doses	60
15	Percent occurrence of mycoflora on the pulp of <i>D. guineense</i> stored in polypropylene bags for three months after irradiation with indicated doses	61
16	Percent occurrence of mycoflora on the pulp of <i>D. guineense</i> stored in jute sacks immediately after irradiation with indicated doses	62
17	Percent occurrence of mycoflora on the pulp of <i>D. guineense</i> stored in jute sacks for three months after irradiation with indicated doses	63

ABSTRACT

The economic distribution and marketing of the lesser known fruit *Dialium guineense* Wild in Ghana has been studied using a structured questionnaire and the rapid appraisal system. The seasonality of the fruit (from January to May) makes it abundant in the peak season and rare or not all for the best part of the year. Some fruits also go to waste in the field owing to underexploitation. This thesis provides information on the resident fungi, mycological quality, sorption isotherm, and the effect of gamma irradiation and packaging on the physico-chemical properties of the fruit and the sensory evaluation of the pulp by a taste panel with the view to showing the economical and nutritional potential of the fruit.

The resident fungi and mycological quality were assessed by the conventional decimal serial dilution technique and plating on three media (PDA, DG 18 and OGYE). Sorption isotherms at $29\pm 1^{\circ}\text{C}$ were determined under simulated Environmental Relative Humidity (20, 55, 65, 75, 85, 95%) provided by glycerol: water mixtures; physico-chemical properties were assessed by the method of the Association of Official Analytical Chemists. Descriptive and preference sensory evaluation were administered using prescribed international methods on a hedonic scale.

The tamarind markets are in the southern sector of the country, namely Greater Accra, Eastern, Central, Ashanti and Volta Regions with the hub at Ho, Abor and Akatsi in the Volta Region. The local names of the fruit varied from one region to another. The fruit was purchased mostly by adult females (48.25%) followed by school children (37.06%) and adult males (14.68%). The produce is marketed predominantly in polypropylene sacks (81.81%) with jute sack taking only 3.63% of the packaging.



The sizes of the fruit as expected were variable. The pulp did not lose moisture at 75% ERH but lost moisture to the surrounding at ERH's 20, 55, and 65% and the same was true for the whole (intact) fruit.

The resident fungi in the pulp belonged to 15 fungal species and 7 genera (*Aspergillus*, *Candida*, *Cladosporium*, *Fusarium*, *Penicillium*, *Neurospora* and *Rhodotorula*). The fungal flora was predominated by *Aspergillus* (*A. alutaceus*, *A. candidus*, *A. flavus*, *A. fumigatus*, *A. niger*, *A. sulphureus*, *A. ustus*) followed by *Penicillium* (*P. digitatum*, *P. expansum*) all of which are being recorded for the first time in the fruit of *D. guineense*. The fruits were also infested with an insect, *Ephestia cautella*, of economic importance. There was a commensurate drastic reduction in the mould count in the fruit as the irradiation dose increased from 0 to 10kGy. The resident fungi were not however completely eliminated with the variable residual species appearing after 3 months storage in the packaging materials (polypropylene, polyethylene and jute). The presence and isolation of mycotoxin-producing species *A. flavus* (Aflatoxins), *A. alutaceus* (Ochratoxins), *P. expansum*, *P. digitatum* (Patulin), *F. verticilloides* (fumonisin) leave much to be desired.

Packaging did not significantly influence pH and ash content; but influenced total titratable acidity with increasing dose and storage time. The crude fat content decreased with prolonged storage and increasing dose of gamma irradiation while there was no interaction between dose applied, packaging material and storage time. The panelists did not find significant ($P>0.05$) difference in the parameters tested for acceptance (colour, sweetness, acidity and tenderness) and they found no differences in the packaging material although they slightly preferred produce kept in jute sacks. The practical implications of these findings are discussed in the light of future promotion and preservation of the pulp for industrial use after gamma irradiation.

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background information

Fruits are generally acceptable as good sources of nutrients and supplement for food in a world faced with the problem of food scarcity. They are known to be excellent sources of nutrients such as minerals and vitamins; they also contain carbohydrates in the form of soluble sugars, cellulose and starch (Nahar *et al.*, 1990). Fruits are a very vital portion of an adequate diet and they serve as food supplement, and an appetizer. For example, dry fruits like apricots, raisins and dates are storehouses of calcium and iron, essential for the strengthening of bones. Custard apple is another excellent source of calcium. Guavas, custard apples, lemons and oranges are effective sources of vitamin C (Romero *et al.*, 1992). Fruits like apple, lemon, orange and pomegranate aid in the proper functioning of the heart. All forms of berries are rich in iron, phosphorus and sodium that are beneficial for blood building and nerve strengthening. Lemons are good for the treatment of liver ailments, indigestion and rheumatism.

The fruits, seeds and leaves of many wild plants already form common ingredients in a variety of traditional native dishes for the rural populace in developing countries (Lock *et al.*, 2005, Humphrey *et al.*, 1993 and Kuhnlein, 1989). However, as a result of the environmental changes sweeping through African societies, wild plants are in danger of disappearing and this may have harmful consequences on the nutritional status of the rural populace (Herzog *et al.*, 1994).

Wild edible fruits are being investigated for their potential use as food supplements in the Sahelian region to increase quality of daily food for the rural population (Glew *et al*, 2005; Okafor, 1981; Getahun, 1974). Research on wild fruits and other wild edible plants is also intended to promote the preservation of these species. In addition to their nutritional value, the preservation of these fruits also has economical benefits, as there is significant trade in some of these wild edible fruits (Ambé, 2001). Even though tropical fruit trees are not the targets of large world markets, they still have considerable importance in local and national economies, being harvested by rural populations for local consumption and commercialisation on a small scale (Bonkougou *et al*, 1998).

Regrettably, the pertinent literature has limited information on the taxonomic accounts of many edible wild plants. Information available were derived from field notes, general oral tradition and those which are found in textbooks. Seyani (1988), presents an account of 180 species of African edible fruits (berries or nuts) distributed in 39 families and 19 species of edible wild flowers in seven families. Okafor (1981) provides information on 171 indigenous woody plants (53 families and 119 genera) of nutritional importance within the forest zone of Nigeria. About 75 wild edible fruit species known by the Malinké ethnic group of Cote d'Ivoire have been inventoried by Ambé (2001).

Examples of well known wild edible fruit species in Africa include *Adansonia digitata* (baobab), *Detarium microcarpum*, *Diospyros mespiliformis* (jackalberry), *Ficus sycomorus* (sycamore fig), *Gardenia erubescens*, *Lannea microcarpa*, *Parkia biglobosa*, *Saba senegalensis*, *Sclerocarya birrea* (marula), *Tamarindus indica* (tamarind), *Vitellaria paradoxa* (= *Butyrospermum parkii*, shea tree), *Ximenia*

americana (sea lemon), *Ziziphus mauritiana* and *Dialium guineense* (Wild), (Lamien-Meda *et al*, 2008).

One of the major limitations of these well known wild fruits is that they are usually seasonal fruits, with only one fruiting season in a year. Gathering for storage has not been attractive economically to farmers in most instances. The black velvet tamarind (*Dialium guineense* Wild) fruit is no exception to this problem as it is seasonal with only one fruiting season lasting for just three (3) to four (4) months. The ripe fruits are available from January till May but the peak period for harvest is between March and April (Okafor, 1975). At the peak harvest period, there is so much ripe fruits which goes to waste as there is presently no industrial product made from the pulp or any method available for preservation. Also, because much of it is not demanded, farmers are not keen in domesticating and raising orchards of the plant (Onwuka and Nwokorie, 2006).

The need for meeting the beverage requirements of Ghana's growing population and development of other sources of fruit wine is even more pressing. Production of non-alcoholic and alcoholic wine from temperate fruits or the reliance on importation of wine are inimical to conserving foreign exchange, stimulating research and establishment of local orchards for beverage production from fruits (Onwuka and Nwokorie, 2006).

One promising source of Ghanaian local raw material for beverage, soft drinks, syrup concentrate and jams is *D. guineense* fruit pulp. The fruit pulp contains a high proportion of vitamin C, sugars and other nutritive components (Irvine, 1961). The pulp can be processed into various food products ranging from jelly to jam and more recently, into non-alcoholic beverage (Onwuka and Nwokorie, 2006). The diet of

many rural and urban dwellers is deficient in proteins, vitamins, essential mineral elements, but high in carbohydrates. The implication is the high incidence of malnutrition and increase in dietary disease, a situation in which children and pregnant and lactating women are most vulnerable (Sadik, 1991). African governments are taking the necessary steps to boost food production by conventional agriculture, and a lot is being done to focus on the possibilities of exploiting the vast numbers of less familiar plant resources of the wild forests (Abdullahi and Abdullahi 2005; Anhwange *et al.* 2004). Many reports on some lesser known seeds and fruits indicate that they could be good sources of nutrient for both man and livestock (Adenkunle and Ogerinde, 2004; Elemo *et al.*, 2002). The fruit of *D. guineense* is no exception and has an economic potential for extensive human exploitation in Ghana.

In Ghana, there is limited research into the nutritional composition, uses or preservation of *Dialium guineense*. However, it has been shown that the fruit could be processed into beverages, soft drinks, alcoholic drinks, syrup/concentrate and jams (Okafor, 1975). Indeed, in their research on the winning potential of three fruits including velvet tamarind, Onwuka and Nwokorie (2006) reported that the black velvet tamarind had rich nutrient content and better wine-making quality than the other fruits they studied. It now remains for scientists to demonstrate that this fruit could be gathered, packaged and preserved during the peak season to be used by the food industry on a sustainable basis.

1.2 Research problem

The contributions of wild fruits, nuts, seeds, vegetables and other classes of edible products to the local diet in developing countries and their potential in overcoming prevailing food shortage problems are enormous.



There is currently little industrial value of this fruit; it is only consumed locally in the fresh state and there are no commercial products made from velvet tamarind in Ghana, although it is used elsewhere. This thesis project sought to provide baseline data on:

- The extent of availability of the fruit nationwide through a survey of its collecting and cultivation localities.
- The potential fungi/microorganisms and insects associated with the fruit and pulp in relation to the storage stability and nutritional characteristics.
- The combination of packaging material and gamma irradiation dose which will prolong the storage shelf-life and organoleptic qualities of the pulp.

1.3 Objectives and scope of the study

The main objective of this project was to use radiation techniques to preserve black velvet tamarind fruits for long term storage. Other specific objectives were:

- To provide information on the extent of availability of the fruit in the southern part of Ghana.
- To determine the resident mycoflora and insects in the tamarind fruit harvested from the field likely to reduce the market value of black velvet tamarind fruit.
- To study the moisture sorption isotherms representative of local tropical conditions (20% - 95% ERH) with the view to predicting the spoilage potential of the fruit stored under ambient conditions before industrial use or marketing.
- To determine the nutritional quality of the pulp.

- To determine an effective dose for radiation processing of black velvet tamarind fruits and preservation of the organoleptic and processing characteristics of the pulp.
- To determine the effect of radiation on mineral nutrients, reducing sugar, crude fat and proteins during storage using physico-chemical and taste panel assessment.
- To determine the best packaging material that will prolong the physico-chemical and microbiological qualities of the fruit during storage in the lean season.

1.4 Relevance of the project

It is anticipated that the use of irradiation technology as a means of preserving the fruits of *Dialium guineense* will provide baseline information on the suitability of the process to preserve the microbiological, physical and chemical characteristics as well as the organoleptic qualities of the pulp. This will be an important springboard to stimulate the economic cultivation of the plant in orchards.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Black velvet tamarind (*D. guineense* Wild) belongs to the family Fabaceae which include herbs, vines, shrubs, trees, and lianas found in both temperate and tropical areas. The Fabaceae comprise one of the largest families of flowering plants, with 630 genera and 18,000 species. The genus *Dialium* belongs to the subfamily Caesalpinioideae and includes over 70 species, majority of which occur in Africa (<http://www.biodiversitylibrary.org/title/1428>, 2009).

This plant is a tree which grows to about twenty (20) meters in height, 0.8m in diameter, low-branching, rarely straight, bearing a compact densely leafy crown but is often shrubby (Okegbile and Taiwo, 1990; Burkill, 1985). Plate 1 shows the habit of the plant in the field.

The fruit coat is brown or black in colour, with the fruit having an ovoid shape and about 2.5 cm in diameter with a velvety skin from which the English name is derived (Plate 2). The seeds are embedded in an orange/red pulp which is sweetly acidic and edible (Lewis, 1978). At maturity, the dry circular pods are cracked open to release the semi-dry fruit pulp (Ubbaonu *et al*, 2005).

The tree also has some medicinal value. The bark has some anodynal or analgesic action and infusions and decoctions are used for treating toothache in Cote d'Ivoire and Ghana (Iwu, 1993; Kerharo and Adam, 1964). The leaves are used for making infusions to treat fevers while a decoction is used in the treatment of dysentery (Savill and Fox, 1967).

The pulp of the fruit is macerated with water to make a refreshing fruit-drink (Burkill, 1985). It ferments to a palatable wine resembling a grape white wine, arising, it is thought, from having the right balance of acids and tannin (Onwuka and Nwokorie, 2006).

The fruit is reported to have a good molluscicidal activity on the freshwater snail, *Bulinus globulus*, giving 100% kill in 100 ppm concentration of extract (Adewunmi and Sofowora, 1980).



Plate 1: Habit of *D. guineense* in the field (X 1/10000)



Plate 2: A bunch of *D. guineense* fruits showing the black velvety exterior of the pod
(X 1/2)

2.2 Distribution of *Dialium guineense*

D. guineense grows in dense savannah forests, shadowy canyons and gallery forests. It is also found in the Sahel region from Senegal to Sudan (Szolnoki, 1985). The tree is found on river banks, swamp margins or bordering areas of seasonal inundation, often locally abundant (Madge, 1994; Keay *et al.*, 1964; Aubreville, 1950).

In Ghana, herbarium records (Table 1) suggests that the tree is found mostly in the south of Ghana. These areas include the Greater Accra Region, Central Region, Volta Region and the Ashanti Region (Taylor, 1960).

2.2.1 Seasonality and economic value

In Nigeria, the tree flowers from September to October and fruits from October to January (Keay, 1989). In Ghana, from September to November, the tree is covered with small white flowers in panicles; the fruit ripens from March to May, but may be earlier and persist longer (Taylor, 1960). The peak period for harvest is between March and April (Okafor, 1975).

D. guineense has many economic uses; the tree is used for timber in the construction industry for building houses and boat construction. In Sierra Leone, the wood is traded as a tumble tree or black tumbler. Since the wood does not crack or split easily, it is used for manufacturing pestles for pounding. It is also used as firewood by blacksmiths (Madge, 1994).

Table 1: Herbarium records of *Dialium guineense* Wild in Ghana.

Locality	Date of Collection	Collector	Herbarium code	Determiner
24m S. Have 4° 11' W, 9° 24' N	1968	C. Geerling and J. Bokdam	GC 1538	C. Geerling and J. Bokdam
2m N. E. Nungua	16/4/1953	D.W. Goodall	GC 15574	Not indicated
Aburi Scarp	22/11/1953	J.K. Morton	Not indicated	Not indicated
Accra	28/11/1902	W.H. Johnson	Not indicated	Not indicated
Accra	9/1/1899	T.W. Brown	GC 371	FR Irvine
Accra Plains	20/10/1953	J.K. Morton	Not indicated	Not indicated
Accra Plains, Legon Gardens	Not indicated	Addo- Ashong	Not indicated	Not indicated
Achimota	12/11/1963	G.K. Akpebla	GC 636	Irvine FR
Achimota	8/9/1936	G.K. Akpebla	GC 601	Irvine FR
Achimota	3/5/1937	H.A. Sampram	GC 99	FR Irvine
Achimota compound, Clarke Bungalow	1928	F.R. Irvine	GC 1006	Not indicated

Table 1 (Cont'd): Herbarium records of *Dialium guineense* Wild in Ghana.

Locality	Date of Collection	Collector	Herbarium code	Determiner
Bamboi	18/10/1958	R. Rose Innes	GC 30654	JB Hall
Brong Ahafo, Sandstone rock near Techiman Alt 32m 7 ^o 35.6 ⁱ N, 1 ^o 51.9 ^j W	27/12/1995	Jongkind C.C.H. and Niewenhuis C.M.J.	CJ 2550	C.C.H. Jongkind
Dodowa road junction and Aburi Scarp	?/10/1952	J.K. Morton	GC 7715	Not indicated
Kete-Krachi, Nsunua	19/12/1956	C.D. Adams	GC 4595	Not indicated
Larteh-Akwapim	2/11/1900	W.H. Johnson	GC 808	Not indicated
Legon-Achimota road	10/3/1955	S.A. Nyako	Not indicated	Not indicated
Legon-Hill	18/3/1956	C.D. Adams	GC 3819	Not indicated
Legon-Hill, Legon	22/10/1955 28/1/1956	C.D. Adams C.D. Adams	GC 5458 GC 3734	Not indicated
Mampong-Ashanti	12/4/1956	A.A. Enti	FH 6085 GC 51	Not indicated

Table 1 (Cont'd): Herbarium records of *Dialium guineense* Wild in Ghana.

Locality	Date of Collection	Collector	Herbarium code	Determiner
Mankessim- Winneba Rd. Forest on hillside near Nakwa/Eisam	15/11/1957	C.D. Adams	GC 4911	JB Hall
Mankrong (Kwahu) near River Afram	10/4/1954	J.K. Morton	GC 4549	JK Morton
Not Indicated	Not indicated	F.N. Howes	GC 1138	Not indicated
Sogakope near Volta	14/11/1957	A.A. Enti	FH 6839 GC 599	Not indicated
Wenchi	October 1932	C. Vigne	2532 Imperial Forests Ins. Herbarium IFIH	Oxford by Hoyle (1953)
Yeji on Lagoon bank	11/4/1967	J.B. Hall	VBS 1203	JB Hall

Source: Ghana Herbarium GC, Department of Botany, University of Ghana.

It is likely that there are some records of *D. guineense* in other localities hitherto unrecorded.

2.2.2 Current knowledge of the nutritive value of *Dialium guineense*

The edible part (pulp) of ripe *D. guineense* fruit is sweet due to the presence of sugars (total sugars 582.1 g kg⁻¹). The fruit is relatively low in protein (61.3 g kg⁻¹) and oil (700 g kg⁻¹) but it is highly acidic (pH 3.3). The seed, however, is mildly acidic (pH 5.5), low in oil (60.1 g kg⁻¹) but a fairly good source of protein (148.8 g kg⁻¹) and minerals (Arogba *et al.*, 1994).

Ubbaonu *et al.*, (2005) have reported that the carbohydrate, ethanol soluble sugars and total soluble solids content of the pulp increase significantly from the 5th week values of 5.80%, 5.30% and 6.25 °Brix to values of 35.92%, 29.21%, and 40.01 °Brix respectively at ripening (17th week). Ubbaonu *et al.*, (2005) also identified that the sugars present were glucose, fructose, maltose and sucrose. They also identified tartaric, citric, malic and ascorbic acids.

The black velvet tamarind pulp is eaten in Ghana by both adults and children, and in Nigeria (where it is known as Icheku or Nchichi) because of its refreshing properties and pleasant scorching taste (Ubbaonu *et al.*, 2005). The mature circular pods are cracked open (manually) to release the semi-dry pulp, which also embeds the seed. There is currently little industrial value attached to this fruit; it is only consumed locally in the fresh state and there are no commercial products made from black velvet tamarind. However, some workers have shown that the fruit can be processed into beverages, alcoholic and non-alcoholic drinks, syrups/concentrates and jams (Okafor, 1975). Studies have also been carried out to examine some physical and chemical changes in the black velvet tamarind fruits at different stages of the fruit development in order to elucidate the appropriate period for harvesting the fruits to



harness the desired properties and the delicate flavour of the pulp which incidentally alters during the fruit development and ripening (Ubbaonu *et al.*, 2005).

The problem black velvet tamarind has is that it is a seasonal fruit lasting for three (3) to four (4) months (Onwuka and Nwokorie, 2006). At the peak harvest period, so much of it is ripen and wastes as there is presently no industrial product produced from it. The microbial and insect deterioration of the fruit in storage has also not been documented to date. Because much of it is not demanded, farmers are not keen on domesticating and raising orchards of the plant. If indeed the fruits can be harvested, dried and preserved in the peak season, there is the potential that the fruit could be tested and proven to be good enough for any large scale production project that can widen the industrial raw material base for the country. The accumulation of information from this baseline study would not only eliminate the post-harvest losses of the fruit but also encourage industrial use of the pulp thus leading to the interest of farmers to expand production of the black velvet tamarind tree.

2.3 Importance of food preservation

The term food preservation refers to any one of a number of techniques used to prevent food from spoiling. Preservation usually involves preventing the growth of bacteria, fungi, and other micro-organisms, as well as retarding the oxidation of fats which cause rancidity. It also includes processes to inhibit natural ageing and discolouration that can occur during food preparation. Some food processing procedures, however, use benign bacteria, yeasts or fungi to add specific qualities and to preserve food (e.g., cheese, wine) (Barbosa-Cánovas *et. al.*, 2003).

Food preservation processes commonly employed are heating to kill or denature micro-organisms (e.g. boiling), oxidation (e.g. use of sulphur dioxide), dehydration (drying), low temperature inactivation (e.g. freezing), irradiation, and many combinations of these methods.

Some preservation methods require the food to be sealed after treatment to prevent recontamination with microbes. Others, such as drying, allow food to be stored without any special containment for long periods.

Food preservation has become an increasingly important component of the food industry as fewer people eat foods produced on their own lands, and as consumers expect to be able to purchase and consume foods that are "out of season". Storage of fruits is critical in fruit industry to avoid economic losses (Khan *et al.*, 2007).

2.4 Irradiation processing of foods to extend shelf-life

Traditional food processing methods such as drying, fermentation, heating, salting and smoking have been used for centuries to improve the quality, quantity, and safety of food. Newer methods such as heat pasteurization, canning, freezing, refrigeration, fumigation, ultrahigh hydrostatic pressure, electrical conductivity heating, and pulsed electrical fields have been added to the many food processing methods. Each of these methods offers specific advantages in protecting our food supplies against destruction, microbial contamination, and spoilage. None is applicable for all types of foods. Several of these methods can cause significant changes in food quality and sensory attributes (Kava, 2007).

Food irradiation, one of the beneficial applications of atomic energy, is an important innovation in food preservation since the development of canning in the 19th century (Subramanian, 2003). Food irradiation is the process of exposing food to ionizing radiation; either high-energy electrons or X-rays from accelerators, or by gamma rays (emitted from radioactive sources as Cobalt-60 or Caesium-137) (ICGFI, 1991) to destroy microorganisms, bacteria, viruses, or insects that might be present in the food. Further applications of irradiation include sprout inhibition, delay of ripening, increase of juice yield, and improvement of re-hydration (Anon, 1991). Exposure of food material to radiation has strong advantages over conventional methods of preservation such as cold storage, fumigation, salting and drying because it does not lead to loss of flavour, odour, texture or quality (Diehl, 1995). Radiation processing could be used for disinfestation of food grains and pulses; inhibition of sprouting in onions, potatoes, garlic, yam and ginger; microbial decontamination of spices; extending shelf-life under recommended conditions of storage; and overcoming quarantine barriers in international trade (Subramanian, 2003).

The radiation doses applied to a product differs from low doses up to 1kGY (for sprout inhibition, insect disinfestation, delay of physiological processes); medium doses up to 10kGY (extension of shelf life and elimination of microorganisms and viruses and improving technological properties of foods etc.) and high doses of between 10 to 50kGY (industrial sterilization and decontamination of food additives and ingredients).

The reasons associated with most decontamination methods aimed at prolonging the shelf-life of foodstuffs encompass five different approaches, namely:

- Reducing the risk of food-borne infections and intoxications.

- Decreasing microbial spoilage.
- Preserving fresh attributes.
- Preserving nutritional quality.
- Avoiding presence of unacceptable levels of toxic residues or formation of unacceptable levels of toxic by-products.

An overwhelming body of scientific data indicates that irradiated food is safe, nutritious, and wholesome (ICGFI, 1991; Diehl, 1995). Health authorities worldwide, including leading national and international scientific organizations, have based their approvals of food irradiation on the results of sound scientific research. Irradiation increases the safety profile and the availability of a variety of foods. The safety of food irradiation has been studied more extensively than that of any other food preservation process and the pertinent literature is replete with examples. The storage or packaging material after irradiation is important in maintaining the integrity of the product from recontamination after irradiation treatment.

2.5 Food packaging

Packaging is used for several purposes including protection of food from contamination and environmental damage and these facilitate transport, define the quantity of food as well as provide information for consumers (Soroka, 2000).

To prevent recontamination, food is usually packaged prior to irradiation. Therefore, the effects of radiation on the food-packaging materials must also be considered when evaluating the safety of irradiated foods. Irradiation can cause changes to the packaging that might affect integrity as a barrier to microbial contamination.

Irradiation might also produce radiolytic products that could migrate into food, affecting odour, taste, and possibly the safety of the food.

A few examples of packaging to improve marketability of products may suffice. For example, the physicochemical characteristics of Persimmon fruits (*Diospyros kaki*) stored in perforated wooden boxes lined with tissue paper, wax paper and polyethylene were reported to be maintained best in polyethylene film whereas other living material did not check shriveling to excessive weight loss (Farooqi *et al.*, 1975). Polyethylene film packaging has been reported to greatly reduce fruit weight loss under uncontrolled room conditions (Golomb *et al.*, 1984). Similarly, packaging storage maintained good organoleptic properties of pear stored in a modified atmosphere (Kolev, 1977). A similar effect was observed for other physico-chemical properties like ascorbic acid and citric acid contents (Ahmed, 1979). Thus critical evaluation of different packaging materials for storage stability of fruits, vegetables, cereals, fish, fish products, meat, etc. is crucial to avoid post harvest losses. This forms part of the study reported in this thesis.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Questionnaire Administration

Sampling questionnaires (Appendix 1) were administered to 55 market women involved in the trade of *Dialium guineense* in five of the ten regions of Ghana (Table 2). There was a grouping response to the questions since most of them had very low educational backgrounds.

Table 2: Specific markets in which questionnaire was administered

Region	Market (s)
Greater Accra	Makola, Kaneshie, Agbogbloshie, Madina
Eastern	Aburi, Adawso, Koforidua, Agomenya
Central	Winneba, Kotokraba, Swedru
Volta	Ho, Akatsi, Abor
Ashanti	Asafo, Kejetia, Kumasi Central market

3.2 Sample Collection

D. guineense fruits were bought from the Akatsi market in the Akatsi District in the south eastern part of the Volta Region of Ghana. This location was chosen because from a questionnaire administered to people involved in the *D. guineense* trade, Akatsi was identified as the major source of the fruits. After the collection, the fruit samples were taken to the Ghana Herbarium GC, Department of Botany, University

of Ghana for identification. All fruits used in this research were ripe and without any visibly identifiable damage or blemish.

3.3 Physical Characteristics, Packaging, Irradiation and Storage of Black Velvet Tamarind fruits

The diameter of the whole velvet tamarind pod was measured using a digital vernier calliper (Mitutoyo, model 500-161U) prior to packaging. The weight of the whole pods and the separated portions (shell, pulp and seed) were also measured. The whole fruits were then packaged in three (3) different packaging materials. These were woven polypropylene sacks, medium-density polyethylene sacks and jute sacks. The packaged fruits were irradiated at the Radiation Technology Centre, Biotechnology and Nuclear Agriculture Research Institute (BNARI) of the Ghana Atomic Energy Commission (GAEC). Samples were irradiated at 2, 4, 6, 8 or 10kGY. These high doses were chosen to ensure effective decontamination against resident moulds. A control experiment was also performed using unirradiated fruits. After irradiation, both the irradiated and unirradiated fruits were stored at ambient temperature (29 ± 1 °C) for three months in the laboratory of the Department of Food Science and Radiation Processing, BNARI-GAEC. Analyses were conducted on the pulp of the fruit after the fruit coat had been removed.

3.4.1 Determination of percentage moisture content of the pulp

The percentage moisture content of both irradiated and non-irradiated (control) black velvet tamarind pulp was determined according to the method of AOAC (1984) on a monthly basis. Five grams (5g) of pulp was accurately weighed into cooled Petri dishes previously heated (130 ± 3 °C). The contents were dried for one (1) hour in an

oven provided with opening for ventilation and maintained at $130 \pm 3^\circ\text{C}$ (1 hour drying period begun when oven temperature was actually 130°C). The dish was covered while still in the oven, transferred to a desiccator, and weighed soon after reaching room temperature. The percentage moisture content was calculated using the formula:

$$\% \text{Moisture} = \frac{M_{(INITIAL)} - M_{(DRIED)}}{M_{(INITIAL)}} \times 100$$

where $M_{INITIAL}$ and M_{DRIED} are the mass of the sample before and after drying, respectively.

3.4.2 Determination of water sorption isotherm of the pulp and the whole fruit under varying equilibrium relative humidities (ERH's) at 30°C

The moisture sorption and desorption under varying ERH's (20, 55, 65, 75, 85, 95%) of unirradiated *D. guineense* was determined by weighing two (2) grams of the pulp and whole fruit and placing it in desiccators containing glycerol : water mixture in a ratio shown in Table 3. The percentage Equilibrium Relative Humidity (ERH) was obtained when the right volume of water was added to glycerol as indicated in Table 3. The upper section of each glass desiccator contained the pulp and whole fruit samples while the lower section contained the formulation of the glycerol : water mixture. The interior the desiccators had a temperature of $26 \pm 1^\circ\text{C}$. The whole setup was kept at room temperature ($30 \pm 1^\circ\text{C}$).

The weight (loss or gain) of the samples was determined every 2 days for 30 days by weighing the samples and a graph drawn of the values (Figs 12 and 13).

TABLE 3: Formulation of glycerol : water mixtures used to establish Equilibrium Relative Humidity in the desiccators

% ERH	Volume of glycerol (ml)	Volume of water (ml)
20	94	6
55	75	25
65	68	32
75	58	42
85	45	55
95	22	78



Plate 3: Photograph showing the arrangement of the black velvet tamarind inside glass desiccators (Note: The glycerol: water mixtures simulating the prescribed ERH's were poured into the bottom of the container) (X 1/100)



3.4.3 Determination of pH of pulp

The pH of the pulp of *D. guineense* was determined every month according to the method of AOAC (1984). Two (2) grams of both irradiated and unirradiated pulp samples were homogenized with 10ml distilled water in a Waring Blender (Model 35BL59, USA) for 15 seconds. Each homogenate was then filtered through a filter paper (Whatman No. 1). The pH of the filtrate was measured using a standard pH meter (Hanna Instruments, Model pH 211, Romania).

3.4.4 Determination of total titratable acidity of pulp

The total titratable acidity of the pulp was determined by using the method described in AOAC (1984). Ten (10) grams of both irradiated and unirradiated black velvet tamarind pulp was homogenized with 100ml of distilled water in a Waring Blender (Model 35BL59, USA) for 15 seconds. The homogenate was then centrifuged at 4000rpm for five (5) minutes. The total titratable acidity was determined by titrating 20ml of the aqueous extract of the black velvet tamarind pulp with 0.1M NaOH using 0.3ml of phenolphthalein as indicator.

3.4.5 Determination of ash content of pulp

Five (5) grams of the pulp of both irradiated and unirradiated *D. guineense* was weighed into shallow crucibles and ignited in a furnace (Carbolite Eurotherm CWF 12/13 with maximum temperature of 1200°C) at 600°C for five (5) hours. The crucibles were removed from the oven and cooled in a desiccator. Soon after

reaching room temperature, the crucibles and its contents were weighed (AOAC, 1990).

3.4.6 Determination of reducing sugar content of pulp

The reducing sugar content in the irradiated and unirradiated black velvet tamarind pulp was determined using the Lane-Eynon method (Anon, 2005). Ten (10) grams of both irradiated and unirradiated black velvet tamarind pulp was homogenized with 100ml of distilled water in a Waring Blender (Model 35BL59, USA) for 15 seconds. The homogenate was then centrifuged at 4000rpm for five (5) minutes. Twenty-five (25) ml of mixed Fehling's solution was pipetted into 250ml volumetric flask. The flask was swirled while heating over a Bunsen flame. The mixed Fehling's solution was boiled moderately for 1.5 to 2 minutes and two (2) drops of methylene blue solution was quickly added. Immediately the homogenate solution of the black velvet tamarind was added via a burette at a rate of about 0.25ml per 15 seconds. The titration was completed within 3 minutes from the time boiling commenced.

$$\text{Percentage of reducing sugars (calculated as sucrose)} = \frac{100 \times A \times a}{P \times V}$$

Where (A = volume (mL) of solution of prepared] of sample; P = weight of sample used (g); V = volume (mL) of sample aqueous solution used in titration; a = amount of sucrose equivalent to 20 mL Fehling's solutions).

3.4.7 Determination of crude fat content of pulp

A nuclear magnetic resonance (NMR) instrument (Oxford Instruments, U.K) was used. The instrument was first autotuned using acetone. The pulp of both the irradiated and unirradiated black velvet tamarind was then poured into a tarred glass tube up to the 4cm mark. The sample mass was then recorded and the tube containing the sample was then transferred into the NMR instrument. The reading of the fat content (%) of the sample was recorded.

3.4.8 Determination of crude protein content of pulp

The crude protein content was determined according to the micro-Kjeldhal method. One (1) gram of both the irradiated and unirradiated black velvet tamarind was weighed into a digestion flask. Two drops of N- catalyst ($K_2SO_4 + G Se$) and 20ml of H_2SO_4 were added, and the solution was boiled briskly for about 2 hours until the solution became clear. The solution was allowed to cool and the volume made up to 100ml by adding distilled water in a volumetric flask. Aliquot solution of 10ml was taken into flat bottom flask and about 15ml of 40 % NaOH was added to make the content strongly alkaline. The content was distilled against 10ml of 2 % boric acid solution containing 2-3 drops of mixed indicator (methyl red and bromocresol green) in a flask. About 50ml of the distillate was collected into the receiving solution after which 0.01N HCl was titrated against it. A blank determination was also done on the reagents. The percent nitrogen and the crude protein content were calculated as follows:

$$\%N = \frac{Vol.(Acid) - Vol.(Blank)}{1g} \times \frac{0.01}{100} \times 14 \times \frac{100}{10} \times 100$$

$$\% Protein = \% N \times 6.25$$

3.4.9 Mycoflora of whole and dehulled fruit before and after irradiation and storage in different packaging materials

Exactly 10g of either pulp, whole fruit, and peel (respectively) were added separately to 100ml of 0.1% peptone in 250ml Erlenmeyer flask. The decimal serial dilution technique was employed up to $1:10^{-3}$. Exactly 10ml of dilution levels were placed in 20ml of different media: Potato Dextrose Agar (PDA), Oxytetracycline Glucose Yeast Agar (OGYE) and Dichloran-Glycerin (DG-18) Agar. The samples were incubated upside down, and left undisturbed in the dark at 25°C for five (5) days. If there was no growth after 5 days of incubation, samples were left intact until growth occurred. Moulds appearing were identified using standard identification manuals (Samson and Reenen-Hoekstra, 1988; Smith, 1960). Identification was confirmed by my supervisor.

3.4.10 Determination of elemental composition of *Dialium guineense*

Instrumental Neutron Activation Analysis (INAA) was used to determine the elemental composition of the unirradiated fruit coat (shell), pulp and seed. The fruit coat and seed samples were ground into a fine powder and the pulp was dried at $103\pm 3^{\circ}\text{C}$ in an oven provided with an opening for ventilation before grinding into a fine powder. Exactly 0.20 g each of the powdered samples was weighed into pre-cleaned irradiation plastic vials. Standard reference materials, namely, 1547 peach leaves and certified reference materials (CRM) were used to validate the concentrations of the elements identified in the samples.

The samples in the plastic vials were irradiated using the Ghana Research Reactor-1 (GHARR-1) which was operated at half power (15.0 kW) with a thermal neutron

flux of $5.0 \times 10^{11} \text{ cm}^{-2} \text{ s}^{-1}$. The irradiation time ranged from 2 minutes to 3 hours depending on the half life of the elements of interest. For short-lived radionuclides, the ground fruit coat (shell), pulp and seed were irradiated for 2 minutes and counted for 10 minutes using thermal neutrons detectors. For medium lived radionuclides, the ground fruit coat (shell), pulp and seed were irradiated for 1 hour with 1 to 2 days decay time and 10 minutes counting time. The detector used in counting was an n-type high purity germanium (Model GR 2518-7500sl, USA). The peak area of the γ -spectrum for samples and standards were evaluated using Windows based software (ORTEC MAESTRO-32). The areas under the peaks were integrated and converted into concentration ($\mu\text{g/g}$) using the relative method of standardization for instrumental neutron activation analysis. Table 4 shows the Nuclear Data (IAEA TECDOC-564) for the elements of interest (IAEA, 1990).



Table 4: Nuclear data (IAEA-TECDOC-564)

Element	Isotope (% Abundance)	Nuclide	Cross-section (barn)	Half-life	γ -Ray energy used (keV)
Al	²⁷ Al (100)	²⁸ Al	0.232	2.241min	1,779.0
Br	⁷⁹ Br (50.69)	⁸⁰ Br	148±4	17.68min	616.8
Ca	⁴⁸ Ca (0.187)	⁴⁹ Ca	1.1±0.2	8.72min	3,084.4
Cl	³⁷ Cl (24.23)	³⁸ Cl	0.428±0.005	37.2min	1,642.7
Co	⁵⁹ Co (100)	⁶⁰ Co	17±2	5.272years	1,173.5, 1,332.5
Cu	⁶⁵ Cu (30.9)	⁶⁶ Cu	2.17±0.03	5.09min	1,039.2
Fe	⁵⁸ Fe (0.28)	⁵⁹ Fe	1.15±0.02	44.5days	192.3, 1,099.3
I	¹²⁷ I	¹²⁸ I	6.2±0.2	25.0min	442.9
K	⁴¹ K (6.73)	⁴² K	1.46±0.03	12.38h	442.9, 1,524.7
Mg	²⁶ Mg (11.01)	²⁷ Mg	0.0382±0.001	9.46min	843.8, 1,014.4
Mn	⁵⁵ Mn (100)	⁵⁶ Mn	13.3±0.2	2.58h	846.8
Na	²³ Na (100)	²⁴ Na	0.530±0.005	14.95h	1,368.6, 2,754.0
V	⁵¹ V (99.75)	⁵² V	4.88±0.04	3.743min	1,434.1

3.5 Hidden insect infestation of fruits

Infested fruits containing the larvae of the insect were put into Kilner jars and allowed to pupate for three weeks until the adults emerged. The insects that infested the fruits were identified at the Department of Animal Biology and Conservation Science of the University of Ghana by Dr. David Wilson.

3.6 Sensory evaluation

Two sensory evaluation sessions were carried out on the irradiated and non-irradiated tamarind fruit (pulp). The first sensory evaluation session was a quantitative descriptive sensory analysis while the second was a preference test (ISO, 2003) (Appendix 2). In all, 60 panelists (all workers of the Ghana Atomic Energy Commission) were used; thirty (30) in each sensory. Panelists were presented with *D. guineense* fruits of all the different irradiation doses from one packaging material at a time. In the descriptive sensory evaluation, panelists were instructed to draw vertical lines across a horizontal unstructured scale, 9.8cm in length. Anchor points were made after every 1.96cm from the left to the right end of the scale. Panelists were instructed to indicate their like or dislike for each attribute using a nine-point hedonic scale ranging from 9 = like extremely through 5 = neither like nor dislike to 1 = dislike extremely for the preference sensory evaluation. Four different attributes (colour, sweetness, acidity and tenderness) were used to grade the overall acceptance. Water was provided to wash the oral cavity after testing each treatment. The serving order of the panelists was randomized.

For the descriptive sensory evaluation, five-point hedonic scales were assigned for the scales for ease of removal, pulp tenderness and sweetness. For ease of removal,

ticks from 0.00 cm-1.96 cm on the scale was interpreted as very difficult; 1.96cm - 3.92cm as slightly difficult; 3.92cm -5.88cm as neither difficult nor easy; 5.88cm - 7.84cm as slightly easy; and 7.84cm -9.80cm as very easy. For pulp tenderness, ticks from 0.00cm -1.96cm on the scale was interpreted as very firm; 1.96cm -3.92cm, slightly firm; 3.92cm -5.88cm, neither firm nor fluffy; 5.88cm -7.84cm, slightly fluffy; and 7.84cm -9.80cm, very fluffy. For sweetness, ticks from 0cm - 3.27cm on the scale was interpreted as very sweet; 3.27 cm – 6.54 cm, slightly sweet; 6.54 cm - 9.80cm, sweet. Ticks for pulp acidity were assigned as follows; ticks from 0.00cm - 1.96cm on the scale was interpreted as very sour; 1.96cm -3.92cm, slightly sour; 3.92cm -5.88cm, neither sour nor acidic; 5.88cm -7.84cm, slightly acidic; and 7.84cm -9.80cm, acidic. The scales for pulp discolouration 1 and pulp discolouration 2 were divided into two sections. For pulp discolouration 1, ticks between 0.00cm and 4.90cm were interpreted as brick-red while those between 4.91cm and 9.80cm were interpreted as orange. For pulp discolouration 2, ticks between 0cm and 4.9cm were interpreted as brown while those between 4.91cm and 9.80cm were interpreted as orange.

For the preference test, the following interpretations were given to the hedonic scale; 9.00 to 9.99 – like extremely; 8.00 to 8.99 – like very much; 7.00 to 7.99 – like moderately; 6.00 to 6.99 – like slightly; 5.00 to 5.99 – neither like nor dislike; 4.00 to 4.99 – dislike slightly; 3.00 to 3.99 – dislike moderately; 2.00 to 2.99 – dislike very much; 1.00 to 1.99 – dislike extremely.

3.7 Statistical analysis

Data was analysed using STATGRAPHICS Plus software and means were separated using the multiple range test. The chosen level of significance was $P < 0.05$. Graphs were drawn using Microsoft Excel on the Microsoft Office 2003 package.

CHAPTER FOUR

4.0 RESULTS

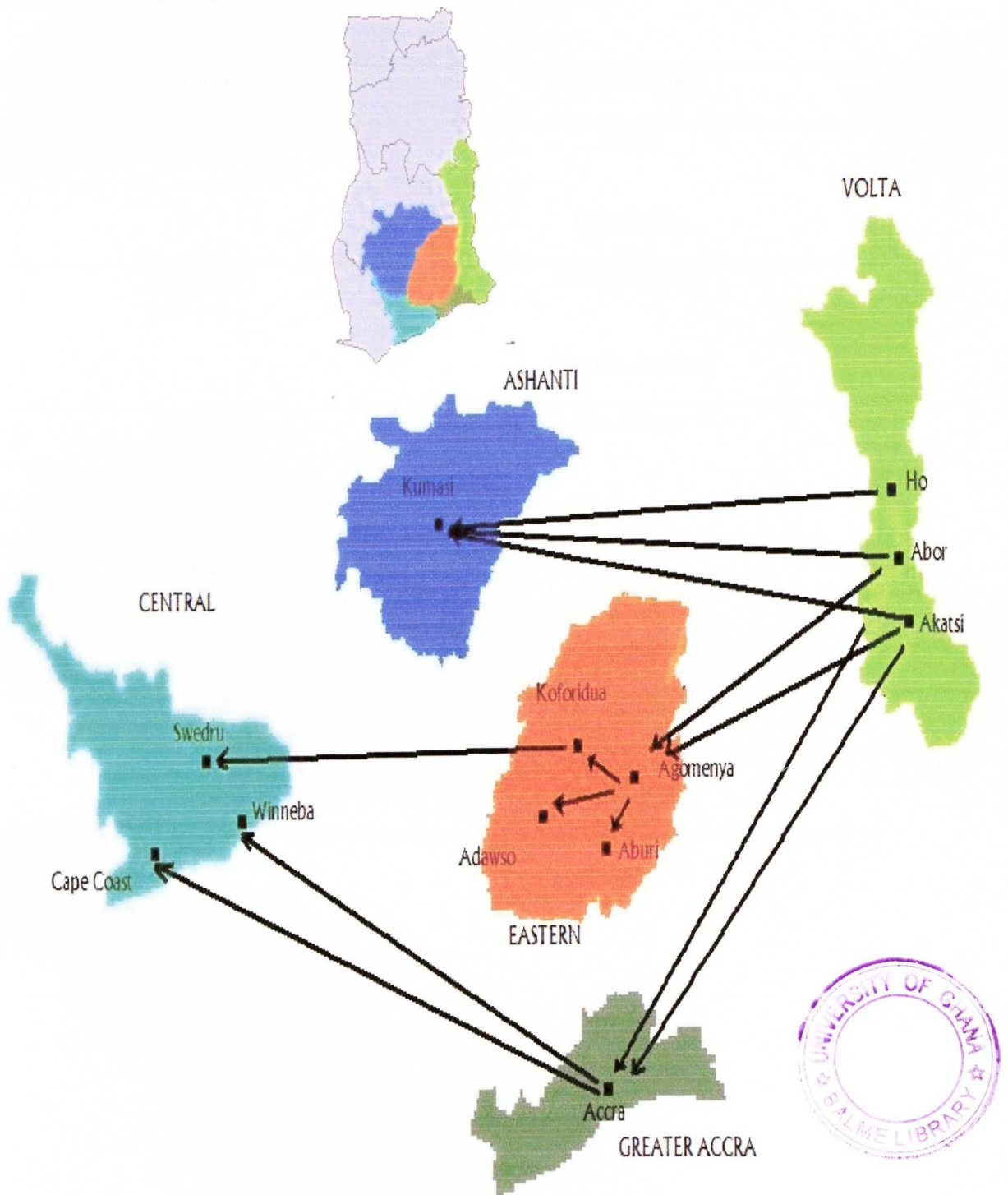
4.1 Rapid appraisal method for assessing and determining sources of *Dialium guineense* on the local markets in Ghana

Questionnaires were administered in five regions of the country, namely Greater Accra, Eastern, Central, Ashanti and Volta. A total of 55 groups of respondents (11 groups in each region) were engaged. A sample of the questionnaire is attached as Appendix 1.

Table 2 shows the specific markets from which fruits of *D. guineense* were purchased. Traders from the Greater Accra Region bought their produce predominantly from the Volta Region (Map 1); those from the Eastern Region purchased *D. guineense* from the Volta Region (Akatsi and Abor) and Agomenya (Eastern Region). Traders from Aburi, Adawso, and Koforidua purchased their velvet tamarind from Agomenya (Map 1).

The local names of the velvet tamarind varied from one region to another as shown in Table 5.

The people who usually patronise the velvet tamarind is shown in Fig. 1. Female adults were those who purchased the fruit most followed by school children and adult males in decreasing order. There was a rapid turnover of the fruit after purchase from the original source. Within 3 weeks, about 200Kg (50Kg x 4 bags) worth of fruit had been sold.



Map 1: Flow of sale of *D. guineense* fruits obtained from markets in the indicated regions of Ghana (Note the movement of produce predominately from Akatsi in the Volta Region)

Table 5: Local names of *D. guineense*

Region	Local name(s)
Greater Accra	Yooyi
	Blackberry
Eastern	Akosua tuntum
	Akosua kokoo
Central	Blackberry
	Asna ba
Volta	Atsitoe
	Atotoe
Ashanti	Akosua tuntum
	Akosua kokoo

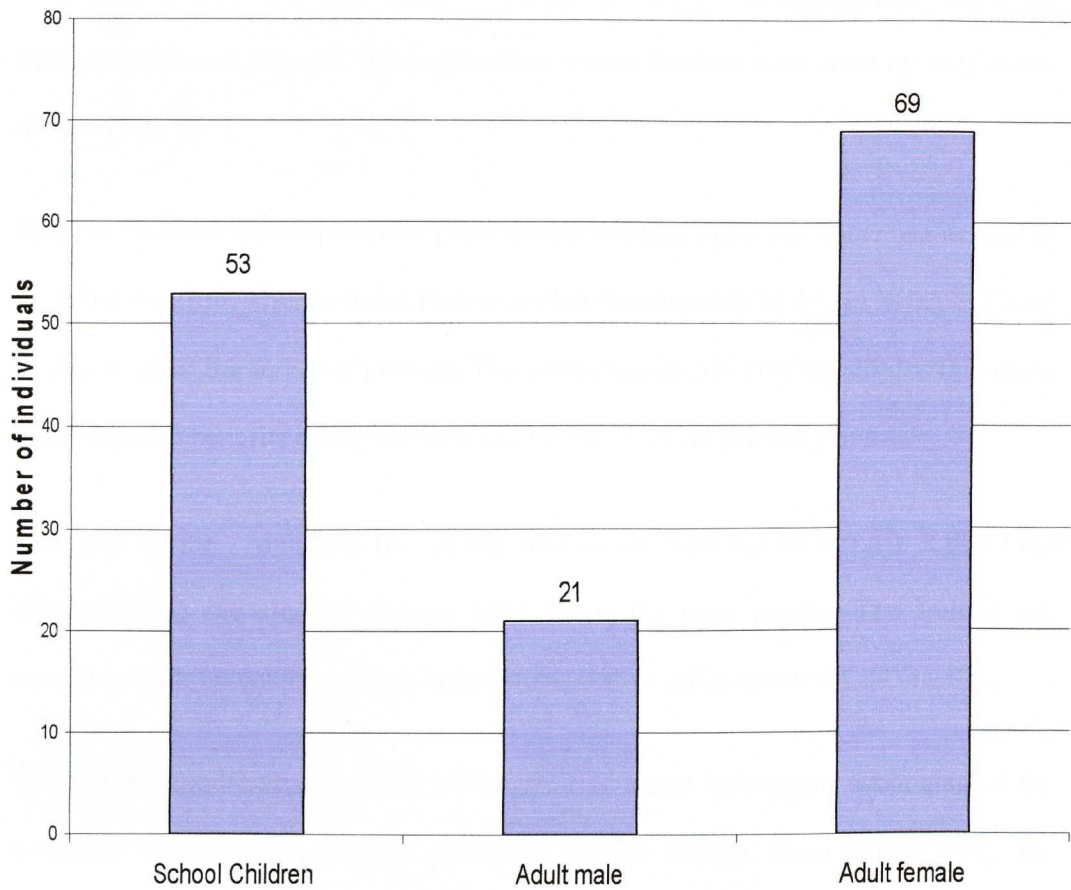


Fig. 1: People who regularly purchase *D. guineense* fruits from the market

The traders preferred packaging the fruit in woven polypropylene sacks for transportation and storage. Cane and rattan woven baskets were used by only a few traders (Fig. 2)

Exactly 74.5% of the respondents (41/55) believed that there was some health benefit in eating the fruit. This included their assertion that the pulp is rich in Vitamin C and is able to clear the throat of phlegm. The remaining 25.5% (14/55) alluded that there was no health benefits eating the fruit except for its sweet sensational taste.

The cost of one “American tin” of the fruit in the lean season is GH¢ 2.00 (Two Ghana Cedis) and slightly cheaper GH¢ 1.5 in the peak season. The fruit is not bought in bulk by consumers but in little handouts of 20Gp to GH¢ 1.00 at a time.

The major post-harvest problem of the fruit is insect infestation accompanied by microbial deterioration. Rodents attack the bags in storage. Even though fungi are ubiquitous and airborne, the traders thought fungi are the least important so far as post-harvest deterioration was concerned. Fig. 3 shows the relative importance of various components of post-harvest losses in the fruits during storage.

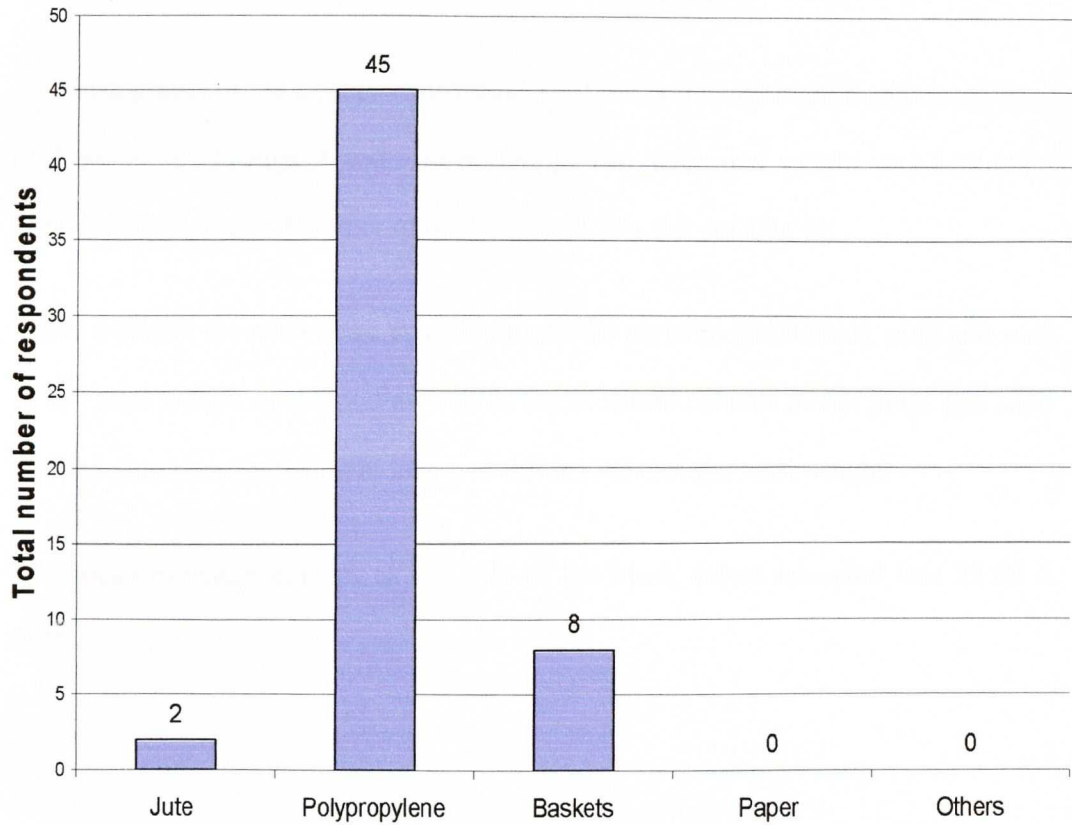


Fig 2: Choice of transporting and storage material for the *D. guineense* fruits

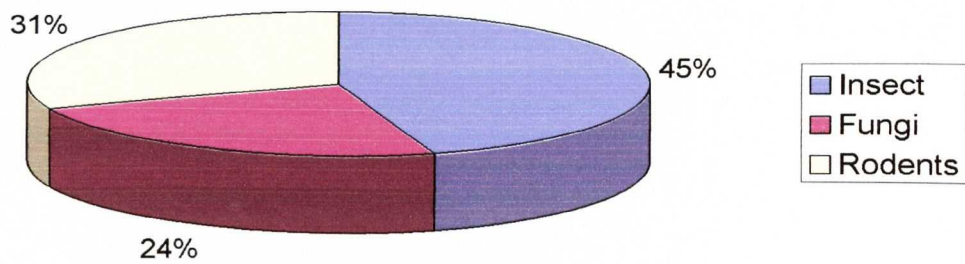


Fig 3: Major sources of post harvest loss in *D. guineense* fruits

4.2 Physical characteristics of the fruit of *D. guineense*

The histograms of the class-size distribution of the pod weight and mean diameter of pod are shown in Figs. 4 and 5 respectively. The mean pod weight was 1.3 ± 0.2 g (Fig 4) and the mean diameter of the pod was 1.70 ± 0.3 cm (Fig 5).

Table 6 shows the percentage contribution of the pericarp (shell/coat), pulp and seed to the total weight of the pod as well as the moisture content of the pulp. The shell and the pulp contributed more than two-thirds (78%) of the total weight.

The mean moisture content of the pulp of the black velvet tamarind was $22.80 \pm 0.8\%$.



Fig 4: Class-size distribution of the pod weight of *D. guineense*

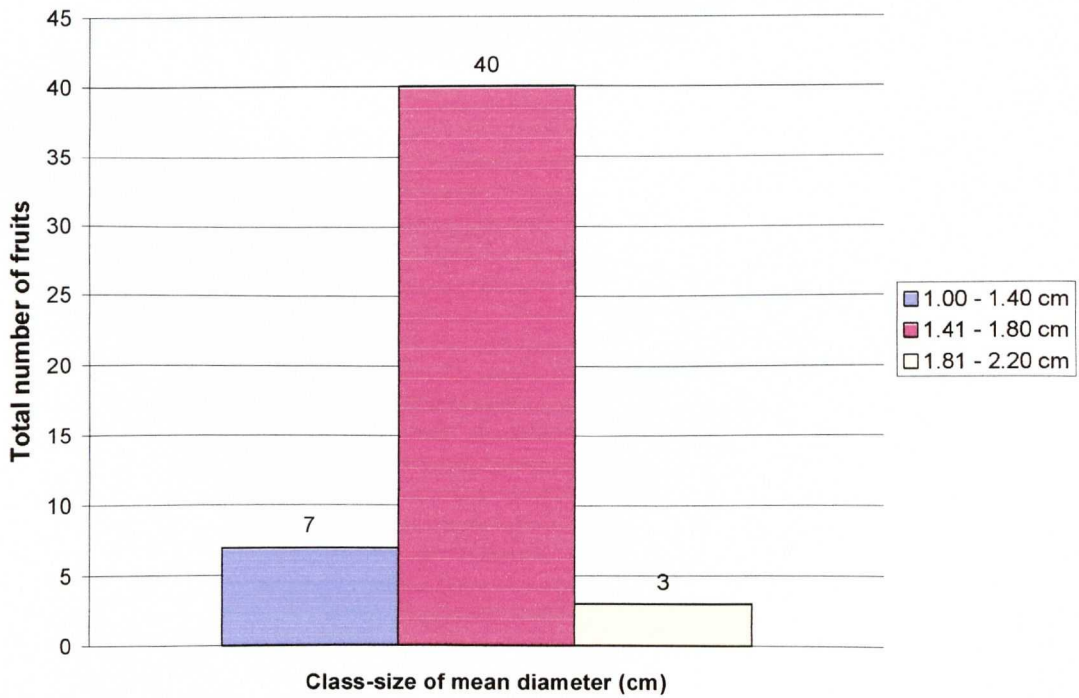


Fig 5: Class-size distribution of the mean diameter of the pod of *D. guineense*

Table 6: Mean values for physical characteristics of *D. guineense* fruit

Fruit part	% of total fruit weight
Shell/Cover	39
Pulp	39
Seed	22
% Moisture Content	22.80 ± 0.8

4.3 Moisture sorption isotherms of whole and de-hulled fruit (pulp) of *D. guineense* at varying equilibrium relative humidities (ERH's)

The set up was as described in the Materials and Methods section and Plate 2. The moisture sorption (at 80% and 95% ERH) and desorption (at 20%, 55% and 65% ERH) isotherms of both the whole fruit (Fig. 6) and the pulp (Fig. 7) followed a near sigmoid curve typical of dehydrated food (Debnatha *et al*, 2002) reaching equilibrium after 4 to 6 days at 20 to 85% ERH in the whole fruit. The moisture content of samples incubated under 95% ERH continued rising (Fig. 6). At ERH's 20, 55 and 65%, there was a decrease (desorption) of moisture to the environment; at 75% ERH, there was hardly any gain or loss in moisture content in both whole fruit and pulp only (Figs. 6 and 7).

The pulp samples incubated at 95% ERH were visibly mouldy after 6 days and were completely colonized by fungi after 10 to 15 days. On the other hand, samples kept at 20 to 75% ERH were apparently devoid of fungal growth.

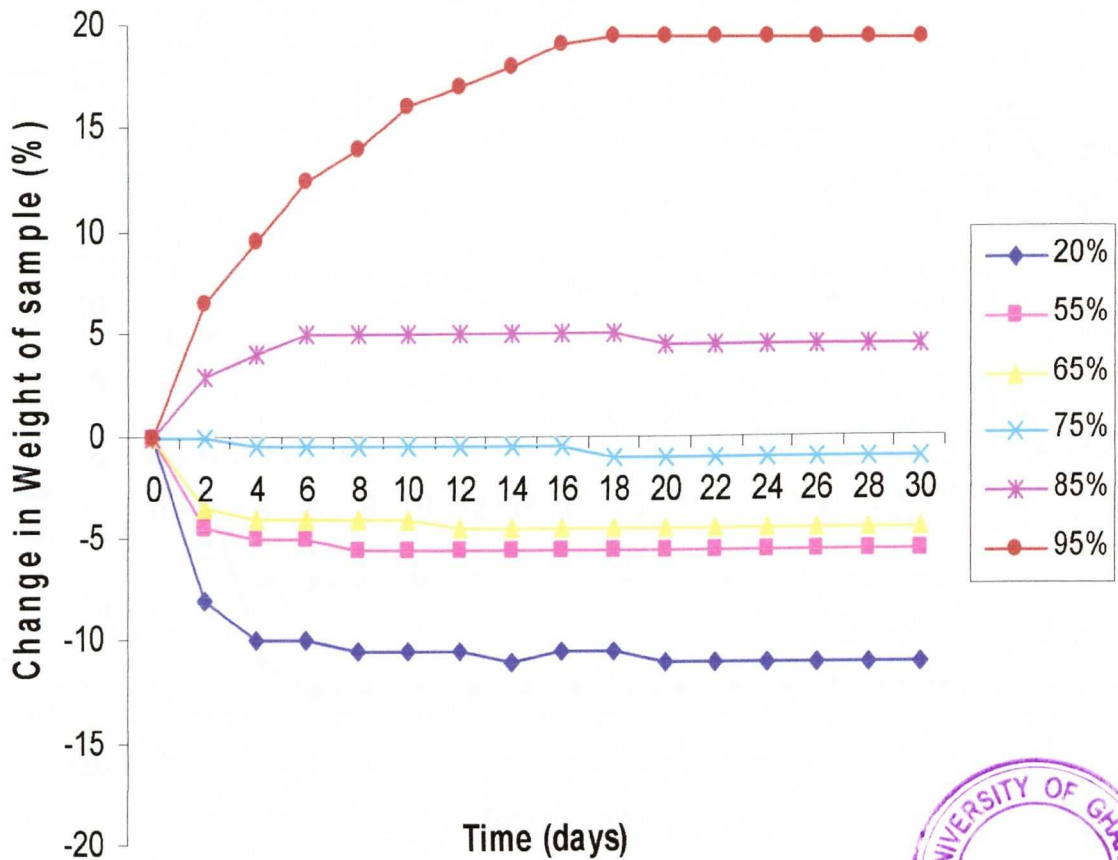


Fig 6: Moisture sorption isotherm of the whole fruit of *Dialium guineense* at different indicated equilibrium relative humidities (ERH's) at $29\pm 1^\circ\text{C}$ for 30 days

(Note the loss in moisture at ERH's 20 to 65%)

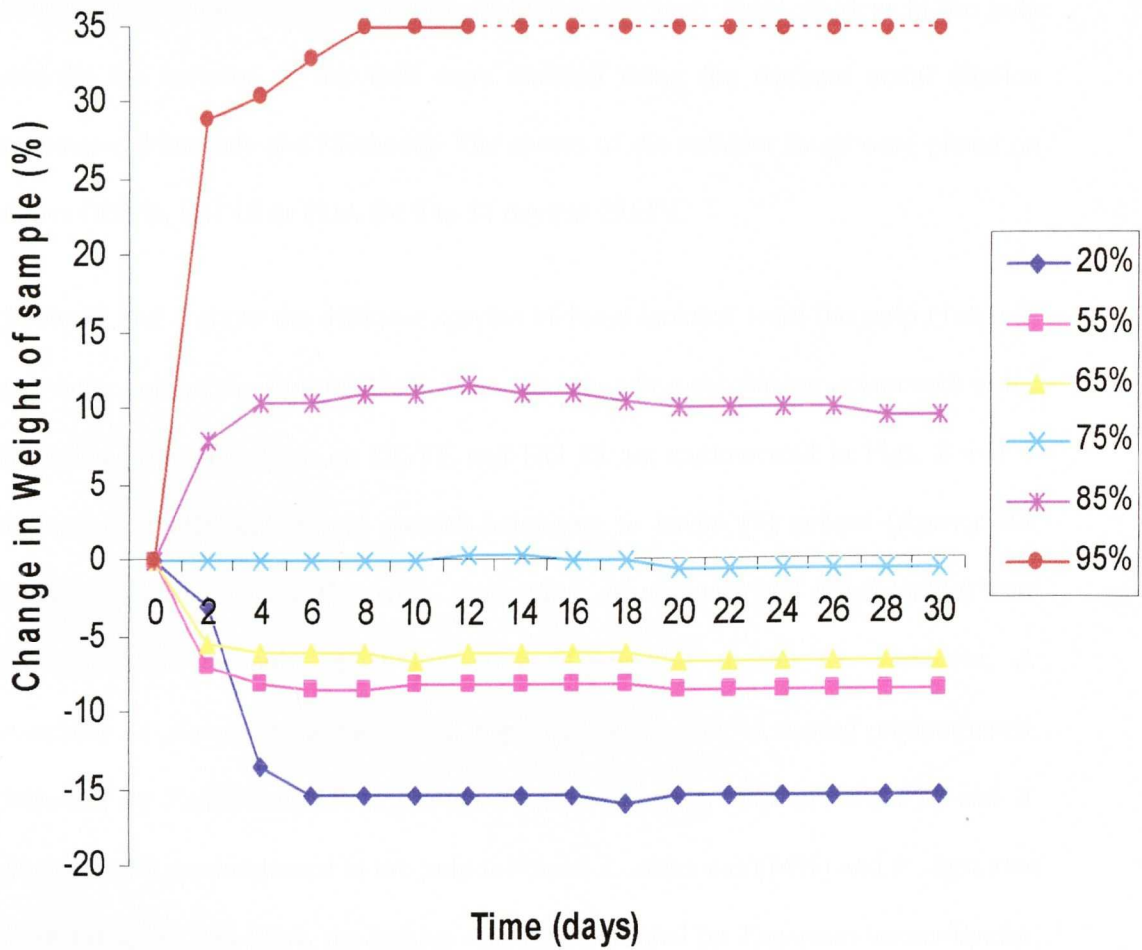


Fig 7: Moisture sorption isotherm of the pulp of *Dialium guineense* at different indicated equilibrium relative humidities (ERH's) at $29\pm 1^\circ\text{C}$ for 30 days. (Note the loss in moisture at ERH's 20 to 65%)

4.4 Mycoflora resident in the fruit of black velvet tamarind (*D. guineense*)

Fungi are ubiquitous and may contaminate products in the field (field fungi) or in storage after harvest (storage fungi). In this experiment, fungi resident in the pulp and on the exterior of the fruit were isolated using the decimal serial dilution technique (Materials and Methods). The spores of the resident fungi were plated on either OGYE, DG 18 or PDA for 7 to 14 days at $29\pm 1^\circ\text{C}$.

Tables 7 and 8 show the different species of fungi isolated from the pulp (Table 7) and outer coat of the fruit (Table 8). The corresponding percentage occurrence of the individual fungal species on OGYE and DG 18 are summarized in Figs. 8 and 9. Fifteen (15) different fungal species belonging to seven (7) genera (*Aspergillus*, *Candida*, *Cladosporium*, *Fusarium*, *Penicillium*, *Neurospora* and *Rhodotorula*) were isolated from the pulp of *D. guineense*. *Aspergillus* species (*A. alutaceus*, *A. candidus*, *A. flavus*, *A. fumigatus*, *A. niger*, *A. sulphureus*, *A. ustus*) predominated, followed by *Penicillium* (*P. digitatum*, *P. expansum*). *A. sulphureus* (25%) and *A. flavus* (20%) predominated in the pulp followed *A. alutaceus* (14%) and *P. digitatum* (14%) (Fig. 8). On PDA, the culture was predominated by *Fusarium verticillioides*, *Candida albicans* and *Rhodotorula* (Table 7).

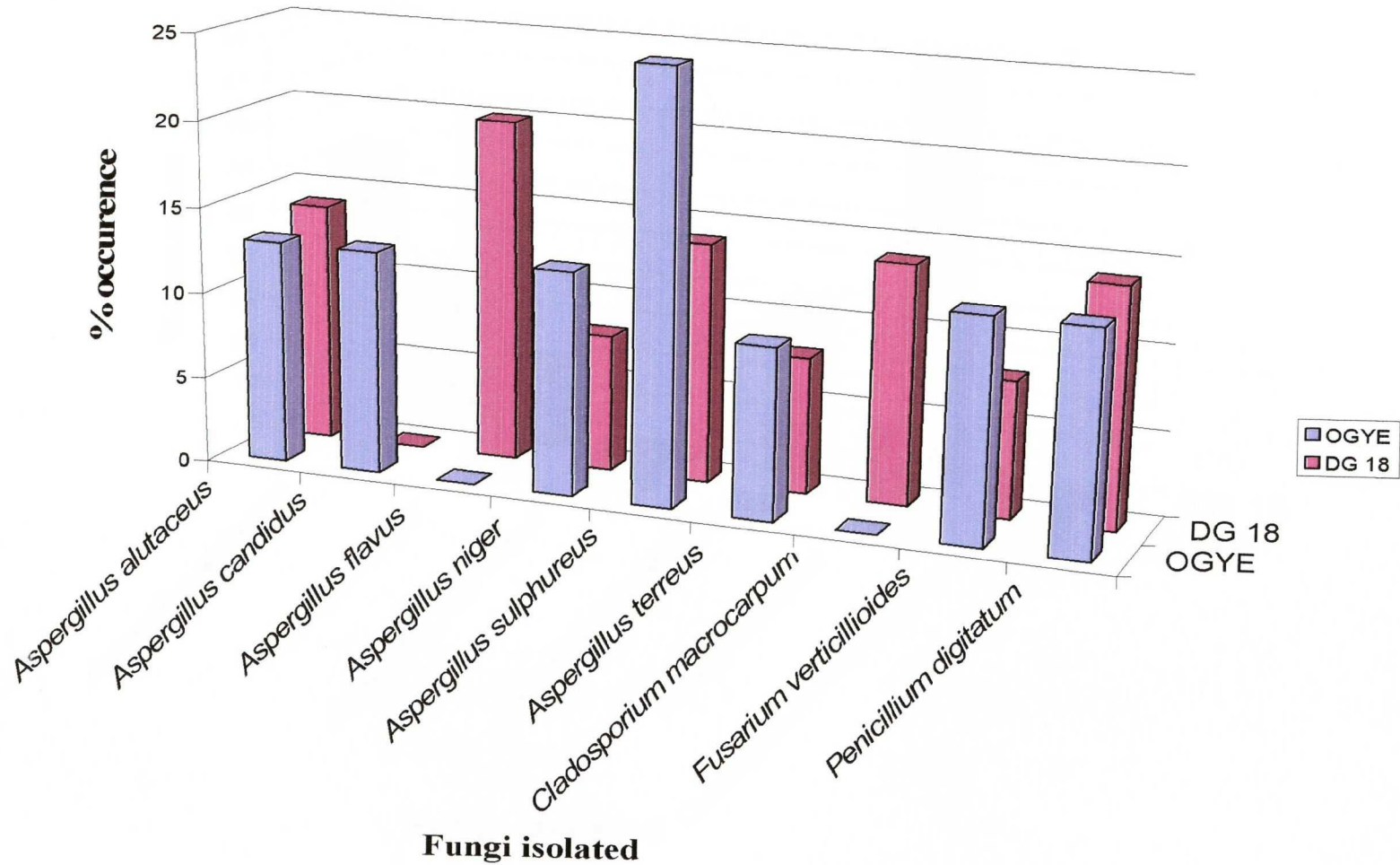


Fig. 8: Percentage occurrence of selected fungi in the pulp of the fruits cultured on either DG 18 or OGYE at $28\pm 1^\circ\text{C}$ for 7 days

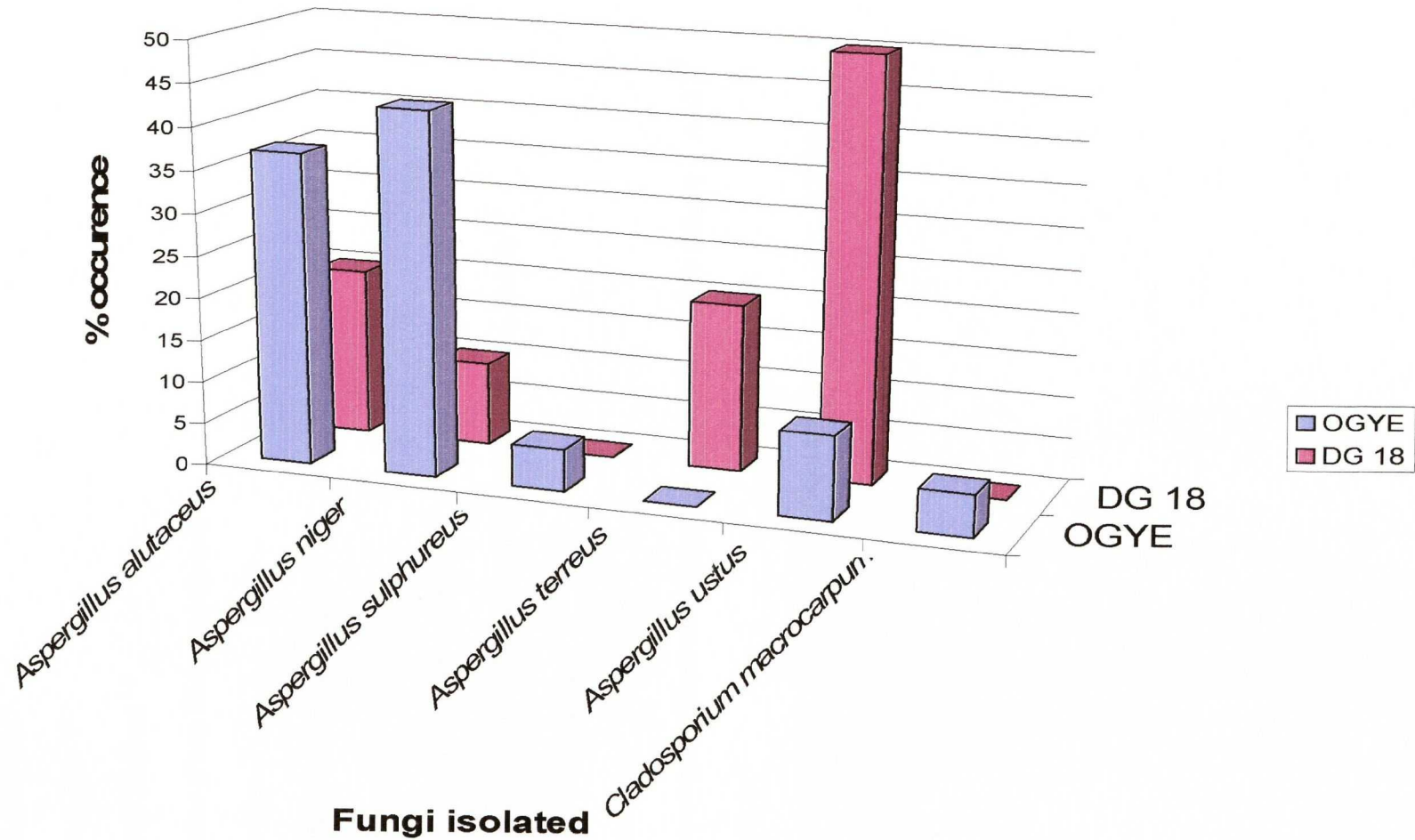


Fig. 9: Percentage occurrence of selected fungi in the outer coat of the fruits cultured on either DG 18 or OGYE at $28\pm 1^\circ\text{C}$ for 7 days

Table 7: Total list of resident fungi isolated from pulp of *D. guineense* at 28±1°C

Aspergillus alutaceus (= *A. ochraceus*) Wilhelm ^{1,2}

A. candidus Link ^{1,2}

A. flavus Lock ²

A. fumigatus Fres ^{1,2}

A. niger van Tieghem ^{1,2}

A. sulphureus (Fres) Thom and Church ¹

A. terreus Thom ²

A. ustus (Bain) Thom and Church ^{1,2}

Candida albicans ³

Cladosporium macrocarpum Preuss ²

Fusarium verticillioides (Sacc) Nierenb (= *F. moniliforme* Sheld) ^{2,3}

Penicillium digitatum Sacc ^{1,2}

P. expansum Link ¹

Neurospora sitophila Shear and Dodge (= *Chrysolinia sitophila* (Mont) v. Arx) ^{1,2}

Rhodotorula sp ³

1: OGYE

2: DG 18

3: PDA

Table 8: Total list of resident fungi isolated from the outer covering of *D. guineense* at 28±1°C

Aspergillus alutaceus (= *A. ochraceus*) Wilhelm ^{1,2}

A. niger van Tieghem ^{1,2,3}

A. sulphureus (Fres) Thom and Church ¹

A. terreus Thom ²

A. ustus (Bain) Thom and Church ^{1,2}

Candida albicans ³

Cladosporium macrocarpum Preuss ²

Rhodotorula sp ³

1: OGYE

2: DG 18

3: PDA

The outer coat of the fruit harboured only eight (8) fungi belonging to four genera (*Aspergillus*, *Cladosporium*, *Candida*, *Rhodotorula*). *Aspergillus* species (*A. niger*, *A. sulphureus*, *A. ustus*, *A. terreus*) were the most abundant. Two yeast species (*Candida albicans* and *Rhodotorula sp*) were isolated on PDA only and were the most predominant.

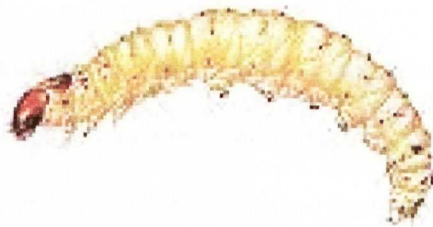
The percentage occurrence of the individual fungal species isolated from the fruit coat is presented in Fig. 9. *A. ustus* (50%) was the most abundant followed by *A. niger* (43%) and *A. alutaceus* (37%). On PDA, *Rhodotorula sp* and *Candida albicans* were the most predominant ($\leq 50\%$).

4.5 Hidden insect infestation of fruits

The Kilner Jar containing 100g of samples kept for three (3) weeks showed signs of the presence of some insect larvae which with time metamorphosed into the adult stage. The insect that infested the fruits of *D. guineense* was identified as *Ephestia* (= *Cudra*) *cautella* (Walker) (Lepidoptera: Pyralidae) (Plate 4).



(A)



(B)

Plate 4: Adult (A) and larva (B) of *E. cautella* (Walker) (X 10)

4.6 Influence of gamma irradiation and packaging on some mycological quality of the pulp of *D. guineense* stored at $29\pm 1^{\circ}\text{C}$ for 3 months

The fruit of the plant is laden with spores of resident fungi belonging to seven genera (*Aspergillus*, *Candida*, *Cladosporium*, *Fusarium*, *Penicillium*, *Neurospora* and *Rhodotorula*) with potential mycotoxin-producing species such as *A. flavus* (Aflatoxins), *A. alutaceus* (Ochratoxin), *F. verticillioides* (Fumonisin), *Penicillium digitatum* and *P. expansum* (Patulin) being present.

The fruits were placed in different packaging sacks (medium density polyethylene, woven polypropylene and jute) and then treated with gamma irradiation at 0, 2, 4, 6, 8, and 10kGY.

Figs. 10 and 11 show the changes in the total number of resident fungi in the pulp before and after gamma irradiation and storage. There was a corresponding inversely proportional decrease in the fungal count as the dose increased (Fig. 10). The reduction in mycoflora was higher in jute than in polyethylene and polypropylene sacks.

The reduction in mycoflora in terms of the fungal species isolated was counted. A dose of 10kGY reduced the number and percentage of individual fungal species encountered (Figs. 12 to 17) initially and after 3 months storage. For example, a dose of 10kGY reduced the fungal colonies on jute sack by 1642% immediately after irradiation while the same dose decreased fungal colonies of produce in polyethylene and polypropylene by 1653% and 347% respectively. There was a commensurate reduction in fungal colonies resident in the fruit as the dose increased from 2 to 8kGY (Fig 10 and 11).

The fungal species succession immediately after irradiation and after 3 months storage are summarized in Figs. 12 to 17. Generally, the resident fungi in the packaging materials varied from one sack to another but were generally predominated by members of the genus *Aspergillus*. Although increasing gamma irradiation up to 10kGY reduced the spectrum of fungal species encountered in the sacks, there was no guarantee that storage under ambient laboratory conditions would not preclude further external contamination of pulp. Thus, in some samples, *A. flavus*, *A. niger*, *A. fumigatus*, *F. verticillioides* and *P. expansum* could still be isolated at 10kGY of gamma irradiation after 3 months storage (Figs. 12 to 17).

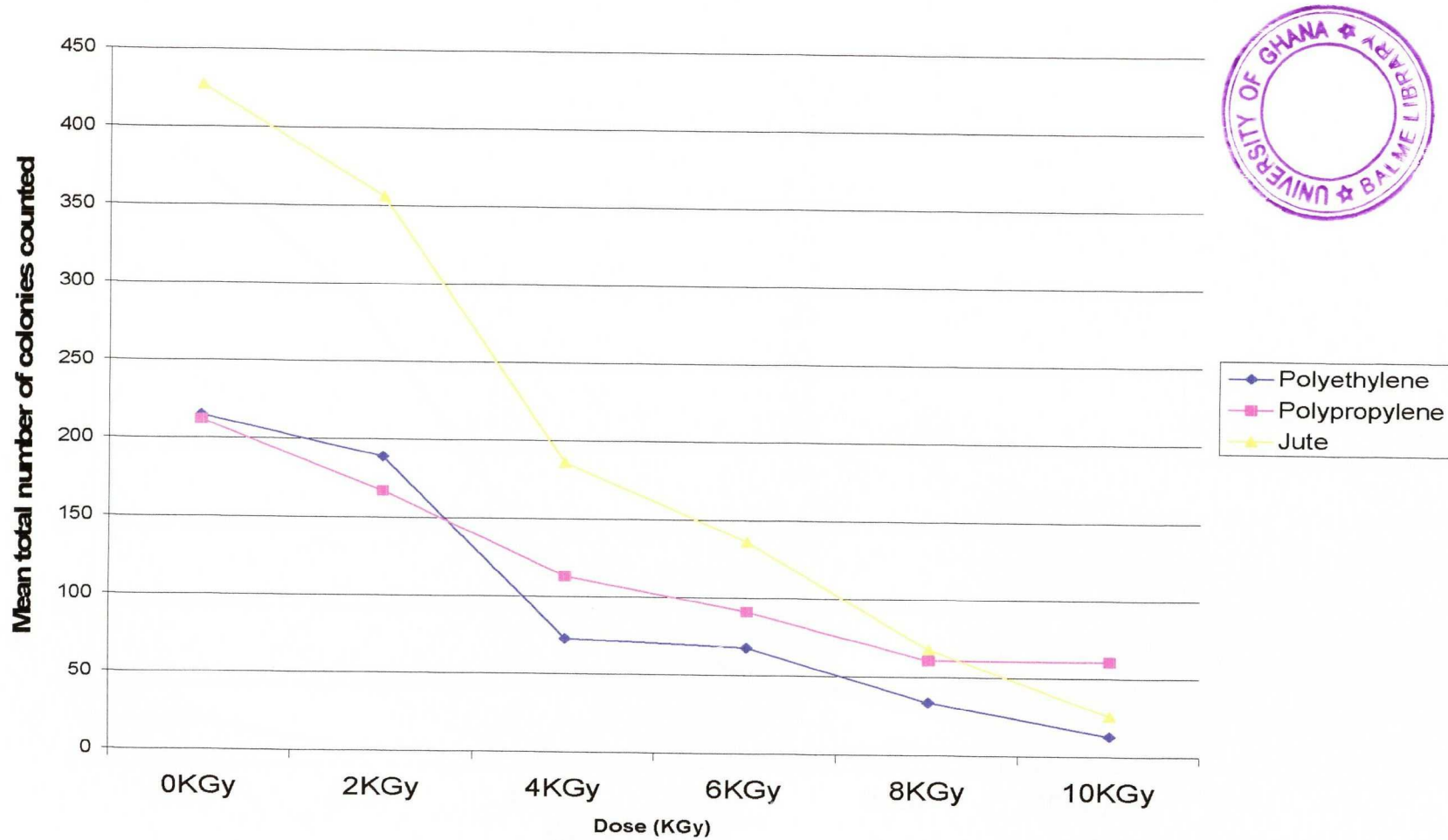


Fig. 10: Changes in the total number of resident fungi in the pulp immediately after gamma irradiation



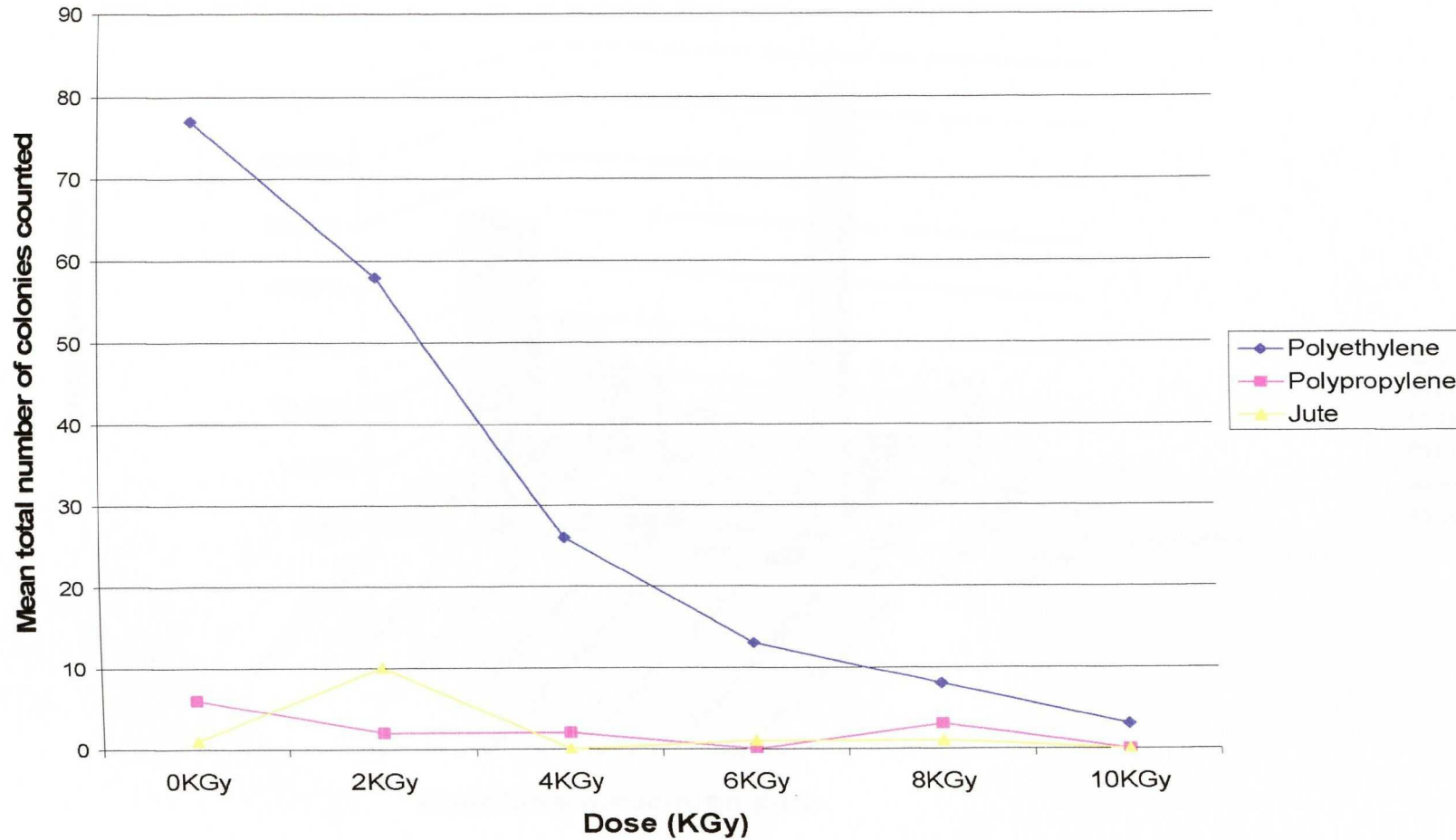


Fig. 11: Changes in the total number of resident fungi in the pulp three months after gamma irradiation

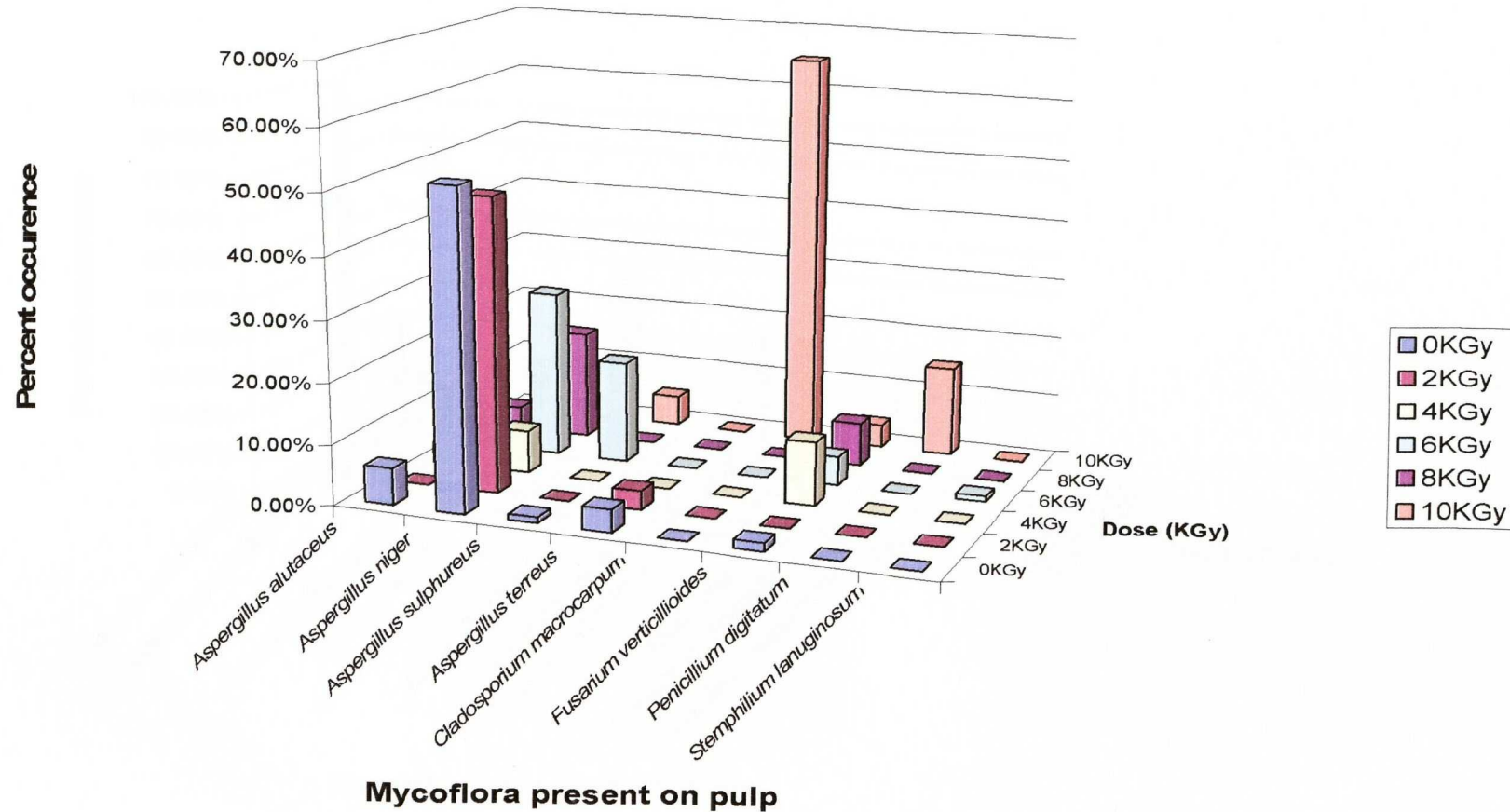


Fig 12: Percent occurrence of mycoflora on the pulp of *D. guineense* stored in polyethylene bags immediately after irradiation with indicated doses

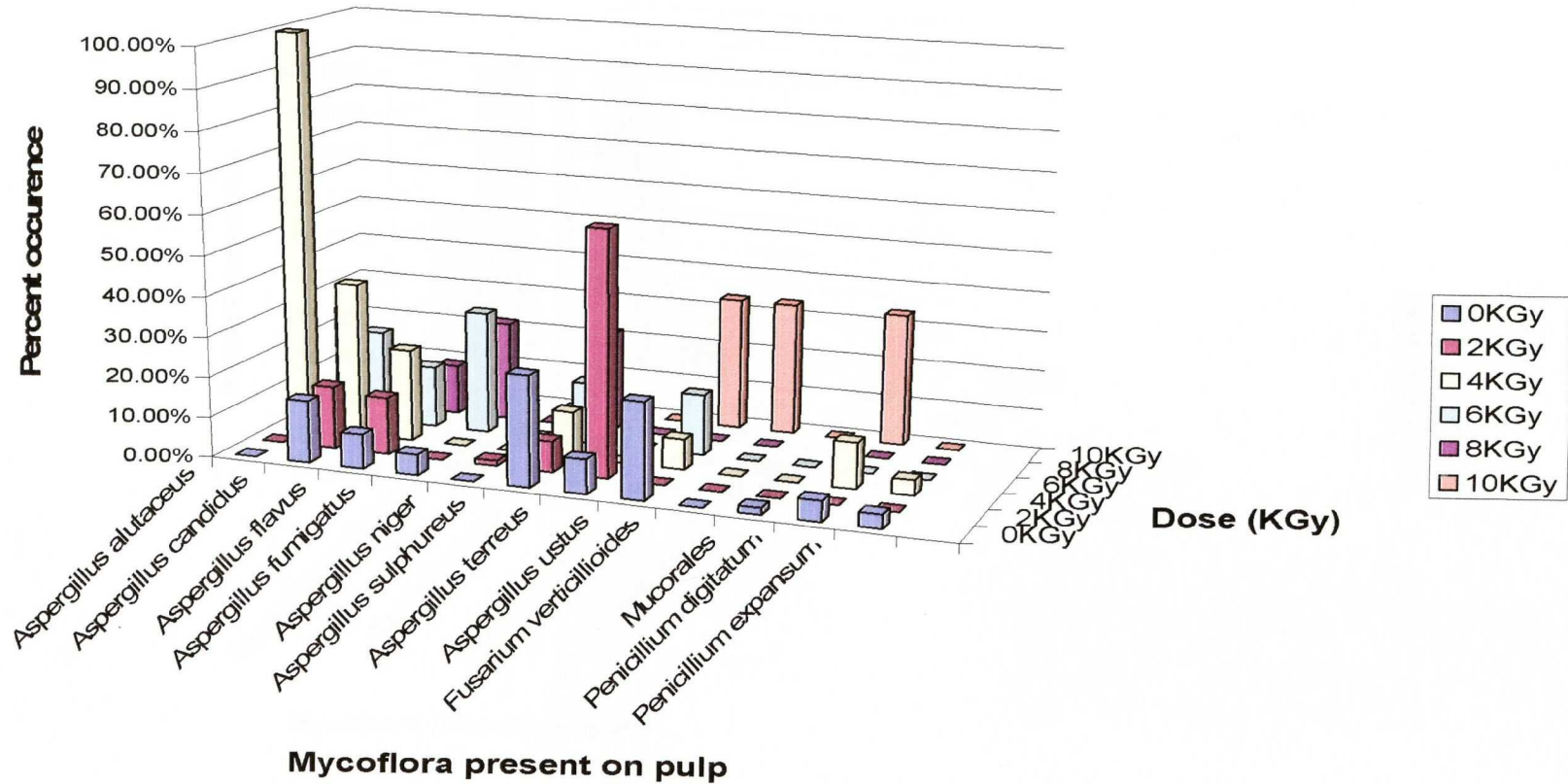


Fig 13: Percent occurrence of mycoflora on the pulp of *D. guineense* stored in polyethylene bags for three months after irradiation with indicated doses

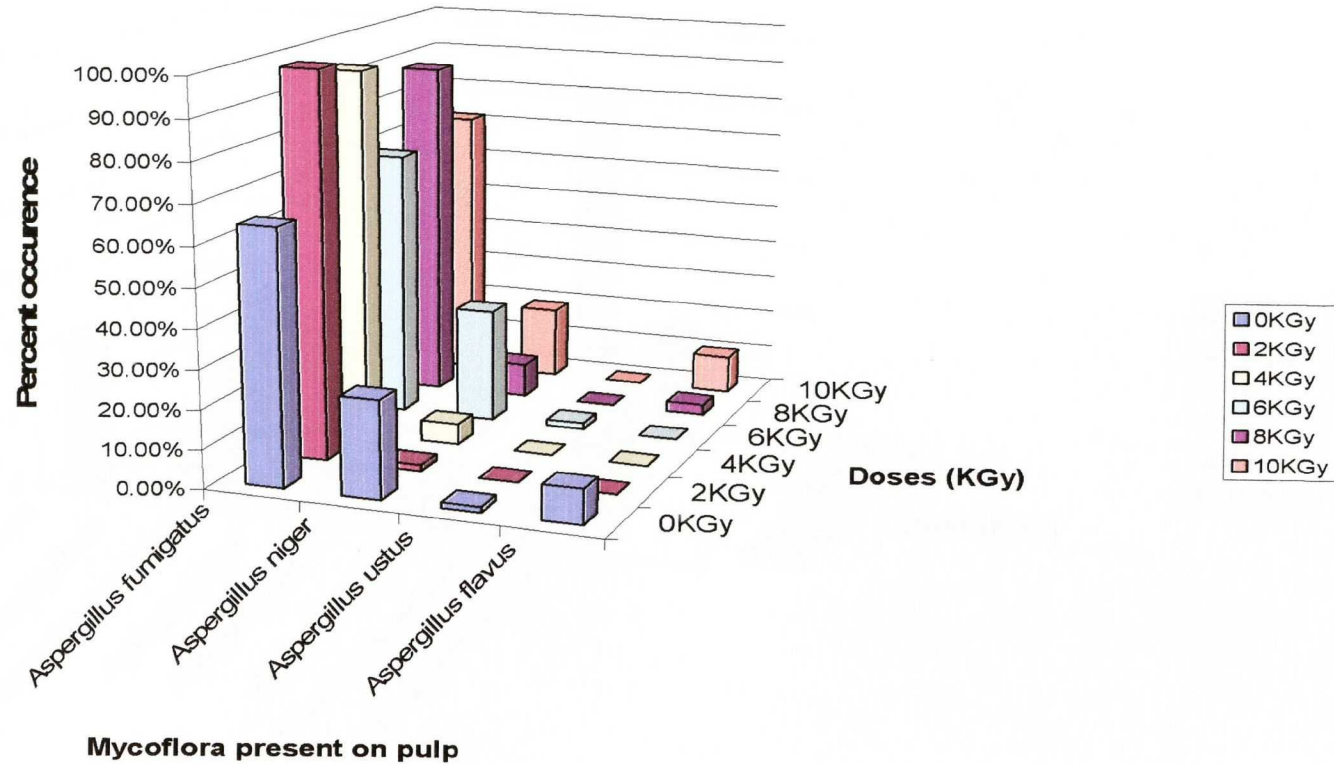


Fig 14: Percent occurrence of mycoflora on the pulp of *D. guineense* stored in polypropylene bags immediately after irradiation with indicated doses

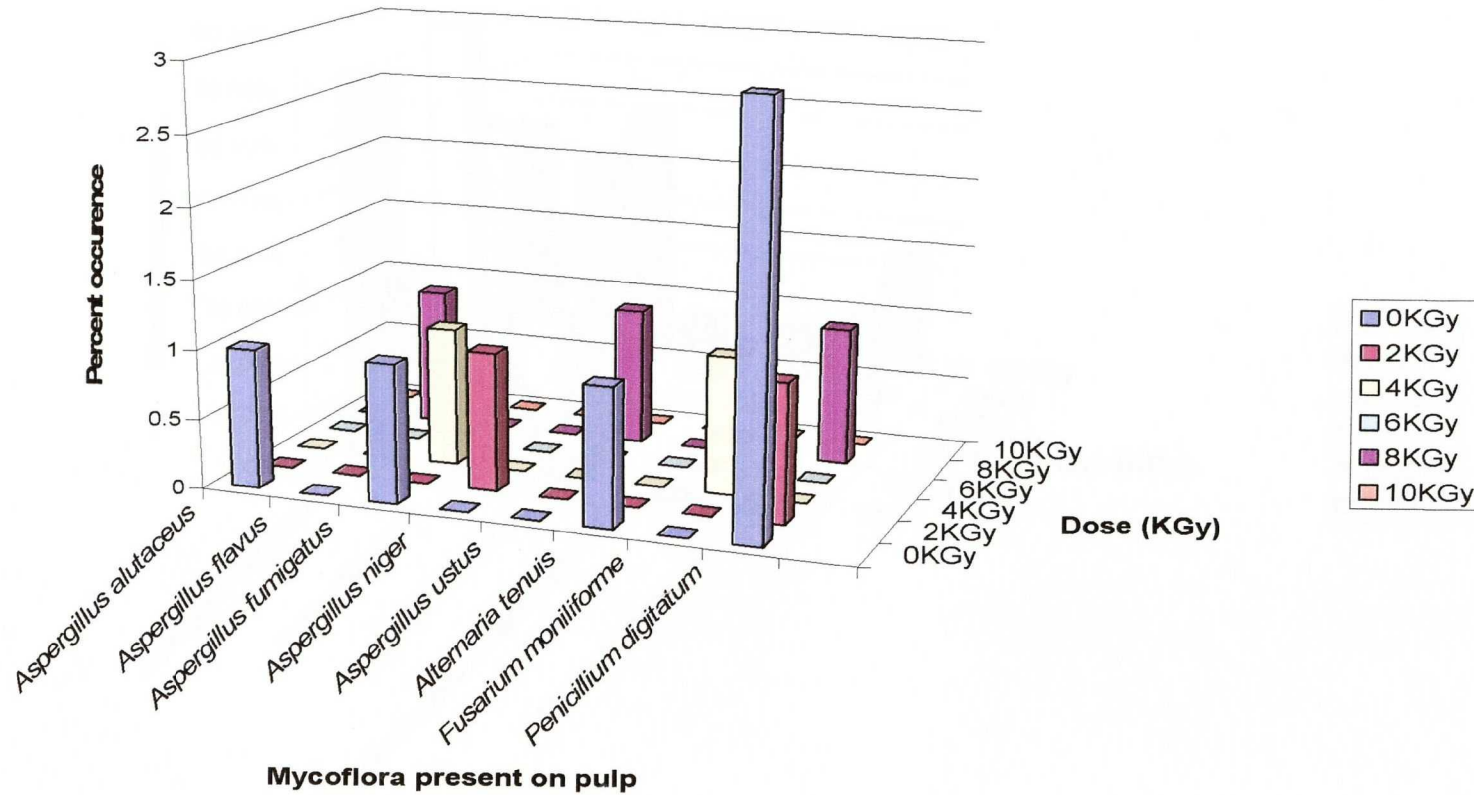


Fig 15: Percent occurrence of mycoflora on the pulp of *D. guineense* stored in polypropylene bags for three months after irradiation with indicated doses

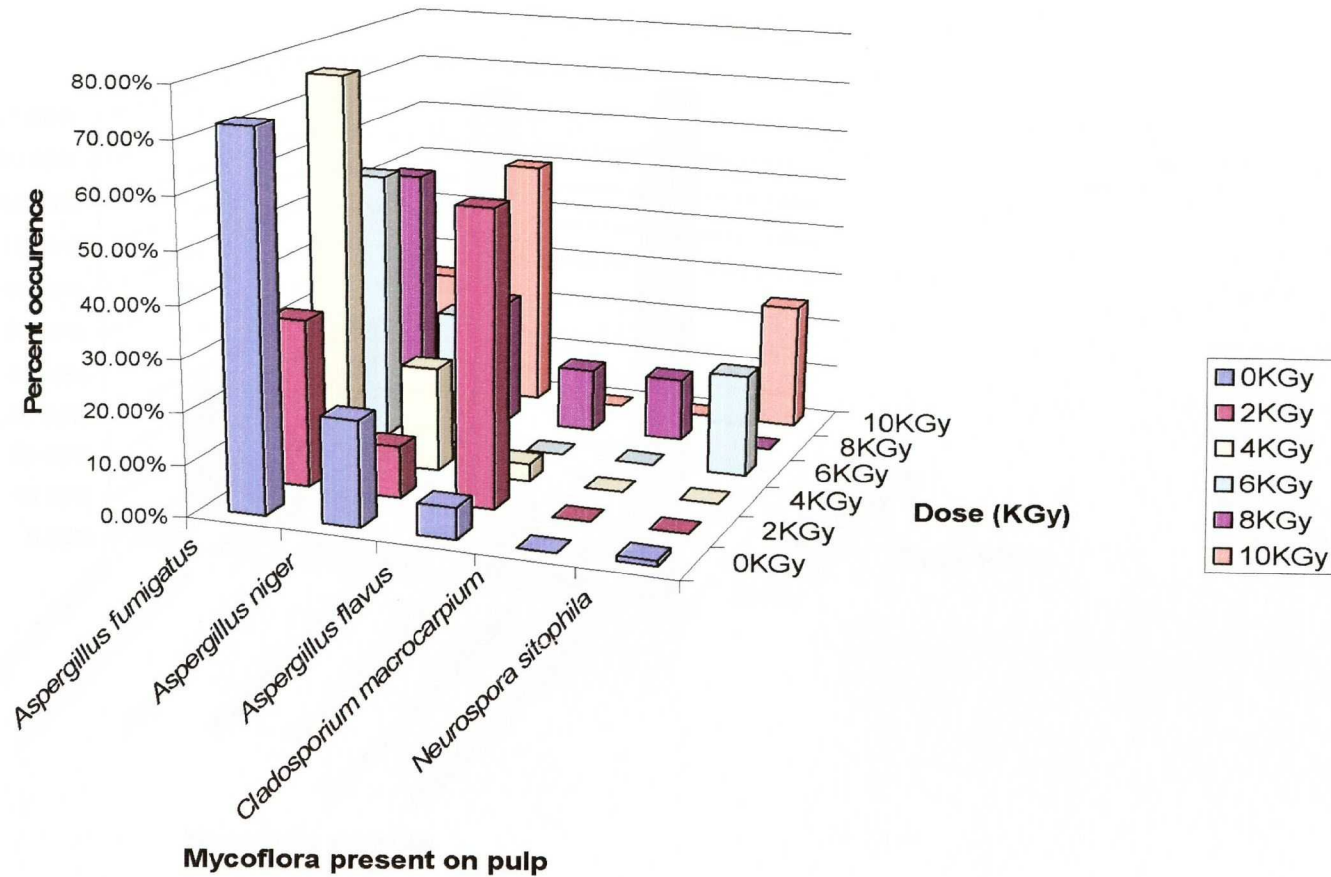


Fig 16: Percent occurrence of mycoflora on the pulp of *D. guineense* stored in jute sacks immediately after irradiation with indicated doses

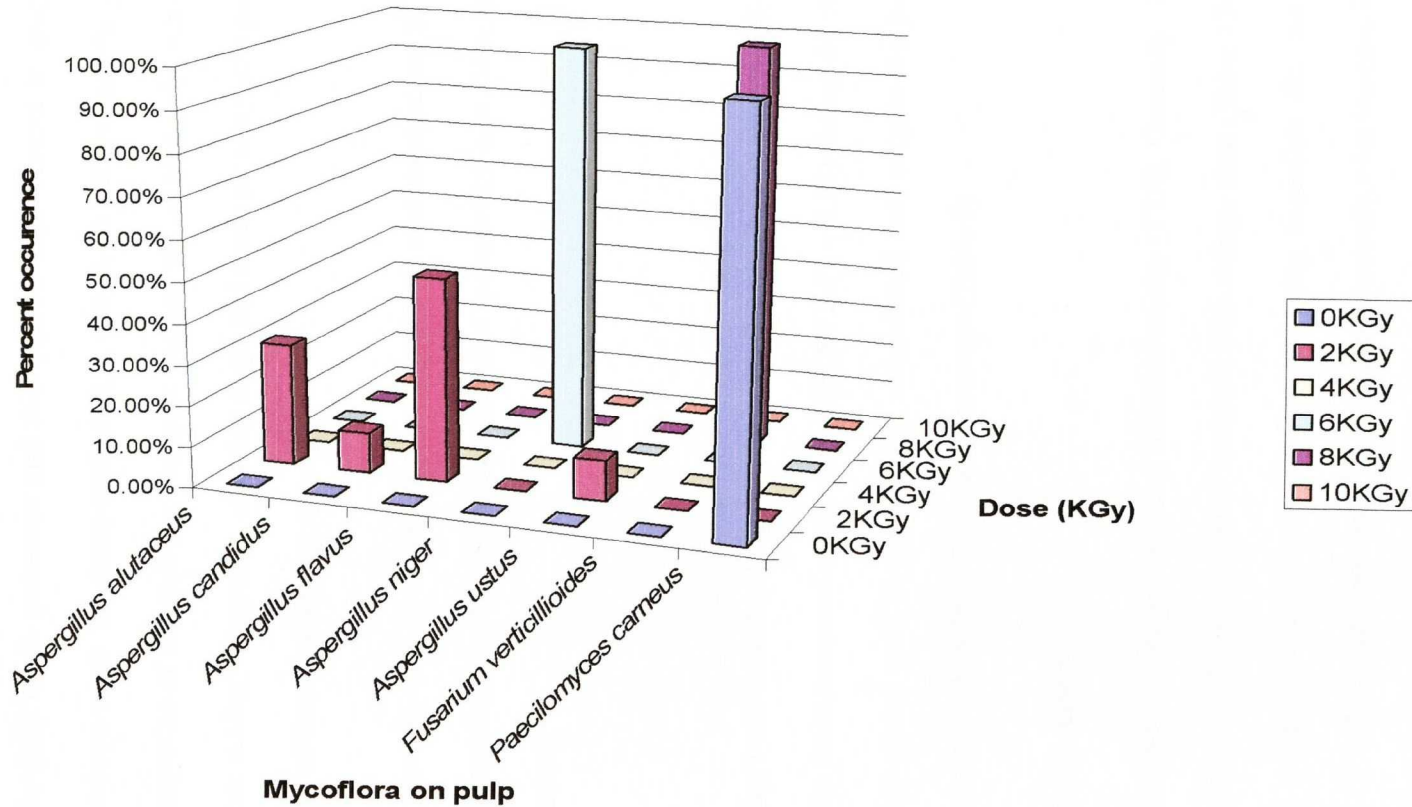


Fig 17: Percent occurrence of mycoflora on the pulp of *D. guineense* stored in jute sacks for three months after irradiation with indicated doses

4.7 Influence of gamma irradiation and packaging on the physico-chemical properties of the pulp of *D. guineense* stored at $29\pm 1^{\circ}\text{C}$ for 3 months

In the previous experiment, irradiation significantly ($P \leq 0.05$) reduced the mould count on the pulp of *D. guineense* and this implied improvement in mycological quality. However, would the doses applied (0, 2, 4, 6, 8, 10 kGY) retain the physico-chemical properties of samples (i.e. moisture content, pH of fruit pulp, ash content, total titratable acidity, fat content, reducing sugar and crude protein) after prolonged storage in the different packaging materials?

The set up was the same as in the previous experiment and standard methods were used in determining the listed physico-chemical parameters (see Materials and Methods section).

pH

The packaging material did not significantly ($P > 0.05$) influence the changes in pH. The pH of the fruits remained on the acid side (pH 3.47 – 3.73) during the three (3) month storage period even at 10kGY. The unirradiated fruit pulp in the jute sack gave the lowest pH (pH 3.46) after 3 months storage (Table 9).

Total Titratable Acidity

The packaging material influenced total titratable acidity (TTA). Generally, TTA of the irradiated (2 to 10 kGY) fruit pulp increased with storage time (Table 10). Even though fruit stored unirradiated in all the packaging materials did not differ significantly ($P > 0.05$). Increase in TTA was commensurate with increase in dose applied.

Ash content

The packaging material did not significantly influence the ash content as all samples kept in the various packaging materials and treated with varying doses decreased in ash content over the period. The loss was more accentuated in samples kept in jute sack treated with 10kGY and stored for three (3) months (Table 11).

Moisture content

The storage bags did not significantly ($P > 0.05$) influence moisture content changes during storage irrespective of the dose applied (Table 12).

Crude fat content

The crude fat content in all the stored samples decreased with prolonged storage (up to 3 months). The fat content decreased with increasing dose and storage period. Fruits stored in polyethylene sacks appear to have a significantly higher fat content ($P > 0.05$) at 10kGY as compared to those stored in polypropylene and jute sack (in decreasing order) (Table 13).

Reducing sugars

There was no interaction between the dose applied, packaging material and period of storage. Each sample behaved differently although there was erratic slight reduction in the reducing sugar content of the samples during storage (Table 14).

Crude protein

There was significant interaction between crude protein of the pulp with dose of irradiation as this parameter decreased with increasing dose up to 10kGY. The

packaging material did not however influence changes during storage time (Table 15) and they behaved independently with the crude protein content.

Table 9: pH of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGY of gamma irradiation

Packaging Material	Storage time (Months)	pH of fruit pulp at indicated doses (kGY)					
		0	2	4	6	8	10
Polyethylene	1	3.59 ^{b_u}	3.57 ^{b_u}	3.69 ^{a_v}	3.7 ^{ab_v}	3.70 ^{a_v}	3.69 ^{a_v}
	2	3.57 ^{b_u}	3.73 ^{a_w}	3.66 ^{a_v}	3.68 ^{a_v}	3.68 ^{ab_v}	3.72 ^{a_v}
	3	3.61 ^{b_u}	3.71 ^{a_v}	3.68 ^{a_v}	3.72 ^{a_v}	3.60 ^{c_u}	3.73 ^{a_v}
Polypropylene	1	3.63 ^{b_u}	3.68 ^{a_{uv}}	3.70 ^{a_v}	3.69 ^{ab_{uv}}	3.69 ^{ab_{uv}}	3.64 ^{b_{uv}}
	2	3.62 ^{b_u}	3.70 ^{a_v}	3.70 ^{a_v}	3.64 ^{b_{uv}}	3.63 ^{bc_u}	3.60 ^{b_u}
	3	3.63 ^{b_{uv}}	3.67 ^{a_{vw}}	3.72 ^{a_{wx}}	3.52 ^{d_u}	3.58 ^{c_u}	3.75 ^{a_x}
Jute Sack	1	3.70 ^{a_v}	3.53 ^{b_u}	3.72 ^{a_v}	3.66 ^{a_{b_v}}	3.71 ^{a_v}	3.72 ^{a_v}
	2	3.70 ^{a_v}	3.75 ^{a_v}	3.70 ^{a_v}	3.60 ^{c_u}	3.63 ^{b_u}	3.71 ^{a_v}
	3	3.47 ^{c_u}	3.52 ^{b_u}	3.51 ^{b_u}	3.50 ^{d_u}	3.56 ^{c_u}	3.62 ^{b_v}

- Initial pH of pulp was 3.72
- Means in the same column which are followed by the same letters (a–c) are not significantly different ($P < 0.05$); means in the same row which are followed by the same letters (u -x) are not significantly different ($P < 0.05$).



Table 10: Total titratable acidity of the fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGY of gamma irradiation

Packaging Material	Storage time Months)	Total titratable acidity (%) of fruit pulp at indicated doses (kGY)					
		0	2	4	6	8	10
Polyethylene	0	1.49 ^a _u	1.47 ^a _u	1.49 ^a _u	1.32 ^a _v	1.42 ^a _u	1.51 ^a _u
	1	1.49 ^a _{vw}	1.51 ^a _{vwX}	1.53 ^a _{wXX}	1.41 ^{ab} _u	1.43 ^{ab} _{uv}	1.59 ^{ab} _x
	2	1.50 ^a _{uvw}	1.51 ^a _{uvw}	1.54 ^a _v	1.41 ^{ab} _u	1.45 ^{ab} _{uv}	1.59 ^{ab} _w
	3	1.51 ^a _{vw}	1.52 ^a _{vw}	1.55 ^a _{vv}	1.43 ^b _u	1.46 ^{abc} _{uv}	1.59 ^{ab} _w
Polypropylene	0	1.47 ^a _u	1.53 ^a _{uv}	1.48 ^a _{uv}	1.57 ^c _{vw}	1.63 ^e _w	1.63 ^{bc} _w
	1	1.48 ^a _u	1.53 ^a _{uv}	1.50 ^a _{uv}	1.56 ^c _{uvw}	1.58 ^{de} _{vw}	1.64 ^{bc} _w
	2	1.50 ^a _u	1.54 ^a _{uv}	1.49 ^a _u	1.57 ^c _{uvw}	1.62 ^e _{vw}	1.66 ^{bcd} _w
	3	1.50 ^a _u	1.53 ^a _u	1.52 ^a _u	1.57 ^c _{uv}	1.63 ^{vw} _{uv}	1.67 ^{bcd} _w
Jute Sack	0	1.70 ^b _v	1.49 ^a _u	1.52 ^a _u	1.70 ^d _v	1.52 ^{bc d} _u	1.70 ^{cd} _v
	1	1.53 ^a _u	1.52 ^a _u	1.52 ^a _u	1.72 ^d _v	1.54 ^{c de} _u	1.73 ^d _v
	2	1.62 ^{ab} _v	1.52 ^a _u	1.51 ^a _u	1.73 ^d _w	1.55 ^{c d} _{uv}	1.75 ^d _w
	3	1.63 ^{ab} _u	1.54 ^a _u	1.51 ^a _u	1.73 ^d _v	1.60 ^{de} _u	1.75 ^d _v

- Means in the same column which are followed by the same letters (a–e) are not significantly different ($P < 0.05$); means in the same row which are followed by the same letters (u -x) are not significantly different ($P < 0.05$).

Table 11: Ash content of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGY of gamma irradiation

Packaging Material	Storage time (Months)	Ash content (%) of fruit pulp at indicated doses (kGY)					
		0	2	4	6	8	10
Polyethylene	1	1.00 ^{c_u}	1.50 ^{b_v}	1.50 ^{b_v}	1.50 ^{b_v}	1.50 ^{b_v}	1.00 ^{c_u}
	2	1.00 ^{c_u}	1.50 ^{b_v}	1.50 ^{b_v}	1.50 ^{b_v}	1.50 ^{b_v}	1.00 ^{c_u}
	3	1.00 ^{c_u}	1.50 ^{b_v}	1.50 ^{b_v}	1.50 ^{b_v}	1.50 ^{b_v}	2.00 ^{a_w}
Polypropylene	1	1.50 ^{b_u}	2.00 ^{a_v}	1.50 ^{b_u}	1.50 ^{b_u}	2.00 ^{a_v}	1.50 ^{b_u}
	2	1.00 ^{c_u}	2.00 ^{a_w}	1.00 ^{c_u}	1.50 ^{b_v}	1.50 ^{b_v}	2.00 ^{a_w}
	3	1.00 ^{c_u}	1.00 ^{c_u}	2.00 ^{b_w}	1.50 ^{b_v}	1.50 ^{b_v}	1.50 ^{b_v}
Jute Sack	1	1.50 ^{b_u}	1.50 ^{b_u}	2.00 ^{a_v}	1.50 ^{b_u}	2.00 ^{a_v}	1.50 ^{b_u}
	2	1.50 ^{b_u}	1.50 ^{b_u}	2.00 ^{a_v}	2.00 ^{a_v}	3.00 ^{c_w}	1.50 ^{b_u}
	3	1.50 ^{b_w}	0.75 ^{d_u}	0.75 ^{d_u}	1.50 ^{b_w}	1.50 ^{b_w}	1.00 ^{c_v}

- Initial ash content of the pulp was 2.25%
- Means in the same column which are followed by the same letters (a–c) are not significantly different ($P < 0.05$); means in the same row which are followed by the same letters (u -x) are not significantly different ($P < 0.05$).

Table 12: Moisture content of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGY of gamma irradiation

Packaging Material	Storage time (Months)	Moisture content (%) of fruit pulp at the indicated doses (kGY)					
		0	2	4	6	8	10
Polyethylene	1	22.07 ^{b_u}	22.35 ^{b_u}	22.18 ^{bc_u}	22.61 ^{a_v}	22.22 ^{bc_{uv}}	22.40 ^{a_u}
	2	22.05 ^{b_{uv}}	22.23 ^{bc_{uv}}	22.16 ^{bc_{uv}}	22.43 ^{ab_{uw}}	21.83 ^{c_v}	22.77 ^{a_w}
	3	22.01 ^{b_u}	21.99 ^{bc_u}	22.48 ^{a_v}	22.25 ^{b_{uv}}	23.12 ^{e_w}	22.77 ^{a_w}
Polypropylene	1	21.96 ^{b_{vw}}	22.30 ^{bc_{wx}}	22.60 ^{a_x}	21.63 ^{c_{uv}}	21.66 ^{cd_{uv}}	21.43 ^{cd_{uv}}
	2	21.77 ^{b_{vw}}	22.00 ^{bc_w}	22.48 ^a	21.31 ^{cd_{uv}}	21.48 ^{cd_v}	21.00 ^{c_u}
	3	21.68 ^b	21.27 ^d	22.33 ^{b_x}	20.96 ^d	21.25 ^d	20.68 ^d
Jute Sack	1	20.72 ^{c_v}	22.33 ^{b_w}	19.28 ^{c_u}	21.99 ^{b_w}	22.34 ^{b_w}	22.32 ^{b_w}
	2	20.58 ^{c_u}	22.31 ^{bc_w}	20.45 ^{d_u}	21.67 ^{c_v}	21.56 ^{c_v}	21.98 ^{b_{vw}}
	3	23.35 ^{d_w}	21.89 ^{c_v}	21.87 ^{c_v}	23.30 ^{e_w}	22.09 ^{bc_v}	21.30 ^{c_u}

- Initial moisture content of the pulp was 22.80%
- Means in the same column which are followed by the same letters (a–e) are not significantly different ($P < 0.05$); means in the same row which are followed by the same letters (u -x) are not significantly different ($P < 0.05$).

Table 13: Crude fat content of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGY of gamma irradiation

Packaging Material	Storage time (Months)	Fat content (%) of fruit pulp at the indicated doses (kGY)					
		0	2	4	6	8	10
Polyethylene	0	1.73 ^a _u	2.03 ^a _u	2.07 ^a _u	2.16 ^a _u	1.76 ^a _u	1.95 ^{ab} _u
	1	1.83 ^b _{uv}	2.09 ^a _u	2.18 ^a _u	2.25 ^a _u	1.52 ^{ab} _v	1.88 ^{ab} _{uv}
	2	1.47 ^{bc} _u	1.88 ^a _u	1.58 ^b _u	2.01 ^a _v	1.43 ^{ab} _u	1.84 ^b _u
	3	1.03 ^{cd} _u	0.91 ^b _u	1.28 ^{bc} _u	1.89 ^a _v	0.86 ^c _u	1.29 ^c _u
Polypropylene	0	1.07 ^{cd} _u	1.05 ^b _u	0.98 ^c _u	1.38 ^b _v	0.89 ^c _u	1.90 ^{ab} _w
	1	0.96 ^d _u	1.00 ^b _u	0.96 ^c _u	1.49 ^b _v	0.89 ^c _u	1.99 ^{ab} _w
	2	0.91 ^d _{uv}	0.88 ^b _u	0.87 ^c _u	1.37 ^{bc} _v	0.84 ^c _u	1.97 ^{ab} _w
	3	1.05 ^{cd} _u	0.88 ^b _u	0.94 ^c _u	0.91 ^{cd} _u	0.82 ^c _u	1.95 ^{ab} _v
Jute Sack	0	1.44 ^{bc} _{uv}	1.08 ^b _u	0.99 ^c _u	1.23 ^{bc} _u	1.16 ^{bc} _u	1.77 ^{bc} _v
	1	1.45 ^{bc} _u	0.92 ^b _v	0.97 ^c _v	1.16 ^{bc} _{uv}	1.13 ^{bc} _{uv}	2.31 ^a _w
	2	1.35 ^{cd} _u	0.91 ^b _v	0.87 ^c _v	1.01 ^{bc} _{uv}	0.84 ^c _v	1.41 ^{bc} _u
	3	0.94 ^d _u	0.92 ^b _u	0.90 ^c _u	0.86 ^d _u	0.86 ^c _u	0.88 ^d _u

- Means in the same column which are followed by the same letters (a–d) are not significantly different ($P < 0.05$); means in the same row which are followed by the same letters (u -w) are not significantly different ($P < 0.05$).

Table 14: Reducing sugar content of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGY of gamma irradiation

Packaging Material	Storage time (Months)	Reducing sugar content (%) at the indicated doses (kGY)					
		0	2	4	6	8	10
Polyethylene	0	11.87 ^a _u	11.90 ^a _u	11.67 ^a _{uv}	11.25 ^a _{uv}	10.43 ^a _v	11.84 ^a _u
	1	11.03 ^a _u	10.02 ^a _u	11.43 ^{ab} _u	11.24 ^a _u	10.32 ^a _u	11.50 ^{ab} _u
	2	10.89 ^a _{uv}	9.87 ^b _u	11.29 ^{ab} _{uv}	11.03 ^a _{uv}	10.15 ^a _{uv}	11.34 ^{ab} _v
	3	10.81 ^a _u	9.80 ^b _u	11.23 ^{ab} _u	10.89 ^a _u	10.06 ^a _u	11.09 ^{ab} _u
Polypropylene	0	11.91 ^a _u	10.43 ^b _v	10.55 ^{ab} _{uv}	11.39 ^a _{uv}	11.25 ^a _{uv}	11.33 ^{ab} _{uv}
	1	11.91 ^a _u	10.22 ^b _v	10.51 ^{ab} _{uv}	11.13 ^a _{uv}	11.09 ^a _{uv}	11.00 ^{ab} _{uv}
	2	11.79 ^a _u	10.18 ^b _v	10.36 ^{ab} _v	11.00 ^a _{uv}	10.92 ^a _{uv}	10.84 ^{ab} _{uv}
	3	11.70 ^a _u	10.17 ^b _v	10.09 ^b _v	10.92 ^a _{uv}	10.87 ^a _{uv}	10.72 ^{ab} _{uv}
Jute Sack	0	11.64 ^a _u	11.18 ^{ab} _u	11.51 ^{ab} _u	10.98 ^a _u	11.34 ^a _u	12.11 ^{ab} _u
	1	11.60 ^a _u	11.00 ^{ab} _u	11.23 ^{ab} _u	10.57 ^a _u	11.14 ^a _u	10.38 ^b _u
	2	11.57 ^a _u	10.96 ^{ab} _u	11.13 ^{ab} _u	10.48 ^a _u	11.01 ^a _u	10.21 ^b _u
	3	11.51 ^a _u	10.85 ^{ab} _u	11.10 ^{ab} _u	10.32 ^a _u	10.89 ^a _u	10.18 ^{ab} _u

- Means in the same column which are followed by the same letters (a–b) are not significantly different ($P < 0.05$); means in the same row which are followed by the same letters (u -v) are not significantly different ($P < 0.05$).

Table 15: Crude protein of fruit pulp before and after 3 months storage in the indicated packaging material treated with 0 to 10kGY of gamma irradiation

Packaging Material	Storage time (Months)	Protein content (%) of fruit pulp at indicated doses (kGY)					
		0	2	4	6	8	10
Polyethylene	0	3.40 ^a _u	2.98 ^a _{vw}	3.13 ^a _{uv}	2.67 ^a _{wx}	2.63 ^a _{wx}	2.58 ^a _x
	1	3.40 ^a _u	2.98 ^a _v	2.89 ^b _v	2.66 ^a _w	2.63 ^a _w	2.57 ^a _w
	2	3.39 ^a _u	2.98 ^a _v	2.89 ^b _v	2.67 ^a _w	2.63 ^a _w	2.57 ^a _w
	3	3.39 ^a _u	2.98 ^a _v	2.89 ^b _v	2.67 ^a _w	2.63 ^a _w	2.57 ^a _w
Polypropylene	0	3.42 ^a _u	2.76 ^a _v	2.50 ^c _v	2.73 ^a _v	2.63 ^a _v	2.61 ^a _v
	1	3.39 ^a _u	2.80 ^a _v	2.49 ^c _w	2.70 ^a _{vw}	2.61 ^a _{vw}	2.61 ^a _{vw}
	2	3.40 ^a _u	2.80 ^a _v	2.47 ^c _w	2.70 ^a _{vw}	2.62 ^a _{vw}	2.61 ^a _{vw}
	3	3.40 ^a _u	2.79 ^a _v	2.47 ^c _w	2.71 ^a _{vw}	2.62 ^a _{vw}	2.61 ^a _{vw}
Jute Sack	0	3.58 ^a _u	2.99 ^a _v	2.83 ^b _{vw}	2.69 ^a _{vw}	2.69 ^a _{vw}	2.59 ^a _w
	1	3.59 ^a _u	2.95 ^a _v	2.83 ^b _{vw}	2.61 ^a _w	2.67 ^a _v	2.59 ^a _w
	2	3.58 ^a _u	2.98 ^a _v	2.83 ^b _{vw}	2.61 ^a _w	2.67 ^a _w	2.56 ^a _w
	3	3.57 ^a _u	2.98 ^a _v	2.83 ^b _{vw}	2.60 ^a _w	2.67 ^a _w	2.56 ^a _w

- Means in the same column which are followed by the same letters (a–c) are not significantly different ($P < 0.05$); means in the same row which are followed by the same letters (u -x) are not significantly different ($P < 0.05$).

4.8 Elemental composition of the fruit coat, pulp and seed of *D. guineense*

The elements in the samples (fruit coat, pulp and seed) were detected using Instrumental Neutron Activation Analysis (INAA) and concentrations were calculated on dry weight basis in triplicate (see Materials and Methods).

Table 16 summarizes the results obtained. Manganese was not present in the fruit coat, but was highest in the pulp ($234 \pm 1.57 \mu\text{g/g}$) as compared to $7.87 \pm 0.03 \mu\text{g/g}$ in the seed. Chlorine was present in all three parts of the fruit (coat, pulp and seed) in varying concentrations; it was highest in the fruit coat ($240.08 \pm 50 \mu\text{g/g}$) and least in the pulp ($180 \pm 24.93 \mu\text{g/g}$). The fruit coat did not contain calcium, sodium and magnesium while the pulp contained calcium in low quantities but high sodium ($332.95 \pm 8.76 \mu\text{g/g}$) and lacking magnesium. The seed contained all the elements detected but lacked potassium. The fruit coat contained potassium in small quantity ($5.64 \pm 0.85 \mu\text{g/g}$) but the pulp was laden with a copious amount of potassium ($549.91 \pm 15.8 \mu\text{g/g}$).

Owing to the time limitation, detection of other long-lived radionuclides like aluminum (Al), copper (Cu), vanadium (V), iron (Fe), cobalt (Co) and bromine (Br) could not be carried out.

Table 16: Elemental composition of the fruit coat, pulp and seed of *D. guineense*

Element	Concentration ($\mu\text{g/g}$)		
	Fruit coat	Pulp	Seed
Manganese	-	23.40 ± 1.57	7.87 ± 0.03
Chlorine	240.08 ± 50	108.05 ± 24.93	131.90 ± 46.66
Calcium	-	0.29 ± 0.02	0.49 ± 0.02
Sodium	-	332.95 ± 8.76	365.58 ± 11.05
Magnesium	-	-	7.87 ± 0.03
Potassium	5.64 ± 0.85	549.91 ± 15.83	-

- Not detected

4.9 Sensory evaluation of the fruit of *D. guineense* treated with gamma irradiation and stored in different packaging materials for 3 months

Studies reported in Section 4.6 suggest that the physico-chemical properties of the fruit pulp was not by and large significantly ($P>0.05$) influenced by the packaging material used and that in some instances, slight reduction in chemical properties were found as the irradiation dose increased.

The irradiated samples of the fruit stored in the different packaging materials were subjected to descriptive and quantitative sensory evaluations. The first sensory evaluation was a quantitative analysis on a nine-point hedonic scale (see Materials and Methods section).

Results obtained are presented in Tables 17 to 22.

Polyethylene sacks

The panelists scored between 3.98 to 4.80 for ease of removal of fruit coat for samples in polyethylene bags and irradiated with 0 to 10kGY of gamma irradiation. There were no significant differences ($P>0.05$) between the irradiated samples and the removal of the fruit coat was neither difficult nor easy.

The mean score of 4.38 to 5.72 connotes neither firm nor fluffy pulp for samples treated with 0 to 8kGY and no significant differences ($P>0.05$) were observed. However, samples treated with 10kGY were put in the pulp tenderness category of slightly fluffy (Table 17) and were significantly different from the rest in tenderness.

The panelists found majority of the fruits to be either brick red or brown as compared to the typical orange colour known for black velvet tamarind. The 0kGY and 8kGY

samples were found to be slightly sweet, whereas all the other samples were neither sweet nor bitter. The panelists scored 0, 2 and 10kGY as slightly acidic (5.88 – 7.84) and scored 4, 6 and 8kGY as neither sour nor acidic (Table 17).

Polypropylene sacks

The panelists scored between 5.23 to 6.53 for ease of removal of samples in polypropylene bags and irradiated with 0 to 10kGY of gamma irradiation. There were no significant differences ($P>0.05$) between the irradiated samples. The removal of the fruit coat was described as neither difficult nor easy (0, 2, 10kGY) or slightly easy (4, 6, 8kGY).

The mean score of 4.58 to 5.68 connotes neither firm nor fluffy pulp for samples treated with 0, 2, 10kGY. Samples treated with 4, 6, 8kGY were put in the category of slightly fluffy (Table 18).

With the exception of samples irradiated at 6kGY that maintained their orange colour, all the other samples were found to be brick red in colour. Panelists found the pulp of all both the irradiated and unirradiated *D. guineense* fruits to be brown as compared to the typical orange colour known for black velvet tamarind. Panelists found three samples (0, 2, 6kGY) to be neither sour nor acidic and the other three (4, 8, 10kGY) to be slightly acidic.

Jute sack

The panelists scored between 5.04 to 6.67 for ease of removal of samples in jute sacks and irradiated with 0 to 10kGY of gamma irradiation. There were significant differences ($P\leq 0.05$) between the irradiated samples. The removal of the fruit coat

was described as neither difficult nor easy (2, 4, 10kGY) or slightly easy (0, 6, 8kGY).

Panelists found no significant difference ($P > 0.05$) between the samples presented in terms of the pulp tenderness. Samples treated with 0, 4, 10kGY gamma irradiation were scored 5.55 to 5.83 which connotes neither firm nor fluffy pulp. Samples treated with 2, 6, 8kGY were put in the category of slightly fluffy (Table 19).

Samples irradiated at 0, 4kGY were described by panelists as having the typical orange colour of black velvet tamarind pulp. All the others were described as brick red. Panelists found three samples (0, 4, 6kGY) to have the orange colour of black velvet tamarind pulp compared to brown; the other three samples (2, 8, 10kGY) were described as brown when compared to the orange colour of black velvet tamarind pulp. Panelists found three samples (4, 6, 10kGY) to be neither sour nor acidic and the other three (0, 2, 8kGY) to be slightly acidic.

Preference analysis

Generally, the panelists neither liked nor disliked the colour of the fruits stored in polyethylene (except 8kGY) and polypropylene (except 4kGY) (Tables 20 and 21). Panelists slightly liked pulp colour of *D. guineense* fruits irradiated at 8kGY and 4kGY and stored in polyethylene bags and polypropylene bags respectively. They also slightly liked the colour of the fruits stored in jute sack (except 8kGY) (Table 22). They neither liked nor disliked the sweetness of the unirradiated samples and the samples irradiated at 2, 4, 8kGY stored in polyethylene bags; they slightly liked samples irradiated at 6kGY and 10kGY. The sweetness of three samples (0, 2, 8kGY) were neither liked nor disliked; the sweetness of the other three (4, 6, 10kGY)

were liked slightly. The panelists “liked slightly” all the samples stored in jute sack, irrespective of the dose of irradiation. Generally, the acidity of the fruit samples was neither liked nor disliked in the polyethylene and the polypropylene bags. For fruits samples stored in jute sacks, panelists slightly liked the acidity of the unirradiated fruits and fruits irradiated at 6, 8kGY; they neither liked nor disliked the acidity of samples irradiated at 2, 4, 10kGY. Panelists slightly liked the tenderness of the unirradiated fruits of both polyethylene bags and polypropylene. Irradiated samples stored in polyethylene bags and polypropylene bags that panelists slightly liked were 6, 10, and 4kGY respectively. They slightly liked all the *D. guineense* fruit samples stored in jute sacks.

Table 17: Descriptive sensory evaluation of samples stored in polyethylene bags

	Ease of removal of coat	Pulp tenderness	Pulp discolouration 1	Pulp discolouration 2	Sweetness	Acidity
0kGY	4.12 ^a	4.59 ^a	4.34 ^{bc}	4.12 ^a	6.01 ^b	6.08 ^a
2kGY	5.15 ^a	5.34 ^a	5.00 ^{bc}	5.15 ^a	5.10 ^b	6.05 ^a
4kGY	3.94 ^a	4.38 ^a	4.13 ^{bc}	3.94 ^a	5.75 ^{ab}	5.00 ^a
6kGY	4.79 ^a	5.72 ^a	5.05 ^c	4.79 ^a	4.59 ^a	5.29 ^a
8kGY	3.98 ^a	4.74 ^a	2.79 ^a	3.98 ^a	6.17 ^{ab}	5.84 ^a
10kGY	4.80 ^a	6.39 ^b	3.73 ^{ab}	4.80 ^a	5.83 ^{ab}	6.31 ^v

- Means in the same column which are followed by the same letters (a–c) letters (or without letters) are not significantly different ($P > 0.05$).

Table 18: Descriptive sensory evaluation of samples stored in polypropylene bags

	Ease of removal of coat	Pulp tenderness	Pulp discolouration 1	Pulp discolouration 2	Sweetness	Acidity
0kGY	5.23 ^a	5.30 ^{ab}	4.91 ^b	3.83 ^a	5.67 ^{ab}	5.45 ^{ab}
2kGY	5.41 ^a	4.58 ^a	4.57 ^{ab}	3.96 ^a	5.77 ^{ab}	5.31 ^a
4kGY	6.53 ^a	6.02 ^{ab}	4.17 ^{ab}	4.06 ^a	5.89 ^{ab}	6.10 ^{abc}
6kGY	6.48 ^a	6.16 ^b	5.29 ^b	3.22 ^a	5.08 ^a	5.14 ^a
8kGY	6.52 ^a	6.72 ^b	3.97 ^{ab}	4.05 ^a	5.69 ^{ab}	6.81 ^{bc}
10kGY	5.66 ^a	5.68 ^{ab}	3.26 ^a	3.85 ^a	6.49 ^b	6.85 ^c

- Means in the same column which are followed by the same letters (a–c) letters (or without letters) are not significantly different ($P > 0.05$).



Table 19: Descriptive sensory evaluation of samples stored in jute sack

	Ease of removal of coat	Pulp tenderness	Pulp discolouration 1	Pulp discolouration 2	Sweetness	Acidity
0kGY	5.92 ^{ab}	5.58 ^a	4.95 ^{bc}	5.40 ^b	6.41 ^{ab}	6.11 ^a
2kGY	5.04 ^a	5.95 ^a	4.56 ^{bc}	4.01 ^a	6.20 ^{ab}	6.15 ^a
4kGY	5.20 ^a	5.55 ^a	5.24 ^c	5.39 ^b	5.18 ^a	5.10 ^a
6kGY	6.36 ^{ab}	6.82 ^a	3.74 ^{bc}	5.12 ^{ab}	6.45 ^{ab}	5.75 ^a
8kGY	6.67 ^b	6.22 ^a	4.54 ^{bc}	4.87 ^{ab}	6.81 ^b	6.34 ^a
10kGY	5.58 ^{ab}	5.83 ^a	2.30 ^a	3.98 ^a	5.95 ^{ab}	5.64 ^a

- Means in the same column which are followed by the same letters (a–c) letters (or without letters) are not significantly different ($P > 0.05$).

Table 20: Preference sensory evaluation of samples stored in polyethylene bags

	Colour	Sweetness	Acidity	Tenderness
0kGY	5.47 ^a	5.60 ^a	5.63 ^a	6.13 ^a
2kGY	5.80 ^a	5.23 ^a	5.27 ^a	5.70 ^a
4kGY	5.93 ^a	5.47 ^a	5.27 ^a	5.60 ^a
6kGY	5.97 ^a	6.17 ^a	5.83 ^a	6.17 ^a
8kGY	6.07 ^a	5.67 ^a	5.67 ^a	5.67 ^a
10kGY	5.53 ^a	6.17 ^a	5.73 ^a	6.37 ^a

- Means in the same column which are followed by the same letters (a–c) letters (or without letters) are not significantly different ($P > 0.05$).

Table 21: Preference sensory evaluation of samples stored in polypropylene bags

	Colour	Sweetness	Acidity	Tenderness
0kGY	5.27 ^a	5.70 ^{ab}	5.33 ^a	6.00 ^{bc}
2kGY	5.80 ^{ab}	5.50 ^a	5.47 ^a	4.93 ^a
4kGY	6.53 ^b	6.50 ^b	5.80 ^a	6.47 ^c
6kGY	5.67 ^{ab}	6.03 ^{ab}	5.43 ^a	5.47 ^{ab}
8kGY	5.63 ^{ab}	5.33 ^a	4.97 ^a	5.43 ^{ab}
10kGY	5.83 ^{ab}	6.47 ^b	5.63 ^a	5.80 ^{abc}

- Means in the same column which are followed by the same letters (a–c) letters (or without letters) are not significantly different ($P > 0.05$).

Table 22: Preference sensory evaluation of samples stored in jute sack

	Colour	Sweetness	Acidity	Tenderness
0kGY	6.57 ^a	6.13 ^a	6.27 ^b	6.77 ^a
2kGY	6.23 ^a	6.00 ^a	5.27 ^a	6.30 ^a
4kGY	6.23 ^a	6.53 ^a	5.97 ^{ab}	6.60 ^a
6kGY	6.27 ^a	6.50 ^a	6.07 ^{ab}	6.53 ^a
8kGY	5.90 ^a	6.80 ^a	6.50 ^b	6.50 ^a
10kGY	6.20 ^a	6.27 ^a	5.90 ^{ab}	6.30 ^a

- Means in the same column which are followed by the same (a–c) letters (or without letters) are not significantly different ($P > 0.05$).

CHAPTER FIVE

5.0 GENERAL DISCUSSION

The black velvet tamarind (*D. guineense*) is one of the lesser known and exploited wild fruits of West Africa. Its seasonal occurrence makes it unattractive for sustained commercial exploitation especially in Ghana. However, data from this thesis indicate that this fruit can be preserved for commercial use when properly harvested.

The preliminary study using the structured questionnaires showed that the fruit is in abundance during the peak season and that women constituted those who purchase the fruit most (48.25%) followed by school children (37.06%) and adult males (14.68%) in decreasing order. This agrees with findings that the urban retail marketing and petty trading have long been dominated by women (Obosu-Mensah, 1999). Indeed, in coastal West Africa, women handle 60 to 90% of the domestic farm produce from the point of origin to the consumer (Obosu-Mensah, 1999). It is interesting to note that school children were the second largest group who purchased the fruit from the market. Recently, McGarry (2008) showed that school children commonly traded their pocket money for fruits of all kind including the black velvet tamarind fruit.

There was another interesting finding. The flow of trading in *D. guineense* showed that all five regions in the southern sector of the country (Central, Greater Accra, Eastern, Ashanti, and Volta) traded in this commodity. This is indicative of its popularity (at least in the peak season). The bulk of the black velvet tamarind was purchased from the Volta Region (Ho, Abor, and Akatsi) and made its way to the Greater Accra, Eastern, and Ashanti Regions and from Greater Accra to the Central

Region (Map 1). This is indicative that there is a well-structured marketing system in the country that needs to be enhanced and preserved for future commercial trading in the product. In future studies, the survey would be extended to the Brong Ahafo, Northern and Upper Regions to ascertain the national utilization of this delicacy in a fruit. Indeed, herbarium records confirm that this plant is found in Bamboi, Techiman, Wenchi and Yeji (Table 1). The local names as found in this study will vary and if extended to the northern sector of the country, one would have an interesting ethnobotanical information on this fruit.

There can be good nutritive value from eating the pulp of the fruit in the fresh state because studies have shown that the fruit is sweet due to the presence of sugars (582.1g kg⁻¹), protein (61.3g kg⁻¹), oil (700g kg⁻¹) and minerals. The fruit also contained glucose, fructose, maltose, sucrose, citric acid, tartaric acid, and ascorbic acid (Ubbaonu *et al*, 2005, Irvine, 1961). Undoubtedly, the school children and the adult males could be enticed by this organoleptic value to purchase the fruit in order to supplement their energy requirements.

If indeed the fruits can be cultivated in commercial quantities and subsequently dried and preserved in the peak season, it can be used for large scale industrial production of various food products ranging from jelly jam, non-alcoholic beverages, soft drinks, syrup concentrates etc (Onwuka and Nwokorie, 2006).

The questionnaire on the transporting and storage of the fruit informed the choice of packaging materials for the subsequent studies using gamma irradiation. Eighty-one point eight one percent (81.81%) of the respondents used woven polypropylene as their packaging material followed by baskets (14.54%) and jute sack (3.63%).

Woven polypropylene and jute sacks were chosen in addition to polyethylene sack for the prolonged storage after the application of gamma irradiation.

The physical characteristics of the fruit showed how variable the pod might be, although mean values for pod weight, diameter, was median (Figs. 4 and 5). The mean pod weight obtained in this study ($1.3 \pm 0.2\text{g}$) was more than double that reported by Ubbaonu *et al.* (2005). It was observed in this present study that a large number of fruits had more than one seed and this might have accounted for the high mean pod weight recorded as compared to the findings of Ubbaonu *et al.* (2005). The observation of the presence of more than one seed in this fruit is not new as there are other similar fruits having up to twelve (12) seeds (<http://www.floridata.com/ref/t/tamarind.cfm>, 2010). The bulk of the weight of the fruit is however contributed by the shell and pulp (78%) and seed (22%) (Table 6).

The grand mean moisture content of the fruit pulp was $22.8 \pm 0.8\%$. This is in contrast with the reported value of 18.2 to 17.75% of the same fruit pulp by Ubbaonu *et al.* (2005). There is information in the pertinent literature that the moisture content of the pulp varies with the developmental stage of the fruit. For instance, Ubbaonu *et al.* (2005) recorded a moisture content of 69.2% during the 9th week of development of the fruit which subsequently decreased to 29.25% at the ripening stage of 17th week of development. Adepoju (2009) and Achoba *et al.* (1992) however found the moisture content of the pulp to be 4.0 and 5.9% respectively. This is a sharp contrast with the previous results and this present study. Presumably, the stage at which the fruits are harvested may determine the moisture content at that stage of development. It is important therefore to harvest the fruits at the period when moisture content is ideal for preservation of the mineral elements and nutritive value of the fruits. Low

moisture content, however, is an indication that there is high dry matter content and that the fruit will have a long shelf-life especially when properly packaged against the influx of external environmental conditions (Eka, 1987).

Equilibrium Relative Humidity (ERH), expressed as a fraction determines the water activity (Christensen, 1962). This fraction, known as water activity (a_w), is the ratio of water vapour pressure of substrate to the vapour pressure of pure water at the same temperature (Scott, 1957). The lower the water activity, the less the water molecules available at the surface of the product for fungal growth and the higher the water activity, the higher the amount of water molecules available for fungal growth or bacterial growth. There is evidence that at ERH less than 75% ($a_w = 0.75$), field and storage fungi do not grow readily. The relationship between water activity and moisture content in a food is often expressed as a sorption isotherm (Palou *et al.*, 1997).

The moisture sorption isotherms of both the pulp and whole fruit followed a near sigmoid curve typical of foods (Debnatha *et al.*, 2002) reaching equilibrium after 4 to 6 days (at 20, 55, 65 and 85% ERH) in the whole fruit and pulp (Figs. 6 and 7). It is worth noting that both pulp and whole fruit stored at ERH's 20, 55 and 65% lost moisture implying that the samples were not expected to grow mouldy.

Samples stored at 75% ERH remained steady in moisture, neither gaining nor losing moisture. Thus, it is anticipated that storage at ambient relative humidity of 20 to 75% would keep the product safe and extend the shelf-life albeit so far as the external aesthetic value is concerned. On the other hand, samples stored at 85 and 95% ERH became mouldy early and was invaded by mould spores after 20 days. This confirms the findings of Wilson and Payne (1994) who stated that food water

activity values of less than 0.70 a_w (less than 70% ERH) are unlikely to support spoilage by microorganisms.

The resident mycoflora of the pulp and the outer coat of the pulp varied (Tables 7 and 8; Figs. 8 and 9). The fifteen (15) different fungi resident in the pulp was predominated by *Aspergillus* species followed by *Penicillium* (Table 7). About half the number of fungi (7) were found on the coat predominated by *Aspergillus* species (Table 8). The preponderance of toxigenic *Aspergillus* (*A. flavus*, *A. fumigatus*, *A. alutaceus*, *A. sulphureus*) and *Penicillium* (*P. digitatum*, *P. expansum*) and *Candida albicans* in the pulp and coat leave much to be desired. *A. flavus* produces aflatoxins B₁, B₂, G₁ and G₂, depending on the strain; *A. fumigatus* produces fumigallin and fumitoxins; *A. alutaceus* forms ochratoxins, kojic acid, penicillic acid and neoaspergillic acid; *P. digitatum* and *P. expansum* form patulin, citrinin and roquefortine C (Samson and Reenen-Hoekstra, 1988). The environmental and cultural conditions for toxin production have been clearly defined by Northolt (1979). On the other hand, *Candida albicans* and related species of *Candida* are Deuteromycetes. They predominantly form yeast-like cells, but there is some formation of mycelium. *Candida* is a normal inhabitant of the mouth, intestinal tract and vagina, and in healthy hosts, lives harmlessly as a saprotroph. It is an opportunistic pathogen, however, and becomes pathogenic in compromised or debilitated hosts (Moore-Landecker, 1996). Factors predisposing the host to infection include malnutrition, unsanitary conditions, the presence of other diseases, or prolonged antibiotic therapy. *Candida albicans* is the causative agent in about 85 to 95% cases of candidosis (Kendrick, 2000; Moore-Landecker, 1996). The presence of these fungi in the fruit of the black velvet tamarind leave much to be desired and



should be decontaminated to extend the shelf-life and quality of the resultant processed products.

There was another storage problem. The harvested fruit was infested by a borer insect *Ephestia cautella*. *E. cautella* has been found infesting benniseed (*Sesame indicum* L.), cassava flour (*Manihot utilissima* Pohl.), cocoa beans (*Theobroma cacao* L.), coffee (*Coffea spp.*), copra (*Cocos nucifera* L.), cotton seed (*Gossypium hirsutum* L.) cowpea (*Vigna unguiculata* Walp), groundnut (*Arachis hypogaea* L.), maize (*Zea mays* L.), millet (*Pennisetum* sp), palm kernel (*Elaeis guineense* Jacq.), and shea butter (*Butyrospermum parkii* Kotschy) (Cornes, 1973). This insect is being recorded for the first time in *D. guineense* fruit in Ghana; the same is true for all the fungi recorded.

The fruits placed in different packaging sacks (medium-density polyethylene, woven polypropylene and jute) and then treated with gamma irradiation (0, 2, 4, 6, 8, 10 kGY) responded differently. Irradiation up to 10kGY drastically reduced the fungal colonies of the fruit in the sacks to different extents. Jute generally had the highest number of colonies compared with other packaging material. This is a new observation because Odamtten and Kampelmacher (1985) have shown using a blotter test that in the absence of an exogenous supply of nutrients, 88% of the sections of jute sacks supports in vivo growth of fungal spores whilst woven polypropylene sacks would not support the growth of contaminating spores. However, there was a commensurate reduction in fungal colonies as the dose increased from 2 to 10 kGY (Figs. 10 and 11). Fungal succession of the pulp before and after irradiation and storage for 3 months in the respective storage sacks varied considerably from one another (Figs. 12 to 17). However, generally, the resident fungi were predominated

by members of the genus *Aspergillus*. Irradiation with 10kGY provided drastic initial mould population reduction but there was no guarantee that storage at ambient laboratory conditions would preclude further external contamination (Figs. 12 to 17). Care must be taken to keep products in a clean environment after irradiation.

Gamma irradiation had variable effect (or no effect) on the physico-chemical properties of the pulp of *D. guineense* after 3 months of storage in the selected packaging materials. The packaging material did not significantly ($P>0.05$) influence the pH, ash content, moisture content, and crude protein of the pulp after 3 months storage (Tables 11 to 13). The packaging material influenced total titratable acidity (TTA), increasing with increasing dose applied. The crude fat in all the stored samples decreased with prolonged storage (Table 13) and with increasing dose. The pH of the fruits remained on the acid side (pH 3.47 to 3.73) in contrast with the findings of Onwuka and Nwokorie (2006) and Ubbaonu *et al.* (2005) who reported a pH range of pH 3.93 and 4.27 respectively. In many fruits, the acidity changes during maturation and ripening, and in some cases like citrus, acidity reduces progressively as the fruit matures on the tree (Barbosa-Canovas *et al.*, 2003). The difference between the present data and that of other workers may be partly attributed to the stage at which the fruits were harvested. Ash content is the inorganic residue remaining after the water and organic matter have been removed by heating in the presence of oxidising agents, which provides a measure of the total amount of minerals within a food (McClements, 2005). Onwuka and Nwokorie (2006) and Achoba *et al.* (1992) reported the ash content of the fruit pulp of *D. guineense* to be between 2.3 and 2.5% which Adepoju (2009) estimated it at 3.2%. Present data from this thesis is between 1.0 and 3.0% (Table 11) after irradiation and storage for 3

months. Steward (2001) reported that irradiation does not significantly alter the elemental composition of food which is in conformity with the present findings.

The crude protein in bags did not vary considerably and was between 2.47 and 3.58%. The crude protein content in the present study is slightly lower than the reported 4.67% by Ajiwe and Umoru (1988). This work has shown that increasing dose results in insignificant lowering of protein content; but storage is not known to affect protein content of food (Joseph *et al.* 2005).

The total titratable acidity (TTA) levels were influenced by dose and packaging material. However, the values obtained were not statistically different from the report of Ubbaonu *et al.* (2005) for the same produce although Onwuka and Nwokorie (2006) reported a lower TTA of 0.33%. Several workers have stated that there are increases in TTA during storage (Azelmat *et al.*, 2006; Zia-Ur-Rehman, 2006; Onigbinde and Akinyele, 1988). The increases have been attributed to the increasing concentration of free fatty acid and phosphate, which results from increased deterioration (Morrison, 1963). The binding of the amino group of amino acids, short chain peptides, and protein, leaving the carboxylic ends free and the presence of acid by-products of advanced Maillard reactions are other possible causes of the increased acidity of the cereal grains stored at elevated temperatures (Fargerson, 1969; Gardner, 1979).

An increase in TTA was attended by a decrease in crude fat content, varying in content with radiation dose increases and packaging material. Fruits stored in polyethylene sacks appeared to have a significantly ($P < 0.05$) higher fat content at 10kGY as compared to those stored in polypropylene and jute sack (in decreasing order) (Table 13).

Interestingly, there was no interaction between dose applied, packaging material and storage time on the reducing sugar content. Each sample behaved differently. It must be emphasised that sugar production from polysaccharides by *D. guineense* starts at the second half of fruit development and that reducing sugars increase steadily while disaccharides decrease during ripening (Vandercook *et al.*, 1980). It is therefore important to harvest at the peak stage of formation of sugars for maximum organoleptic benefits.

The elemental composition of the fruit coat, pulp and seed showed the presence of manganese, chloride, calcium, sodium, magnesium and potassium in varying quantities (Table 16). The fruit pulp contained all but magnesium; the seed contained all but potassium while the fruit coat lacked manganese, calcium, sodium and magnesium.

Calcium is a very important mineral and very essential in human diet. It plays a role in the formation of bones and is also essential for blood clotting and muscle contraction (Wardlaw and Smith, 2006). Its presence in the pulp albeit low is advantageous to the consumer. Potassium and sodium were found in high quantities in the fruit pulp ($549.91 \pm 15.83 \mu\text{g/g}$ and $332.95 \pm 8.67 \mu\text{g/g}$ respectively) and function in the fluid balance and nerve impulse transmission within cells. Although magnesium was found in the seed only, it is a very important micronutrient required for bone formation and aids enzyme action and nerve heartbeat functions (Witney and Rolfes, 2005). The fruit pulp of *D. guineense* undoubtedly has nutritive value (Tables 11 to 16) and should serve as a good nutrient supplement for the consumer. Recent studies in Ghana have shown that the Volta clam (*Galatea paradoxa*) mantle contains these same elements which were found suitable for human consumption

based on the WHO Safety Reference Standards (Amoah *et al.*, 2010). Owing to time limitation, detection of other long-lived radionuclides like Aluminium (Al), Copper (Cu), Vanadium (V), Iron (Fe), Cobalt (Co) and Bromine (Br) could not be carried out. Future studies will consider these elements because it is known that heavy metal toxicity can result in damaged or reduced mental or central nervous functions, lower energy levels, and damage to blood composition, lungs, kidneys, liver and other vital organs (Seeley *et al.*, 1998; Fox, 1999). Long term exposure may result in slowly progressing physical, muscular and neurological degenerative processes that mimic Alzheimer's disease (Seeley *et al.*, 1998; Fox, 1999). Parkinson's disease, muscular dystrophy and multiple sclerosis and long-term ingestion or contact with some metals or their compounds may even result in cancer.

In the concluding chapter of this thesis, the sensory evaluation of the pulp treated with varying doses (0 to 10kGY) of gamma irradiation and stored in different packaging materials was carried out with the view of ascertaining the market acceptability by the consumer. Studies reported in Section 4.6 suggest that the physico-chemical properties of the fruit pulp was not by and large significantly ($P>0.05$) influenced by the packaging material and that in some instances, slight reduction in chemical properties were found as the irradiation dose was increased. The descriptive and quantitative sensory evaluations also showed variable responses (Tables 17 to 22). However, the panelists liked all the *D. guineense* fruits irradiated up to 10kGY and stored in jute sacks. The critical point is the dose necessary to reduce fungal load and possible mycotoxin production during storage. In this case, packaging in any of the bags will not influence preference by consumers since the sensory evaluation results point to this. The recommended dose of 10kGY will also

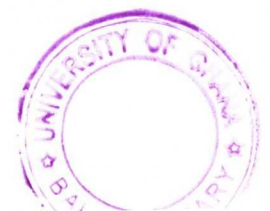
cater for the insect infestation as this dose is well beyond that usually used in disinfestations by gamma irradiation.

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

The black velvet tamarind (*D. guineense*) is a very important fruit tree in West Africa and be classified among the lesser known fruits, although it is eaten by natives of West Africa and Ghana in particular for its refreshing properties and pleasant scorching taste. Recent studies have shown that this fruit can be used in producing beverages, alcoholic and non-alcoholic drinks, syrup concentrates and jams (Okafor, 1975). The fruit contains low protein, oil, is mildly acidic, sugar (glucose, fructose, maltose, sucrose), tartaric acid, citric acid, malic acid and ascorbic acid (vitamin C) (Onwuka and Nwokorie, 2006). Data from this thesis also indicate that the fruit contains ash, crude fat, reducing sugar, crude protein, manganese, chloride, calcium, sodium, magnesium and potassium. The economic and nutritive value of this plant cannot therefore be discounted and must be given the necessary attention to increase cultivation in orchards in the country. The flow of sale of the fruits in Ghana shows that the marketing network is already well-established at least in the southern sector of the country, with the Volta Region (Ho, Abor and Akatsi) serving as the hub for the distribution.

The fruits undoubtedly are laden with fungal spores (15 fungal species belonging to 7 genera and predominated by *Aspergillus*) of pathological importance, in the field before and after harvest and storage. This thesis has shown that the storage humidity above 75% ERH may be detrimental to the quality of the stored fruit and the moisture content of the pulp was critical in the extension of its shelf-life. Irradiation with gamma rays from a Co⁶⁰ source up to 10kGY is required to reduce considerably the fungal load and that a dose of 10kGY would not



significantly alter the nutrient and organoleptic qualities of the fruit in storage. The packaging material also did not seem to significantly ($P>0.05$) influence the storage stability of the pulp for human consumption. The preference of the taste panelists for produce kept in jute sacks is interesting and worth following up, although the other two sacks were also good packaging materials by the analytical method used.

The methods used in this thesis to assess the mycological quality of the pulp are in conformity with the WHO Standards as spelt out in their Technical Report Document Series 598 (1976). Furthermore, there is approval by the WHO/IAEA/FAO Expert committee (1980) on wholesomeness of irradiated food that a dose up to 10kGY would not be harmful for human consumption.

What remains now is a further survey for collating and strengthening the production, marketing and preservation of the fruit during the lean season so that gamma irradiation could be used to extend the shelf-life for the fruit which will be used in industrial and small-scale production of beverages, jams, fruit drinks etc. If this is achieved, this thesis would have contributed as a baseline study and springboard for the entrepreneurs to take up the challenge.

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APPENDIX 1

Questionnaire administered to people involved in the *Dialium guineense* trade

Market:

Locality: Region:

1. Where do you get your fruit of Black tamarind from

Region GA ER CR WR BA AR VR NR UE UW

For each region, state the Town/District.....

2. What is the local name of this plant?

.....

3. When is the peak season of this fruit on the market?

.....

4. Who are those who normally buy this fruit from you?

School children Adult male Adult female Others (specify)

5. How is the purchase of the fruit on the market?

Bad Good Very Good Average

c. When the fruits are going out of season

d. Throughout the season

e. Others (Specify)

6. How are the black tamarind fruits delivered? In

Jute Polypropylene Baskets Paper Others

7. How many bags do you buy at a time?

1 2 3 4 5 More

How long does it take to exhaust your stock?

8. Do you know of any health benefits from eating the fruit on a regular basis?

Yes No

9. If yes, state the benefit.....

10. What is the cost of one American tin of the Black Tamarind?

Lean season..... Peak season.....

11. List the major post-harvest losses that you encounter in order of importance

a. Insects b. Fungi c. Others (Specify)

12. When during the season does each post harvest loss occur?

a. At the start of the season

b. During the peak of harvest

Pulp discolouration 2



Brown

Orange

Sweetness



Very sweet

Sweet

Acidity



Very sour

Not acidic

Any comments:

.....

.....

.....

b. Questionnaire administered in the preference sensory evaluation

SENSORY EVALUATION OF BLACK VELVET TAMARIND

Panelist Number: Date:

Name: Gender: Age:

Sample Codes: 416 391 275 678 263 502

You are presented with black tamarind samples. Please observe and taste the samples in the order presented from left to right and indicate your like or dislike for each sample based on the attribute stated using the nine-point scale below as a guide. You can rinse your mouth / chew a piece of the biscuit provided between samples.

- 9 – Like extremely 6 – Like slightly 3 – Dislike moderately
 8 – Like very much 5 – Neither like nor dislike 2 – Dislike very much
 7 – Like moderately 4 – Dislike slightly 1 – Dislike extremely

Attribute	416	391	275	678	263	502
Colour						
Sweetness						
Acidity						
Tenderness						

Any comments:

.....

APPENDIX 3

a. ANOVA for the moisture content of *D. guineense*

Source of variation	d.f	s.s	m.s.	v.r.	F pr.
Month	3	23.8454	7.9485	27.31	<.001
Packaging	2	7.2973	3.6487	12.53	<.001
Dose	5	24.7639	5.6793	18.87	<.001
Month X Packaging X Dose	30	12.0799	2.0133	6.92	<.001
Residual	132	38.4224	0.2911		
Total	172	106.4089			

b. ANOVA for the fat content of the pulp of *D. guineense*

Source of variation	d.f	s.s	m.s.	v.r.	F pr.
Dose	3	6.4748	1.2950	7.81	<.001
Month	2	4.1236	1.3745	8.29	<.001
Packaging	5	8.2243	1.0754	9.73	0.02
Dose X Month X Packaging	30	1.0891	0.0726	0.44	0.964
Residual	132	19.8869	0.1657		
Total	172	39.7987			

c. ANOVA for the pH of the pulp of *D. guineense*

Source of variation	d.f	s.s	m.s.	v.r.	F pr.
Dose	3	0.054956	0.010991	3.55	0.005
Month	2	0.192300	0.064100	20.71	<.001
Packaging	5	0.150781	0.030054	7.85	<.001
Dose X Month X Packaging	30	0.118467	0.007898	2.55	0.002
Residual	132	0.371400	0.003095		
Total	172	0.887903			

d. ANOVA for the total titratable acidity of the pulp of *D. guineense*

Source of variation	d.f	s.s	m.s.	v.r.	F pr.
Dose	3	0.315920	0.063184	9.03	<.001
Month	2	0.028324	0.009441	1.35	0.262
Packaging	5	0.091857	0.038235	4.89	0.07
Dose X Month X Packaging	30	0.018538	0.001236	0.18	1.000
Residual	132	0.840083	0.007001		
Total	172	1.294723			

e. ANOVA for the ash content of the pulp of *D. guineense*

Source of variation	d.f	s.s	m.s.	v.r.	F pr.
Dose	3	2.1875	0.4375	3.84	0.003
Month	2	16.2431	5.4144	47.54	<.001
Packaging	5	13.7395	3.9042	23.71	<.001
Dose X Month X Packaging	30	2.3403	0.1560	1.37	0.173
Residual	132	13.6667	0.1139		
Total	172	48.1770			

f. ANOVA for the crude protein content of the pulp of *D. guineense*

Source of variation	d.f	s.s	m.s.	v.r.	F pr.
Dose	3	14.34256	2.86851	40.00	<.001
Month	2	0.14481	0.04827	0.67	0.570
Packaging	5	13.98670	1.14004	21.73	0.040
Dose X Month X Packaging	30	1.12702	0.07513	1.05	0.412
Residual	132	8.60557	0.07171		
Total	172	38.20666			

g. ANOVA for the reducing sugar content of the pulp of *D. guineense*

Source of variation	d.f	s.s	m.s.	v.r.	F pr.
Dose	3	5.505	1.101	0.71	0.620
Month	2	6.092	2.031	1.30	0.277
Packaging	5	3.127	0.942	0.96	0.043
Dose X Month X Packaging	30	16.356	1.090	0.70	0.781
Residual	132	186.996	1.558		
Total	172	218.076			

APPENDIX 4

ANOVA table for ease of removal of fruit coat of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	37.9885	5	7.5977	0.99	0.4270
Within groups	1338.88	174	7.6947		
Total (Corr.)	1376.87	179			

ANOVA table for pulp tenderness of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	88.7832	5	17.7566	2.58	0.0278
Within groups	1196.11	174	6.87422		
Total (Corr.)	1284.9	179			

ANOVA table for pulp discolouration 1 of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	107.587	5	21.5174	3.48	0.0051
Within groups	1076.24	174	6.18529		
Total (Corr.)	1183.83	179			

ANOVA table for pulp discolouration 2 of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	39.6129	5	7.92259	1.26	0.2813
Within groups	1089.94	174	6.26404		
Total (Corr.)	1129.56	179			

ANOVA table for sweetness of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	55.0568	5	11.0114	1.52	0.1863
Within groups	1261.51	174	7.25003		
Total (Corr.)	1316.56	179			

ANOVA table for acidity of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	38.5707	5	7.71413	1.09	0.3688
Within groups	1233.78	174	7.0907		
Total (Corr.)	1272.35	179			

ANOVA table for case of removal of fruit coat of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	55.1644	5	11.0329	1.60	0.1614
Within groups	1196.88	174	6.87863		
Total (Corr.)	1252.05	179			

ANOVA table for pulp tenderness of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	83.0923	5	16.6185	2.07	0.0714
Within groups	1397.13	174	8.02948		
Total (Corr.)	1480.22	179			

ANOVA table for pulp discolouration 1 of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	78.4413	5	15.6883	1.96	0.0876
Within groups	1395.96	174	8.02273		
Total (Corr.)	1474.4	179			

ANOVA table for pulp discolouration 2 of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	14.7391	5	2.94783	0.39	0.8547
Within groups	1312.78	174	7.54473		
Total (Corr.)	1327.52	179			

ANOVA table for sweetness of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	30.9496	5	6.18992	0.86	0.5111
Within groups	1256.36	174	7.22045		
Total (Corr.)	1287.31	179			

ANOVA table for acidity of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	86.5479	5	17.3096	2.34	0.0439
Within groups	1288.53	174	7.40532		
Total (Corr.)	1375.07	179			

ANOVA table for ease of removal of fruit coat of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	61.4774	5	12.2955	1.65	0.1502
Within groups	1299.6	174	7.46895		
Total (Corr.)	1361.08	179			

ANOVA table for pulp tenderness of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	33.9963	5	6.79927	0.88	0.4989
Within groups	1351.52	174	7.76736		
Total (Corr.)	1385.52	179			

ANOVA table for pulp discoloration 1 of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	171.057	5	34.2114	5.09	0.0002
Within groups	1170.27	174	6.72567		
Total (Corr.)	1341.32	179			

ANOVA table for pulp discoloration 2 of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	63.2877	5	12.6575	1.77	0.1204
Within groups	1241.15	174	7.13307		
Total (Corr.)	1304.44	179			

ANOVA table for sweetness of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	47.1518	5	9.43036	1.36	0.2414
Within groups	1205.73	174	6.92949		
Total (Corr.)	1252.88	179			

ANOVA table for sweetness of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	30.2558	5	6.05117	0.81	0.5420
Within groups	1295.34	174	7.4445		
Total (Corr.)	1325.6	179			

ANOVA table for preference evaluation for colour of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	8.96111	5	1.79222	0.47	0.7977
Within groups	662.433	174	3.80709		
Total (Corr.)	671.394	179			

ANOVA table for preference evaluation for sweetness of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	21.5167	5	4.30333	1.23	0.2973
Within groups	609.033	174	3.50019		
Total (Corr.)	630.55	179			

ANOVA table for preference evaluation for acidity of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	8.8	5	1.76	0.51	0.7690
Within groups	601.4	174	3.45632		
Total (Corr.)	610.2	179			

ANOVA table for preference evaluation for tenderness of samples stored in polyethylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	15.5611	5	3.11222	0.74	0.5922
Within groups	728.767	174	4.18831		
Total (Corr.)	744.328	179			

ANOVA table for preference evaluation for colour of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	25.5778	5	5.11556	1.34	0.2516
Within groups	666.733	174	3.8318		
Total (Corr.)	692.311	179			

ANOVA table for preference evaluation for sweetness of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	30.4444	5	6.08889	1.85	0.1059
Within groups	573.2	174	3.29425		
Total (Corr.)	603.644	179			

ANOVA table for preference evaluation for acidity of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	11.8278	5	2.36556	0.68	0.6355
Within groups	601.033	174	3.45421		
Total (Corr.)	612.861	179			

ANOVA table for preference evaluation for tenderness of samples stored in polypropylene

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	41.9833	5	8.39667	2.25	0.0514
Within groups	648.967	174	3.72969		
Total (Corr.)	690.95	179			

ANOVA table for preference evaluation for colour of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	6.73333	5	1.34667	0.44	0.8171
Within groups	527.467	174	3.03142		
Total (Corr.)	534.2	179			



ANOVA table for preference evaluation for sweetness of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	12.9611	5	2.59222	0.78	0.5681
Within groups	581.1	174	3.33966		
Total (Corr.)	594.061	179			

ANOVA table for preference evaluation for acidity of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	26.2278	5	5.24556	1.36	0.2398
Within groups	668.767	174	3.84349		
Total (Corr.)	694.994	179			

ANOVA table for preference evaluation for tenderness of samples stored in jute sack

Source	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
Between groups	4.86667	5	0.973333	0.30	0.9110
Within groups	560.133	174	3.21916		
Total (Corr.)	565.0	179			