

**KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY,
KUMASI**

COLLEGE OF AGRICULTURE AND NATURAL RESOURCES

FACULTY OF AGRICULTURE

DEPARTMENT OF HORTICULTURE

**EFFICACY OF FOUR BOTANICALS AND TWO CHEMICAL
FUNGICIDES**

**IN THE CONTROL OF CROWN ROT DISEASE OF BANANA (*Musa spp*
AAAA) cv. Medium Cavendish**

**A THESIS SUBMITTED TO THE SCHOOL OF GRADUATE STUDIES,
KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY
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**FOR THE AWARD OF
MASTER OF SCIENCE IN POSTHARVEST PHYSIOLOGY.**

EMMANUEL OPOKU AMPOMAH

DECEMBER, 2009

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BY

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DECEMBER, 2009

DECLARATION

I hereby declare that except for references to other people's work which have been duly acknowledged, this write-up submitted to the Board of Postgraduate Studies, Kwame Nkrumah University of Science and Technology, Kumasi, is the result of my own investigation and has not been presented for any degree elsewhere.

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DEDICATION

I dedicate this Thesis to my parents and Cynthia Atakora (Mrs).
God richly bless you.

KNUST



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ABSTRACT

Botryodiplodia theobromae was isolated and identified as the main causative agent or pathogen involved in crown rot disease of bananas collected from Fremponso and Volta River Estate Limited of the Atiwa and Asuogyaman districts in the Eastern region of Ghana. *Aspergillus niger* and *Aspergillus flavus* were isolated and identified as part of the crown rot organisms from infected crowns of bananas and these were nonpathogenic when inoculated into healthy matured crowns of green bananas. Different concentrations of botanical and chemical fungicides were prepared and tested for their effectiveness on Medium Cavendish banana inoculated with *Botryodiplodia theobromae* to find out which treatment best controls crown rot disease. There were significant differences ($P < 0.05$) in the effectiveness of these treatments in controlling the crown rot pathogen. The rot pathogen, *Botryodiplodia theobromae* when inoculated into healthy hands of Medium Cavendish banana and treated with botanical and chemical fungicides had effect on postharvest quality parameters such as total soluble solids and total titratable acidity. Pulp to peel ratio, pH and percentage weight loss were not affected by these treatments. Medium Cavendish banana inoculated and treated with *Moringa oleifera* (leaf extract), *Azadirachta indica* (seed extract) and *Cassia alata* (leaf extract) developed the disease in day 3. Medium Cavendish banana treated with *Zingiber officinale* (rhizome extract), ShavitF71.5WP and Mancozeb developed crown rot in days 5, 6 and 8 respectively. ShavitF71.5WP at a concentration of 2.0g/l and Mancozeb at a concentration of 7.0g/l offered good protection against crown rot disease of banana. *Zingiber officinale* (rhizome extract) at a concentration of 66.67% w/v was the most effective of the botanicals tested against crown rot disease. ShavitF71.5WP was superior to Mancozeb.

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CHAPTER ONE

1.0 INTRODUCTION

Bananas are monocotyledons and belong to the family Musaceae. They are tree-like perennial herbs, two to nine metres tall with an underground rhizome or corm, a pseudostem composed of leaf sheaths and a terminal crown of leaves through which an inflorescence emerges. Some seven to nine months after planting of a sucker, an inflorescence is formed at the base of the pseudostem. About one month later, this inflorescence emerges through the centre of the leaf crown, and fruits may be ready for harvest 90-150 days after inflorescence emergence (Seymour *et al.*, 1993).

Banana is an important food crop for man, especially in the tropics. The annual world production is about 45 million tonnes, 20 million tonnes of dessert cultivars and the rest culinary types. Banana is the only tropical fruit which is exported in large quantities, total export being in the order of 6-7 million tonnes per annum (Bose and Mitra, 1990). The highest concentration of production is in tropical America and the largest markets are in North America, Europe, and Japan. From a socio-economic view point, bananas are acceptable crops to grow, eat and sell, and they can also provide reliable family income and job opportunities (Robinson, 1996). Export banana is an important dollar earner of exporting countries (Picq *et al.*, 1998). In Ghana, banana exported to the European Union Countries were of the order of \$10,330,000 (GEPC, 2006).

Based on 2002 United Nation's Food and Agriculture Organization (FAO) statistics, Clay (2004) puts forth the following figures: Banana cultivation takes up 4.1 million hectares, globally producing 67.5 million metric tonnes of fruits.

The flavour, texture, convenience, ease of eating and nutritional value has made dessert bananas very popular. Banana is a useful source of vitamins A, C and B₆ and has twice the concentration of potassium compared with other ripe fruits (Nakasone and Paull, 1998).

Bananas are used in special diets where ease of digestibility, low fat, no cholesterol, minerals (high potassium, low sodium) and vitamin content are required. The fruit does not cause digestive disturbances; it readily neutralizes free acids in the stomach and does not give rise to uric acid. Special diets for babies, the elderly and patients with stomach problems, gout and arthritis often contains banana because of the above attributes (Nakasone and Paull, 1998).

Unfortunately, this important crop has a number of problems that affect its production and shelf life.

Diseases of the fruit can cause severe problems if they are not controlled. The most virulent disease of banana is black Sigatoka which is found worldwide, except for subtropical and Caribbean. Panama disease has caused severe disruption of banana production where susceptible cultivars such as 'Gros Michel' are grown. The two other major diseases are the bacterial disease 'Moko' and viral bunchy-top disease (Nakasone and Paull, 1998).

Postharvest diseases including anthracnose, crown rot and finger rot cause significant losses in revenue to farmers and traders. Crown rot disease, often described as disease complex caused by several fungi, affect banana trade of most producing countries (Dadzie and Orchard, 1997). Two or more fungi may attack the crown simultaneously or successively and cause tissue rotting which results in deterioration of banana hands

during ripening. Banana fruits infected by crown rot organisms are rejected on the market.

Reports on postharvest losses of fruits caused by diseases may be significantly high in a number of producing countries. For example 20% of banana produced in Sri Lanka are lost to postharvest factors including diseases (Ranasinghe *et al.*, 2005). Though figures on incidence and losses due to postharvest diseases on fruits are not readily available in Ghana, observations on markets and in homes suggest that losses could be significantly high. These losses may be causing significant financial losses to traders particularly those who export the commodity. Crown rot disease is very common in Ghana and its control definitely can increase financial earnings of traders and inflows of foreign exchange into the country.

Postharvest diseases of banana, particularly crown rot are controlled through the use of chemical fungicides such as thiabendazole and imazalil (Robinson, 1996). In recent years, integrated disease management systems that reduce dependence on chemical fungicides are being emphasized in several importing countries (Anonymous, 2001).

This study was, therefore, conducted basically in an attempt to find safer methods of controlling crown rot disease. Plant parts of certain species suspected or reported to possess fungicidal properties were investigated to establish the extent to which they can protect banana against crown rot disease.

The following objectives were set in for study:

- to determine the causal organism(s) responsible for crown rot disease through inoculation experiments, and

- to determine the efficacy of *Azadirachta indica* (neem seed extract), *Moringa oleifera* (leaf extract), *Cassia alata* (leaf extract) and *Zingiber officinale* (rhizome extract) in controlling crown rot disease.

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CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 BOTANY AND DISTRIBUTION

The banana is important in the humid tropical lowlands, with year-round production. Edible bananas are derived from either *Musa acuminata* (A) or *Musa balbisiana* (B) or a combination of both. Cultivars are diploid or triploid, with some new tetraploid developed by breeding. Most dessert bananas are AA or AAA, with the triploid AAA being the most important in the trade (Nakasone and Paull, 1998).

M. acuminata has seeded fruit. Dessert cultivars were developed from it via parthenocarpy and sterility, aided by human selection and vegetative propagation. *M. balbisiana* also has a wild seeded fruit, is more suited to drier areas and occurs from India to New Guinea and the Philippines, though absent from central Malaysia. It was similarly taken into cultivation, with selection of natural diploid, triploid and tetraploid hybrids. Bananas are now cultivated throughout the tropics and subtropics (Nakasone and Paull, 1998).

2.2 HARVESTING AND FRUIT CARE

According to Gowen (1995), the major problems affecting bananas during and after harvest are:

- the susceptibility of the mature fruit to physical damage caused during transport and marketing, by repeated handling of the bunches of fruit before dehanding and of the hands of fruits in boxes afterwards,
- decay of ripe banana due to attack by pathogenic fungi, which develop from infections which can be established at any time before or after harvest, and

- uneven and unpredictable ripening of the fruit which fails to meet consumer demand.

Gowen (1995), further stated that physical damage leads to unsightly blackening of both unripe and ripe bananas and if severe, causes bruising and breakdown of the edible pulp. It also provides one of the major avenues for invasion by those fungi which cause decay. The damage and decay can be a major problem in the marketing of bananas in industrialized importing countries due to downgrading in quality and price. Superficial skin discoloration is of less significance in domestic marketing in producing countries, where the eating quality of the fruit is more important than its appearance. In order to understand the harvesting and postharvest practices used in the commercial trading of banana, it is necessary to have some idea of the development of the fruit and the physical and chemical changes which occur after harvest.

Additionally, since the pre-harvest and postharvest environments of banana affect its quality and marketable life, it is important to understand these factors and their interactions. Pre-harvest factors affecting postharvest performance of bananas include the cultivar grown, soil and climatic conditions, cultivation practices and damage by pests and diseases. The export of banana fruit from the tropics to remote destinations has been in progress for over 100 years (Burden, 1969, Thompson, 1981 and Gowen 1995). The postharvest practices necessary to minimize losses are, therefore, very well developed and sophisticated.

2.3 MATURITY AND HARVESTING

2.3.1 Harvest Maturity

Banana is a climacteric fruit, which means it has a development phase during which, among other things, the fruit increases in size and accumulates carbohydrates in the form of starch. Growth ceases when the fruit is fully mature and the ripening phase is initiated. The initiation of ripening in climacteric fruits such as bananas is marked by a rapid rise in respiration rate of the fruit to a peak after which the rate gradually falls as ripening progresses. Bananas have short shelf life after ripening, and so those which have to be transported over long distances are harvested in the green condition and transported under refrigeration to their destination. They are then ripened under controlled conditions by the importer or distributor. The stage of maturity for harvesting the fruit depends on the market for which it is intended and is determined in terms of the marketable life required. Bananas to be marketed locally can be harvested at a more mature stage than those which are to be exported (Gowen, 1995).

Bananas are harvested at 75-80% maturity for long distance transport taking some three weeks, at 90% for regional and Caribbean island transport time of two weeks and fully mature for local domestic marketing within one week. If the fruits are to mature at harvest they may well split during handling, especially if they have recently been irrigated or it has rained. Mature fruit may also ripen prematurely during transport (Burden, 1969 and Thompson, 1981).

2.3.2 Maturity Indices

According to Gowen (1995), there are no universally recognized objective methods which can be used to determine when bananas should be harvested. Much research has been carried out on the subject but positive results have mostly been location specific

or lack the potential for practical application under production conditions, particularly where production is in the hands of small-scale growers.

Some indices which have been used are:

- age of the bunch after emergence from the pseudostem
- the angularity of the fruit fingers in cross-section
- ratio of the fruit peel to pulp
- measurement of the finger length or diameter
- firmness of fruit
- Brittleness of the flower end of the bunch.

2.3.2.1 The age of the bunch after emergence

This is measured from the time when the fruit bunch is first visible on its emergence from the pseudostem and is called shooting. For a given combination of cultivar and growing conditions, the approximate time between the emergence of the bunch and harvest can be estimated. In this case the bunch can be marked at the time of shooting and subsequently harvested at the appropriate time. In Southern Ecuador, Ghana and some other production areas, this is done by having a coloured stripe down the side of the polyethylene film bunch cover. This cover is placed on the bunch at the time of shooting. The colour of the stripe is changed weekly so that the age of the bunch can be readily known by the colour shown (Burden, 1969 and Thompson, 1981).

2.3.2.2 The angularity of the fingers

Determination of the maturity stage of bananas by visual inspection of the fullness of the fingers is one of the most commonly used methods. In the early stages of development the fingers are clearly angular in cross-section, but as growth progresses,

the angularity decreases and the fruit becomes more rounded. The final degree of roundness varies between cultivars, but it is a reasonably precise method and, with experience, is practical to use especially for small-scale growers. Although farmers generally know and commonly use this method it is not always applied in practice (Burden, 1969 and Thompson, 1981). In some countries farmers harvested less mature fruit, even for local markets, because they have cash-flow problems and harvest as soon as the ripening room operator will accept the fruit (Thompson, 1981).

Various terms are used in producer countries to classify the angularity of the fruit, and marketing agents for export companies require farmers to harvest only a particular grade. In the Caribbean islands, grades classified from light three-quarters to round- full have long been used as the basis for judging where to cut bunches for export. The maturity of hands in the bunch varies slightly, those at the proximal end of the bunch being more mature than those at the distal end, so the estimate of maturity is based on the fullness of fruit of the middle hand. In Latin America, where large scale production is centered on plantation units of around 400ha, in which uniform production conditions are achieved, maturity is assessed by the harvester using specially designed spring caliper to measure the thickness of a standard finger of the outer whorl of the second hand from the distal end of the bunch (Stover and Simmonds, 1987).

2.3.2.3 Pulp to peel ratio

This is used as an indication of fruit maturity, but since it is a destructive method which can only be applied by taking representative samples from the bunch, it is of limited practical application for farmers. In India, a minimum pulp: peel ratio of 1:1 has been accepted as being the acceptable level of maturity for ripening Dwarf Cavendish banana (Burden 1969; Delal *et al.*, 1970 and Thompson, 1981).

2.3.2.4 Diameter and length of banana fingers

Fruit maturity is related to the diameter of the fingers and by measuring predetermined fingers on each bunch with a pair of calipers, an estimate of the bunch's maturity can be made. This parameter also varies between seasons. Therefore, it is often difficult to establish standards even on one banana plantation. Finger length has also been used. This method is sometimes used to assess the maturity of banana hands or the bunch before harvest, but the practice is not wide spread. It is more commonly employed to grade hands of bananas during packing station operations. There is no standard method of measuring the length of banana finger. Some measure the outside curve from the junction of the pulp and fruit stalk to the tip of the fruit, others take this measurement on the inner curve of the fruit and yet others measure in a straight line from the fruit stalk to the tip. Where this method is used, it is necessary to standardize the measurement to particular fingers as done for angularity (Burden, 1969; Stover, 1972 and Thompson, 1981).

2.3.2.5 Firmness of fruit

A method of evaluating the maturity or ripeness of fruit is by determining its firmness using a penetrometer to measure the pressure required to break the skin. The firmness changes during development and can be used to determine the maturity of a bunch using representative samples (Burden, 1969 and Thompson, 1981). Firmness can be measured objectively using instruments such as Magness-Taylor or an Effagi pressure tester, but this requires destructive testing of representative samples. Non-destructive sonic

techniques for measuring fruit firmness have also been developed. These methods do not appear to have found wide application (Burden, 1969 and Thompson, 1981).

2.4 HARVESTING METHODS AND TRANSPORT OF FRUIT TO PACKAGING STATIONS

2.4.1 Field Dehanding

According to Thompson (1981), dehanding of banana is the cutting of the hands of fruit from the main bunch stem. This involves cutting through the crescent-like pad of tissues which attaches the hand to the main stem. The cut surface exudes latex for sometime after dehanding and is also liable to infection by fungi which cause a disease known as crown rot. Field dehanding of bananas was developed in the Windward Islands during the early 1970's, mainly in response to the high level of damage suffered when bunches of fruit were moved from the field to packing stations. This damage occurred to a greater or lesser extent with all growers, but was particularly serious in bananas harvested from small mountainside plots.

The technique involved dehanding the bunch either whilst still attached to the pseudostem or when they were cut and hung from a frame beside the plant. The former method was more effective in reducing damage but the latter easier in practice.

Originally, the hands were packed into specially designed plastic field boxes, usually lined with leaves, for transport to pack house, but subsequently the hands were picked directly into fibreboard boxes in the field ready for export. This was only achieved when the problem of latex staining of the fruit and crown were overcome. These problems were normally dealt with by washing the hands and treating them with chemicals at the

packing station. In the field they were both controlled by first laying the hands crown downwards on the banana leaves to prevent the latex getting onto the fruit and placing a pad of absorbent paper impregnated with alum (potassium aluminium sulphate) and fungicide (thiabendazole or benomyl) on the cut crown of each hand soon after dehanding. This helps to coagulate the latex to prevent it staining the fingers and prevent crown rot becoming established (Burden, 1969 and Thompson, 1981).

Additional problem associated with field dehanding and packing was infestation of boxed fruit by insects mainly cockroaches and ants. Where this is a problem it has been dealt with by fumigation of the boxed fruit with the broad spectrum insecticide permethrin, usually during ripening in importing country. Field dehanding is now an established practice with many growers and has resulted in an increase in the percentage of top quality fruit marketed (Burden, 1969 and Thompson, 1981).

2.5 PACKING STATION OPERATIONS

Dehanding fruit in the field and packing them directly into fibreboard boxes for export eliminates the need for a packing station and has the advantage of saving the capital and operating costs of such a facility. The system has, however, found limited application and all other systems involve the use of a packing station. This may vary in design and construction from a simple pole and thatch structure to a purpose designed permanent building. The operations required for preparing and packing bananas for export will be the same irrespective of the type of structure. They include dehanding, washing, selection and grading, fungicide application and packing (Burden, 1969 and Thompson, 1981).

2.5.1 Dehanding

It is usual for bananas to arrive at the packing station as bunches. Where a cableway is installed the bunches are brought right to the dehanding site still suspended from the rolling hook on which they were placed when harvested. If they have been by other means they should not be stacked on the ground, but immediately hung from an overhead structure at the correct height. In all cases the dehanding site should be close to the end of the wash tank. Usually the bunch is hang from the proximal end, with the largest hands uppermost. When dehanding, the crown should be cut close to the main stem of the bunch. It should be evenly cut, leaving as much as possible of the crown attached to the hand, otherwise its outer fingers may be detached during subsequent handling. The design of the knife used for dehanding varies considerably in different countries. A curved knife, with the inside curve sharpened, is usual but in some countries a curved chisel type of knife is preferred. In all cases the knife must be very sharp to give a clean smooth cut in a single movement. As bunches of bananas are dehanded, the hands must be placed immediately in the wash tank (Burden, 1969 and Thompson, 1981).

2.5.2 Washing, Selection and Grading

Hands of the fruits removed from the bunch are immediately placed in the wash tank to remove dirt and latex which exudes from the cut surface of the crown. There should be a flow of water through the tank to avoid the accumulation of dirt and fungal spores, which may infect the crown. In Jamaica this flow is sometimes achieved by diverting water from a stream through the tank, alternatively pumps can be used to keep up the flow of fresh water. If there is no continuous fresh water supply, chlorine (at 100mg/l active chlorine) and for alum (10g/l) are normally added to recirculated water to remove

latex and destroy microorganisms. Normally, the flow of water is from the dehanding end of the tank, so that the hands of bananas move along to the far end, where workers select and grade prior to fungicide application. Separate washing and de-latexing tanks are sometimes used, in which case the dehanding fruits are washed for 4 minutes and delatex for 10 minutes. More commonly a single tank is used, in which the hands are left for 15-20 minutes to remove latex. Latex has no physiologically detrimental properties but if left on the surface of the fruit causes unsightly staining, which can affect the fruit's market value (Burden, 1969 and Thompson, 1981).

2.5.3 Packaging

The final handling of the bananas in the packing station involves packing the hands in boxes in which they are to be marketed. Since the 1960s, virtually all bananas exported outside their production region have been packed in fibreboard boxes holding an average weight of about 14 or 18 Kg depending on market preference. The boxes are usually fully telescopic slotted type, often with a divider to improve the stacking strength. They are made up by glueing or stapling. The box dimensions used vary, but now tend to conform to international standard modular dimension for palletized handling. Boxes must be very strong to withstand stacking pressures, which can be as much as 250 Kg in some break bulk stacks. It is essential that the box design should allow for good ventilation of the contents. Ventilation holes should be as large as permissible and located to give free air flow through the contents of close stacked boxes. This will maintain an even temperature throughout the fruit during refrigerated shipment. However, ventilation holes can affect the stacking strength of fibreboard boxes so a compromise should be reached to maximize ventilation and stacking strength

(Burden, 1969 and Thompson, 1981).

Most exporters use polyethylene film liners to help reduce moisture loss from the bananas and provide some protection from damage during handling and transport. This procedure varies with different producers and the distance the bananas must be transported. Some fold sheets of polyethylene around the fruit; others pack all box contents in a sealed bag under partial vacuum, and a few use individual small bags, each containing a single hand of fruit. Vacuum packing is commonly used where the transport time is long because it can modify the carbondioxide and oxygen levels around the fruit, which is said to extend their green life. Polyethylene film packing has also been used to delay ripening of boxed bananas during transport, as an alternative to refrigeration. The hands of green fruit sealed in polythene bags with an ethylene absorbent (potassium permanganate) remained in the green, hard condition for up to 18 days at ambient temperatures during transport from North Queensland to Auckland, New Zealand (Scott *et al.*, 1971). The bananas should be packed in a regular pattern in the box in such a way that the hands of fruit do not move and damage each other when the box is handled. The pack should be full and tight enough to prevent the content moving, but not so full that the bananas are damaged by pressure when the boxes are stacked (Burden, 1969 and Thompson, 1981).

2.6 TRANSPORT OF BOXED BANANA

The packed boxes are usually loaded onto refrigerated lorry and taken to a ship where individual boxes are placed directly into a refrigerated holds. This method is called break bulk. In some cases the boxes are loaded onto pallets at the pack houses and the pallets transported and loaded onto ships. Palletization means that fewer boxes can be loaded into ship's holds. Refrigerated containers (reefers) are used, the banana boxes

being loaded directly into 20 or 40 foot insulated containers at the pack house and then transported to ship by road. The containers transported by sea to the importing country and then by road directly to the ripening rooms. In some cases the containers are loaded (stuffed) at the docks of the exporting country. Reefers are used in some countries for export bananas, but break bulk shipment is cheaper and is usually preferred where reefer ships are available. Green bananas that are preclimacteric are shipped under refrigeration to prevent the initiation of ripening before they arrive at their destination (Burden, 1969 and Thompson, 1981).

Stacking boxes of fruit in ships holds or containers must be done with great care to ensure adequate ventilation to all boxes. This is particularly important where palletization is used because with a solid stack of boxes on a pallet the air will tend to go around it leaving some of the boxes inadequately cooled. Techniques to ensure air is directed through the boxes include stapling strips of fibreboard between pallets or even having inflatable bags jammed around the pallets (Burden, 1969 and Thompson, 1981).

Bananas are shipped under a controlled temperature of 13-14°C and are provided with regular changes of fresh air throughout the voyage to avoid damage due to accumulation of carbondioxide. Neither the bananas nor the ship's hold are pre-cooled prior to loading the boxes into the ship. The practice has been attempted in the Caribbean, but it was found that the temperature in the hold rose quickly to ambient during loading. Also, workers were reluctant to move in and out of the cold rooms, believing that it adversely affects their health. The ships' refrigeration has capacity for rapidly cooling the fruit, which do not appear to be adversely affected. It is desirable, however, that when bananas are shipped in reefer containers with integral or clip-on refrigeration units, both the

boxes of fruit and the containers should be pre-cooled and loaded very quickly, because many of the containers' refrigeration units are designed only to maintain cool temperatures, not for the rapid removal of field heat (Burden, 1969 and Thompson, 1981).

2.7 FRUIT MATURATION OF BANANA

Fruit maturation is an important postharvest criterion essential in the screening of banana because, the stage of maturation at which any fruit is harvested greatly influences the green-life or the ability of that fruit to be stored for long periods and its final eating quality. Every fruit will develop its full characteristic flavour, taste and colour during storage if it is picked during an optimum period. Fruits harvested at an early stage of maturity are more susceptible to shriveling and mechanical damage and are of poor quality upon ripening, despite having a long storage life. On the other hand, harvesting at an advanced stage of maturity is unsuitable for fruits intended for long distance shipment due to their shorter storage life (Dadzie and Orchard, 1997).

Therefore, it was important to carry out harvesting at the right maturity stage to suit the purpose. Maturity at harvest is an important factor affecting quality perception and the rate of change of quality during postharvest handling. By knowing the stage of maturation of banana, it would be possible to schedule harvesting, handling and marketing operations efficiently (Kader, 1994).

2.8 GREEN-LIFE

Bananas are usually harvested at matured green stage and stored. During storage, they remain firm and green without any significant changes in skin colour, texture or

composition for an extended period of time depending on the temperature, humidity and age at harvest before the commencement of ripening. This well-defined period after harvest, during which fruits remain green and firm, is referred to as the pre-climacteric life or green-life (Dadzie and Orchard, 1997).

Once the green-life of the fruit has ended and ripening has been initiated, it is irreversible and any fruit in this condition would be over ripe during the marketing process. Their green-life potential will play a significant role in the overall acceptability of the fruit. Bananas which have long green-life or remain green for a long time after harvest, or ripen slowly, will facilitate marketing of the fruit and reduce postharvest losses (Dadzie and Orchard, 1997).

2.9 STORAGE AND RIPENING

2.9.1 Storage

Bananas can be stored at a temperature slightly above 13°C and a relative humidity of 85 to 95% for about three weeks and is ripened in a week or two at 16.5-21°C. Banana fruit becomes blackened at lower temperatures and should not be placed in a refrigerator. In producing countries, the banana is carried either by rail or by road in unrefrigerated carriage. On the other hand, the produce for overseas trade is carried in refrigerated ships, the banana being kept in a cool air circulation at about 11-13.5°C. Premature ripening is probably the biggest single cause of loss during storage. Experiments indicate that ripening can be delayed, that is, storage life can be prolonged by keeping the fruit in relatively high concentration of carbondioxide and low concentration of oxygen. Dipping of bananas in 200ppm thiabendazole has been approved and recommended as postharvest treatment (Bose and Mitra, 1990).

After 4 weeks of storage at $14.5^{\circ}\text{C} \pm 1^{\circ}\text{C}$ and 80-90% RH, 64% fruits of Robusta were fully ripe and Dwarf Cavendish was overripe. A double coating of 12% wax emulsion prolonged the storage life of Dwarf Cavendish banana by 10-12 days at 14.5°C . From a storage study, Bose and Mitra (1990) found that Dwarf Cavendish banana can be stored at 14.4°C with 80-90% RH for 25 days. Simple polythene bagging was adequate for extending storage life by about a week at warm ambient temperature. While longer storage could cause ripening of green bananas due to ethylene accumulation, a vermiculite/cement block soaked in KMnO_4 solution removes ethylene and naturally extends storage life.

Recommended storage conditions for two banana cultivars (Pisang Rastali and Pisang Embun) was 20°C with continued ethylene removal and low (5-10% v/v) CO_2 concentration with relative humidity of about 80 percent. Observations were that application of ethylene was the best method to hasten ripening without loss in fruit quality or flavour. A pre-storage dip in E-9267 emulsifiable mineral oil at 0.4% was found effective in reducing fruit decay and also prolonging storage life (Bose and Mitra, 1990). Treating the fruits with benzimidazole fungicides controlled *Asperillus niger* and prolongs shelf-life for 8 days. Dipping of fruits in 1.5 or 2.5 percent talprolong solution delayed yellow colour development by 4 to 8 days and changes in pH and total acidity when stored at 30°C or at $20-24^{\circ}\text{C}$ after treatment (Bose and Mitra, 1990).

2.9.2 Ripening

Bananas are not usually allowed to ripen on the tree as it takes a long time to reach that stage. Moreover, the fruit peel splits, fruit ripens unevenly and fails to develop good

colour and aroma; hence the marketable quality deteriorates. Therefore, banana needs to be ripened artificially. On arrival at the destination, the banana bunches are immediately sold to wholesale dealers who store the fruits in loose heaps in godowns and ripen them in lots per the need of the retailer dealers. In tropical conditions, fruits for local consumption are harvested and ripened by hanging the bunches in a shady place. The predominant carbohydrate of green banana was found to be starch which hydrolysed to sucrose, glucose and fructose on ripening. The starch hydrolysis did not commence until respiration had increased to the two thirds of the climacteric peak and at about the peak of ethylene production. The starch degradation is accompanied by an increase in sucrose content followed by glucose and fructose formation during ripening (Bose and Mitra, 1990).

The ripening process for the export dessert-type bananas are normally carried out by specialists under controlled conditions just before distribution and marketing to consumers. Under controlled ripening conditions, ethylene is supplied from compressed gas cylinders, ethylene generators or ethylene-generating chemicals, such as ethephon (Nakasone and Paull, 1998).

Commercially, bananas are treated with about 100 ppm ethylene for about 24h under controlled temperature and humidity conditions and ventilation to prevent carbondioxide build-up. Temperature control allows fruit to be ripened on a specific schedule to colour stage 3 for distribution from 4 days at 19°C to 10 days at 14.5°C. The humidity control has a significant impact on the final skin colour developed and flesh softening (Nakasone and Paull, 1998).

2.9.2.1 Assessment of changes that occur during ripening

Physiologically mature bunches are harvested and the fingers of the second hand are used in assessing changes during ripening of banana. However, fruits from the third hand may be included if there are not enough sample fruits. Fruits are ripened naturally at ambient temperature or by exposure to ethylene (1ml/litre) for 24-48 hours at relative humidity of 90-95% (Dadzie and Orchard, 1997).

2.9.2.2 Peel and pulp colour changes

The disappearance or loss of peel green colour and the corresponding increase in yellowing of the peel during ripening are the obvious manifestations in banana. The loss of green colour is due to degradation of the chlorophyll structure. External changes in peel colour during ripening often reflect changes in pulp colour (Dadzie and Orchard, 1997).

Fruits are classified according to peel colour, by visually matching the peel colour of the fruits against a colour chart. Also the use of colour measuring devices could give a good indication of the changes that occur (in the peel and pulp) during ripening or the stage of ripeness of the fruit. Peel and pulp colour charts may be developed for each hybrid to help standardize the stages of ripeness (Dadzie and Orchard, 1997).

2.9.2.3 Conversion of starch into sugar

The most striking postharvest chemical change which occurs during the post-harvest ripening of banana is the hydrolysis of starch and the accumulation of sugar (i.e. sucrose, glucose and fructose) which are responsible for the sweetening of the fruit as it ripens. In dessert banana (e.g. Cavendish) the breakdown of starch and the synthesis of sugar

are usually completed at full ripeness (peel colour stage 6-7), while in plantain this breakdown is slower and less complete and continues in over-ripe and senescent fruits (Dadzie and Orchard, 1997).

Different methods and procedures for assessing starch (or sugar) content during ripening of fruits including banana has been described by various researchers, however, most of these methods are complex, time consuming and require trained personnel and expensive technology. Hence, an easy, rapid and inexpensive method of estimating the starch content of the fruit could serve as a useful indicator of ripeness. The starch iodine test is a simple, rapid and inexpensive method of visually assessing the conversion of starch to sugar during fruit ripening (Dadzie and Orchard, 1997).

2.9.2.4 Changes in pulp to peel ratio

Pulp to peel ratio is a good and consistent index of ripening of banana. Pulp to peel ratio increases in response to ripeness (i.e. peel colour score). Changes in pulp to peel ratios during ripening of banana indicate differential changes in moisture content of the peel and pulp. The increase in pulp to peel ratio during ripening is related to sugar concentration in the two tissues. During ripening, there is a rapid increase in the sugar concentration in the pulp compared to the peel thus contributing to a differential change in osmotic pressure. The peel loses water both by transpiration to the atmosphere and also to the pulp by osmosis, thereby contributing to an increase in the fresh weight of the pulp as the fruit ripens. This results in an increase in the pulp to peel ratio during ripening (Stover and Simmonds, 1987).

2.9.2.5 Changes in pulp firmness

Under normal storage conditions, bananas undergo significant textural transformations as they pass through the ripening process. The crisp, hard and green fruit turns into a yellow fruit with tender and soft internal pulp at the optimal ripening stage, and becomes mushy as it advances towards senescence. The loss of firmness during ripening leads to lower quality and higher incidence of mechanical damage during handling and transportation. Loss of pulp firmness during ripening varies with cultivar/hybrid. Pulp firmness is often inversely related to ripening, implying that, as ripening progressed, pulp firmness declined (Dadzie and Orchard, 1997).

Generally, the triploid cultivars are usually firmer than the tetraploid hybrids (Dadzie, 1994). Loss of firmness or softening during ripening has been associated with two or three processes. The first is the breakdown of starch to form sugar. The second is the breakdown of the cell walls or reduction in the middle lamella cohesion due to solubilisation of pectic substances. The third is the movement of water from the peel to the pulp during ripening due to the process of osmosis (Dadzie and Orchard, 1997).

2.9.2.6 Changes in total soluble solids content

During ripening of banana the total soluble solids content increases. However, the magnitude of increase is dependent on cultivar/hybrid. In most ripe fruits, including banana, sugar forms the main component of soluble solids. Since the amount of sugar in fruits usually increases as the fruit matures and ripens the soluble solids content of the fruit can be a useful index of stage of ripeness. Soluble solids content vary between cultivar and between stages of ripeness. For instance, in some hybrids soluble solids contents increase to a peak and then decline (the drop in total soluble solids may be due

to the conversion of sugar in the pulp to alcohol). While in some hybrids, total soluble solids continue to increase with ripening (Dadzie and Orchard, 1997).

2.9.2.7 Changes in pulp pH and total titratable acidity

Pulp pH and total titratable acidity are important postharvest quality attributes in the assessment of fruit ripening quality. In most banana cultivars/hybrids, there is a rapid decline in pulp pH in response to increasing ripeness. However, the magnitude of decline is cultivar dependent. Generally, when fruits are harvested at matured green stage, the pulp pH is high but as ripening progresses pH drops. Thus the pulp pH could be used as an index of ripening. Usually organic acids decline during ripening as they are respired or converted to sugar. Organic acids are important in giving a desired sugar-to-acid balance which results in pleasing fruit taste during ripening. Acidity measured as titratable acidity in the pulp tissues of most banana show large increases during ripening or as ripening progresses. Therefore, total titratable acidity could be used as an index of ripening (Dadzie and Orchard, 1997).

2.9.2.8 Changes in peel and pulp moisture and dry matter content

According to Dadzie and Orchard (1997) peel and pulp moisture and dry matter content are important postharvest parameters in the evaluation of the ripening quality of new banana. During ripening, the moisture content of the peel decreases whereas that of the pulp increases; this is because the peel loses water both to the atmosphere and to the pulp. In most cultivars/hybrids, the dry matter content of the peel and pulp during ripening does not change significantly.

2.9.2.9 Changes in respiration rate and ethylene production

During ripening of banana there is a tremendous increase in the amount of ethylene produced. This increase is usually accompanied by an increase in respiration rate of the fruit (a phenomenon which is called climacteric). The rate of respiration and ethylene production usually depends on storage temperature, age of fruit and cultivar/hybrid (Dadzie and Orchard, 1997).

2.9.2.10 Changes in phenolics during ripening

Banana fruits can contain high levels of phenolic compounds especially in the peel. Tannins are perhaps the most important phenolic from the point of view of fruit utilization because they can give fruit an astringent taste. As fruit ripens their astringency becomes lower which seems to be associated with a change in structure of the tannins, rather than a reduction in their levels, in that they form polymers (Burden, 1969 and Thompson, 1981). Phenolics are also responsible for the oxidative browning reaction when the pulp of fruit especially immature fruit is cut. The enzyme polyphenoloxidase is responsible for this reaction (Burden, 1969 and Thompson, 1981).

2.9.2.11 Changes in pigment during ripening

There are normally two types of pigment in the peel of bananas: chlorophylls and carotenoids. The change in colour of ripening fruits is associated with the breakdown of chlorophylls with carotenoid level remaining relatively constant (Seymour, 1985). The colour change is thus largely an unmasking of the yellow pigments. Cavendish banana cultivars example Grand Nain, Valery and Dwarf Cavendish, can fail to completely degreen when they ripened at 25°C and above (Dadzie and Orchard, 1997). This can result in bananas which are ripe in every other respect remaining green and the higher

the temperature over a range of 25-35°C results in higher residual chlorophyll levels. Recent studies of the effect of temperature on degreening of Cavendish bananas failed to reach a definitive conclusion, although there was some indication that it was related to thylakoid ultrastructure (Dadzie and Orchard, 1997).

2.9.2.12 Changes in Flavour and Aroma

Changes in phenolics, carbohydrates and acids during ripening, contribute to the development of the flavour of bananas. However, many other chemicals such as amyl esters and butyl esters gave the fruit the distinctive banana flavour and aroma and a fruity flavour and aroma respectively. The exact relationship between the chemistry and the flavour and aroma of bananas has not yet been fully determined. However, besides the volatile compounds above, other esters as well as aldehydes, alcohols and ketones have been associated with fruit flavour and their production rates can increase during ripening (Burden, 1969 and Thompson, 1981).

2.10 POSTHARVEST DISEASE

Postharvest diseases can cause serious losses of fruits both in terms of quantity and quality. Fruits infected with disease have no market value. There are many postharvest diseases of banana such as crown rot, anthracnose, and finger rot. However, only the important disease such as crown rot is discussed (Dadzie and Orchard, 1997).

2.10.1 Crown Rot Disease

Crown rot is a disease which occurs in all banana growing regions. It is potentially the most serious postharvest problem, especially where dehanding and boxing of fruit is not carried out in modern centralized plants. A number of fungi have been isolated from

decaying crowns and the frequency of isolation varies between regions (Waller, 1981; Johanson and Jeger, 1993). The most common pathogens associated with crown rot are *Colletotrichum musae* (*Gloesporium musarum*), *Fusarium roseum*, *Fusarium semitectum* and *Botryodiplodia theobromae*. Other species include *Cephalosporium* sp., *Verticillium theobromae*, *Ceratocystis paradoxa* and *Phomopsis* sp. (Dadzie and Orchard, 1997). In addition, more than a dozen other fungi have been found in crown rot affected tissues.

In its natural state, the tough skin of banana protects the fruit against fungal diseases. But when the hands are cut from the stalk, the massive open wound created is an ideal weak spot for crown rot fungi to enter and grow. Crown rot fungi are everywhere, in the form of microscopically small spores. Fungal spores on the fruit in the field are carried along, after harvesting bunch, to the packinghouse. Spores follow the fruit right into delatexing baths, where they are drawn deeply into the weak spot, the wound on the crown tissue due to dehanding. Spores also remain on the outside of the fruits and are packed (Dadzie and Orchard, 1997). The spores germinate and cause rot of the crowns.

2.10.2 Symptoms

According to Dadzie and Orchard (1997) symptoms of crown rot are characterized by:

- Softening and blackening of tissues at the cut crown surface.
- White, grey or pink mould may form on the surface of the cut crown.
- Infected tissue turns black and the rot may advance into the finger stalk.
- Severely affected fingers may fall from the crown.

2.11 BOTANICALS USED AS TREATMENT

2.11.1 The Neem Tree

Neem is a member of the mahogany family, Meliaceae. It is currently known by the botanic name *Azadirachta indica* A. Juss. Previously, however, it was known by several names and some botanists formerly lumped it together with at least one of its relatives. The result is that the older literature is so confusing that it is sometimes impossible to determine just which species is being discussed (NRC, 1992).

2.11.2 Description

Neem trees are attractive broad-leaved evergreens that can grow up to 30m tall and 2.5m in girth. Their spreading branches form rounded crowns as much as 10m across. Neem trees remain in the leaf except during extreme drought, when the leaves may fall off. The short, usually straight trunk has a moderately thick, strong furrowed bark. The roots penetrate the soil deeply; at least where the site permits, and particularly when injured, they produce suckers. This suckering tends to be especially prolific in dry localities (NRC, 1992).

For example, it easily withstands pollarding (repeated lopping at heights above 1.5m) and its topped trunk resprouts vigorously. It is also freely coppiced (repeated lopping near-ground level). Regrowth from both pollarding and coppicing can be exceptionally fast because it is being served by a root system large enough to feed a full-grown tree. The small, white bisexual flowers are borne in axillary clusters. They have a honey like scent and attract many bees. Neem honey is popular and reportedly contains no traces of azadirachtin. The fruit is smooth, ellipsoidal drupe, up to almost 2cm long. When ripe, it is yellow or greenish yellow and comprises a sweet pulp enclosing a seed. The

seed is composed of a shell and a kernel (sometimes two or three kernels), each about half of the seed's weight (NRC, 1992).

Neem tree normally begins bearing fruit after 3-5 years, becomes fully productive in 10 years and from then on can produce up to 50kg of fruits annually. It may live for more than two centuries (NRC, 1992).

2.11.3 Distribution

Neem is thought to have originated in Assam and Burma (where it is common throughout the central dry zone and the Siwalik Hills). However, the exact origin is uncertain: some say neem is native to the whole Indian subcontinent: others attribute it to dry forest areas throughout all of South and Southeast Asia including Pakistan, Sri Lanka, Thailand, Malaysia and Indonesia. It is in India, however, that the tree is most widely used. It is grown from the southern tip of Kerala to the Himalayan hills in tropical to subtropical regions, in semiarid to wet tropical regions, and from sea level to about 700m elevation. It is now well established in at least 30 countries, particularly those in the regions along the Sahara's southern fringe, where it has become an important provider both for fuel and lumber. Although widely naturalised, it has nowhere become a pest. Indeed, it seems rather well domesticated. It appears to thrive in villages and towns. Over the last century or so, they have also been established in Fiji, Mauritius, the Caribbean and many countries of Central and South America (NRC, 1992). In some cases it was probably introduced by indentured labourers who remembered its value from their days of living in India's villages. In other cases it has been introduced by foresters. In the continental United States, small plantings are prospering in southern Florida and exploratory plots have been established in southern California and Arizona (NRC, 1992).

2.11.4 Propagation

The tree is easily propagated both sexually and vegetatively. It can be planted using seeds, seedlings, saplings, root suckers or tissue culture. However, it is normally grown from seed either planted directly on the site or transplanted as seedlings from a nursery. The fruit drops from the trees by itself; the pulp, when wet can be removed by rubbing against a coarse surface and after washing with water, the clean, white seeds are obtained. In certain nations, Togo and Senegal, for example, people leave the cleaning to fruit bats and birds, which feed on the sweet pulp and then spit out the seeds under the trees. It is reputed that neem seeds are not viable for long. It is generally considered that after 2-6 months in storage they will no longer germinate. However, some recent observations of seeds that had been stored in France indicated that seeds without endocarp had an acceptable germinative capacity (42 percent) after more than 5 years (NRC, 1992).

2.11.5 Growth

The tree is said to grow almost anywhere in the lowland tropics. However, it generally performs best in areas with annual rainfalls of 400-1,200mm. It thrives under the hottest conditions, where maximum shade temperature may soar past 50°C, but it will not withstand freezing or extended cold. It does well at elevations from sea level to perhaps 1000m near the equator. The tree also grows well on some acid soils. Indeed, it is said that the fallen neem leaves, which are slightly alkaline (pH 8.2) are good for neutralizing acidity in the soil. On the other hand, neem cannot stand wet feet and quickly dies if the site becomes waterlogged (NRC, 1992).

Neem often grows rapidly. Weeds seldom affect growth. Except in the case of very young plants, neem can dominate almost all competitors. In fact, they themselves may become weeds. They spread widely under favourable site conditions, since the seeds are distributed by birds, bats and baboons. For the same reason, natural regeneration under old trees is often abundant (NRC, 1992).

2.11.6 Effects of Neem on Fungi

Azadirachtin found in neem has demonstrated antifungal activity. Should this prove widely applicable, the availability of a natural fungicide that can be grown, extracted and applied by farmers themselves could be of great consequence to worldwide agriculture and food supply. Fungi attack crops in countless numbers and forms and they are constantly evolving enemies of farms and forests. Many fungi can reach epidemic proportions, a few have no cure and some can make certain crops impossible to grow. And despite the best of modern science, they still threaten wheat, corn, rice and other plants that feed the world. Not a lot is known about neem's practical use against rots, smuts, wilts, mildews, die-backs, and other fungal plant diseases. However, several tests have indicated considerable promise (NRC, 1992).

In one test, neem oil protected the seeds of chickpeas against the serious fungal diseases *Rhizoctonia solani*, *Sclerotium rolfsii* and *Sclerotinia sclerotiorum*. It also slowed the growth of *Fusarium oxysporum* but did not kill it. In addition, neem cake incorporated into the soil completely blocked the development of the resting forms of *Rhizoctonia solani* thereby interfering with the long-term survival of this devastating fungus. In another, neem-seed extracts showed beneficial effects against leaf fungi (NRC, 1992).

2.12 *Moringa oleifera*

Moringa oleifera Lam. (Horse-radish tree or Drumstick) is a medium-sized (about 10 metres high) tree belonging to the Moringaceae family. The Moringaceae is a single genus family with 14 known species, of these only *M. oleifera* (synonym *M. pterygosperma* Gaertn.) is the most widely known species and is planted in the whole tropical belt (Maroyi, 2006). The tree is indigenous to northern India and Pakistan commonly known as the 'horse-radish' tree, arising from the use of the root by Europeans in India as a substitute for horse-radish, *Cochlearia armoracia* (synonym *Armoracia rusticana*). Like *C. armoracia*, the roots of moringa are pungent and were commonly used as a condiment or garnish. Such a practice would not now be recommended as the root has been shown to contain 0.105% alkaloids, especially following ingestion. The other widely used common name is 'drumstick' tree, arising from the shape of the pods, resembling the slender and curved stick used for beating the drum (Oliver-Bever, 1986).

Common name of *M. oleifera* in Malabar is Moringo and this is the origin of the generic name. The early herbarium specimens document it as an ornamental tree, planted in public parks and private gardens. It has also been grown by Indian families as a vegetable. But moringa is now a permanent feature on the menu of the Tonga people of Zimbabwe, with its leaves being used as a spice when preparing food. *M. oleifera* may have been introduced in Zimbabwe during the European occupation or possibly long before Arab traders (Maroyi, 2006).

The highest concentrations of moringa tree are found in Binga District, where it is known as *Zakalanda* by the locals. It is believed that the tree was brought to Binga by the Indian traders using the Zambezi River in their search for gold, ivory and other items. It is now widely cultivated in several parts of the country and is naturalized in

many areas including the Zambezi valley. Although moringa tree is essentially not indigenous to Zimbabwe, it has become part of the traditional diet in Binga District and many other places. Reliable information regarding its utilization is crucial. Unfortunately in Zimbabwe, such data are often lacking, despite the economic importance of *M. oleifera*. Very little research has been done on the species although it is widely used by the rural poor as a food resource (Maroyi, 2006).

2.12.1 Description

Short, slender, deciduous, perennial tree, to about 10 m tall; rather slender with drooping branches; branches and stems brittle, with corky bark; leaves feathery, pale green, compound, tripinnate, 30–60 cm long, with many small leaflets, 1.3–2cm long, 0.6–0.3cm wide, lateral ones somewhat elliptic, terminal one obovate and slightly larger than the lateral ones; flowers fragrant, white or creamy-white, 2.5cm in diameter, borne in sprays, with 5 at the top of the flower; stamens yellow; pods pendulous, brown, triangular, splitting lengthwise into 3 parts when dry, 30–120cm long, 1.8cm wide, containing about 20 seeds embedded in the pith, pod tapering at both ends, 9-ribbed; seeds dark brown, with 3 papery wings. Main root thick. Fruit production in March and April in Sri Lanka (Duke, 1983). The leaves contain the potent antibiotic and fungicide pterygospermin (Oliver-Bever, 1986).

2.12.2 Germplasm

Reported from the African and Hindustani Centers of Diversity, Moringa or cultivars thereof is reported to tolerate drought, laterite, sand, fungus, bacteria and mycobacteria (Duke, 1983). Several cultivars are grown: 'Bombay' is considered one of the best, with

curly fruits. Others have the fruits 3-angled or about round in cross-section. In India, 'Jaffna' is noted for having fruits 60–90 cm, 'Chavakacheri murunga' 90–120cm long. ($2n = 28$) (Duke, 1983).

2.12.3 Distribution

Native to India, Arabia, and possibly Africa and the East Indies; widely cultivated and naturalized in tropical Africa, tropical America, Sri Lanka, India, Mexico, Malabar, Malaysia and the Philippine Islands(Duke, 1983).

2.12.4 Ecology

Ranging from subtropical dry to moist through tropical very dry to moist forest life zones, Moringa is reported to tolerate annual precipitation of 4.8 to 40.3dm annual temperature of 18.7 to 28.5°C and pH of 4.5 to 8. Thrives in subtropical and tropical climates, flowering and fruiting freely and continuously. It grows best on dry sandy soil and is drought resistant (Duke, 1983).

2.12.5 Cultivation

In India, the plant is propagated by planting limb cuttings 1–2m long, from June to August, preferably. The plant starts bearing pods 6–8 months after planting but regular bearing commences after the second year. The tree bears for several years (Duke, 1983).

2.12.6 Harvesting

Fruit or other parts of the plant are usually harvested as desired according to some authors, but in India, fruiting may peak between March and April and again in

September and October. Seeds are gathered in March and April and oil obtained from the seeds (Duke, 1983).

2.13 *Cassia alata*

Cassia alata L. (family: Leguminosae) is a shrub, employed in traditional medicine in many parts of India and West Indies for the treatment of various ailments. The plant is native to Southeast Asia, Fiji, Northern Australia, Africa and Latin America. It is known as gelenggang (Malaysia), Mbai Ni Thangi (Fiji), Ringworm bush (Australia), Ketepeng Badak (Indonesia) and Te'elango (Tonga). It grows aggressively in areas with high water table and sunlight. Often, it forms thickest and also, may be grown as ornamental. Decoctions of the leaves, flowers, bark and wood are used in treating skin diseases such as eczema, pruritis, itching and in constipation. The flowers are also used in the treatment of bronchitis and asthma (Elysha Nur *et al.*, 2002).

Cassia alata is used in different ways and for different purposes. The bioactivity of this plant includes antibacterial, antifungal, antimicrobial, diuretic, laxative, analgesic and choleretic (Elysha Nur *et al.*, 2002).

2.13.1 Chemical Constituents

Cassia alata plants contain many diverse constituents such as alkaloids, lectins, saponins, cyanogenic, glycosides, isoflavones and phytoestrogens (Reezal *et al.*, 2002).

Two new glycosides were isolated from the seeds of *Cassia alata* namely chrysoeriol-7-

0-(2''-0-beta-D-mannopyranosyl)-beta-D-allopyranoside and rhamnetin-3-0-(2''-0-beta-

D-mannopyranosyl)-beta-D-allopyranoside (Reezal *et al.*, 2002).

2.14 *Zingiber officinale*

Ginger is one of the most traded spices globally. Ginger derives its name from Sanskrit word stringa-vera, which means "with a body like a horn". The spice is used extensively for culinary and medicinal application. Though often thought of as the root of the plant, it actually is the rhizome of the *Zingiber officinale* plant. The spice was initially found in China, but later the production extended to other Asian countries including India (Anonymous, 2009).

2.14.1 Origin and Distribution

Ginger is native to southern parts of China but now it is cultivated in all the tropical and sub tropical regions of the world. Nearly half of the world production comes from India. Other major producers are Brazil, Jamaica, Nigeria, Thailand, Australia and Fiji (Anonymous, 2009).

2.14.2 Chemical Composition

The oil content of the ginger is responsible for its fragrance. The main chemical constituents include sesquiterpenoids and zingiberene. Some other constituents includes β -sesquiphellandrene, bisabolene, farnesene and a small monoterpenoid fraction like β sphelladrene, cineol, and citral. Pungency is determined by non-volatile phenylpropanoids and diarylheptanoids (Anonymous, 2009).

2.14.3 Description of Ginger

Rhizome of Ginger is knobbly and fleshy that is covered in rings. This part is used in food and medicine. Rhizomes grow underground but they are not roots but stem. Purple

flowers grow directly from the rhizomes and are generally 30 cm long. Fruits of ginger are red in colour and have three chambers with number of small seeds (Anonymous, 2009).

2.14.4 Medicinal Properties

Ginger is also said to possess certain medicinal properties which aids in digestion and cure digestion related problems, diarrhoea and stomach cramps. Ginger is effective against nausea related to both motion sickness and morning sickness and in relieving pain and reduces inflammation. It reduces arthritis, rheumatism and muscle spasms. Ginger also circulates the blood, removes toxins, cleanse the bowels and kidneys and aids in all skin related problems (Anonymous, 2009).

It is said that it also aid in the treatment of asthma, bronchitis and other respiratory problems (Anonymous, 2009).

2.15 Mancozeb (Chemical Fungicide)

Mancozeb is classified as a contact fungicide with preventive activity. It inhibits enzyme activity in fungi by forming a complex with metal-containing enzymes including those involved in production of adenosine triphosphate (ATP). Mancozeb is used to protect many fruit, vegetable, nut and field crops against a wide spectrum of fungal diseases, including potato blight, leaf spot, scab on apples and pears, and rust on roses. It is also used for seed treatment of cotton, potatoes, corn, safflower, sorghum, peanuts, tomatoes, flax and cereal grains (Anonymous, 2005).

2.16 Shavit F71.5WP (Chemical Fungicide)

Shavit F71.5 WP is systemic fungicides with protective and curative action against powdery mildew. Shavit F71.5 WP is a fungicide combining the systemic properties of triadimenol with the contact properties of folpet. Triadimenol belongs to the family of triazole fungicide and as such it is an inhibitor of biosynthesis of ergosterol. Triadimenol moves upwards in the vascular system and is distributed to give full protection to new shoots. On the other hand folpet acts as a protectant and has a contact action. It binds to sulphur-hydrogen bond, thereby interfering with respiration process in fungi (Anonymous, 2010).

3.0 MATERIALS AND METHODS

The work was conducted at the Pathology Laboratories of Crops Research Institute (CRI) of the Centre for Scientific and Industrial Research (CSIR) at Fumesua in Kumasi from August 2008 to March 2009. The work was divided into two parts. The first part involved the isolation and identification of crown rot pathogens from Atiwa and Asuogyaman districts in the Eastern region of Ghana.

The second part involved the development of control methods (basically the use of botanical and chemical fungicides) and the effect of these treatments on the quality (total soluble solids, pH, total titratable acidity, weight loss and pulp to peel ratio) of Medium Cavendish banana.

3.1 EXPERIMENT ONE: COLLECTION AND IDENTIFICATION OF CROWN ROT ORGANISMS

This aspect of the experiment dealt with preparation of potato dextrose agar (PDA) for culturing the organisms, isolation of organisms from infected crowns of banana samples and pathogenicity test to confirm the rot causing organism (s).

3.1.1 Preparation of Potato Dextrose Agar (PDA) Medium

Two hundred grams (200g) of Irish potato were washed freely under running water. The tubers were cut into smaller pieces and boiled until the pieces were soft. The soft pieces were mashed and distil water added to make one litre suspension. Twenty grams (20g) of glucose and twenty grams (20g) agar were added to the potato suspension and stirred using a magnetic stirrer. The mixture was next sterilized by autoclaving at 121°C and a pressure of 100 p. s. i. for 15 minutes (Johnston and Booth, 1983).

After autoclaving, the medium was allowed to cool. The medium was next dispensed into 9cm diameter Petri dishes in a sterile Lamina Flow Chamber. The media poured into the Petri dishes were stored in polythene bags in a refrigerator and used when required.

3.1.2 Isolation of Fungal Organisms from Infected Crowns

Banana fruits with infected crowns were collected from several locations in the Atiwa and Asuogyaman districts in the Eastern region of Ghana. Pieces of crown tissues were cut from hands of banana. The pieces obtained were surface sterilized in a 5% sodium hypochlorite solution for five minutes. After sterilization the pieces were washed three times with sterile distilled water. The pieces were dried in a sterile Lamina Flow Chamber, placed on PDA in Petri dishes and after that incubated at 28°C in an incubation room. The pieces were observed daily for fungal growth. Fungal tissues that grew from the pieces were subcultured to obtain pure single fungal species.

The fungal isolates were maintained on potato dextrose agar through regular subculturing. Morphological and cultural characteristics including shape, size and colour of conidia of the isolates were examined under the microscope after incubation at 28°C for 14 days on potato dextrose agar. The organisms were identified as *Botryodiplodia theobromae*, *Aspergillus flavus* and *Aspergillus niger*.

3.1.3 Pathogenicity Test

After obtaining pure cultures of the fungal isolate, fresh healthy mature green Medium Cavendish banana bunches were obtained from Banana Market in Kumasi. The bunches were dehanded into smaller hands containing 10 fingers each. The cut surface of the hands were washed with distilled water to remove the latex and thereafter sterilized with 90% sodium hypochlorite solution. Circular discs were removed from the crown region of Medium Cavendish banana hand with cork borer (No. 5). A disc of 14-days old cultures of the isolates were used to plug the holes created in the crown region. The inoculated banana hands were incubated at room temperature and daily observation was

made. A re-isolation was made on potato dextrose agar (PDA) from the rotted portion of the inoculated banana hands and the isolates were compared with the original cultures of the fungi for confirmation as the rot causing organism. Complete Pathogenicity test showed that, the isolates of *Aspergillus flavus* and *Aspergillus niger* were nonpathogenic whereas that of *Botryodiplodia theobromae* were pathogenic.

3.2 EXPERIMENT TWO: TEST TO ESTABLISH EFFICACY OF BOTANICAL AND CHEMICAL FUNGICIDES

This aspect of the experiment also dealt with preparation of botanical and chemical fungicides for controlling the organisms isolated, assessment of disease incidence and studies of physico-chemical parameters of treated banana samples during storage.

3.2.1 Preparation of Botanical Extracts

Fresh leaves of *Moringa oleifera* and *Cassia alata*, seeds of *Azadirachta indica* and rhizome of *Zingiber officinale* (rhizome) were each washed with tap water before 100g of each sample was weighed and blended with 150ml of distilled water and then filtered with clean cheese cloth to obtain the extracts (Okigbo and Nmeko, 2005; Nwachukwu and Osuji, 2008). The extracts were applied on the crowns of Medium Cavendish banana hands immediately after they were prepared.

3.2.2 Preparation of Chemical Fungicide

3.2.2.1 ShavitF71.5WP (a.i., Metalaxyl)

ShavitF71.5WP fungicide was prepared according to the manufacturer's (Makhteshim Chemical Works Limited, Israel) instruction by dissolving 2.0g of the powder in a litre of water.

3.2.2.2 Mancozeb (a.i., Mancozeb)

Mancozeb fungicide was prepared according to the manufacturer's (Jiangsu Holly Corporation, China) instruction by dissolving 7.0g of the powder in a litre of water.

3.2.3 SOURCE OF BANANA FOR STUDY

Mature green Medium Cavendish banana bunches which were unbruised and healthy were purchased from a Banana market in Kumasi and quickly transported in the early hours of the morning to the Plant Pathology Laboratory of Crops Research Institute, Fumesua, Kumasi.

3.2.4 PREPARATION OF BANANA HANDS AND APPLICATION OF TREATMENTS

Banana bunches brought to the laboratory were dehanded with a sharp knife. The hands were sorted for uniformity of shape, colour, size and the absence of physical defects. Afterwards, they were washed with distilled water to remove the latex and allowed to air dry. Banana hands were divided into 24 samples having 8 treatments with 3 replications. Inoculation wounds created on the banana crowns were applied with either Moringa, Neem, Cassia or Ginger extracts using a 1000ppm micropipette. 14-day old isolate of *Botryodiplodia theobromae* was inserted into the inoculation wound and extracts of the botanicals were re-applied to the crowns of the banana hands. The procedure was repeated for Shavit F71.5WP and Mancozeb samples. Uninoculated and untreated banana samples were used as control.

Banana hands which were treated were assessed during ripening for disease severity using a crown rot index developed by Sarananda and Wijeratnam (1994) where; 0= no

rot, 1= 25% of the crown affected, 2= 50% of the crown affected, 3= 75% of the crown affected and 4= 100% of the crown affected with rot extending into the finger stalk.

The length travelled by the pathogen from the crown region to the peel of the finger was also measured and recorded in centimetres.

3.2.5 STUDIES OF PHYSICO-CHEMICAL CHANGES DURING RIPENING OF

BANANA

Fruit samples from each treatment were assessed daily for determination of total soluble solids, pH and total titratable acidity.

3.2.5.1 Total soluble solids

Pulp was cut into pieces and thirty grams (30g) of the pulp was measured using a toploading electronic balance (HD-801). The weighed banana pulp was blended with 90ml distilled water using a kitchen blender.

The mixture was then filtered using a clean cheese cloth to obtain the filtrate. A single drop of the filtrate was placed on the prism of a hand-held refractometer (DIGIT-080) (AOAC, 1990).

3.2.5.2 pH

Pulp was cut into pieces and thirty grams (30g) of the pulp was measured using a toploading electronic balance (HD-801). This was blended with 90ml distilled water using a kitchen blender.

The mixture was then filtered using a clean cheese cloth to obtain the filtrate. The filtrate was then poured into test tubes and the pH meter (HM-16S) electrode washed in distilled water and placed in the filtrate. The pH value of the filtrate was read and recorded after the reading has stabilized (AOAC, 1990).

3.2.5.3 Total titratable acidity

Pulp was cut into pieces and thirty grams (30g) of the pulp was measured using a toploading electronic balance (HD-801). The weighed banana pulp was blended with 90ml distilled water using a kitchen blender.

The mixture was then filtered using a clean cheese cloth to obtain the filtrate. 25ml of the filtrate was pipetted into 200ml conical flask and 25ml of distilled water was added. Four drops of phenolphthalein indicator was added and titrated against 0.1N Sodium hydroxide (NaOH) (AOAC, 1990).

3.2.5.4 Weight loss

Weight loss was measured by taking the initial weight of each treated samples (start of experiment) and the weight at each observation period. Weight loss was calculated as percentage of the initial weight.

$$\text{Weight loss (\%)} = \frac{W1 - W2}{W1} \times 100$$

Where W1 = initial weight, and W2 = final weight.

3.2.5.5 Pulp to peel ratio

Pulp to peel ratio of treated banana samples were determined by weighing the pulp and peel separately using a top-loading electronic balance (HD-801). The pulp to peel ratio was calculated as: pulp weight/peel weight.

3.3 EXPERIMENTAL DESIGN

Completely randomised design (CRD) with three replications was used and each replicate consisted of one hand of unbruised, healthy banana with 10 fruits. The results obtained were analysed using SAS computer package and the difference between treatments means were separated using least significance difference at 5%.

4.0 RESULTS

4.1 EXPERIMENT ONE: COLLECTION AND IDENTIFICATION OF CROWN

ROT ORGANISMS

Three organisms; *Botryodiplodia theobromae*, *Aspergillus flavus* and *Aspergillus niger* were identified. Out of these three organisms, *Botryodiplodia theobromae* was determined to be the rot causing organism.

4.1.1 Characterization of Fungal Isolates from Crown Rot Tissues

4.1.1.1 *Botryodiplodia theobromae*

Three fungal species were isolated from the crown rot tissues. The first isolate produced greyish black cultures. This fungus produced pycnidia on old cultures. The pycnidia formed by this fungus could be seen with the naked eye as black projections on culture plates (Plate 1). The spores were oval and compared to other spores were relatively big. Mature spores were dark and each has a single septum (Plate 2). Young, immature spores were colourless and unicellular (Plate 3). The presence of the mature spores allowed a proper identification of this fungus. The fast and constantly growing mycelium of the fungus was snow-white at first, becoming greyish black over time.



Plate 1: *Botryodiplodia theobromae* on PDA 14 days old culture showing pycnidia

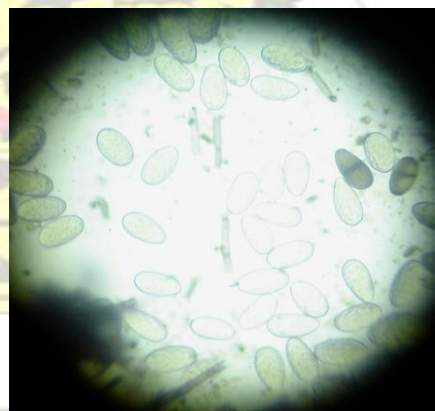


Plate 2: Mature and immature spores of *Botryodiplodia theobromae*



Plate 3: Immature spores of
Botryodiplodia theobromae

4.1.1.2 *Aspergillus flavus*

The second fungal isolate produced olive green cultures containing millions of spores (Plate 4). Spores produced by the isolate were globose to subglobose and green in colour under the microscope (Plate 5).

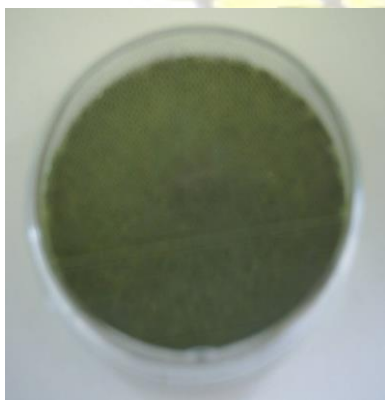


Plate 4: *Aspergillus flavus* in culture
10 days



Plate 5: Spores of *Aspergillus flavus* after

4.1.1.3 *Aspergillus niger*

The third isolate produced black cultures (Plate 6). The spores of this isolate were black when observed under the microscope (Plate 7).



Plate 6: *Aspergillus niger* in culture after 10 days

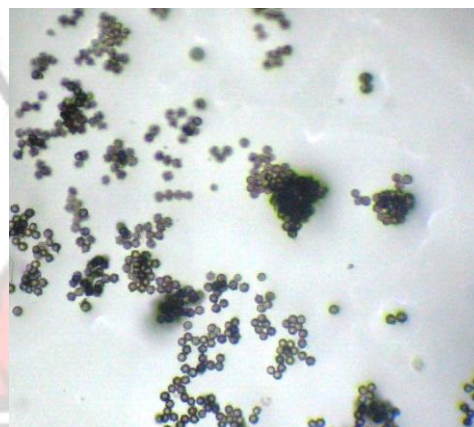


Plate 7: Spores of *Aspergillus niger*

4.1.2 PATHOGENICITY TEST

In the pathogenicity test, matured green banana inoculated with *Botryodiplodia theobromae* developed symptoms of crown rot disease five days after incubation. However, *Aspergillus flavus* and *Aspergillus niger* inoculated singly on healthy banana did not show any symptoms of the disease.

4.2 EXPERIMENT TWO: TEST TO ESTABLISH EFFICACY OF BOTANICAL

AND CHEMICAL FUNGICIDES

Of the four botanical fungicides, that is Moringa (*Moringa oleifera*) extract, Neem (*Azadirachta indica*) extract, Cassia (*Cassia alata*) extract and Ginger (*Zingiber officinale*) extract tested against *Botryodiplodia theobromae*, Ginger extract was able to

control the rot causing organism. Shavit F71.5WP (a.i., Metalaxyl) and Mancozeb (a.i., Mancozeb) as chemical fungicides were able to control *Botryodiplodia theobromae* but Shavit F71.5WP was found to be superior to Mancozeb.

4.2.1 Efficacy of Botanical and Chemical Fungicides

Table 4.1a showed the area of the crown region covered by the rot pathogen of treated bananas. Samples treated with Moringa extract, Neem extract and Cassia extract had 25% of their crown region affected with the rot pathogen by day 3 whereas samples treated with Ginger extract also had 25% of its crown region affected with the rot pathogen by day 4. The area of the crown region affected with the rot pathogen for samples treated with botanical fungicides increased with storage days.

Table 4.1a: Daily development of crown rot on treated banana

Days	1	2	3	4	5	6	7	8	9	10
Treatments										
Uninoculated and untreated	0.00	0.00	0.00	1.00	2.00	2.00	3.00	3.00	3.00	3.00
Inoculated and untreated	0.00	0.00	0.00	1.00	3.00	3.00	4.00	4.00	4.00	4.00
<i>Moringa oleifera</i> (leaf extract)	0.00	0.00	1.00	2.00	3.00	3.00	4.00	4.00	4.00	4.00
<i>Azadirachta indica</i> (seed extract)	0.00	0.00	1.00	2.00	4.00	4.00	4.00	4.00	4.00	4.00
<i>Zingiber officinale</i> (rhizome extract)	0.00	0.00	0.00	0.00	1.00	2.00	2.00	3.00	4.00	4.00
<i>Cassia alata</i> (leaf extract)	0.00	0.00	1.00	2.00	4.00	4.00	4.00	4.00	4.00	4.00
Mancozeb	0.00	0.00	0.00	0.00	0.00	1.00	2.00	3.00	4.00	4.00
ShavitF71.5WP	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.00	2.00	2.00

Scale: 0= no rot, 1=25% of the crown affected, 2=50% of the crown affected, 3=75% of the crown affected and 4=100% of the crown affected with rot extending into the finger stalk (after Sarananda and Wijeratnam, 1994)

For samples treated with Mancozeb, there was no rot on the crown region until day 6 where 25% of the crown region was affected with the rot pathogen.

Similarly, samples treated with ShavitF71.5WP also had no rot on the crown until day 8 where 25% of its crown region affected with the rot pathogen. Table 4.2a indicated that infection of the pedicel by the rot pathogen was evident at day 5 for samples treated with Moringa extract, Neem extract and Cassia extract whereas that of Ginger extract was evident at day 8 and thereafter infection increased with storage days. For samples treated with Mancozeb and ShavitF71.5WP, pedicel infection was evident at days 9 and 10, respectively (Table 4.2a). Plates 8 to 15 show Medium Cavendish banana hands that have been inoculated, treated and incubated.



Plate 8: Uninoculated and untreated
(7 days in storage)



Plate 9: Inoculated and untreated banana
(7 days in storage)



Plate 10: Neem treated banana

(7 days in storage)



Plate 11: Cassia treated banana

(7 days in storage)



Plate 12: Ginger treated banana (7 days in storage)



Plate 13: Moringa treated banana (7 days in storage)



Plate 14: Mancozeb treated banana (7 days in storage)



Plate 15: Shavit F71.5WP treated banana (7 days in storage)

There were significant differences ($P < 0.05$) among samples treated with botanical fungicides. Treatment of banana samples with Ginger extract were the most effective as mean crown rot score was 1.60 with a mean infection (0.46cm) of the rot pathogen extending into the pedicel. However, samples treated with Moringa extract, Neem extract and Cassia extract were less effective as mean crown rot scores were 2.43, 2.70 and 2.70, respectively, and there were no significant differences ($P > 0.05$) in their performance in controlling the crown rot pathogen. Infection of the rot pathogen into the pedicel of samples treated with Moringa extract, Neem extract and Cassia extract were

1.37cm, 2.41cm and 3.39cm respectively (Table 4.1b and 4.2b).

Table 4.1b: Mean crown rot disease severity score

Treatments	
Uninoculated and untreated	1.66b
Inoculated and untreated	2.30a
<i>Moringa oleifera</i> (leaf extract)	2.43a
<i>Azadirachta indica</i> (seed extract)	2.70a
<i>Zingiber officinale</i> (rhizome extract)	1.60b
<i>Cassia alata</i> (leaf extract)	2.70a
Mancozeb	1.33b
ShavitF71.5WP	0.50c

CV% = 15.53, Grand mean = 1.90

Means with the same letter are not significantly different ($P < 0.05$)

Mean separation by Least Significant Difference

By the 10th day crown rot infection had developed well beyond one centimetre into the pedicel for banana samples treated with Moringa, Neem, Cassia and Ginger extracts, Inoculated and untreated and Mancozeb with the exception of Shavit F71.5WP which was less than one centimetre. Those that were uninoculated and untreated had no crown rot infection (Table 4.2a).

Table 4.2a: Crown rot infections (cm) into pedicels of treated banana

Days	1	2	3	4	5	6	7	8	9	10
Treatments										
Uninoculated and untreated	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Inoculated and untreated	0.00	0.00	0.00	0.00	0.39	1.02	2.43	3.07	4.21	6.35
<i>Moringa oleifera</i> (leaf extract)	0.00	0.00	0.00	0.00	0.43	0.51	1.81	2.52	3.50	4.95
<i>Azadirachta indica</i> (seed extract)	0.00	0.00	0.00	0.00	1.61	2.39	3.41	4.35	5.62	6.71
<i>Zingiber officinale</i> (rhizome extract)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.47	1.63	2.53
<i>Cassia alata</i> (leaf extract)	0.00	0.00	0.00	0.00	1.99	2.82	4.97	6.40	7.94	9.79
Mancozeb	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.71	1.22
ShavitF71.5WP	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.82

There was significant difference ($P<0.05$) between banana samples treated with Ginger extract and Inoculated and untreated but no significant differences ($P>0.05$) were observed between Ginger treated samples and Uninoculated and untreated.

Samples treated with ShavitF71.5WP and Mancozeb were effective as mean crown rot scores were 0.50 and 1.33 respectively. Infection of the rot pathogen into the pedicel was 0.08cm for samples treated with ShavitF71.5WP and 0.19cm for samples treated with Mancozeb, indicating that ShavitF71.5WP was more effective than Mancozeb.

Table 4.2b: Effect of treatments on mean crown rot infection (cm)

Treatments	
Uninoculated and untreated	0.00d
Inoculated and untreated	1.74c
<i>Moringa oleifera</i> (leaf extract)	1.37c
<i>Azadirachta indica</i> (seed extract)	2.41b
<i>Zingiber officinale</i> (rhizome extract)	0.46d
<i>Cassia alata</i> (leaf extract)	3.39a
Mancozeb	0.19d

ShavitF71.5WP	0.08d
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CV% = 24.06, Grand mean = 1.21

Means with the same letter are not significantly different ($P < 0.05$)

Mean separation by Least Significant Difference

4.2.2 Effect of Treatments on Total Soluble Solids

Table 4.3a showed the daily total soluble solids changes of treated banana samples. Starch conversion to sugar started from day 4. There were increases in total soluble solids for all treatments until the 7th or 8th day when the values started declining. The Inoculated and untreated recorded the highest total soluble solids of 15.00 by day 6 earlier than all the other treatments. Banana samples treated with Neem extract (15.00), Cassia extract (15.00) and Uninoculated and untreated recorded their highest total soluble solids at day 7. Samples treated with Moringa extract (14.00) and Ginger extract (15.00) recorded their highest total soluble solids at day 8. Banana samples treated with Shavit F71.5WP (13.00) and Mancozeb (12.00) also recorded their highest total soluble solids at days 9 and 10 respectively.

With the passage of time total soluble solids for most treatments had increased and reaching their peak and thereafter a decline in the total soluble solids.

The mean total soluble solids of treated samples are represented in Table 4.3b. Banana samples treated with botanical fungicides showed significant differences ($P < 0.05$). Samples treated with Cassia extract recorded the highest mean total soluble solids of 8.50 and was significantly different from Ginger extract but was not significantly different ($P > 0.05$) from Neem extract, Moringa extract, Uninoculated and untreated and Inoculated and untreated. However, there were no significant differences ($P > 0.05$) in mean total soluble solids between ShavitF71.5WP and Mancozeb but significant differences ($P < 0.05$) were observed when compared to Uninoculated and untreated.

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Table 4.3a: Daily total soluble solids (°Brix) of banana fruits

Days	1	2	3	4	5	6	7	8	9	10
Treatments										
Uninoculated and untreated	0.00	0.00	0.00	8.00	12.00	13.00	14.00	13.00	14.00	11.00
Inoculated and untreated	0.00	0.00	0.00	8.00	13.00	15.00	15.00	14.00	12.00	11.00
<i>Moringa oleifera</i> (leaf extract)	0.00	0.00	0.00	0.00	7.00	12.00	13.00	14.00	13.00	12.00
<i>Azadirachta indica</i> (seed extract)	0.00	0.00	0.00	3.00	11.00	14.00	15.00	14.00	11.00	10.00
<i>Zingiber officinale</i> (rhizome extract)	0.00	0.00	0.00	0.00	2.00	9.00	13.00	15.00	14.00	12.00
<i>Cassia alata</i> (leaf extract)	0.00	0.00	0.00	5.00	13.00	13.00	15.00	14.00	14.00	11.00
Mancozeb	0.00	0.00	0.00	0.00	3.00	7.00	9.00	11.00	10.00	12.00
ShavitF71.5WP	0.00	0.00	0.00	0.00	0.00	7.00	9.00	11.00	13.00	12.00

Table 4.3b: Effect of treatments on mean total soluble solids

Treatments	
Uninoculated and untreated	8.50a
Inoculated and untreated	8.80a
<i>Moringa oleifera</i> (leaf extract)	7.10abc
<i>Azadirachta indica</i> (seed extract)	7.80ab
<i>Zingiber officinale</i> (rhizome extract)	6.50bc
<i>Cassia alata</i> (leaf extract)	8.50a
Mancozeb	5.20c
ShavitF71.5WP	5.20c

CV% = 15.26, Grand mean = 7.20

Means with the same letter are not significantly different ($P < 0.05$) Mean separation by Least Significant Difference

4.2.3 Effect of Treatments on pH pH changes in banana samples treated with botanical and chemical fungicides, Inoculated and untreated as well as Uninoculated and untreated are shown in Table 4.4a. Banana samples treated with Cassia extract recorded the highest mean pH of 5.02, followed by Uninoculated and untreated, with Neem extract and ShavitF71.5WP recording the lowest mean pH of 4.88. However, the differences observed among all the treatments were not significant ($P > 0.05$) (Table 4.4b).

Table 4.4a: Daily pH of treated banana samples

Days	1	2	3	4	5	6	7	8	9	10
Treatments										
Uninoculated and untreated	5.05	5.08	5.02	4.84	4.77	4.74	4.85	4.96	5.20	5.41
Inoculated and untreated	5.05	4.99	4.97	4.51	4.61	4.74	4.80	4.90	5.18	5.33
<i>Moringa oleifera</i> (leaf extract)	5.05	5.03	4.85	4.87	4.90	4.88	4.84	4.82	4.95	5.10
<i>Azadirachta indica</i> (seed extract)	5.05	5.03	4.95	4.80	4.62	4.70	4.77	4.87	5.00	5.03
<i>Zingiber officinale</i> (rhizome extract)	5.05	5.11	5.06	5.02	4.89	4.81	4.82	4.87	5.00	5.20
<i>Cassia alata</i> (leaf extract)	5.05	5.09	5.11	4.74	4.71	4.76	5.00	5.09	5.20	5.45
Mancozeb	5.05	5.00	5.01	5.00	4.96	4.80	4.83	4.74	4.84	4.93
ShavitF71.5WP	5.05	4.96	5.04	4.98	5.12	4.82	4.74	4.66	4.66	4.83

Table 4.4b: Effect of treatments on mean pH

Treatments	
Uninoculated and untreated	4.99a
Inoculated and untreated	4.90a
<i>Moringa oleifera</i> (leaf extract)	4.92a
<i>Azadirachta indica</i> (seed extract)	4.88a
<i>Zingiber officinale</i> (rhizome extract)	4.97a
<i>Cassia alata</i> (leaf extract)	5.02a

Mancozeb	4.91a
ShavitF71.5WP	4.88a

CV% = 1.36, Grand mean = 4.94

Means with the same letter are not significantly different ($P > 0.05$) Mean separation by Least Significant Difference

4.2.4 Effect of Treatments on Total Titratable Acidity

There was a rise and fall in total titratable acidity of banana samples during the experimental period of 10 days as shown in Table 4.5a.

Table 4.5b showed the mean effects of the various treatments on total titratable acidity of banana. The results showed significant differences ($P < 0.05$) among the treatments. Banana samples treated with Cassia extract recorded the highest mean acidity of 0.19 and was not significantly different ($P > 0.05$) from Ginger extract, Mancozeb and Uninoculated and untreated but significantly different ($P < 0.05$) from samples treated with Neem extract which recorded the lowest mean acidity of 0.15. There were no significant difference ($P > 0.05$) between samples treated with Mancozeb and ShavitF71.5WP as their mean acidity were similar.

Table 4.5a: Total titratable acidity (milliequivalent) of banana fruits

Days	1	2	3	4	5	6	7	8	9	10
Treatments										
Uninoculated and untreated	0.14	0.19	0.17	0.20	0.27	0.21	0.17	0.16	0.20	0.14
Inoculated and untreated	0.14	0.15	0.14	0.23	0.27	0.16	0.14	0.15	0.13	0.15
<i>Moringa oleifera</i> (leaf extract)	0.14	0.17	0.19	0.17	0.21	0.19	0.16	0.17	0.18	0.17
<i>Azadirachta indica</i> (seed extract)	0.14	0.15	0.14	0.15	0.24	0.18	0.14	0.16	0.13	0.16
<i>Zingiber officinale</i> (rhizome extract)	0.14	0.17	0.17	0.16	0.24	0.21	0.18	0.20	0.17	0.17

<i>Cassia alata</i> (leaf extract)	0.14	0.22	0.19	0.23	0.27	0.21	0.14	0.19	0.20	0.15
Mancozeb	0.14	0.16	0.15	0.16	0.20	0.17	0.18	0.23	0.20	0.22
ShavitF71.5WP	0.14	0.16	0.14	0.15	0.16	0.16	0.20	0.26	0.20	0.21

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Table 4.5b: Effect of treatments on mean total titratable acidity

Treatments	
Uninoculated and untreated	0.18ab
Inoculated and untreated	0.16cd
<i>Moringa oleifera</i> (leaf extract)	0.17bcd
<i>Azadirachta indica</i> (seed extract)	0.15d
<i>Zingiber officinale</i> (rhizome extract)	0.18abc
<i>Cassia alata</i> (leaf extract)	0.19a
Mancozeb	0.18abc
ShavitF71.5WP	0.17cd

CV% = 5.85, Grand mean = 0.18

Means with the same letter are not significantly different ($P < 0.05$)

Mean separation by Least Significant Difference

4.2.5 Effect of Treatments on Percentage Weight Loss

Percentage weight loss from all treatments increased with storage days as shown in Table 4.6a.

Table 4.6b showed the mean effect of the various treatments on mean percentage weight loss of banana samples. The results showed no significant differences ($P>0.05$) among the treatments. Samples treated with Neem extract recorded the highest mean percentage weight loss of 8.34. Banana samples treated with Moringa extract recorded the lowest mean percentage weight loss of 6.74.

Samples treated with ShavitF71.5WP (7.50) and Mancozeb (6.94) also recorded percentage weights loss but there were no significant differences ($P>0.05$).

Table 4.6a: Cumulative weight loss (%) of banana fruits

Days	1	2	3	4	5	6	7	8	9	10
Treatments										
Uninoculated and untreated	0.00	1.05	2.47	4.17	5.43	6.68	7.93	9.14	10.71	12.37
Inoculated and untreated	0.00	1.20	2.79	4.98	6.31	8.01	9.75	11.54	13.33	14.94
<i>Moringa oleifera</i> (leaf extract)	0.00	1.22	2.53	4.38	5.75	7.37	9.02	10.68	12.38	14.10
<i>Azadirachta indica</i> (seed extract)	0.00	1.29	2.95	5.15	7.07	9.16	11.27	13.45	15.53	17.61
<i>Zingiber officinale</i> (rhizome extract)	0.00	1.32	3.07	5.13	6.72	8.42	10.08	11.93	13.70	15.65
<i>Cassia alata</i> (leaf extract)	0.00	1.17	2.69	4.75	6.55	8.60	10.67	12.75	14.98	17.55
Mancozeb	0.00	1.27	3.08	5.01	6.40	7.84	9.28	10.72	12.15	13.73

ShavitF71.5WP	0.00	1.20	2.97	5.10	6.67	8.46	10.03	11.82	13.58	15.25
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Table 4.6b: Effect of treatments on mean percentage weight loss

Treatments	
Uninoculated and untreated	5.99a
Inoculated and untreated	7.28a
<i>Moringa oleifera</i> (leaf extract)	6.74a
<i>Azadirachta indica</i> (seed extract)	8.34a
<i>Zingiber officinale</i> (rhizome extract)	7.60a
<i>Cassia alata</i> (leaf extract)	7.97a
Mancozeb	6.94a
ShavitF71.5WP	7.50a

CV% = 14.56, Grand mean = 7.30

Means with the same letter are not significantly different ($P > 0.05$)

Mean separation by Least Significant Difference

4.2.6 Effect of Treatments on Pulp to Peel Ratio

With the passage of time samples treated with botanical and chemical fungicides showed an irregular pattern of increase and decrease in pulp to peel ratio during storage (Table 4.7a).

Table 4.7b showed the mean effect of the various treatments on pulp to peel ratio of banana samples. There were no significant differences ($P > 0.05$) among the treatments.

Samples treated with Ginger extract recorded the highest mean pulp to peel ratio of 1.85 with Neem extract recording the lowest mean pulp to peel ratio of 1.65 (Table 4.7b).

Table 4.7a: Pulp to peel ratio of banana fruits

Days	1	2	3	4	5	6	7	8	9	10
Treatments										
Uninoculated and untreated	1.42	1.32	1.47	1.58	1.71	1.63	1.78	2.31	2.30	2.36
Inoculated and untreated	1.42	1.44	1.60	1.84	1.75	1.51	1.71	2.44	2.40	2.19
<i>Moringa oleifera</i> (leaf extract)	1.42	1.35	1.53	1.63	1.68	1.66	1.64	2.32	2.47	2.50
<i>Azadirachta indica</i> (seed extract)	1.42	1.25	1.41	1.52	1.63	1.90	1.63	2.51	1.86	1.39
<i>Zingiber officinale</i> (rhizome extract)	1.42	1.31	1.40	1.54	1.57	1.75	1.63	2.53	2.86	2.53
<i>Cassia alata</i> (leaf extract)	1.42	1.36	1.44	1.60	1.66	1.81	1.79	2.61	2.42	1.98
Mancozeb	1.42	1.48	1.54	1.58	1.52	1.68	1.62	2.08	2.20	2.11
ShavitF71.5WP	1.42	1.36	1.60	1.52	1.54	1.54	1.75	2.10	2.29	2.23

Table 4.7b: Effect of treatments on mean pulp to peel ratio

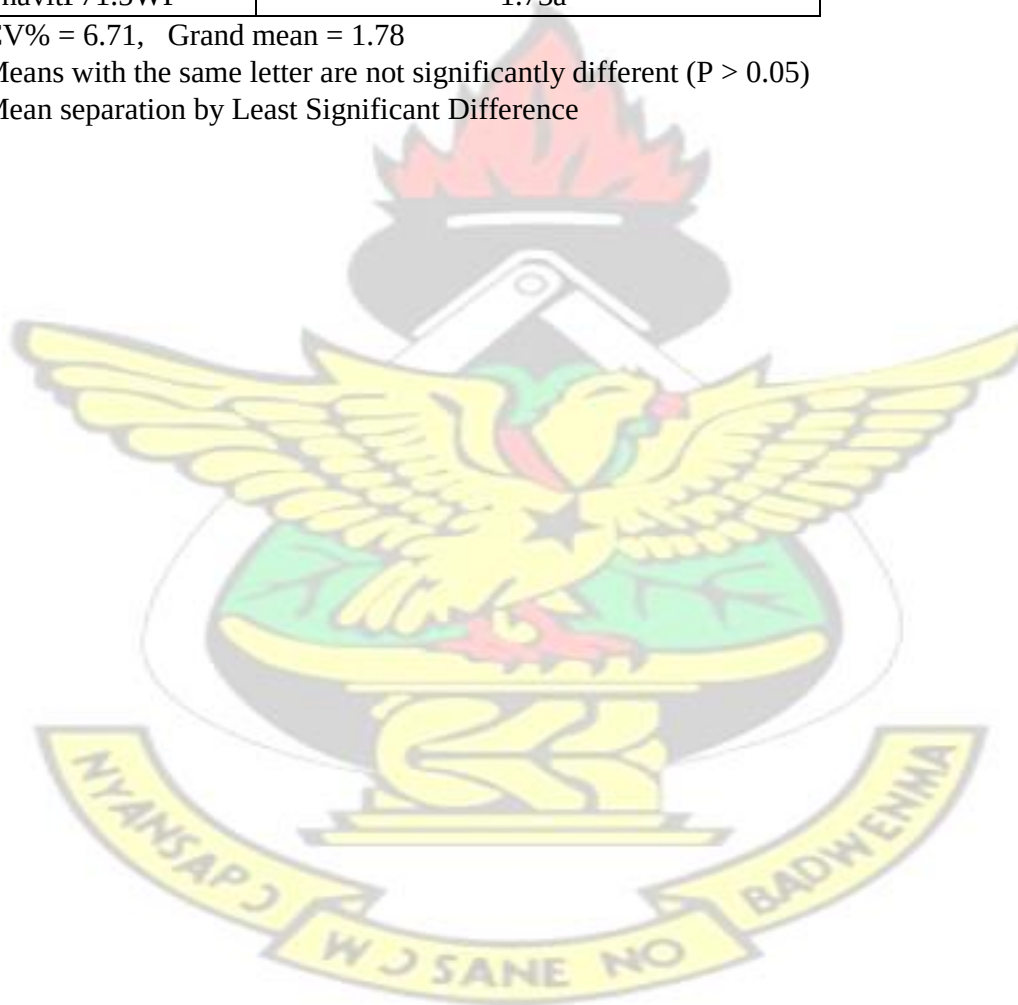
Treatments	
Uninoculated and untreated	1.79a

Inoculated and untreated	1.83a
<i>Moringa oleifera</i> (leaf extract)	1.82a
<i>Azadirachta indica</i> (seed extract)	1.65a
<i>Zingiber officinale</i> (rhizome extract)	1.85a
<i>Cassia alata</i> (leaf extract)	1.81a
Mancozeb	1.72a
ShavitF71.5WP	1.73a

CV% = 6.71, Grand mean = 1.78

Means with the same letter are not significantly different ($P > 0.05$)

Mean separation by Least Significant Difference



5.0 DISCUSSION

5.1 EXPERIMENT ONE: COLLECTION AND IDENTIFICATION OF CROWN

ROT ORGANISMS

5.1.1 Fungal Isolates

Three isolates; *Botryodiplodia theobromae*, *Aspergillus flavus* and *Aspergillus niger*, all very common pathogens, were isolated and identified using culture and spores characteristics from the crown rot tissues of the banana (Mathur and Kongsdal, 2003; Kunz, 2007). *Botryodiplodia theobromae* has been isolated from crown rot tissues by other workers (Amusa *et al.*, 2003 and Ocran, 2005). It is, therefore, not surprising that this fungus (*Botryodiplodia theobromae*) was isolated from crown rot tissues. When inoculated into healthy matured green banana samples, it was able to cause the crown rot disease. This fungus is likely to be the main cause of crown rot in the zones of the Eastern region of Ghana where the bananas were collected from. The *Aspergillus flavus* and *Aspergillus niger*, however, did not cause the disease when inoculated into healthy crowns of banana samples. These may, therefore, be existing as saprophytes on the crown.

5.2 EXPERIMENT TWO: TEST TO ESTABLISH EFFICACY OF BOTANICAL

AND CHEMICAL FUNGICIDES

5.2.1 Efficacy of Botanical and Chemical Fungicides

Antifungal properties in Moringa extract, Neem extract, Cassia extract and Ginger extract were investigated against *Botryodiplodia theobromae* isolate from infected banana with crown rot disease and these botanical fungicides were compared with two chemical fungicides; ShavitF71.5WP (a.i., Metalaxyl) and Mancozeb (a.i., Mancozeb).

Several works have shown that Moringa extract, Neem extract and Cassia extract have fungicidal properties (Oliver-Bever, 1986; NRC, 1992 and Elysha Nur *et al.*, 2002). In this work, however, the fungicidal properties in these three botanicals were not effective when investigated against the rot pathogen.

Moreover, samples which were affected by the crown rot disease had reduced fruit quality, shelf-life and marketability.

The rate of spread of the rot pathogen on crown treated with Ginger extract was not as rapid when compared to the other three botanicals (Moringa, Neem and Cassia extracts) and was able to reduce the incidence of the rot pathogen. Of the four botanicals investigated against the rot pathogen, Ginger extract demonstrated the greatest fungicidal activity whereas the other three botanicals failed to offer the needed protection. Control was possible with Ginger extract due to the fact that, sesquiterpenoids and zingiberene which were found in Ginger extract inhibited the growth of the rot causing pathogen (Anonymous, 2009).

ShavitF71.5WP and Mancozeb, however, gave better control of crown rot disease than any of the four botanicals. The effectiveness of ShavitF71.5WP was due to the fact that it interferes with the respiration processes of the rot pathogen (Anonymous, 2010). Mancozeb inhibits enzyme activity in the rot pathogen by forming a complex with metal-containing enzymes including those involved in production of adenosine triphosphate (Anonymous, 2005). It is, therefore, not surprising that the two products suppressed the development of the pathogen, slowing down the rot process. From the results of the experiment, when the two chemicals were used as postharvest treatment for crown rot disease of bananas, ShavitF71.5WP was found to be superior to

Mancozeb.

5.2.2 Effect of Treatments on Total Soluble Solids

Conversion of starch to simple sugars was faster in samples treated with botanical extracts. Most of the treated banana had increasing trends in total soluble solids and attained a peak and, thereafter, declined. Banana treated with ShavitF71.5WP and Mancozeb, however, experienced gradual conversion of starch to simple sugars. Rathore *et al.* (2007) working on mango found similar trends in total soluble solids. The increase in total soluble solids could be due to alteration in the cell wall structure and breakdown of complex carbohydrates into simple sugars during storage.

The increase and decrease in total soluble solids could be attributed to the hydrolytic changes in starch and conversion of starch to sugars. These are important ripening processes in mango and other climacteric fruits including banana and further hydrolysis decreased the total soluble solids during storage (Kays, 1991 and Kittur *et al.*, 2001).

Ranasinghe *et al.* (2005) reported that Embul banana treated with essential oils of clove, cinnamon bark, cinnamon leaf and benomyl (chemical fungicide) had no effect on total soluble solids at 28°C for 14 days. This study, however, indicated that total soluble solids can be significantly affected by botanical and chemical fungicides.

5.2.3 Effect of Treatments on pH

At harvest, pulp pH is between 5.4 and 6.0 and during ripening pH decreases to 4.0 at the fully ripe stage and increase gradually thereafter (John and Marchal, 1995). Similar trend was found in the present study. However, treatments did not have any effect on pH. In general there was a slow decline in pH in response to ripening and, thereafter,

increased slightly. Works by Ranasinghe *et al.* (2005) showed that Embul banana treated with essential oils of clove, cinnamon bark, cinnamon leaf and benomyl (chemical fungicide) did not have any effect on pH at 28°C for 14 days.

5.2.4 Effect of Treatments on Total Titratable Acidity

In general, levels of acids decline during ripening, presumably due to their utilization as respiratory substrates and during ripening sugar levels within the fruit tend to increase due to the mobilization of starch reserves within the fruit (Tucker, 1990).

All banana samples treated with botanical and chemical fungicides, showed inconsistent patterns of increase and decrease in total titratable acidity with storage days. This trend of inconsistency was also reported by Caussiol (2001), that total titratable acidity increased as the banana fruit ripened and then decreased as the fruit became overripe. However, analysis of variance for total titratable acidity showed that the treatments effect was significant. Ranasinghe *et al.* (2005) working on Embul banana treated with essential oils of clove, cinnamon bark, cinnamon leaf and benomyl (chemical fungicide) found that these treatments had no effect on total titratable acidity at 28°C for 14 days. The significant differences observed in the present study for total titratable acidity could be due to the crude extracts of the botanical fungicides used and the composition of the chemical fungicides. These properties in the botanical and chemical fungicides might have influenced the total titratable acidity.

5.2.5 Effect of Treatments on Percentage Weight Loss

Treatment of banana with Neem extract recorded the highest mean percentage weight loss but was not significantly different from the other treatments. In general, percentage

weight loss of all treatments increased with days. This could be attributed to loss of moisture from the pulp and peel (Stover and Simmonds, 1987).

Furthermore, unripe banana is mainly composed of starch which represents 20-25% of the fresh weight of the pulp and 3% of the fresh weight of the peel (Seymour *et al.*, 1993). Starch is converted into sucrose, glucose, fructose and maltose which have lower molecular weight (Dadzie and Orchard, 1997).

In the present study however, analysis of variance for percentage weight loss indicated that the treatments effect was not significant.

5.2.6 Effect of Treatments on Pulp to Peel Ratio

Pulp to peel ratio of all treated samples were above one. Ramma *et al.* (1999) reported that pulp to peel ratio of 1 and above was considered to be very good indication of banana harvest maturity and ripening which ensures acceptable eating quality of banana. The results of the present study showed that pulp to peel ratio of 1 and above were observed even though there were fluctuations in these ratios with days.

These changes in pulp to peel ratio observed in the treated samples could be attributed to the rise in water content of the pulp during ripening and this is partly linked to the respiratory breakdown of carbohydrates. It could also be attributed to osmotic migration of water from the peel to pulp because of higher concentration of sugar in the pulp (John and Marchal, 1995). The results also indicated that treatments effect on pulp to peel ratio of banana samples were not significant during the storage period.

6.0 SUMMARY, CONCLUSION AND RECOMMENDATION

Botryodiplodia theobromae, *Aspergillus niger* and *Aspergillus flavus* were the main fungal organisms isolated from infected bananas collected from the Fremponso and Volta River Estate Limited of the Atiwa and Asuogyaman districts in the Eastern region of Ghana. *Botryodiplodia theobromae* when inoculated caused healthy crowns to rot. Meanwhile *Aspergillus niger* and *Aspergillus flavus*, however, did not cause crown rot when inoculated into healthy matured hands of banana.

In evaluating the effectiveness of the four botanical fungicides in controlling crown rot disease of Medium Cavendish banana, it was realized from the experimental results that *Moringa oleifera* (leaf extract), *Azadirachta indica* (seed extract) and *Cassia alata* (leaf extract) were not effective in suppressing the rot causing organisms. *Zingiber officinale* (rhizome extract) could, however, suppress the growth of the rot organism and thus the development of crown rot disease. The chemical fungicides were more effective in controlling the rot pathogen than the botanicals.

ShavitF71.5WP was found to be superior to Mancozeb in controlling crown rot disease. Total soluble solids, total titratable acidity, pH, percentage weight loss and pulp to peel ratio were the main physico-chemical parameters studied in relation to application of treatments to banana samples and subsequent storage.

Comparison of treatment means indicated significant differences for total soluble solids and total titratable acidity while there were no significant differences for pH, percentage weight loss and pulp to peel ratio. This shows that the treatments could influence the total soluble solids and total titratable acidity but not pH, percentage weight loss and pulp to peel ratio.

Exporters in the banana trade could adopt ShavitF71.5WP and Mancozeb in addition to already documented standard fungicides for use as a postharvest dips for the treatment of crown rot disease.

Alternatively, *Zingiber officinale* (rhizome extract) as a botanical also has the potential and, therefore, could offer some meaningful protection to banana crowns when applied immediately after dehanding.

In conclusion, the occurrence of crown rot disease is closely linked to poor cultural and disease management practices in banana fields, unclean packing houses and improper postharvest handling. The disease could be a serious problem for handlers who fail to manage them properly. The results obtained with *Zingiber officinale* (rhizome extract) could lead to development of eco- friendly, safe and convenient means of reducing postharvest losses in banana.

It is recommended that the concentration of *Zingiber officinale* (rhizome extract) be varied to determine the most effective level of its efficacy in controlling crown rot disease of banana. Further experiment could be done with other botanicals with fungicidal properties.

REFERENCES

- Amusa, N. A., Ashaye, O. A., Oladepo, M. O. and Kafaru, O. O. (2003). Pre-harvest deterioration of Sour sop (*Annona muricata*) at Ibadan Southwestern Nigeria and its effect on nutrient composition. *African Journal of Biotechnology*, 2 (1), 23-25.
- Anonymous (2001). *Promote Sustainable Agriculture in Ghana*. The Organic farmer. 6 (4), Pp 4-5.
- Anonymous (2005). Mancozeb. (<http://www.sinoharvest.com/products/Mancozeb.shtml>; Date accessed: 10-03-2008)
- Anonymous (2009). Ginger. (<http://www.spice-trade.com/ginger>; Date accessed: 22-04-09)
- Anonymous (2010). (<http://www.hektas.com.tr/DesktopDefault.aspx?TabId=316&llacId=4980&DListTip=1>; Date accessed: 20-12-10)
- AOAC (Association of Official Analytical Chemists) (1990). Official Methods of Analysis. Washington, D.C.
- Bose, T. K and Mitra, S. K (1990). *Fruits: Tropical and Subtropical*. NAYA PROKASH 206 Bidhan Sarani. Calcutta 700 006 India. Pp 132-5.
- Burden, O. J. (1969). Control of postharvest diseases in banana. *Q. Agric. J.*, 95, 621-624.
- Caussiol, L. (2001). Postharvest quality of conventionally and organically grown banana fruit. MSc Thesis, Cranfield University.
- Clay, J. W. (2004). *World Agriculture and the Environment: A Commodity-byCommodity Guide to Impacts and Practices*. Covelo, CA, USA: Island Press. (<http://site.ebrary.com/lib/brown/Doc?id=10064697&pp=250>; Date accessed 12-0308)
- Dadzie, B. K. (1994). Six monthly reports for the INIBAP/FHIA/NRI (ODA Holdback) project on post-harvest cooking banana and plantain characterization (April - September, 1994).
- Dadzie, B.K. and Orchard, J.E. (1997). Routine Postharvest Screening of Banana/Plantain: Criteria and Methods. Pp 15,21,26,29 and 51.
- Delal, V. B., Thomas, P., Nagaraja, N., Shah, G. R and Amla, B. L. (1970) Effects of wax coating on bananas of varying maturity. *Indian Food Packer*, 24, 36-40.
- Duke, J.A. (1983). *Handbook of Energy Crops*. Unpublished. (http://www.hort.purdue.edu/newcrop/duke_energy/Moringa_oleifera.html; Date accessed: 20-03-08)

- Elysha Nur, I., Somchit, M. N. and Abdul Rahim, M. (2002). In Vitro Antibacterial Activity and Effect of *Cassia alata* in Livers of Mice. In: Omar, R., Ali Rahman, Z., Latif, T. and Adam, J. H (Eds). Proceedings of the Regional Symposium on Environment and Natural Resources, 10-11th April 2002, Hotel Renaissance Kuala Lumpur, Malaysia. 1:509-515.
- GEPC (Ghana Export Promotion Council) (2006). Value of Main Agricultural Non-Traditional Exports. <http://www.countrystat.org/gha/cont/pxwebquery/ma/081ctr015/en> (Date accessed 15-05-08)
- Gowen, S. (1995). Bananas and Plantains. Chapman and Hall, 2-6 Boundary Row, London SE 1 8HN. Pp 403-406.
- Johanson, A. and Jeger, M. J. (1993). Use of PCR detection of *Mycosphaerella fijiensis* and *M. musicola*, the causal agents of Sigatoka leafspots in banana and plantain. *Mycological Res.*, 97,670-674.
- John, P. and Marchal, J. (1995). Ripening and biochemistry of the fruit. In: Gowen, S. *Bananas and plantains*, Chapman and Hall, 2-6 Boundary Row, London SE 1 8HN.
- Johnston, A. and Booth, C. (1983). Plant Pathologist's Pocketbook (2nd ed.). CAB, England. Pp 397.
- Kader, A. A. (1994). Fruit maturity, ripening and quality relationships. *Perishables Handling Newsletter* 80: 2.
- Kays, S. J. (1991). Postharvest physiology of perishable plant products. *Vas Nostrand Rein Hold Book*, AVI Publishing Co. Pp 149-316.
- Kittur, F. S., Habibunnisa, N. S. and Tharanathan, R. N. (2001). Polysaccharide-based composite formulations for shelf-life extension of fresh banana and mango. *Eur. Food Res. Tec.* 213: 306-311.
- Kunz, R. (2007). Control of postharvest disease (*Botryodiplodia* sp.) of *Rambutan* and *Annona* Species by Using a Bio-Control Agent (*Trichoderma* sp.). <http://www.icuc-iwmi.org/files/Publications/Robert%20report%20290807.pdf> (Date accessed: 10-2-08)
- Maroyi, A. (2006). The utilization of *Moringa oleifera* in Zimbabwe. http://www.jsda-africa.com/Jsda/Summer_2006/PDF/ARC_UtilizationMOeifera.pdf (Date accessed: 10-2-08)
- Mathur, S. B. and Kongsdal, O. (2003). *Common Laboratory Seed Health Testing Methods for Detecting Fungi*. International Seed Testing Association, Switzerland. Pp 134-5.
- Nakasone, H. Y. and Paull, R. E. (1998). *Tropical Fruits*, CAB International Wallingford Oxon OX 10 8 DE, UK. Pp 103-130.

- NRC (National Research Council) (1992). *Neem: A tree for Solving Global Problems*. National Academy Press, Washington, D.C. Pp 23, 25, 26 and 53.
- Nwachukwu, E. O. and Osuji, J. O. (2008). Evaluation of plant extracts for antifungal activity against *Sclerotium rolfsii* causing cocoyam cormel rot in storage, *Research Journal of Agriculture and Biological Sciences*, 4(6):784-787.
- Ocran, J. K. (2005). Effect of postharvest hot water and fungicide application on the severity of crown rot disease and quality of Cavendish bananas (*Musa spp*). MSc Thesis, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana.
- Okigbo, R. N. and Nmeko, I. A. (2005). Control of yam tuber rot with leaf extracts of *Xylopi aethiopia* and *Zingiber officinale*. *Africa Journal of Biotechnology* 4(8), Pp 804-807.
- Oliver-Bever, B. (1986). *Medicinal Plants in Tropical West Africa*. Cambridge University Press, Cambridge. Pp 46-47.
- Picq, C., Fouré, E. and Frison, E. A. (1998). Banana and food security: The banana production, a major economic issue for food security. International symposium, Douala, Cameroon, 10-14, November 1998. Pp 424-5.
- Ramma, I., Beni Madhu, S. P. and Peerthum, S. (1999). Postharvest quality improvement of banana. Food and Agricultural Research Council, Réduit, Mauritius.
<http://www.gov.mu/portal/sites/ncb/moa/farc/amas99/s53.pdf>(Date accessed: 10-03-08)
- Ranasinghe, L., Jayawardena, B. and Abeywickrama, K. (2005). An integrated strategy to control postharvest decay of Embul banana by combining essential oils with modified atmosphere package. *International Journal of Food Science*. 40:97-103.
- Rathore, H. A., Masud, T., Sammi, S. and Soomro, A. H. (2007). Effect of storage on physico-chemical composition and sensory properties of mango (*Mangifera indica* L). *Pakistan Journal of Nutrition*, 6 (2) 143-148.
- Reezal, I., Somchit, M. N. and Abdul Rahim, M. (2002). In vitro antifungal properties of *Cassia alata* (Gelenggang Besar). In: Omar, R., Ali Rahman, Z., Latif, T. and Adam, J. H (Eds). Proceedings of the Regional Symposium on Environment and Natural Resources, 10-11th April 2002, Hotel Renaissance Kuala Lumpur, Malaysia. 1:509-515.
- Robinson, J. C. (1996). *Bananas and Plantains*, University Press Cambridge. Pp184, 215and 216.
- Sarananda, K.H. and Wijeratnam, R.S.W. (1994). Controlled atmosphere storage of Embul bananas. *Tropical Agricultural Research*, 6, 17-22.

Scott, J., Wills, R. B. H. and Rippon, L. E. (1971). The use of polyethylene bunch covers during growth as a retardant to the ripening of bananas. *Trop. Agric. Trinidad*, 48, 163-168.

Seymour, G. B., Taylor, J. E. and Tucker, G. A. (1993). *Biochemistry of Fruit Ripening*. Chapman and Hall, 2-6 Boundary Row, London SE 1 8HN. Pp 83 and 89.

Seymour, G.B. (1985). The effects of gases and temperature on banana ripening. PhD Thesis, University of Reading.

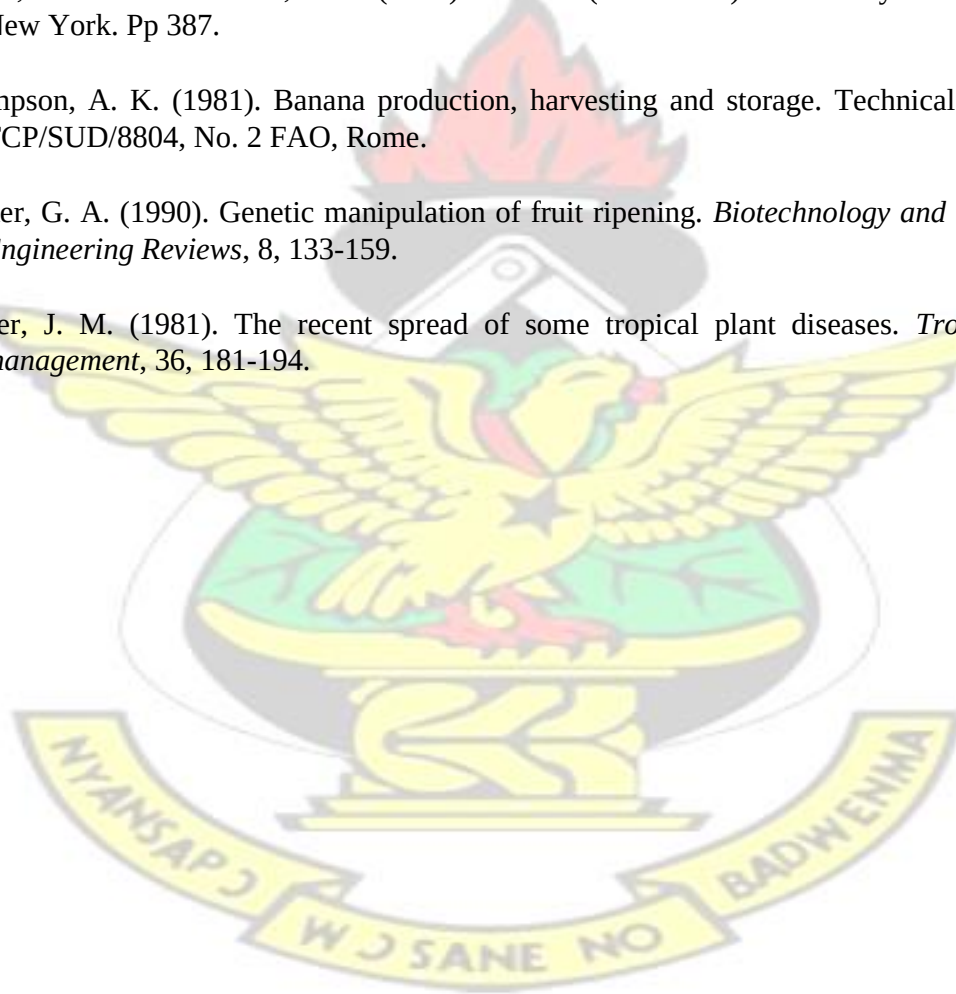
Stover, R. H. (1972). *Bananas, Plantains and Abaca Diseases*. Commonwealth Mycological Institute, Kew.

Stover, R. H. and Simmonds, N. W. (1987). *Bananas* (3rd edition). John Wiley & Sons, Inc. New York. Pp 387.

Thompson, A. K. (1981). Banana production, harvesting and storage. Technical Report TCP/SUD/8804, No. 2 FAO, Rome.

Tucker, G. A. (1990). Genetic manipulation of fruit ripening. *Biotechnology and Genetic Engineering Reviews*, 8, 133-159.

Waller, J. M. (1981). The recent spread of some tropical plant diseases. *Trop. Pest management*, 36, 181-194.



APPENDICES

Appendix 1.0 ANOVA for the effect of treatments on crown rot infection

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	32.50315196	4.64330742	55.09	<.0001
Error	16	1.34863667	0.08428979		
Corrected Total	23	33.85178863			

Appendix 1.1 ANOVA for the effect of treatments on percentage weight loss

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	11.46132717	1.63733245	1.45	0.2534
Error	16	18.06479067	1.12904942		
Corrected Total	23	29.52611783			

Appendix 1.2 ANOVA for the effect of treatments on pulp to peel ratio

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	0.09659917	0.01379988	0.97	0.4844
Error	16	0.22752267	0.01422017		
Corrected Total	23	0.32412183			

Appendix 1.3 ANOVA for the effect of treatments on total soluble solids

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	44.40000000	6.34285714	5.25	0.0029
Error	16	19.32000000	1.20750000		
Corrected Total	23	63.72000000			

Appendix 1.4 ANOVA for the effect of treatments on pH

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	0.05649733	0.00807105	1.78	0.1612
Error	16	0.07264800	0.00454050		
Corrected Total	23	0.12914533			

Appendix 1.5 ANOVA for the effect of treatments on total titratable acidity

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	0.00263496	0.00037642	3.50	0.0180
Error	16	0.00172200	0.00010763		
Corrected Total	23	0.00435696			

Appendix 1.6 ANOVA for the effect of treatments on crown rot disease severity

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	12.44958333	1.77851190	20.33	<.0001
Error	16	1.40000000	0.08750000		
Corrected Total	23	13.84958333			

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