DISTRIBUTION AND OCCURRENCE OF MANGO ANTHRACNOSE (*Colletotrichum gloeosporioides.*Penz and Sacc) AROUND JIMMA, SOUTHWESTERN ETHIOPIA

M.Sc. Thesis

By

Ayantu Tucho

May, 2014

Jimma, Ethiopia

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Submitted to the School of Graduate Studies, Jimma University College of Agriculture and Veterinary Medicine In Partial Fulfillment of the Requirements for the Degree of Master of Science in Plant pathology

By

Ayantu Tucho

May, 2014

JIMMA UNIVERSITY

APPROVAL SHEET
School of Graduate Studies

As thesis research advisor, I hereby certify that I have read and evaluated this thesis prepared, under my guidance, by **Ayantu Tucho**, entitled "Distribution and Occurence of Mango Anthracnose (*Colletotrichum gloeosporioides.penz. and sacc)* at Jimma, Southern Ethiopia**"**. I recommend that it be submitted as fulfilling thesis requirement.

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As member of the board of Examiners of the M.Sc. Thesis Open Defense Examination, we certify that we have read, evaluated the thesis prepared by **Ayantu Tucho** and examined the candidate. We recommended that the thesis could be accepted as fulfilling the thesis requirement for the Degree of Master of Science in plant pathology.

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Chairperson Signature

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DEDICATION

I dedicate this thesis manuscript to my beloved father, **Tucho Dano Dhunfa** and my mother **Bakane Gula Hunde** who showed me that education is the most important gift that parents can provide to their children and for nursing me with affection and love and for their dedicated partinership in the succès of my life.

STATEMENT OF THE AUTHOR

First, I declare that this M.Sc Thesis is my genuine work and has not been submitted to any other institution anywhere for award of any academic degree, diploma, or certificate. All sources of the materials used in this Thesis have been duly acknowledged. This thesis has been submitted in partial fulfillment of the requirements for the award of the degree of master in plant pathology at Jimma University, Ethiopia. Brief quotations from this Thesis are allowable without special permeation provided that an accurate acknowledgment of the source is made.

Name: Ayantu Tucho Signature: _____

Place: JUCAVM, Jimma, Ethiopia Date of Submission: _____

BIOGRAPHICAL SKETCH

The author, Ayantu Tucho, was born at A/Dullacha, in Horro Guduru Wollega Zone, on June 12,1987 from her mother Bekane Gula and her father Tucho Dano. She attended and completed grade 1 to 8 at Gitilo Naajo Elementary school (1995-2003) and grade 9 to 12 at Shambu senior secondary school (2004-2008), in Horo Guduru Wallaga Zone, Ethiopia. She joined Haramaya University in October, 2008 and graduated with a B.Sc.degree in **crop production and protection** in July10, 2010. The Author was employed by Ethiopian Minstry of Education in September, 2011 and served as assistant lecturer in Jigjiga University for one year. She joined the school of graduate studies in September, 2012, at Jimma University College of Agriculture and Veternary Medicine (JUCAVM) to pursue her graduate study in Plant pathology.

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Wishing all Holy blessings from Jesus Christ and be considered in his eternal government.

LIST OF ACRONOMYS AND ABBREVATIONS

ANOVA	Analysis of Variance
CSA	Central Statistical Agency
°C	Degree Celsius
FAO	Food and Agriculture Organization
FAOSTAT	Food and Agricultural Organization Statistical Division
JUCAVM	Jimma University College of Agriculture and Veterinary Medicine
M	Meter
Mt	Metric tone
Km	Kilo meter
LSD	Least significant difference
m.a.s.l	Meter above sea level
MM/yr	Millimeter per year
SAS	Statistical Analysis Software
SW	Southwestern
GPS	Geographical Position System
NaOCl	Sodium hypochlorate
PDA	Potato dextrose agar
E.C	Ethiopian calendar

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ABSTRACT

Mango (Mangifera indica L.) is grown throughout the tropics and subtropics of the world and its production and productivity is limited by several biotic and abiotic factors. Mango anthracnose, caused by Colletotrichum gloeosporioides is considered as the most important mango disease in the humid tropics that contribute significantly to pre-harvest and post- harvest fruit losses. Despite its important, research work addressing the distribution and occurrence of mango anthracnose both in the field and at market in mango producing areas of Jimma Zone, SW Ethiopia is not yet documented. The current study was conducted to determine the distribution and occurrence of mango anthracnose (C. gloeosporioides penz.) around Jimma, south west Ethiopia. The study was conducted in three potential mango producing woredas (Gomma, Seka chokorsa, and Kersa woreda) and Jimma town of Jimma Zone in the SW Ethiopia. Twelve (12) kebeles from all woredas was assessed. Assessments were performed at three positions per tree (upper, middle and lower). Isolation was done to confirm the causal pathogen. Pathogenecity of the identified C. gloeosporioides was tested on detached leaf and fruits of mango. From all assessed PAs significantly higher (83.5%) incidence on the leaf was recorded on the lower tree canopy at Bulbulo kebele of Gomma woreda and the lowest (26.3%) anthracnose incidence was recorded in Kitto kebele of Jimma area on the upper tree canopy. Higher percentage of mango anthracnose incidence on the leaf was recorded at Gomma woreda (72.1%) whereas lower percentage of mango anthracnose incidence was recorded at Kersa Woreda (41%). Mango anthracnose incidence on the fruit was higher in Gomma Woreda (74%) and lower in Kersa Woreda (36.24%). The severity of mango anthracnose was the highest in Kasohixi Kebele of Gomma woreda (80.5%) at the lower tree canopy and the lowest at Marewa kebele of Kersa woreda (30.3%) at the upper tree canopy. The incidence and severity was high (95.3% and 82%) in Agaro market respectively and lower disease incidence and severity (70.66% and 64%) was recorded in Jimma market. The fungus was, identified to be Colletotrichum gloeosporioides. Generally from this study mango anthracnose (C. gloeosporioides) was 100% prevalent in the assessed three waredas and one urban area. The incidence was higher on the fruits than the leaves. The disease was more severe in the market place than in the farmers' fields. However, to get full picture of the prevalence of this disease and to design appropriate control methods, it is advisable to conduct similar assessments in different mango growing agro ecologies and along mango value chain.

Key words: Incidence, Severity, Disease prevalence, Tree canopy, Mango, *Colletotrichum gloeosporioides*

1. INTRODUCTION

Mango (*Mangifera indica L.)* is grown throughout the tropics and subtropics of the world and it belongs to the family *Anacardiaceae (*Bally, 2006*)*.It is native to India and southern Asia. By virtue of its wide ecological range, delicious taste, superb flavor, very high nutritive and medicinal value as well as great religio-historical significance, it is called the "King of the fruits" (Lakshmi *et al.*, 2011). It is the most popular and commonly eaten fruit among millions of people in tropical areas and especially the developed countries. Apart from its economic importance, it is forest and environmentally friendly to fight against drought, use as shade and fire wood. Mangos are a highly nutritious fruits containing carbohydrates, proteins, fats, minerals, and vitamins, in particular vitamin A (beta carotene), vitamin B1, vitamin B2, and vitamin C (ascorbic acid) (Bally, 2006, Biniyam, 2010). Mango fruits contribute immensely to diet especially in the tropics and have been observed to be higher in vitamin C than citrus fruits (Charles *et al., 2012*).

The crop is grown in over 87 countries in the world, with developing countries account for about 98% of total production while, developed countries account for 80% of world import trade. Among internationally traded tropical fruits, mango ranked second only to banana both in quantity and value and fifth in total production among major fruit crops worldwide. The world production of mangoes is estimated to be over 26 million tons per annum (FAO, 2009). India ranks first among the world's mango producing countries, accounting for 54.2% of the total mangoes produced worldwide. Other prominent mango producing countries include China, Thailand, Indonesia, Philippines, Pakistan, and Mexico. In Nigeria, the total area dedicated to mango production is estimated to be 126,500 hectares with a production output of 734000 metric tons in 2007 and place the country in the ninth position of top mango producing countries of the world and highest producer in Africa.

Mango (*M. indica* L.) is a perennial tree which can live more than fifty years and it is also the leading fruit produced in most parts of eastern and south-western Ethiopia both in area coverage and quantities produced (Yeshitla, 2004). There are also ample garden mango trees in different parts of the country at farmer's holdings. The livelihood of most of these farmers is highly supplemented by the sale of mango fruits. According to agricultural survey of Ethiopia central statistical agency (CSA,2012/13), about 61,972.60 hectares of land is under fruit crops in Ethiopia. Bananas contributed about 58.11% of the fruit crop area followed by mangoes that contributed 14.21% of the area.

According to FAOSTAT, (2010) the total cultivated area for mango in Ethiopia is not more than 12, 000 hectares. The highest annual production estimated in the past five years is 180,000 MT and more area coverage is expected in the south-western and other parts of the country due to more conducive climatic and edaphic factors. At present, very little mango is exported from Ethiopia with only 4 tons exported in 2006 at a value of less than US$1000 (Binyam Teshome, 2010). The types of locally available varieties are not well known. But varieties like Tommy Atkins, Kent and Keitt apple are under cultivation in the Upper Awash Agro Industry.

The quality of fresh fruits depends on the post harvest handling during harvesting, transportation and storage (Wiersinga and Jager, 2009). Compared with several temperate fruits, the tropical and subtropical fruit such as mango, banana and papaya presents greater problems in storage and transportation because of their perishable nature (Baldwin, *et al.,* 1999). Growing and marketing of fresh produce in Ethiopia is complicated by post harvest losses both in terms of quantity and quality between harvest and consumption. The post harvest losses of perishable commodities are estimated to be as high as 50% in Ethiopia (FAO, 2009). The production, marketing and consumption of mango, banana and papaya fruits are restricted due to improper handling, inadequate transport and storage facility, disease problems, and sensitivity to low storage temperature (Baldwin, *et al.,* 1999).

In recent years, mango production and quality has been declining due to the occurrence of a variety of abiotic and biotic factors, some leading to the development of sudden death of trees (Masood *et al*., 2009; Masood *et al*., 2011). According to Yeshitela (2004) even if the farmer's livelihood is highly supplemented with the income from their mango trees, there is a declining trend in yield and quality of mango due to old age, poor management , seedling originated nature of the trees and different pre and post harvest disease of mango. Even though the amount of mango production and cultivation area of the country is not well known, according to Kader (2009) the current post-harvest loss of mango fruits in Ethiopia is more than 26.3%, in areas where rain is prevalent this can reach to 35%.

Fruit of mango can be attacked by a number of pathogens including fungi, bacteria, algae and insects such as fruit flies. Fungal disease is one of the most important causes of post harvest losses in mango. Major post-harvest fungal problems on mangoes in Africa include anthracnose caused by *Colletotrichum gloeosporioides* (Swart, 1999), stem-end rot caused by *Lasiodiplodia theobromae* and soft brown rot caused by *Botryosphaeria rhodina* (Johnson *et al*., 1995). Anthracnose caused by *Colletotrichum gloeosporioides* Penz. and Sacc. is the most serious disease widely distributed in all mango growing regions of the world (Smoot and Segall, 1963; Sangeetha and Rawal.,2009) and is a major constraint to the expansion of export trade of mango (Jeger and Plumbley, 1988). Mango anthracnose can reduce fruit quality and cause between 30 to 60% of harvest losses (Vega, 2001).Anthracnose is one of several fruit diseases that affect pre- and post-harvest quality (Ploetz, 2003). Among the postharvest diseases of mango, anthracnose is the most prevalent in humid growing areas. The incidence of this disease can reach almost 100% in fruit produced under wet or very humid conditions (Arauz, 2000).

Conidia are dispersed by rain splash and infection requires free moisture (Jeffries *et al*., 1990). As appressoria age, they become melanized. Melanization strengthens the appressorium and facilitates penetration of the cuticle by infection pegs that the appressoria produce. The presence and prevalence of melanized appressoria have been used to predict when infection is possible and anthracnose control measures are needed (Fitzell and Peak, 1984; Dodd *et al*., 1991).

Post harvest anthracnose disease occurrence directly affects the marketable fruit rendering it worthless. Post harvest anthracnose occurrence is directly linked to the field phase where initial infections usually start on young twigs, leaves and later spreads to the flowers causing blossom blight, destroys the inflorescences and finally prevent fruit set (Nelson,2008). For instance in areas where rain is prevalent during flowering and fruit set, anthracnose can cause destruction of the inflorescences and infection and drop of young fruits where this can obviously lead to serious losses, reaching up to 35% of the harvested fruits (Martinez *et al*., 2009). The disease incidence from different countries has been reported to be 32% in South Africa (Sanders *et al*., 2000), 64.6% in Costa Rica during 1990 (Arauz *et al*., 1994).

Mango production is limited by some biotic factors in humid forest region of Ethiopia despite its economic importance. Fruit anthracnose disease was commonly found associated with mango fruits produced in the humid forest region of Jimma zone, Southwestern Ethiopia. The fruits rot so quickly after harvest due to this anthracnose rendering marketable fruits unattractive and worthless. It has been reported that anthracnose is presently the most common and most important field and postharvest disease of mango widely distributed in all mango-growing regions of the world (Sangeetha and Rawal, 2009). This disease has made mango production non-attractive to farmers and home gardeners in Jimma zone, Southwestern Ethiopia. However, research work addressing the distribution and occurrence of mango anthracnose both in the field and at market in mango producing areas of Jimma Zone, SW Ethiopia is not yet documented. Therefore the current research was initiated to study the distribution and occurrence of mango anthracnose (*Colletotrichum gloeosporioides*) both in the field and market in Jimma Zone.

OBJECTIVES

General objective

- To determine the distribution and occurrence of mango anthracnose (*C. gloeosporioides* penz.*)* around Jimma, south west Ethiopia.

Specific objectives

- To assess mango anthracnose (*Colletotrichum gloeosporioides*) disease incidence and severity in farmer's field of mango producing districts around Jimma, SW Ethiopia.

- To assess mango anthracnose (*Colletotrichum gloeosporioides*) disease incidence and severity in main market places of mango producing districts around Jimma, SW Ethiopia.

2 LITERATURE REVIEW

2.1 Botany and history of mango

Mango was belongs to the kingdom *Planteae*, division *Angiospermae*, class *Magnoliopsida*, order *Sapindales*, family *Anacardiaceae*, genus *Mangifera* and species *indica*, consisting of numerous species of tropical fruiting trees in the flowering plant family. The mango is indigenous to India, and it is cultivated in many tropical and subtropical regions of the world. Mango is one of the most extensively exploited fruits for food, juice, flavour, fragrance and colour. In several cultures, its fruit and leaves are ritually used as floral decorations at weddings, public celebrations and religious ceremonies (McGovern and LaWarre, 2001).

There are at least 62 species in the genus of which 15 bear edible fruit (Litz, 1994). Kaur *et al.* (1980) indicated that the mango tree is believed to have evolved as a canopy layer species in the tropical rain forest of south and south-east Asia. Litz (2003) mentioned that the mature mango trees can attain a height of 30 meters with a crown radius of 10m and can survive for more than 100 years and still fruiting. The root system consists of a long, vigorous tap root and abundant surface feeder roots. In deep soil the taproot descends to a depth of 6m, and the profuse, wide-spreading feeder roots also send down many anchor roots which penetrate for several meters. The tree is an arborescent evergreen one with simple, alternate, oblong ovate to oblong lanceolate leaves, 15–35 cm long and 6–16 cm broad; when young they are orange-pink, rapidly changing to a dark glossy red, then to dark green as they mature. They are spirally arranged and produced in flushes (Litz, 2003).

According to Litz (2003) the flowers are borne on terminal pyramidal panicles 10–40 cm long, glabrous or pubescent; the inflorescence is rigid and erect and is widely branched, usually densely flowered with hundreds of small flowers, 5-10 mm in diameter. The flowers are small, monoecious and polygamous. Both male and perfect flowers are found within a single inflorescence; the pistil aborts in male flowers.

The 10 ratio of male to perfect flowers is strongly influenced by environmental and cultural factors. The flowers have four to five sepals that are ovate to ovate oblong and also highly pubescent (Litz, 2003).

Litz (2003) further stated that there are four to five petals 5–10 mm long, with a mild sweet odour suggestive of lily of the valley, oblong to ovoid to lanceolate and also thinly pubescent. The floral disc is four to five-lobbed, fleshy and large, and located above the base of the petals. There are five large, fleshy nectaries that form a five lobed receptacle. Although there are four to five stamens, only one or two of them are fertile; the remainder are sterile staminodes that are surmounted by a small gland. In addition, two or three smaller filaments arise from the lobes of the nectaries. The stamens are central and that it is believed the flowers are cross–pollinated by flies (Litz, 2003).

Litz (2003) describes the mango fruit as a large, fleshy drupe, containing edible mesocarp of varying thickness. The fruit is highly variable in size, shape and colour, and may be yellow, orange, red or green when ripe, depending on the cultivar. When ripe, the unpeeled fruit gives off a distinctive resinous sweet smell. Chlorophyll, carotenes, anthocyanins and xanthophylls are all present in the fruit, although chlorophyll disappears during ripening whereas anthocyanins and carotenoids increase with maturity (Lakshminarayana, 1980). Fruit colour at maturity is genotype–dependent. The exocarp is thick and glandular. The mesocarp can be fibrous or fibre–free with flavor ranging from turpentine to sweet. In its centre is a single flat oblong 11 pit that can be fibrous or hairy on the surface, depending on the cultivar. Inside the pit 1-2mm thick is a thin lining covering a single seed, 4-7cm long, 3-4cm wide, and 1cm thick. The seed contains the plant embryo. The endocarp is woody (Litz, 2003).

The fruit vary in shape (kidney, round, oblong, oval) and weight and ranges from a few gram to 2.5 kg. Although the fruit will ripen on the tree, it is usually picked green for shipment. The crop is considered mature when the shoulder of the fruit broadens (fills out) and some fruits on the tree have begun to change color from green to yellow. Prior to this color change, the fruit is considered mature when the flesh near the seed changes color from white to yellow.

2.2 Nutritional and medicinal value of mango

Mangos are extremely nutritious and contain carbohydrates, proteins, fats, minerals, vitamins: vitamin A (beta carotene), B1, B2, and vitamin C (ascorbic acid) (Bally, 2006).These nutrients play a crucial role in human nutrition thus the health of the individual. Mangos also make important seasonal contributions to the diet of many countries in the tropics especially African countries that primarily have a starch (carbohydrates)-based diet. Diarra *et al.* (2010) reported that Mango-Seed Kernels is a good source of soluble carbohydrates. Mangoes are used in processing of various products such as juice, purée, slices or pickles but the fresh mango consumption are more preferable.

Ripe mangos fruits are rich sources of vitamin A and are used to treat vitamin A deficiencies such as night blindness. Also drinks made from the infusion of fresh mango leaves have been used to treat diabetes and dried mango seed ground into powder is used to treat diarrhea. Diarrhea and throat disorders are treated by bubbling the bark extracts mixed with water (Bally, 2006). Some other uses of the mango includes its use in agro forestry and environmental practices such as livestock shelter, home gardens, fence post, wind breaks and animal feeds. Other uses include: flavorings in which its puree is used to give flavor to many foods such as drinks, ice cream, wines, teas etc., honey (from its nectar), and making leafy vegetables from boiled young leaves and used for tannin/dye (Martin et al., 1998).

Naturland (2001) reports the importance of mango that ripened fruits are eaten fresh everywhere, and used to make juice or marmalade, dried and made into candy. Also all leftovers from the fruits can be used as animal feed. For instance, the young leaves are very good as cattle feed, because they have a protein content of 8-9% and high calcium (Ca) content as well. The bark and leaves of mango trees can also be used as a dye for cloth. The wood from mango trees is highly suitable for making charcoal which is widely used in rural areas in Africa as a source of fuel (Naturland, 2001).

2.3 Production and Productivity of Mango in Ethiopia and Beyond

Mango is a common garden tree throughout the tropics. Chomchalow and Songkhla (2008) mentioned that mango is native to Indo-Myanmar region and has been cultivated for more than 4000 years. Nowadays, mango is cultivated throughout the tropical and subtropical world for commercial fruit production, as a garden tree, and as a shade tree for livestock. In 2009, India, Mexico, Thailand, Brazil, and Pakistan produced the largest amount of fresh mango for export among the mango growing countries. In fact, Asia was the main exporter with 46.27% of global mango production in 2009 (FAOSTAT, 2012).

Productivity depends on a number of factors, including quantity of previous crop, weather and soil conditions, altitude, control of pests and diseases, fertilization and cultivar. Even in the case of the same cultivar, yields vary greatly because mango is grown under widely varying agro climatic conditions and cultural practices. Mango production predominates in dry and wet tropical low land areas $23^{0}26'$ North and South of the equator, on the Indian subcontinent, Southeast Asia and Central and South America (Litz, 1997). Mango is grown in at least 87 countries but no where it is so greatly value as in India where 40% of total fruits grown in India is only mango. India ranks first among world's mango producing countries accounting for 57.18% of the total world mango production of 19.22 million tons (Krishnan, et al, 2009). India's contribution to the world's mango production is the highest *i.e.*, 15,64200 MT whilst only 0.3 per cent (47,149 MT) is exported, compared to South Africa whose total production is 38,000 MT and 32.5% of it *i.e.*, 12,341MT is being exported, being the highest in terms of export among the other countries (Krishnan, et al, 2009).

Mango production is also impaired by a number of insects and diseases (Anwar *et al*., 2011). With an increase in Ethiopian mango crop production and considering the current postharvest loss of mango fruits of 26.3%, there is not only a need but also a potential for the fruit to be processed into various product types, consequently increasing the market potential of the mango fruit (Kader and Truneh, 2009). The production share of mango was found to be next to that of banana. According to the FAO's *2009* Food Market Analysis of Tropical Fruits, mangoes

dominated world production of tropical fruit at 31.5 million metric tons, comprising a full 40 percent of global tropical fruit output.

In recent years, mango production and quality has been declining due to the occurrence of a variety of abiotic and biotic factors, some leading to the development of sudden death of trees (Masood *et al*., 2009; Masood *et al*., 2011). Abiotic factors affecting mango production include nutrients deficiency, drought, temperature fluctuations, mechanical injuries and improper management practices i.e. ploughing and intercropping (Ploetz, 2003; Malik *et al.*, 2004; Nafees *et al*., 2010; Masood *et al*., 2012). Mango is now cultivated throughout the tropical and subtropical world for commercial fruit production, as a garden tree, and as a shade tree for stock. The season for mangoes in Ethiopia starts in January to April and from September to November. Mangoes are mainly produced in west and East of Oromia, SNNPR, Beneshangul and Amhara.

Table 1 Estimate of area, production and yield of Mango fruits in Ethiopia (Binyam Teshome, 2010)

Year	Number of holders	Area in m^2	Production in quintal	Yield (kg/ha)
2003/04 (1996) E.C	350,067	4,96400	292,283.00	5888
2004/05 (1997) E.C	414,574	5,81400	301,715.00	5189
2005/06 (1998) E.C	463,868	5,40031	547,291.24	10406
2006/07 (1999) E.C	558,976	6,79610	626,111.83	9408
2007/08 (2000) E.C	695,030	6,73083	484,360.97	7196
2008/09 (2001) E.C	716,447	6,05100	441,582.00	7297

2.4 Occurrence and importance of mango anthracnose

Colletotrichum is one of the most economically important plant pathogenic genera causing anthracnose of fruits and leaves of a wide range of hosts worldwide, and particularly in the tropics and subtropics (Sutton 1992, Hyde *et al*., 2009a, 2010). The above-ground parts of plants and fruit trees can be affected by anthracnose disease and in the case of fruit infection results in reduction in quantity and/or quality and post harvest losses (Phoulivong *et al*., 2010a).The causal agent of mango anthracnose is *Colletotrichum gloeosporioides* which is a major and important fungal pathogen of mangoes. It has been reported that major losses occur from flowering till fruit set and also after harvest. Anthracnose is very common in wet, humid, warm weather environment and favor infections in the field. Warm, moist temperature further aids postharvest anthracnose development in mangoes (Nelson, 2008).

Colletotrichum species are cosmopolitan and it has been shown that multiple species can infect a single host, while a single species can infect multiple hosts (Cai et al. 2009, Hyde et al. 2009). Fungus-host relationships are broad, imprecise and often overlapping (Freeman *et al*., 1996), It is also believed that *Colletotrichum* species may adapt to new environments (Sanders and Korsten 2003), leading to serious cross infection problems in plant production. The study of pathogenic variability of *Colletotrichum* species is therefore important and the understanding of the host range of a particular pathogen may help in efficient disease control and management (Whitelaw-Weckert *et al*., 2007).

2.5 The genus Colletotrichum species

The genus *Colletotrichum* is classified into Eumycota to the major sub division of Deuteromycotina, class coelomyoetes; order Melanconiales (Agrios, 2005) and family melanconiaceae (Illingwoth *et al.,* 1991). The taxonomy of the species of the genus *Colletotrichum* is frequently revised and is still in a state of confusion (Bonde *et al.,* 1991). Representative of the genus *Colletotrichum* are ubiquitous and often polygamous causing a variety of disease symptoms commonly known as anthracnose on fruits, leaves and stems, die – back on branches, root rot, leaf spot, blossom rot, fruit and seedling branches of a wide range of

crops. The genus has also been recorded worldwide both as pre- harvest and post harvest causes of crop loss (Jeffries *et al.*, 1990).

2.6 Epidemiology

Moist conditions and high humidity are primary factors in the spread and development of anthracnose. Conidia produced on branch terminals, mummified inflorescences, flower bracts and leaves (most important) are significant sources of inoculum (Dodd *et al*., 1991; Fitzell and Peak, 1984). They are produced most abundantly when free moisture is available, but also at relative humidity as higher as 95%. Conidia are dispersed by rain splash and infection requires free moisture (Jeffries *et al*., 1990). As appressoria age, they become melanized. Melanization strengthens the appressorium and facilitates penetration of the cuticle by infection pegs that the appressoria produce.

The presence and prevalence of melanized appressoria have been used to predict when infection is possible and anthracnose control measures are needed (Fitzell and Peak, 1984; Dodd *et al*., 1991). Small fruit can develop minute brown spots and abort if infected early in their development. Once an appressorium is formed and fruit exceed 4 – 5 cm in diameter, infections stop development. Quiescent infections renew development once concentrations of preformed fungal inhibitors in fruit decline during the ripening process. On larger (especially ripening) fruit, lesions can form anywhere, but linear smears that radiate from the stem end to the apex are common. Lesions on fruit are superficial and extend into the flesh only after large portions of the fruit surface are affected. Nonetheless, even superficial disease development results in serious aesthetic damage and rejection of fruit along the marketing chain.

A study of the genetic diversity in the population of the mango anthracnose pathogen in Florida showed that there might be exceptions to the aforementioned general pattern of the disease cycle (Gentotti and Davis, 1993). Molecular analysis on isolates of *C. gloeosporioides* from different mango tissues revealed variation in patterns of pectic degrading enzymes. They concluded from the study that the fungus on mango was genetically diverse, suggesting variation in ability to

cause disease in different tissue by different isolates. Other related work also indicated that the mango population of *C. gloeosporioides* (Hyden *et al*., 1994). Mango fruit can also be infected with conidia from isolates of *Colletotrichum* sp. from other host plants such as avocado, papaya and citrus (Freeman and Shabi, 1996). The epidemiological significance of these potential inoculums sources, on the disease cycle, still need to be assessed.

Generally genetic and geographical data seem to suggest that the mango population of *C. gloeosporioides* was disseminated through the world from a single source as endophytes. An increased understanding of the origins and diversity of *C. gloeosporioides* on mango would have relevance to future research on host and control strategies across regions and locations. Termination of fungal quiescence on climacteric fruits appears to be related to the reduction of antifungal compounds or the production of ethylene by the ripening fruit (Prusky, 1996). As mango fruit ripens, there is reduction in the concentration of phenolic compounds, which are active against *C. gloeosporioides in vitro*. Similar systems have been found with avocado anthracnose (Pruskey and Keen, 1993). The involvement of ethylene in the termination of quiescence strongly suggests that *Colletotrichum* sp. must have coevolved to develop a mechanism to use the host's ripening hormones as signal to reactivate the infection process. This mechanism may prevent contact of the pathogen with host tissue that has high level of antifungal compounds. Resistance to the pathogen in mango fruit tissue is advantageous to the host during seed development, but not afterwards because the ripe needs to be destroyed by invading saprophyte or weak pathogens to help liberate the fruit to germinate in rich organic substrate. Therefore, there is evolutionary value in allocating chemical defense to the immature fruit not to the ripe fruit, as in apparently the case in mango (Pruskey and Keen, 1993).

2.7 Disease cycle

Dissemination: spores (conidia) of the pathogen are dispersed passively by splashing rain or irrigation water.

Inoculation: spores land on infection sites (panicles, leaves, branch terminals and fruits). Infection and pathogen development: on immature fruits and young tissues, spores germinate and penetrate through the cuticle and epidermis to ramify through the tissues. On mature fruits, infections penetrate the cuticle, but remain quiescent until ripening of the climacteric fruits begins.

Symptom and disease development: black, sunken, rapidly expanding lesions develop on affected organs.

Pathogen reproduction: sticky masses of conidia are produced in fruiting bodies (acervuli) on symptomatic tissue, especially during moist (rainy, humid) conditions. Many cycles of disease can occur as the fungus continues to multiply during the season.

Pathogen survival: the pathogen survives between seasons on infected and defoliated branch terminals and mature leaves. Ascospore production in dry leaves on the ground has been reported, but the role of the sexual stage in the disease cycle is unclear. Since conidia are formed abundantly in the mango canopy, this is considered to be the primary source of inoculum. In the field, *C. gloeosporioides* produces conidia on lesions on leaves, twigs, panicles, and mummified fruit. Conidia can be rain-splashed to other leaves or flowers and cause secondary infections; thus the disease is polycyclic in these organs. Developing fruit can be infected, and some isolates can cause pre harvest fruit loss. In the case of postharvest anthracnose, developing fruit are infected in the field, but infections remain quiescent until the onset of ripening, which occurs after harvest. Once the climacteric period of the fruit starts, lesions begin to develop. There is no fruit-to-fruit infection; hence postharvest anthracnose is a monocyclic disease.

A study of genetic diversity in the population of the mango anthracnose pathogen (Aruaz, 2000) showed there may be exceptions to this general pattern of the disease cycle. Mango fruit can be infected with conidia from isolates of *Colletotrichum* spp. from other host plants such as avocado, papaya, banana, coffee, and citrus. The epidemiological significance of these potential inoculum sources on the disease cycle has not been assessed. The lifecycle of anthracnose

diseases involves essentially production of spores on susceptible hosts, dispersal of spores, penetration of host tissue, initiation of an infection process within the cells, development of lesions, formation of bristly spores and dispersal usually by water-splash, air currents, insects or other forms of contact (Nelson, 2008).

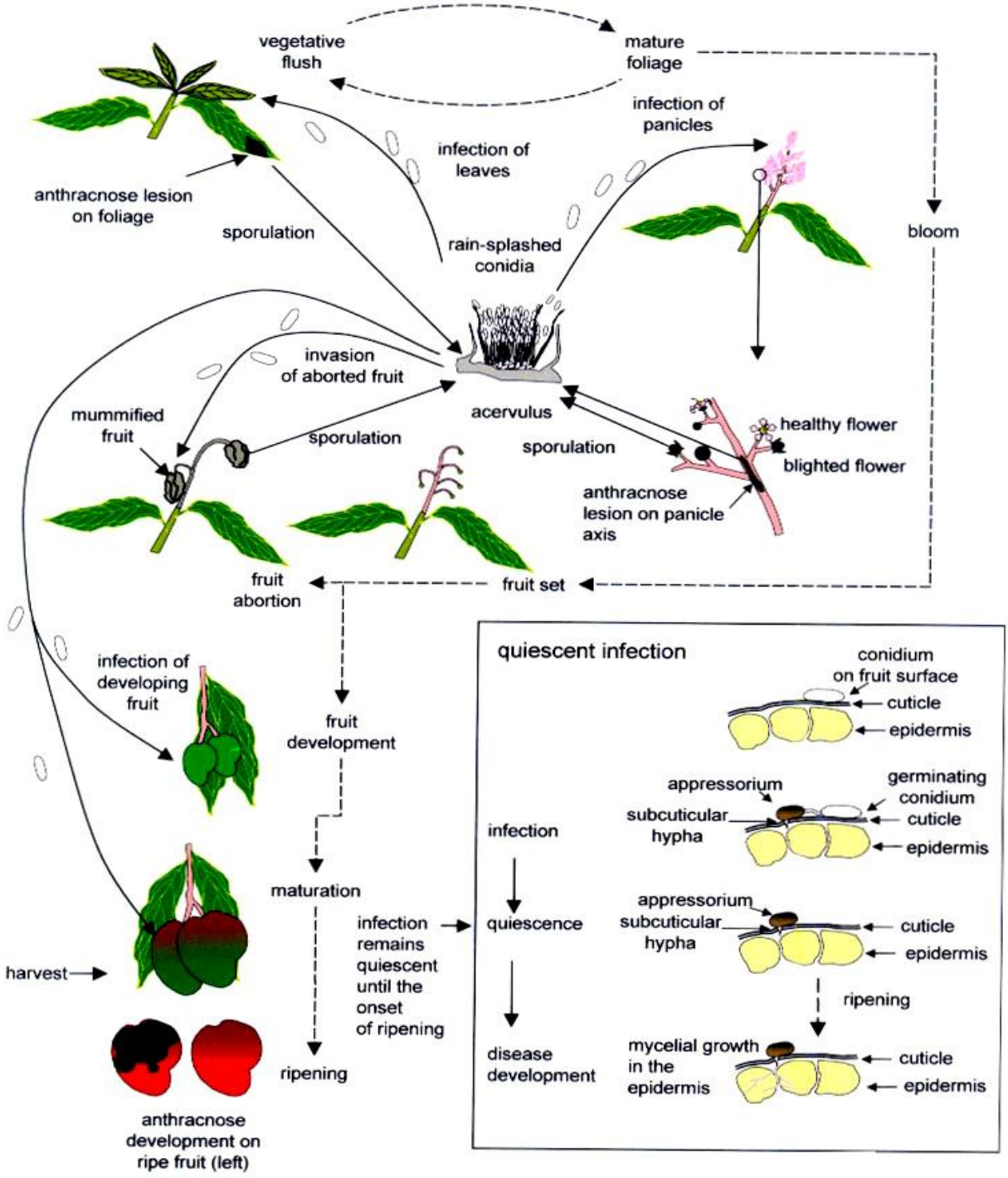


Figure 1 Anthracnose disease cycle (Arauz, 2000).

2.8 Distribution of mango anthracnose

Mango anthracnose is the most serious disease widely distributed *in* all mango growing regions of the world. It is a major constraint to the expansion of export trade of mango. Worldwide mango anthracnose is recognized as the most important field and post-harvest disease (Ploetz and Prakash, 1997). It was first reported from Puerto Rico (Collins, 1903) and later from Hawaii (Higgins, 1906), Florida (Fawcett, 1907), Cuba (Cardin, 1910), Philippines (Wester, 1911), Columbia (Taro, 1929), South Africa (Doidge, 1932), India (Stevens and Pierce, 1933), Brazil (Bitancounrt, 1938), United States (Traub and Robinson, 1938) and Pakistan (Sattar and Mallik, 1939).

2.9 Symptoms of anthracnose

The symptoms of anthracnose occur on the leaves, petioles, twigs, flower clusters and fruits of mango (Nelson, 2008). On the leaves, wounds begin as small, angular, brown to black spots that can expand to form extensive dead areas and can drop off during dry weather. On flowers, the symptoms are small black or dark-brown spots which can extend and kill flowers before fruit production severely decreasing yield. Ripe fruits affected by anthracnose develop sunken, major, dark brown to black decay spots before or after picking and fruits can drop from trees prematurely thus causing pre harvest infection of the fruit. The fruit spots can finally enter deep into the fruit, resulting in extensive fruit rotting. The majority of green fruit infections remain latent and largely undetectable until ripening. Fruits that appear healthy at harvest can develop significant anthracnose signs rapidly upon ripening (Nelson, 2008).A second symptom type on fruits consists of a “tear stain” symptom, a linear necrotic regions on the fruit that may or may not be associated with superficial cracking of the epidermis, exhibiting an “alligator skin” effect and even causing fruits to develop wide, deep cracks in the epidermis that extend into the pulp lesions on stems and fruits may produce prominent, pinkish orange spore masses under wet conditions (Nelson, 2008).

According to Johnson et al. (1995) and Arauz (2000), postharvest infections of anthracnose show a rounded black to brown lesion with a rounded border on the fruit surface. Lesions are normally confined to the peel but in severe situations, the fungus can enter the pulp. In the advanced stages of the disease, the fungus produces acervuli and lots of orange or pink masses of conidia in the lesions (Freeman *et al*., 1998; Arauz, 2000).

A B

Figure 2 Symptom of mango anthracnose on fruit (A) and the leaf (B) (**Ashutosh *et al.*, 2012)**

2.10 Damage caused by mango anthracnose

Anthracnose diseases, particularly those caused by *Colletotrichum* (Gloeosporium) or Glomerella fungi are very common and destructive on numerous crop and ornamental plants. Although severe everywhere, anthracnose diseases cause their most significant losses in the tropics and subtropics. Pre- and post-harvest losses of many high-value crops are substantial in the tropics due to various diseases caused by *C. gloeosporioides*. Flower infection on mangoes (blossom blight) can destroy flowers and young fruit and cause complete crop failure. Fruit infection may cause premature fruit drop, but major fruit losses occur during ripening when quiescent (dormant) infections break out and cause spreading black lesions. Anthracnose of other fruits also causes major post-harvest losses. Heavy infections cause rapid rotting, and even light infections which cause mainly cosmetic damage will shorten fruit storage life. Because of variability between seasons and locations, overall figures for losses are difficult to give, but it is clear that in many mango growing areas losses of up to 50% of the crop at the various stages of the disease would not be uncommon. (Nelson, 2008).

2.11 Biology and Ecology of mango Anthracnose

Anthracnose is a common name of plant diseases characterized by black lesions, usually sunken, caused by certain imperfect fungi that produce spores, e.g. *Colletotrichum*, *Gloeosporium* and some closely-related *Sphaceloma* species. The anthracnose pathogen reaches its most serious dimension at high moisture and warm temperature. For example *C. gloeosporioides* has an optimum of 25-29°C but it will also survive at temperatures as low as 4°C. Spore germination, dispersal and infection require relative humidity near 100%. However, in drier situations disease expression can occur when latent infections are activated through aging or tissue damage.

The anthracnose diseases are primarily transmitted through seed, but also through infected plant parts. Rain splash will also disperse spores within crop canopy. The pathogen persists on and in seed, crop residues, and weed hosts. Anthracnose is caused by fungi that produce conidia within black fungal fruiting bodies called acervuli (Waller, JM. 1992).

2.12 Management practices of mango anthracnose

A number of options are available for the management of mango anthracnose under field condition. Among these, orchard sanitation and pruning of dead twigs and branches which may harbour the fungus are the principal control measures used to reduce the source of a new infection cycle. The widespread occurrence of the inoculum of the fungus makes it impossible to control the disease by pruning and the removal of dropped leaves alone. To be more successful, the above mentioned measures have to be supplemented by spray applications using Mancozeb, copper oxychloride, Maneb, Propineb, Benomyl etc. cultivar selection and cultural or agronomic practices are also other options of managing mango anthracnose. The pre-harvest management can be achieved by regular spraying of trees from flowering time onwards with mancozeb (at recommended label rates every 14 days) to reduce the level of infection in the developing fruit. Copper sprays recommended for the control of mango scab will also control anthracnose with only a one day withholding period (Pitkethley and Conde, 2007). Post-harvest treatments are available for control of anthracnose in mango fruit. Prochloraz is used as a cold non re-circulating spray.

Hot water dips used to control fruit flies will also control anthracnose and stem end rots. Hot benomyl dips will control anthracnose and are useful where stem end rots are a problem (Pitkethley and Conde, 2007). Control of post harvest anthracnose disease can be achieved from field management and after harvest treatments or preferably a combination of both.

Biocontrol agents have also been considered in management of post harvest diseases of fruit and vegetables as a viable alternative to the use of present day synthetic fungicides (Pang *et al*., 2002). However, when these methods are applied as treatment alone, they prove to be less effective and inconsistent in their commercial applicability (Droby *et al*., 2001), limiting their

acceptance as an alternative to synthetic fungicides. Post harvest biological control of mango anthracnose has been attempted with limited and varying results (Arauz, 2000).Lack of efficacy in these methods and negative perception in the public towards synthetic chemicals lead to growing interest towards natural alternatives, particularly of plant origin.

Silva *et al.* (2008) studied several extracts from *A. eupatoria*, *Petiveria* sp. *D. lanata, P. lanceolata* and *S. rebaudiana* afforded very promising results to be used for the control of *C. gloeosporioides*. The most active extract was that from *O. manjorona* which inhibited 96% of *C. gloeosporioides* spore germination. One member of the *Amaryllidaceae* family *Polianthes tuberosa* L. was evaluated against the mycelial growth of *C. gloeosporioides* on potatodextrose-agar medium. Pandey *et al.* (2009) observed that 17 plant extracts checked the radial growth of the pathogens, however leaf extract of *Azadirachta indica* was found more effective in inhibiting the redial growth of the pathogen followed by *Moras alba* which is equally effective in inhibiting the radial growth of *C. gloeosporioides.* Leaf extract of *Syzygium communi* and *Lantana camara* were comparatively less effective against all the isolates of *C. gloeosporioides* Penz. and Sacc.

Excessive use of benomyl, thiophanate- methyl and thiobendazole as pre-and post-harvest sprays has led to a reduction in effectiveness in certain areas where pathogen resistance to fungicides has been reported (Spalding, 1982). Indiscriminate use of the chemicals is not only hazardous to people but also disrupt the natural ecological balance by killing the beneficial soil microbes (Ansari, 1995). The integration of a number of practices aiming to reduce or eliminate negative side effects caused by chemicals used for controlling major mango diseases is the most realistic option for solving the problem (Chowdury and Rahim, 2009).

3 MATERIALS AND METHODS

3.1 Description of Study Area

The study was conducted in the potential mango producing districts of Jimma Zone, south western Ethiopia from April to June; in production season of 2013. The study woredas were selected based on their mango production potential and area proximity. The ecological description of the study sites are indicated in Table 2.

Table 2 Location and climatic characteristics of the study woredas (source: Agricultural offices of the respective woredas (2013)

Study woredas	Location	Altitude(m.a.s.l)	Annual rain fall(mm)	Mean minimum temperature(^{o}c)	Mean maximum temperature(^{o}c)
Seka Chokorsa	$7^0$36'41"N and $36^0$44'12" E	1580-2560	1800-2300	15	25
Kersa	$7^0$38'-$7^0$54'30"N and $36^0$38'-$36^0$53'E	1600-2400	1587	10	32
Gomma	7 50'35" -7 51'00"N and 36"35'30"E	1387-2870	800-2000	12.4	28.4
Jimma	$7^0$138^{0}56N and $35^0$5237^{0}37E	1700- 1730	1637	11.43	26.2

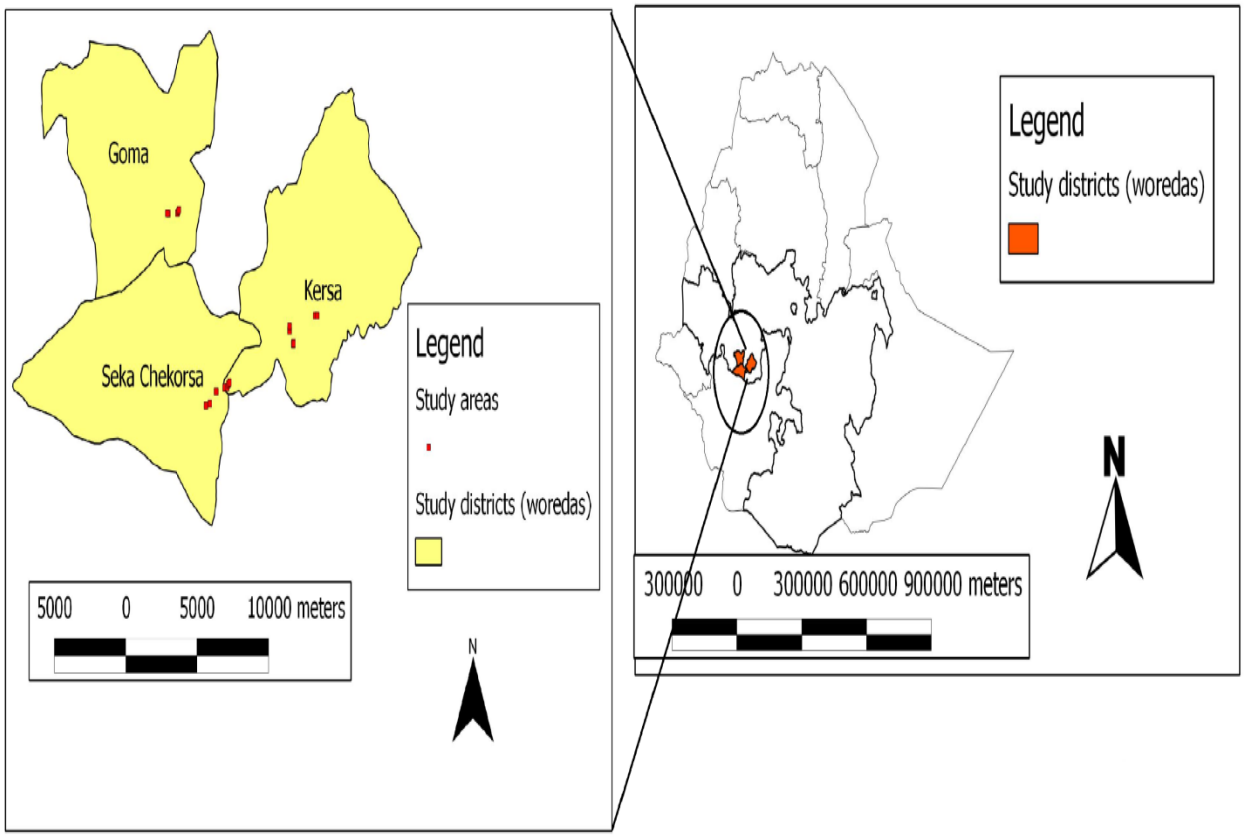


Figure 3 The map of the study area

3.2 Sampling Method

The survey was conducted in three major mango growing districts and one urban mango production area. These were Seka Chokorsa, Kersa, Gomma and Jimma town. Purposive sampling method was used for selecting woredas, kebeles within wareda and mango orchard. Random sampling method was used for selecting mango tree within orchard. Three kebeles per each waredas and six trees per plot were assessed. Longitude, latitude and altitude data from surveyed areas were recorded using Global Positioning Systems (GPS). In addition, cultural practices such as age of crops and cropping pattern were noted. For the post harvest assessment, four markets such as Beshishe which is located in Jimma, Agaro which is located in Gomma,

Sarbo which is located in Kersa and Seka markets which is located in Seka chokorsa woreda were assessed and traders were the source of the sampled fruits.

3.3 Assessment of disease incidence, severity and Prevalence

Designed questionnaires were used for oral interview of farmers, extension agents and experts. A total of six farmers were interviewed in each kebele by contacting the farmers face to face. For detail contents of the questionnaires, see appendix 6. The survey was conducted along the direction from Jimma town to the respective waredas. At each plot six trees were randomly selected and used for the disease incidence and severity assessment on leaves and fruits. Assessments were performed at three positions per tree (upper, middle and lower). The post harvest disease incidence and severity assessments were made at market place. For this 15 (fifteen) fruits from five traders were randomly selected and replicated three times for each market and assessed.

3.3.1 Disease Incidence

A systematic field survey of mango anthracnose was carried out in the three selected mango-growing districts and one urban area around Jimma to determine mango anthracnose frequency or occurrence and severity. Disease incidence on the fruit and leaves was measured using the following formula.

$$\textbf{Disease incidence}(\mathbf{I}) = \frac{\text{number of infected fruit (leaf)}}{\text{Total number of assessed fruit (leaf)}} \text{x}100$$ (Charles *et al., 2012*)

$$\textbf{Percent of occurrence (Prevalence)} = \frac{\text{number of field with infected mango}}{total} \text{x}100$$

3.3.2 Disease Severity

Disease Severity on the fruits was estimated based on percent area covered by lesions of the disease. Disease severity on plant parts was recorded using a five point rating scale (Corkidi, *et al.*, 2006; fig. 4). The assessment was done at physiological maturation stage of the fruit.

Percentage disease index (PDI)

Based on the numerical ratings given above a 'Percent disease index' for fruit anthracnose was calculated using the formula (Mayee and Datar, 1986):

$$PDI = \frac{\text{Sum of numerical ratings}}{\text{No. of plants scored X Maximum score on scales}} \times 100$$

Infected area	infected area	infected area	infected area	infected area
0-1%	1-5%	6-9%	10-49%	50-100%
No disease	slight disease	moderate disease	severe disease	very severe disease

Figure 4 The scale used for the assessment of field evaluation of severity of mango anthracnose (**Corkidi, *et al.*, 2006).**

3.4 Isolation

Isolation was done to confirm up the causal pathogen was anthracnose or other fungus. Isolation was done by cutting several small sections 3-5 mm^2 from the margin of the infected lesion so that they contain both diseased and healthy looking tissue of mango and surface sterilized by sodium hypochlorate (NaOCl) for about 15 to 30 seconds; the sections were taken out aseptically

one by one and at regular intervals to surface sterilize each at different times (Agostini and Timmer, 1992). The sections were washed in three changes of sterile water and then blotted dry on clean sterile paper towels then three pieces of tissue were placed per petridish with a freshly-prepared potato dextrose agar medium (PDA). The Petri dishes were then kept for 7 days in an incubator under a temperature of 28 °C, and then fungal growth was examined under a binocular and compound microscope.

3.4.1 Culturing, sub-culturing and culture preservation

Single spore isolation was carried out to obtain pure culture of *C. gloeosporioides* isolate, the purified culture was sub-cultured, Fungal culture was inoculated to test tube slant containing PDA and incubated at 28^0C for 7 days and at 5^0C for further use and then the pure culture was sub cultured from test tube slant and kept for 7 days at 28^0C and then used for pathogenicity test.

3.4.2 Pathogenecity tests

3.4.2.1 Preparation of spore suspension

Pathogenecity test of the identified *C. gloeosporioides* was conducted on detached leaf and fruit of mango. The suspension of conidia was prepared by suspending mycelia scraped from 7 days old of *C. gloeosporioides* penz. Separately in 3 milliliter sterile distilled water and shaking vigorously for 3 minutes (*Charles et al, 2012*). The resulting suspension was filtered through 2-layer cheesecloth. The concentration of spore suspension was adjusted to $1x10^6$ spores or conidia/millimeter using haemacytometer.

Figure 5 Preparation of fungal suspension

3.4.2.2 Fruit wounding technique (pin-plick inoculation)

Three mango cultivars (Tommy Atkins, Apple mango and local cultivar) were used. Inoculation was performed following the method of Sun et al. (2008). Fifteen green matured mango fruits, five fruits from each variety were randomly collected, thoroughly washed and disinfected in 70% ethanol and 1% NaOCl and arranged in randomized complete design(CRD) with three replication. . The disinfected fruits were then rinsed in four changes of sterile distilled water and air before inoculation. The fruits were each pierced with sterilized needle in three places; then 0.02mlspore containing $1x10^6$ per milliliter spore suspension of fungal isolates was placed on the wounded portion of the fruit by using pipette, sealed in moist plastic box with sponge which was sprayed with sterilized water to maintain at least 95% relative humidity (Than et al. 2008a), and incubated for 7 days in incubator at 28^oC. Control fruits were inoculated with sterile distilled water. Anthracnose symptoms were evaluated after 7 days.

Figure 6 Inoculation of mango fruits with isolate of mango anthracnose collected and isolated from different mango growing areas of Jimma zone, SW Ethiopia

3.4.2.3 Detached leaf technique (DLT)

Detached new leaves free from anthracnose symptom were collected, washed, and surface sterilized. The leaves were then sprayed with the spore solution of fungal isolate and placed on five larger plastic Petri dishes lined on the inside with moist tissue paper, covered with moist paper towels and incubated for 7 days at 28^{o}C until symptom appearance.

Figure 7 .The leaves of mango before inoculation by the pathogen

3.4.3 Re-isolation of isolated fungal pathogens

The causative organisms in the diseased parts were re-isolated on potato dextrose agar as described under 3.4 above. The characters of the re-isolated pathogens were compared with their original isolates.

3.5 Statistical data analysis

Data were first checked for various ANOVA assumptions. For incidence and severity data, an arsine square root transformation was performed to stabilize the variance. The field survey data for mango anthracnose was analyzed by using three stage nested design; mango tree was nested under assessed kebeles. Woreda was introduced as random effect factor in the model while kebeles and mango tree as fixed factors. The post harvest mango anthracnose data was analyzed using one way ANOVA in Minitab v. 16. The main and interaction effects of anthracnose disease response variables across location were determined using the proc GLM of SAS soft ware version 9.2(SAS institute, 2008). Mean separation was carried out using LSD test at 5% level of significance. The model for predicting the response variable of the field was expressed as:

$$Y_{ijkl} = \mu + \rho_i + \gamma_{j(i)} + \beta_{k(ij)} + e_{l(ijk)}$$

Where: - μ= is the overall mean

$Y_{ijkl=}$ is the dependent variable observed in replication **l** of treatment **ijk**,

$P_{i=}$ is the effect from the random factor of different location,

$\gamma_{j(i)}$ =is the effect of fixed factor of different kebeles with in a location and

$\beta_{k(ij)}$ =is the effect of fixed factor of selected different mango tree with in selected kebeles, and

$e_{l(ijk)}$ = is the error effect.

The model used for one way ANOVA for the market values were indicated as follows:-

$$Y_{ij} = \mu + \rho_{i+} e_{ij}$$

Where: - μ = is the overall mean

Y_{ij} = is the dependent variable observed in replication **i** of treatment **ij**,

P_i = is the effect from different markets

e_{ij} = is the error effect.

4 RESULT AND DISCUSION

Survey results indicated wide distribution of mango anthracnose across the different agro ecologies of study areas. The incidence and severity of mango anthracnose at farmer's field and main markets varied across the agro-ecology of the study areas, and these findings were presented and discussed as follows.

4.1 Constraints of mango production in the study areas

Survey results indicated that there are many constraints associated with mango production in the study areas. Among these, extreme environmental conditions, mango anthracnose and bacterial blight were some of the prominent problems. From all the respondents about 64.4% said that the production of mango was affected by mango anthracnose which blackens the fruits thereby predispose them to pre-mature dropping before harvest. About 27.7% and 2.7% of the respondents said that major problems of mango production were environmental condition and bacterial blight, respectively.

Table 3 Problems associated with mango production in the study areas

Constraints	Frequency of respondents	Percentage (%)
Mango anthracnose	50	64.4
Bacterial blight	2	2.7
Environmental condition	20	27.7
Total	72	100

4.2 Management strategy used in study area on mango anthracnose

Management of mango anthracnose disease associated with mango production, 6.9% of the respondents use combinations of inter cropping ,timely planting and removing infected plants, 16.6 % of the respondents use chemicals and 76.4% of the respondents did not use any kind of practices to control the problems (Table 4). Generally, according to the responses of the farmers, there was lack of cultural practices such as sanitation, pruning and different cropping pattern to control diseases associated with mango production in study area including mango anthracnose disease. Particularly, there was no practices of removing dead or diseased wood, additional growth flushes to allow more light penetration into the leaf canopy and control of tree height to facilitate cultural management practices such harvesting.

Table 4 Control strategies used in surveyed area on mango anthracnose

	Control	Frequency of respondents	Percentage (%)
Mango anthracnose	Cultural practice	5	6.9
	Chemical	12	16.6
	Other	55	76.38
	Total	72	100

4.2 Variety produced and Environmental condition for Anthracnose

Different variety of mango were produced in the study areas, from these about 83.3% respondents produce local variety, 6.9% of respondents produce Tommy Atkins and 9.7% of the respondent produce Apple mango variety (table 5). These results showed most of the respondents produce local variety of mango which is more than 20 years old. Different mango varieties introduced to these study areas by the community themselves were from unknown sources or with no information regarding their management and thus constraints such as failure to set fruit, extended periods on fruits setting, diseases and pests, etc are very common. All the

respondents said the diseases on mango were more prominent during humid and wet condition than hot and dry condition.

Table 5 Variety produced and Environmental conditions favoring mango Anthracnose in the study areas

Variety produced	Frequency of respondents	Percentage (%)	Environmental condition favoring mango anthracnose	Frequency of respondents	Percentage (%)
Local variety	60	83.3	Humid and wet condition	72	100
Tommy Atkins	5	6.9	Hot and dry condition	0	-
Apple mango	7	9.7	Other time	0	-
Total	72	100	Total	72	100

4.3 Incidence of mango anthracnose on the leaves

The three way interaction between location, kebele and tree canopy; the two way interactions between kebele and location, and the main effect of location were significant (Appendix Table 1). From all assessed kebeles significantly higher (83.5%) incidence was recorded on the lower tree canopy at Bulbulo kebele of Gomma district and the lowest (26.3%) anthracnose incidence was recorded in Kitto kebele on the upper tree canopy (Table 6). According to the review of (Johnson, 2008) the infection of *C. gloeosporioides* was from conidia spread by water and other movement in canopy. The disease may be more prevalent on leaves and fruit from lower west side of trees (in Southern hemisphere and Anthracnose is most noticeable in the lower branches. Often the very top portions of the tree escape infection and appear quite healthy in comparison to the lower sections of the tree. High disease incidence at Gomma might be due to high humidity, rain fall and heavy dew during critical infection periods which greatly increase the disease

incidence in the study areas as observed during assessment. Nelson (2008) stated that wet, humid and warm weather conditions favor anthracnose infections in the field and warm and humid temperature favor postharvest anthracnose development.

Table 6 The interaction effect of kebeles with tree canopy position on mango anthracnose incidence on leaf

Location	Kebele	Tree canopy	Incidence on the leaf (%)
Gomma	Kaso hixi	Upper	70.33^{abcd}
		Middle	71^{abcd}
		Lower	81.833^{ba}
	Bulbulo	Upper	71.667^{abcd}
		Middle	70^{abcde}
		Lower	**83.5^{a}**
	Agaro01		62.50^{def}
		Upper	61.667^{def}
		Middle	77.167^{ab}
		Lower	
Jimma town	Kitto		26.333^{m}
		Upper	**29.000^{lm}**
		Middle	37.833^{ijklm}
		Lower	
	Qoci	Upper	35.500ijklm
		Middle	29.833^{klm}
		Lower	37.333^{ijklm}
	Wuhalimat	Upper	62.5^{cdef}
		Middle	61.333^{def}
		Lower	71.167^{abcd}
Kersa	Ankaso	Upper	32.167^{jklm}
		Middle	33.500^{ijklm}
		Lower	43.833^{ghijk}
	Girma	Upper	33.333^{ijklm}
		Middle	37.500^{ijklm}
		Lower	47.833^{fghi}
	Marawa	Upper	37.000^{ijklm}
		Middle	38.667^{ijklm}
		Lower	61.667^{def}
Seka chokorsa	Saka	Upper	63.5^{cde}
		Middle	55.50^{efgh}
		Lower	68.5^{bcde}
	Gibe	Upper	41.167^{hijkl}
		Middle	45.333^{ghij}
		Lower	39.333^{ijklm}
	Qacama	Upper	68.33^{bdce}
		Middle	57.167^{defg}
		lower	68.333^{bcde}
P value			0.0390
CV (%)			20.92703
LSD			3.4097

Means followed by a different letter are significantly different at ($\alpha = 0.05$)

4.4 Incidence of mango anthracnose on the fruit in the field

The distribution of mango anthracnose on the fruit was different from kebele to kebele statistically (Table 7). The highest incidence was recorded in Kaso hixi kebeles of Gomma wareda (85%), but there was no statistically significant difference between Agaro 01 and Kitto kebeles of Gomma wareda and Jimma area, respectively. The lowest incidence on mango fruit was recorded in Ankaso kebele of Kersa woreda (34 %). This is most probably due to absence of favorable environmental condition for the pathogen. The highest disease incidence was recorded in Kaso hixi kebele of Gomma woreda most probably due to lack of sanitation, farmers did not prune the damaged stems from infected plants and they tended to ignore debris of diseased stems and fruits around the farm. Some reports on other crops considered conidia produced from debris or dead leaves as the main source of *C. gloeosporioides* inoculums which could rapidly initiate an epidemic once favorable condition for dispersal and infection occurred (Masanto *et al*., 2009).

Several studies conducted under field conditions found that the dispersal of those conidia was highly influenced by water, primarily rain splash (Alemayehu *et al*., 2010). Often the elevation of an area can be categorized into three altitude groups: Lowlands (< 1500 m a s l), intermediate (1500-2000masl) and highlands (> 2000masl). Accordingly, the current study areas have fallen between 1500-2000m, which was considered as intermediate altitude. Both anthracnose incidence and severity were significantly higher ($P<0.0001$) in intermediate altitudes. Anthracnose incidence and severity were 85% and 83.33%, respectively in the intermediate altitude. This result was in agreement with the finding of Alemayehu *et al*. (2010) on incidence and severity of sorghum anthracnose in Ethiopia who indicated the highlands had a lower disease incidence and severity, indicating that fields with severe anthracnose infection are consistently located in areas below 2000 m.a.s.l.

Table 7 Mean incidence of mango anthracnose on the fruit across Assessed Kebeles

Location	Kebeles	Incidence on fruit (%)
Gomma	Kaso hixi	(67.21)85[a]
	Bulbulo	(50.36)59.3[cd]
	Agaro01	(61.89)77.8[ab]
Kersa	Girma	(38.88)39.5[e]
	Marawa	(36.45)35.3[e]
	Ankaso	(35.67)34[e]
Seka	Gibe	(50.77)60[cd]
	Saka	(46.43)52.5[d]
	Qacama	(36.75)35.8[e]
Jimma area	Qoci	(60.94)76.4[b]
	Wuhalimat	(54.15)65.7[c]
	Kitto	(63.44)80[ab]
LSD	-	7.95
CV (%)	-	18.9
P Value	-	p <.0001

The data in the bracket are transformed data. Means followed by a different letter in the column are significantly different at ($\alpha = 0.05$)

4.5 Severity of mango anthracnose on the fruit

The ANOVA showed significant ($p<0.0256$) interaction effect between Location, kebeles and tree canopy (Appendix Table 3). The severity of mango anthracnose was highest in Kaso hixi Kebele of Gomma woreda (80.5%) at the lower tree canopy and the lowest at Marewa kebele of Kersa Woreda (30.3%) at the upper tree canopy (Table 8). The higher severity of mango anthracnose on fruit at Kaso hixi kebeles on the lower tree canopy might have been due to the fact that anthracnose is most noticeable in the lower branches where there is more humidity. Often the very top portions of the tree escape infection and appear quite healthy in comparison to the lower sections of the tree. Lack of sanitation and cultural practice on trees was also other factor for the highest severity. In addition, mango fruiting in the zone usually coincides with rainy seasons and high humid conditions associated with high temperatures, which favor infection and colonization of the crop by fungal pathogens and which in turn predisposes mango production to serious fungi attack. If orchards were free of bushes, there would be greater aeration within the orchards, which will in turn, reduce temperature and more sunlight penetration, which could reduce the relative humidity within the orchards but this was not observed in present study. There was lack of adequate spacing of trees, pruning and removal of dead leaves and branches which increase humidity and temperature in the orchards. Due to this, the disease was more severe in the assessed woredas. In this study the severity of mango anthracnose on fruit across kebeles were significantly different among the mango growing areas and among studied kebeles; there was no area completely free from the presence of mango fruits anthracnose. This was in agreement with (Pitkethley and Conde, 2007) who state that anthracnose disease is found in all mango growing areas of the world.

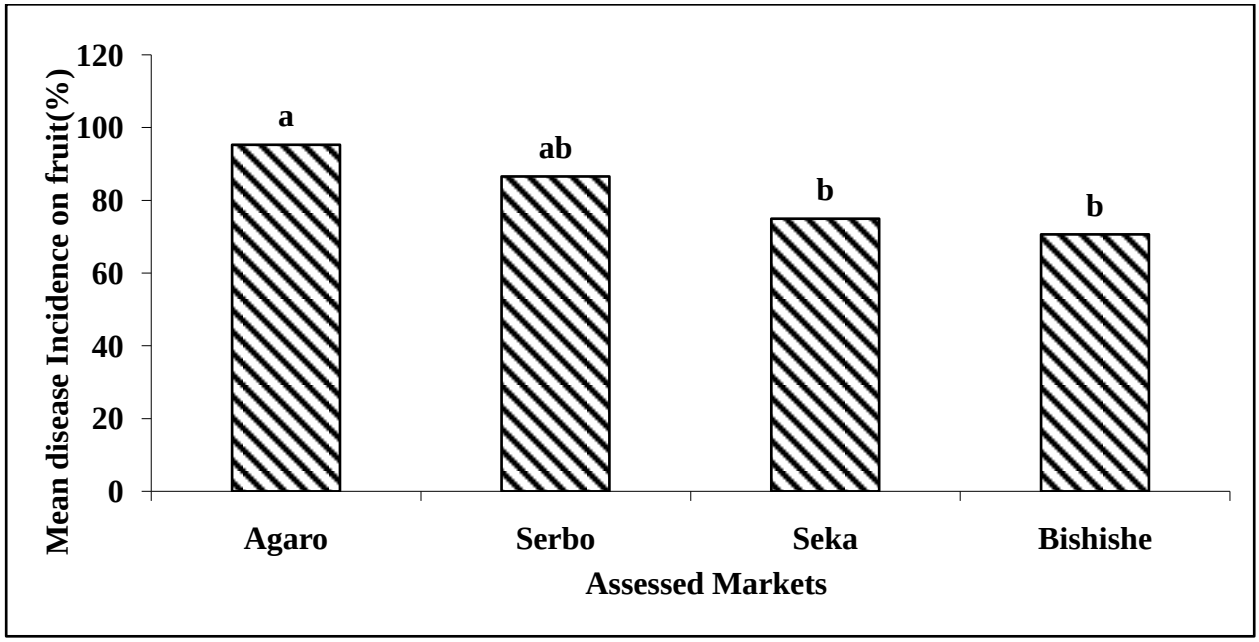
Table 8 Interaction effect of kebele with tree canopy on severity of mango anthracnose on fruit

Location	Kebele	Tree canopy	Severity on the fruit (%)
Gomma	Kaso hixi	Upper	63.333^{bc}
		Middle	55.833^{cdef}
		Lower	80.5^{a}
	Bulbulo	Upper	55.000^{cdefg}
		Middle	50.500^{cdefgh}
		Lower	66.667^{bc}
	Agaro01	Upper	57.667^{cde}
		Middle	62.333^{bcd}
		Lower	72.500^{ab}
Jimma town	Kitto	Upper	48.333^{efghij}
		Middle	47.000^{efghijk}
		Lower	50.500^{cdefgh}
	Qoci	Upper	38.667^{ijklmn}
		Middle	37.333^{jklmn}
		Lower	39.500^{ijklmn}
	Wuhalimat	Upper	44.000^{fghijkl}
		Middle	38.667^{ijklmn}
		Lower	45.000^{fghijkl}
Kersa	Ankaso	Upper	34.167^{lmn}
		Middle	35.167^{klmn}
		Lower	37.667^{jklmn}
	Sarbo	Upper	50.333^{cdefgh}
		Middle	44.333^{fghijkl}
		Lower	53.833^{cdefgh}
	Marawa	Upper	30.333^{mn}
		Middle	34.833^{klmn}
		Lowe	38.167^{ijklmn}
Seka chokorsa	Saka	Upper	41.333^{ghijklm}
		Middle	40.000^{fghijk}
		Lower	43.000^{ijklmn}
	Gibe	Upper	41.333^{ijklm}
		Middle	40.333^{fghijkl}
		Lower	42.000^{fghijkl}
	Qacama	Upper	40.000^{jklmn}
		Middle	41.000^{ijklm}
		Lower	47.833^{efghij}
P value		P=0.0256	
CV (%)		21.3	
LSD		3.3723	

Means followed by a different letter are significantly different at ($\alpha = 0.05$)

4.6 Incidence of mango anthracnose at the market

The incidence of mango anthracnose significantly varied from market to market (Appendix Table 4). Incidence values as high as 95.3%, 86.6%, 75% and70.66% in Agaro, Serbo, Seka and Beshishe market respectively were recorded (Fig. 8). The incidence of mango anthracnose was higher in the market than at field condition. This could be attributed to fruit softening during the ripening process, the natural defense mechanisms break down, and latent infections of anthracnose develop into black lesions that rot the whole fruit in days. Post harvest anthracnose is the major reason for losses of mangos during storage and transport. As explained by Leonard and Williams (2012) the incidence of this disease can reach almost 100% in fruit produced under wet or very humid conditions. Postharvest disease development is a major constraint to the quality and shelf life of mango fruit there by limiting its domestic and export marketing (Bally et al., 2009). Like other fresh commodities, mango has also been found prone to postharvest fruit decay due to rapid disease development during storage and ripening (Prusky et al., 2009).



Bars copped with the same letter(s) are not significantly different at $p<0.05$

Figure 8 Incidence of mango anthracnose disease across different markets in SW Ethiopia

4.7 Severity of mango anthracnose in the market

The result of the study revealed that statistically there was no significant difference in anthracnose severity among the assessed market places (P> 0.05) (Appendix Table 5). The mean values of the mango anthracnose severity at the market were 82%, 75.3%, 74% and 64% in Agaro, Serbo, Seka and Jimma market, respectively. Disease severity would be even greater at consumer level where ripe fruits might be stored for one or more days from purchase and the fungi keep on decaying the fruits. Son-Quang (2002) reported that postharvest losses of mango could go up to 97%, depending on cultivars, locations, cultural practices and environment.

4.8 The relationship of anthracnose disease in the farmers field and in the market

From this study the percentage of disease incidence and severity on the fruit was higher in main markets than that of farmer's field (Fig.9). For instance, the highest incidence and severity of the disease were recorded in farmers' fields of Gomma woreda and Agaro market (the large local market in Gomma woreda). The incidence was higher in farmers' fields in Gomma woreda (74%) and Agaro market (95%) respectively and the severity was also highest in farmers' fields in Gomma woreda and Agaro market by 63% and 82% respectively. So there is a relationship between farm and market indicating that fruits in the market are largely brought from the farmer's field to the local market and those fruits were already infected in the field and disease development severely increased when brought in to the market. The highest disease incidence and severity recorded in the market places might be due to infections which occur before harvest and then remain quiescent until sometime during ripening and poor postharvest handling practices. Anthracnose, which is the most serious postharvest disease of a wide range of tropical and sub-tropical fruits such as mango, banana, papaya and avocado, is an example of a disease arising from quiescent infections established prior to harvest (Johnson, 2009).

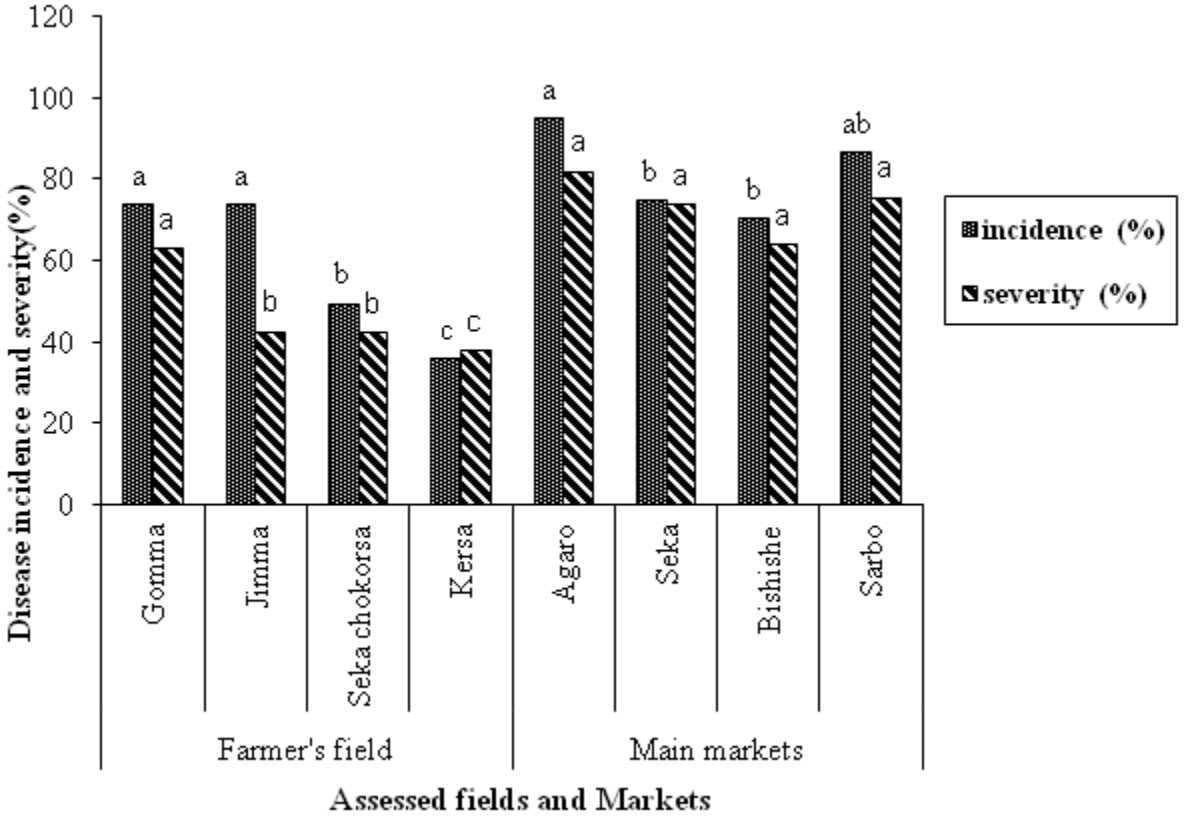


Figure 9 The comparison of disease incidence and severity in farmer's field and in main markets

4.9 Isolation of the causal pathogen

Inoculation of solidified Potato dextrose agar with small cut pieces of lesions from the symptomatic mango fruits and incubation at temperature that fluctuated between 28 and 30^{o}C for 7 days produced mixed fungal growth, which was later sub-cultured to obtain pure cultures. Some of the pure cultures obtained, on Potato dextrose agar, had colonies that were whitish to dark grey with thick to sparse lawns of aerial mycelium when viewed from the top of Petri dishes (Fig.10a) and were greenish to orange or dark brown centre bordered by creamy surrounding when viewed from the reverse side of the Petri dish (Fig. 10b). When viewed under the microscope, conidia were observed to be hyaline; single celled and cylindrical with obtuse ends (Fig. 10c). The fungus was, morphologically identified to be *Colletotrichum*

gloeosporioides. On PDA (Potato-Dextrose-Agar) medium, the fungus (*C. gloeosporioides*) grew well with grayish white to dark grey and produce aerial mycelium ranging from a thick mat to sparse tufts. Conidia are hyaline, unicellular and either cylindrical with obtuse ends or ellipsoidal with a rounded apex and a narrow truncate. This result was in agreement with Charles *et al., 2012.*

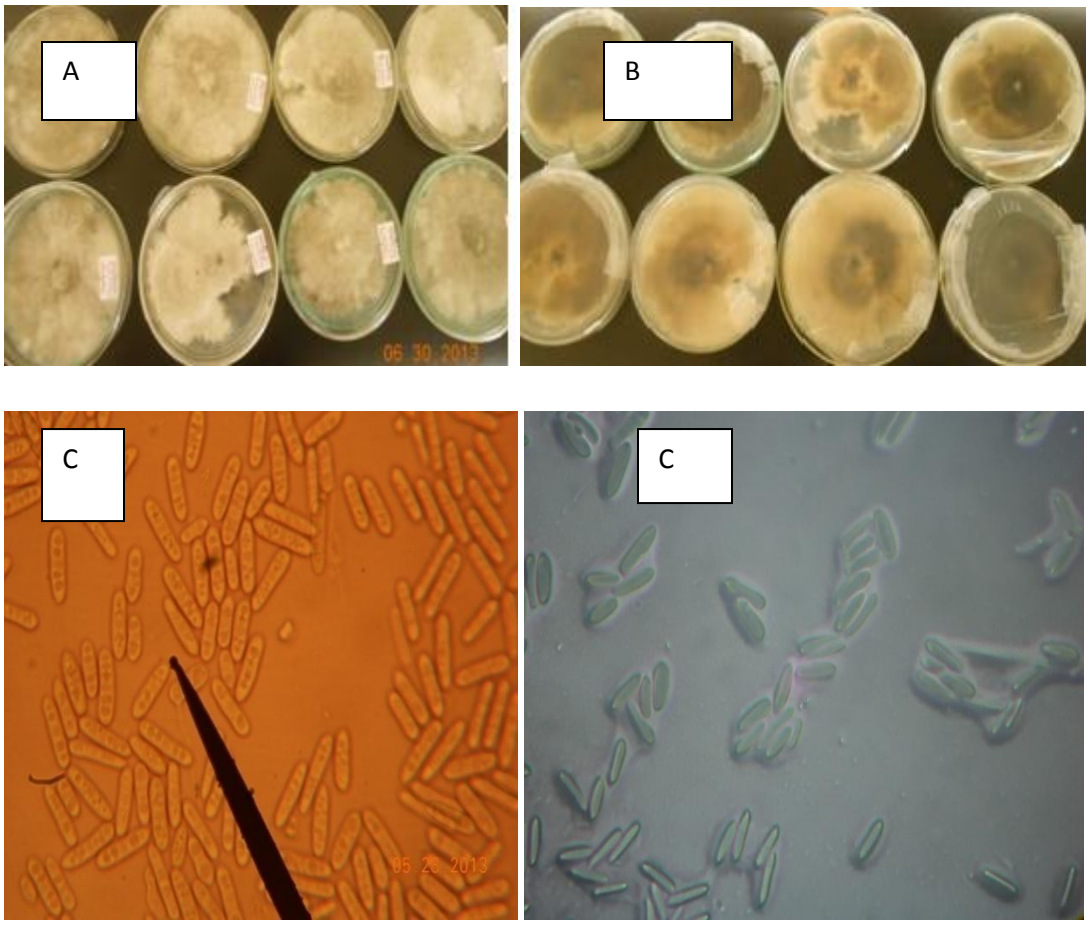


Figure 10 *Colletotrichum gloeosporioides* the causal agent of mango fruit anthracnose disease (a) top view of colony in a Petri dish (b) reverse view and (c) Microscopic view

4.10 Pathogenecity test

Pathogenecity test was carried out separately for mango anthracnose (*Colletotrichum gloeosporioides)* isolated from symptomatic mango fruits. The inoculated fruits showed anthracnose disease symptom typical of those observed after 7 days old on both healthy leaf and

fruits of mango (Fig. 11). This test was in agreement with (Than *et al*., 2008; Sangeetha and Rawal, 2009; Jayasinghe and Fernando, 2009) who confirmed the pathogenicity test on detached mango fruit.

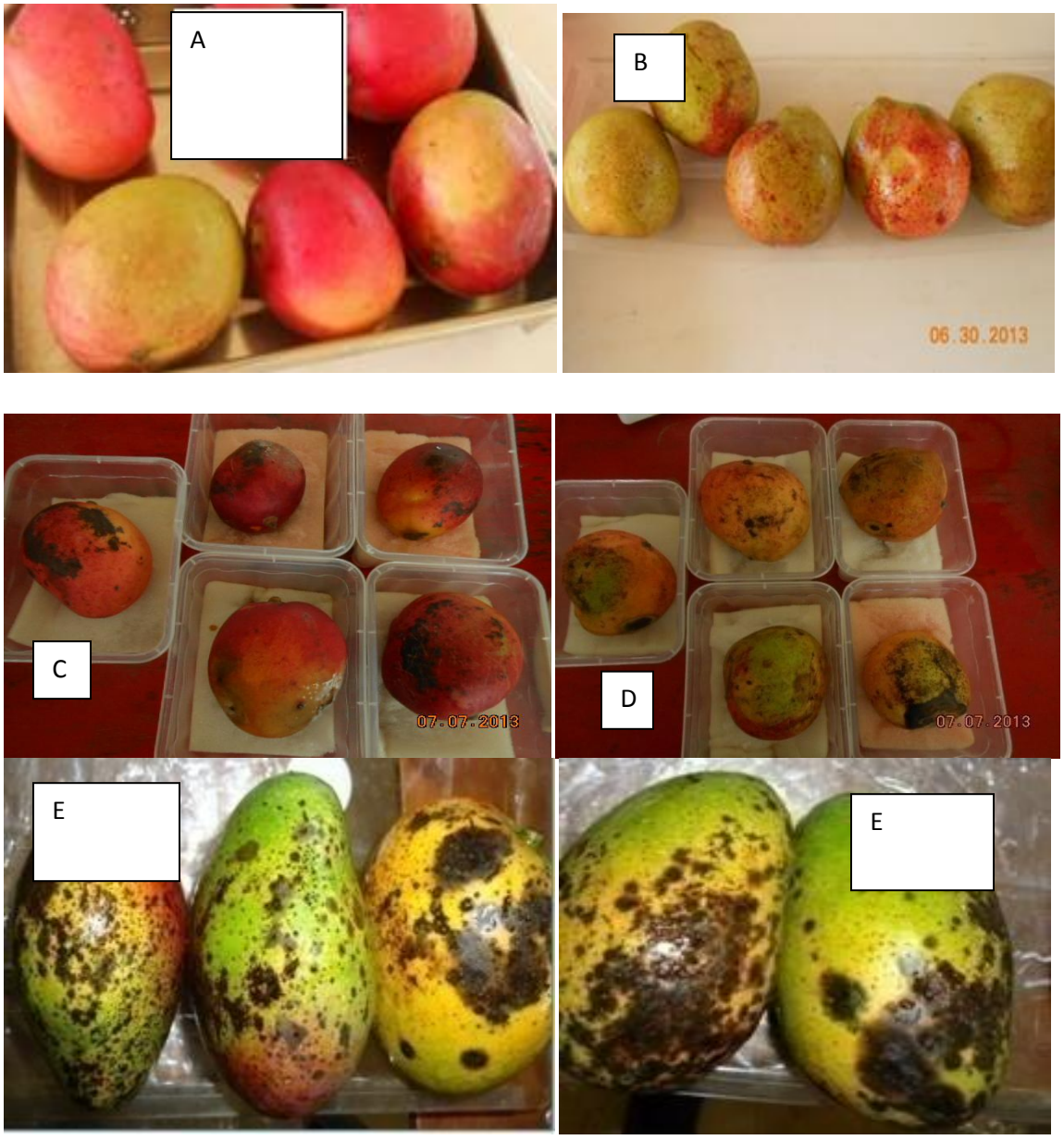


Figure 11 Tommy Atkins variety (A), Apple (B) variety before inoculation and tommy atkins variety(C), apple variety (D) and local variety (E) after inoculation with symptom.

5 SUMMARY AND CONCLUSION

Mango (*Mangifera indica L.)* is grown throughout the tropics and subtropics of the world and it belongs to the family *Anacardiaceae.* In recent years, mango production and quality has been declining due to the occurrence of a variety of abiotic and biotic factors, some leading to the development of sudden death of trees. Major Pre and post-harvest fungal problems on mangoes in Africa include anthracnose caused by *Colletotrichum gloeosporioides.* Anthracnose, caused by the fungus *Colletotrichum gloeosporioides*, is undoubtedly the most common and widespread fungal disease of mango and is a major factor limiting production in areas where conditions of high humidity prevail. The fungus invades inflorescences, fruits, leaves and twigs. This study was to assess the distribution and occurrence of mango anthracnose *(Colletotrichum gloeosporioides)* both on farmer's field and in main markets in Jimma area, South Western Ethiopia.

In the present study, three potential mango producing waredas and one urban area namely, Kersa, Gomma, Seka chokorsa waredas and Jimma area respectively were assessed. The distribution of mango anthracnose (*Colletotrichum gloesporides P*enz Sacc) in four waredas and 12 kebeles were assessed. During study data such as incidence on the leaf and fruit at upper, middle and lower tree canopy in the field, severity on fruit at upper, middle and lower tree canopy in the field, Incidence and severity on fruit on the market, altitude, Latitude of the study area, cropping pattern, cultural practices performed by local community for controlling the disease (oral interview) and the environmental condition during assessment were recorded.

The results of the study showed higher mango anthracnose incidence on the leaf at Gomma woreda (72.2%) and lower (41 %) at Kersa woreda. The mean incidence on the leaf varied from one woreda to other wareda. The result from the study reveals that the mean incidence of mango anthracnose on the fruit in the field was higher in Gomma Woreda (74%) and lower in Kersa Woreda (36.24%). The anthracnose incidence on fruit was higher at the lower tree canopy (62.48%) and lower at the upper tree canopy (54.87%) but statistically there was no significantly difference among the three tree canopy. The severity of mango anthracnose was higher in

Gomma Woreda (63.1 %) and lower in Kersa Woreda (38.1 %). The severity across Woreda was significantly different from each other. The disease assessment was done at four markets. Accordingly, higher incidence (95.3%) and severity (82.0%) was recorded at Agaro market and the lower incidence (70.66%) and severity (64%) was recorded in Jimma (Bishishe) market.

Generally from this study mango anthracnose (C. *gloeosporioides*) was 100% prevalent in the assessed three waredas and one urban area. When the incidence of mango in the field on leaf and fruit in the woredas was compared the highest incidence was recorded on the fruit (74%) and the lowest disease incidence was recorded on the leaf of mango (72%). The disease was more severe in the market than in the field, due to latent infection occurs when a fungus starts to germinate. Infection initiates and then halts for an undefined period which infection is continued. Thus, it presents a dormant phase in the parasitic relationship and appear on the fruit after harvest. Although anthracnose was found prevalent in all mango-growing areas surveyed, the occurrence and severity was probably more influenced by environmental conditions and cultural practices.

The fungus invades panicle, twigs, leaves and fruits but this work was conducted only on the leaves and fruit. So to know the distribution of this disease on the panicle and twigs further research is needed. In addition, to get full picture of the prevalence of this disease and to design appropriate control methods, it is advisable to conduct similar assessments in different mango growing agro ecologies and along mango value chain.

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7. APPENDICES

Appendix Table 1 Analysis variance for mango anthracnose incidence on the leaf.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Location	3	23534.59871	7844.86624	9.53	<.0001
Canopy	2	30109.24042	6021.84	22.31	1.000
kebele(location)	11	29302.10206	2663.82746	20.98	<.0001
mango(locati*kebele)	54	29302.10206	365.86536	2.88	<.0001
Kebele(loca)*canopy	22	16994.64	128.74	2.00	0.0390
Error	142	20581.6667	144.9413	-	-

Appendix Table 2 Analysis variance for mango anthracnose incidence on the fruit

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Location	3	29023.6064	9674.5355	35.45	<.0001
Canopy	2	662.46377	132.49275	1.07	0.3775
kebele(location)	11	104434.1947	9494.0177	34.78	<.0001
mango(locati*kebele)	54	49779.2788	921.8385	3.38	<.0001
Kebele(loca)*canopy	22	16994.64734	128.74733	2.00	0.0890
Error	142	20581.6667	144.9413	-	-

Appendix Table 3 Analysis variance for mango anthracnose severity on the fruit

Source	DF	Type III SS	Mean Square	F Value	Pr > F
location	3	6592.38426	2197.46142	25.09	<.0001
canopy	2	345.30435	69.06087	0.76	0.5790
kebele(location)	11	21437.37963	1948.85269	22.25	<.0001
mango(locati*kebele)	54	8494.02778	157.29681	1.80	0.0033
Kebele(loca)*canopy	22	1606.69	87.92	0.73	0.0256
Error	142	12436.500	87.5809	-	-

Appendix Table 4 Analysis variance for fruit incidence of mango anthracnose on the market

Source	DF	SS	MS	F	P
C1	3	1121.7	373.9	4.31	0.044
Error	8	694	86.7	-	-
Total	11	1815.7	-	-	-

S = 9.314 R-Sq = 61.78% R-Sq(adj) = 47.44%

Appendix Table 5 Analysis variance for fruit severity of mango anthracnose on the market

Source	DF	SS	MS	F	P
C1	**3**	**497**	**165.7**	**2.77**	**0.111**
Error	**8**	**478.7**	**59.8**		
Total	**11**	**975.7**			

S = 7.735 R-Sq = 50.94% R-Sq (adj) = 32.54%

Appendix 6 Questionnaire

This questionnaire is prepared to get feedback from the farmers on mango production practice and cultural practices used for disease management on mango in selected districts of Jimma. I would appreciate for all the cooperation made.

Please introduce yourself and the objectives of the study to the interviewee very politely. Complete the questionnaire by circling the letter of the choice and filling in the open ended questions very patiently. One question may have more than one answer. Please don't forget to thank the interviewee after completing the interview.

Date: _____

Name of district: _____

Agro-ecological location _____

Name of interviewee: _____

Name of interviewer: _____

Problems on mango and management practices

1. What are the constraints of production or problems of mango in this area?

A. vertebrate pest B. diseases C. environmental condition

2. How can you control the disease occur on the crop (on mango)?

A. cultural practice B. chemical practice C. other

3. Is there local/ modern technologies available to:-

A. Prevent B. Eradicate C. Control D. Not available

4. If the technology for the above question available are the technologies:-

A. Economically feasible B. Not feasible C.Not applicable

5 what kind of variety do you grow, from where you brought and by what criteria do select the variety? A. local B.Vandyke C. Tommyatkins D.Apple mango E.other

6 when did this disease start to affect your crop? A. humid condition B. Hot condition C. other time

How long have you practiced mango production? _____ years

8. Is mango consumed in your family? A. Yes B. No

9. If yes, Experience in mango consumption? _____ Years

10. Do you have information about mango anthracnose? A, Yes B, No

11. For the above question if the answer is yes, what is the amount of loss due to this disease?

_____

_____

12. How can you scout or asses the presence of disease on mango?

_____

_____

13. How can you produce mango?

_____

_____

14. Is there cultural practice to control mango anthracnose? A .yes B. No

15. If the answer for the above question is yes, what kinds of cultural practice do you apply when cultivating mango?

16. Have you use fungicide for controlling mango anthracnose? A, Yes B, No

17. If the answer for the above question is yes what is the name of the fungicide and from where you got it?