

## Citrus Production, Constraints and Management Practices in Ethiopia: The Case of *Pseudocercospora* Leaf and Fruit Spot Disease

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### Abstract

Citrus is economically important fruit crop in Ethiopia. However, its production is seriously constrained by various diseases including *Pseudocercospora* leaf and fruit spot. Surveys were conducted between June 2012 and May 2013 in the main citrus production areas of the country to assess the spread of the disease, and to determine its incidence and severity. A total of forty-nine citrus orchards in twenty-eight districts were surveyed. Random sampling techniques were used for data collection. The results showed that the disease had prevailed and widely spread in the districts assessed in the wet humid areas of the south, southwest, northwest, and north central parts, but not in the low altitude drier areas of the central rift valley and the eastern region of the country. Accordingly, 63.3% of the orchards surveyed were infected with the disease. The overall mean incidences of the disease on leaves of sweet orange, mandarin, lemon and lime were 36.2, 21.5, 17.1 and 16.3% while disease severity ratings were 2.6, 2.3, 2.0 and 1.7, respectively. Similarly, the average incidences and severities on fruits were 63.8, 29.4, 18.0 and 16.7%, and 4.0, 3.0, 2.0 and 2.0, in the same order. However, disease incidences in the different orchards ranged from zero to 76.7% on leaves and from zero to 100% on fruits. Disease severity ratings also varied from one to five on both leaves and fruits. In general, citrus orchards in the south and southwest Ethiopia that are known to have high rainfall and humidity conditions were more severely affected by the disease than those in the northwest and north central parts of the country. In these areas, respondents indicated that many farmers completely abandoned their citrus trees due to this disease. The causative agent was also identified to be the fungus *Pseudocercospora angolensis* based on cultural and morphological characteristics, and pathogenicity tests of representative isolates obtained from infected leaf and fruit samples.

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### Article Information

#### Article History:

Received : 28-04-2014

Revised : 16-05-2014

Accepted : 18-05-2014

#### Keywords:

Citrus

Leaf and fruit spot disease

*Pseudocercospora*

*angolensis*

Ethiopia

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## INTRODUCTION

Citrus (*Citrus* spp.) is one of the most economically important fruit crops grown by smallholders and commercial farmers in Ethiopia (Seifu, 2003; Kassahun *et al.*, 2006; Mohammed, 2007). The total area coverage and the annual production of citrus were estimated 5,947 ha and 77,087 tons, respectively (CSA, 2011 and 2012). However, citrus production and productivity in Ethiopia is seriously threatened by various diseases including leaf and fruit spot disease (Eshetu, 1999; Seifu, 2003; Mohammed, 2007; Sisay, 2007), which is caused by the fungus *Pseudocercospora angolensis* (Carvalho and Mendes] Crous and Braun) (Seif and Hillocks, 1993; Kuate, 1999; Pretorius *et al.*, 2003).

*Pseudocercospora* leaf and fruit spot (PLFS) disease has been reported as very destructive on citrus species in twenty-two tropical African countries and Yemen since its first observation in Angola and Mozambique in 1952 (Seif

and Hillocks, 1993; Kuate, 1999; Mohammed, 2013). Due to the traditional trade of infected fruit and planting material, and the wind-borne dispersal nature of the fungal spores, PLFS disease is a potential threat to the Mediterranean basin and South Africa (Seif and Hillocks 1993; Kuate, 1999), and to the major producing American and Asian countries that supply more than 70% of the world's citrus production (Ndo *et al.*, 2010).

The PLFS disease affects virtually all citrus species (Kirk, 1986; Kuate, 1999; Dewdney and Timmer, 2009). It became a serious citrus production constraint due to its impact on yield, quality and international trade. The disease occurs at all development stages and indiscriminately attacks leaves, fruits and young twigs. It causes devastating lesions that result in considerable premature leaf and fruit drop, and blemish fruits that remain on the tree. It can weaken trees and jeopardize

production in severe conditions (Eshetu, 1999; Timmer *et al.*, 2003; Mohammed, 2007). The disease can cause 50 to 100% yield loss; and a single lesion usually renders the fruit unsalable (Seif and Hillocks, 1993, 1999; Kuate *et al.*, 2002; Chung and Timmer, 2009). It also affects the yield and quality of essential oils extracted from citrus fruit peels (Kuate *et al.*, 2003).

In Ethiopia, characteristic symptoms of PLFS disease were first observed in the southern part in late 1980s (Yimenu, 1993). In early 1990s, similar disease symptoms were evident in the southwest areas. In some of these areas, entire fruit loss occurred and many farmers uprooted their citrus trees (Eshetu, 1997, 1999). Later, the disease was reported in the northwest part of the country (Yigzaw and Gelebelu, 2002; Kassahun *et al.*, 2006; Mohammed, 2007). Eshetu (1999) carried out a survey in southwest Ethiopia on sweet oranges and reported 100% incidence, and 43% and 65% severity on leaves and fruits, respectively. Mohammed (2007) also conducted a survey in south, southwest and northwest Ethiopia and reported incidences that ranged from 64.5 to 98.6% on leaves and from 78.6 to 94.2% on fruits, and severity which varied from 52.4 to 84.0% on leaves and from 65.6 to 90.4% on fruits.

Knowledge on the pathogen population including its biology and ecology is required for appropriate monitoring and development of effective management strategies to combat the disease (Sisay, 2007; Amata *et al.*, 2009). Management efforts are also based on the economic importance of the disease and its geographical distribution (Garnsey *et al.*, 1998; Hughes *et al.*, 2002; McDonald and Linde, 2002; Mohammed, 2007). However, the available information on the biology of the fungus and the epidemiology of PLFS disease is very limited (Eshetu, 1999; Kuate, 1999; Mohammed, 2007). In Ethiopia, there is no information on the current occurrence and distribution, and the extent of damage due to citrus PLFS disease. Therefore, this study was initiated to assess the present spread of PLFS disease, and to determine its incidence and severity in the major citrus growing areas of the country.

## MATERIALS AND METHODS

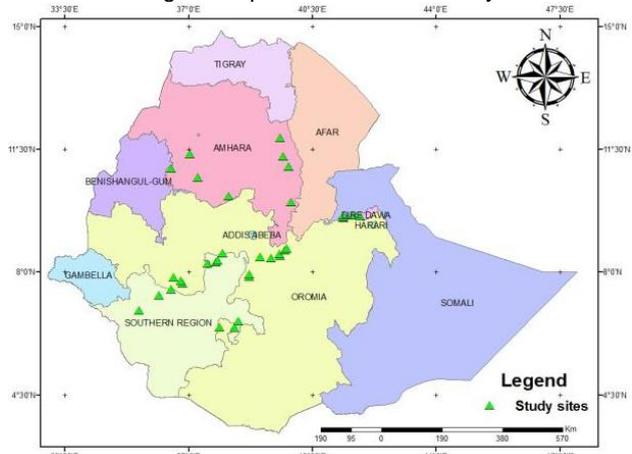
### Survey Area

Field surveys were conducted between June 2012 and May 2013 on various citrus species in the main citrus growing areas of Ethiopia. The geographic distribution of citrus orchards surveyed in the country is indicated in figure 1. A total of forty-nine citrus orchards and nurseries were surveyed in twenty-eight districts in the South Nations, Nationalities and People (SNNP), Oromia, Amhara and Somali Regional States, and the Dire Dawa City Administration where nearly the entire citrus production of the country is located. Citrus orchards, including backyards of small farmers, groves of Farmers' Association and Federal Prison Administration of Ethiopia, private and public medium and large scale commercial farms, fruit nurseries of the Ministry of Agriculture (MoA), and citrus foundation blocks in the research centers were surveyed.

### Assessment of Citrus Orchards in Ethiopia

During the surveys, detailed questionnaire was used to collect general information about each citrus orchard from the farmers, development workers, horticulture experts, and researchers. Each citrus orchard considered in the

survey completed one questionnaire. The questionnaire was compiled in three sections. The first section dealt with geographical and climatic aspects of the major citrus production areas. The second section covered the history of each citrus orchard, including farm size, rootstock and scion source, cultivars planted, orchard and/or tree age, ownership, flora composition around the orchard, soil type and application of fertilizer. The third section comprised the major diseases and insect pests, and the management practices used in each citrus orchard. Information collected from the different citrus orchards were combined for each question and summarized for the main parameters to give an overview of citrus production and field management practices in the country.



**Figure 1:** Map of Ethiopia showing the areas covered in surveys carried out in 2012 and 2013 to assess the occurrence and spread of *Pseudocercospora* leaf and fruit spot disease of citrus.

### Incidence and Severity of PLFS Disease of Citrus in Ethiopia

Sampling was done in the forty-nine citrus orchards to map the distribution of PLFS disease in the country. The prevalence, incidence and severity of the disease were assessed based on the characteristic disease symptoms that were visually determined in the field on randomly selected citrus trees and plant parts. The geographic coordinates of the locations were taken using Global Positioning System (GPS 72H, Garmin Ltd., Taiwan) receiver to map the disease distribution.

Disease prevalence was determined by the number of sites surveyed showing PLFS disease symptoms, expressed as the percentage proportion of the total number of locations surveyed (Amata *et al.*, 2009). Disease incidence and severity were determined by randomly taking five to ten representative citrus trees diagonally across the field in each orchard surveyed (Seif and Hillocks, 1999; Mohammed, 2007). The incidence and severity of each selected tree were determined and the mean of each site was calculated. Disease incidence on the foliage was estimated by counting visibly infected and total number of leaves on eight randomly selected terminal shoots from the upper and lower halves of the canopy in the four quadrants of each selected tree, and expressed as a percentage. Incidence on fruits was similarly assessed using 5 to 40 randomly selected intact fruits per tree depending on availability. Incidence was calculated based on the presence or absence of visible disease symptoms on each fruit. Assessment of severity on the foliage and fruits was done on the same samples

used for disease incidence. Severity on leaves was estimated using the percentage leaf area infected based on a one-to-five scoring scale where 1 = no symptoms, 2 = 1-25%, 3 = 26-50%, 4 = 51-75% and 5 = above 75% (Amadi, 2008; Ezeibekwe, 2011). A similar scoring system was used for fruit severity, where 1 = healthy, 2 = less than 5%, 3 = 5-20%, 4 = 21-50% and 5 = above 50% of fruit surface affected (Seif and Hillocks, 1999).

#### Isolation and Identification of the Causative Agent

Infected leaf and fruit samples were taken from trees showing PLFS disease symptoms. Samples were placed in labeled transparent plastic bags, covered with brown paper bags, kept in cool ice box containers and then transported to the laboratory for isolation and identification of the causative agent. In the laboratory, the samples were washed in tap water and surface sterilized in 70% ethanol followed by 5% Clorox, each for one minute, and then rinsed three times with distilled and autoclaved water. Sterilized leaves or fruit peels were cut, and four to six leaf discs or peel pieces were placed on each Petri dish containing potato dextrose agar (PDA) in five replicates and incubated at 25±1°C. After five days of incubation, cultures were examined for the development of the causative agent. Cultures were purified using hyphal tipping onto fresh PDA medium and were incubated for four to seven days at 25±1°C. The causative agent was characterized based on its phenotypic characteristics by visual observation and using a stereomicroscope.

Pathogenicity tests were conducted on apparently healthy, young leaves taken from actively growing citrus seedlings. Leaves were washed in distilled water and surface sterilized in 5% Clorox. They were rinsed repeatedly with sterile distilled water. Two sterilized leaves were placed, by keeping the abaxial side up, in each Petri dish containing water agar (1%). Inoculation of leaves was carried out by placing drops of conidial suspension, with concentration of 10<sup>5</sup> to 10<sup>6</sup> conidia mL<sup>-1</sup> (Eshetu, 1999; Seif and Hillocks, 1999). The Petri dishes were sealed with parafilm to maintain high relative humidity. Control leaves were inoculated with sterile distilled water. Cultures were incubated at 26°C for two weeks, and inoculated leaf samples were examined daily for disease symptom development. The re-isolation procedure was carried out from experimentally infected samples to demonstrate Koch's postulate. The re-isolated cultures were examined for cultural and morphological comparisons with the original cultures to confirm that it was indeed the same pathogen we had seen in the original culture.

For further genetic study, the fungal cultures were stored at 5°C in test tubes with slant PDA medium at the Biotechnology Laboratory of the Holetta Agricultural Research Center, Ethiopia.

## RESULTS AND DISCUSSION

### Assessment of Citrus Orchards in Ethiopia

To give an overview of the citrus production status and the field management practices used in the different citrus orchards in the country, categories of the general information and the representative answers given by the respondents of each citrus orchard considered, and the field observations made during the surveys are described in this section.

### Geographical Locations and Climatic Conditions

The areas covered in the surveys lie between 6.417° and 11.831° North latitude respectively at Guangua town (Dilla area) in the south and at Woldiya town in the northern part, and 35.593° and 41.862° East longitude at Bebeke in the southwest and at Dire Dawa in the eastern Ethiopia, respectively (Figure 1). The locations surveyed have altitudes ranging from 900 meters above sea level (m.a.s.l) at Bebeke to 2000 m in Agaro area, with a wide range of soil types (Table 1).

According to the climatic information obtained from the orchards surveyed (Table 1), the mean minimum temperatures ranged from 10°C at Nura Era in the central rift valley to 21°C at Lado around Lake Abaya in the south. Similarly, the mean maximum temperatures varied from 21°C at Bikolo in the northwest to 37.5°C at Nura Era. The optimum temperature to induce flowering in citrus is between 13°C and 24°C with a tolerance range of ±3°C (Davies and Albrigo, 1994). The results showed that almost all citrus orchards surveyed have temperature ranges which are ideal for citrus production. The mean annual rainfalls for the areas surveyed ranged from 316 mm at Nura Era to 1750 mm at Bebeke. The major citrus producing areas have long growing seasons with low precipitation, but most of them are with river, lake or ground water supply. However, the period of the rainy season and the precipitation intensity and distribution vary from area to area. The mean annual rainfalls of the main citrus producing areas were between 316 mm and 500 mm (Table 1), which necessitates the application of supplementary irrigation water. However, long, wet rainy seasons were reported to favor the development of high disease pressure and increase preharvest fruit loss of citrus (Seifu, 2003; Sisay, 2007).

### Citrus Production in Ethiopia

Citrus farming in Ethiopia is a mixed agriculture (Seifu, 2003; Sisay, 2007), in which, many horticultural, field and forage crops, and shade trees are grown with and around citrus plantings. According to the information obtained from the surveys, several fruit crops (mainly mango, banana, avocado and papaya), vegetable crops (primarily tomato and onion), coffee, and maize were the major ones integrated in most of the citrus plantings and/or grown around citrus orchards. The orchards considered in the surveys represented plantings with ages ranging from 2 to 70 years old. About 60% of the citrus orchards surveyed constituted younger plantings, 20 and less years of age while the rest 40% of the orchards had plantings of over 20 years old.

### Area Coverage and Ownership of the Orchards

The land holdings of citrus orchards surveyed varied from 0.15 ha by the smallholder farmers to 10,030 ha for Bebeke Coffee Estate farm. The area coverage of citrus plantation for these orchards ranged from a few citrus trees in the backyards of smallholders to 1,000 ha at Nura Era citrus farm. About 83.7% of the orchards surveyed had citrus plantings of 10 ha or less, and 12.2% of the orchards were with citrus plantings between 10 and 100 ha. Only a few citrus orchards had area coverage greater than 100 ha.

The ownership of the citrus orchards considered in the survey was also assessed. Nearly half of the orchards (24 out of 49) were owned by smallholder farmers. Eighteen orchards were state-owned (some of them were in the

**Table 1:** Description of the geographic locations and climatic conditions of citrus orchards surveyed in 2012 and 2013 in Ethiopia

Citrus Orchards	Altitude (m.a.s.l)	Mean temperature range (°C)	Mean annual rainfall (mm)	Relative humidity (%)	Soil type
<b>MIDROC, Horizon Plantation S.C.</b>					
Bebeka Coffee Estate	900-1200	15-30	1750	No data	Vertisol
Nura Era	1100-1205	10-37.5	316	44.3	Clay to sandy loam
<b>Upper Awash Agro Industry Enterprise</b>					
Merti	1100	11-34	383	No data	Black soil
Abadeshka-Jeju	1100	11-34	383	No data	Black soil
<b>Erer Gota Citrus Farms</b>					
Erer	1120	18-27	425-500	No data	Sandy loam
Fetuli	1180	18-27	425-500	No data	Sandy loam
Gota	1120	18-26	425-500	No data	Sandy loam
Hurso	1130	No data	No data	No data	Sandy loam
<b>Federal Prison Administration Citrus Farms</b>					
Ziway	1680	13-28	600	52	Sandy loam
Shewarobit	1320	16-30	425	44.5	Black heavy soil
Tibila, AfricaJuice Tibila S.C.	1240	16-36	700	No data	Clay loam
Gibe, Ethiopian Seed Enterprise	1100	15-35	800	No data	Vertisol
<b>Government Fruit Nurseries</b>					
Guangua (Dilla area)	1620-1680	18-24	1000	No data	Sandy soil
Bikolo	1900	12-21	828	No data	Brown soil
Chagni	1750	15-28	No data	No data	Red to brown soil
Finote Selam	1800	14-26	No data	No data	Red soil
Dejen (Kurar)	1600	No data	No data	No data	No data
Harbu	1560	No data	No data	No data	Black soil
<b>Research Foundation Blocks</b>					
Melkassa Agricultural Research Center	1550	13-28	750	77	Sandy loam
Jarre, Hayk Research Sub-Center	1700	14-28	500	No data	Clay loam
Jimma University	1780	14-30	1150	No data	Red soil
Tony Farm, Haramaya University	1280	18-28	595	42.2	Sandy loam
<b>Citrus Farms of Private Investors</b>					
Ethioflora	1600	13-28	543	52	Sandy soil
Woldiya	1760	No data	800	No data	Brown soil
Koka	1595	No data	No data	No data	Sandy loam
<b>Orchards of Smallholders</b>					
Aleta Wendo	1900	12-26	1400	No data	Red soil
Abeshege (Welkite)	1830-1860	10.3-25.9	1244	No data	Red to brown soil
Kebena	1780	10-26	1240	No data	Vertisol
Ginbo	1440-1500	No data	No data	No data	Red brown sandy loam
Lado (Lake Abaya)	1280	21-32	900	No data	Clay black soil
Senbo	1440	No data	No data	No data	Black soil
Agaro	1680-2000	No data	No data	No data	Red to Brown soil
Yebu	1620-1640	11.6-27.1	1640	No data	Red soil
Goro (Woliso)	1860	No data	1260	No data	Black soil
<b>Association, Harbu Tropical Fruits Propagator Farmers</b>					
	1600	No data	No data	No data	Black soil

process of privatization), while six orchards were owned by private companies. One orchard was owned by Farmers' Association. However, in terms of area coverage, the ownerships of the private companies, the government and Farmers' Association represented 1115.1, 503.52 and 8.5 ha, respectively. The smallholder farmers in total contributed only 1.0 ha of citrus plantings because they grow a few or several citrus trees in their backyards.

#### Citrus Species and Varieties

Different citrus species and varieties are cultivated in the country. The information obtained from the surveys showed that sweet orange, mandarin, lime, lemon, grapefruit, citrus hybrids, sour orange and citron were produced in 48, 18, 15, 13, 5, 3, 3 and 2 of the 49

orchards surveyed, respectively (data not shown). The most dominant sweet orange variety produced in the country was Valencia (in 61.1% of the locations considered) followed by unknown variety (58.3%), Washington Naval (47.2%), Hamlin (44.4%), Pineapple (38.9%), and Jaffa (16.7%). Algerian Tangerine (in 36.1% of the locations), Fairchild (30.6%) and Dancy (27.8%) were the most widely produced mandarin varieties. Among limes and lemons, UCR Meyer (in 25% of the locations), Bears (22.2%), Allen Eureka (13.9%) and Mexican Lime (11.1%) varieties were produced. Citrus hybrids, grapefruit and citron varieties were the least produced.

Citrus trees that were grown by smallholder farmers were directly from seeds and were not grafted. However,

citrus trees in state and private owned orchards, government nurseries and research foundation blocks were grafted. The original sources of scions and rootstocks of most old orchards were unknown, but 33.3% of the respondents did not have available information. The lack of information in this regard could complicate management and breeding programs aimed at improving citrus production (Sisay, 2007). However, the major sources of the recent citrus plantations included University of California at Riverside, Melka Sedi Farm in the Middle Awash, Upper Awash Agro Industry Enterprise, Research Centers, ICRISAT, and local growers. The most commonly used rootstocks were Sour orange, Volkameriana and Troyer Citrange.

### Irrigation and Fertilization Practices

Moisture is a limiting factor for good quality citrus production. The mean annual rainfalls reported for the major citrus orchards during the surveys were below 500 mm (Table 1). These citrus orchards (24 out of 49) used the nearby rivers or lakes for irrigation purposes. The frequency of irrigation water application in most of these orchards averaged once or twice a month using the traditional double ring basin method of irrigation (Seifu, 2003; Sisay, 2007). Application of surface water at wider intervals creates moisture stress during early spring while the tree is at flowering stage. This could result in excessive drop of flowers and immature fruits, followed by smaller crop yield. Drought followed by good rains could also lead to out-of season flowering and fruit setting. Saturated and poorly drained citrus orchard soils can contribute to root rot and tree dieback, which may ultimately result in total yield loss (Sisay, 2007). The improper use of the irrigation system can create a direct contact between tree bark and surface water, which results in increased soil borne disease infections and eventually tree dieback (Caruso and Wilcox, 1990; Oudemans, 1999).

According to the information obtained from the respondents, fertilizer application was not practiced in most of the citrus orchards, particularly those orchards owned by small-scale farmers (71.4%, or 35 out of 49). The big citrus producing orchards (18.4%, or 9) reported applying DAP and Urea chemical fertilizers to their citrus trees. The use of both chemical fertilizers and animal manure was reported by Ethioflora, Melkassa and Hurso citrus orchards. Application of animal manure and compost to fertilize citrus trees was reported by Woldiya and Bikolo citrus orchards, respectively. However, fertilizer application in all citrus orchards in the country was not based on soil and plant tissue nutrient analyses.

### Preharvest Diseases and Disorders of Citrus

Many of the respondents differentiated between citrus insect pests and diseases during production. However, they have difficulties to identify diseases incited by pathogens from abiotic disorders. According to the information obtained from the respondents and field observations made during the surveys, most citrus orchards had suffered from complex of diseases (Table 2). Virus and virus-like diseases including tristeza, greening and exocortis, and unknown diseases were reported to cause tree decline and dieback at Erer Gota, Hurso, Tony, Nura Era, Merti, Melkassa, Koka, Ziway, Ethioflora, Shewarobit, Harbu, Gibe, Ginbo, and Bebeka citrus orchards. In these orchards, symptoms of leaf

yellowing, mottling and cupping, dying of twigs, stunting, and deterioration of trees were clearly observed.

Citrus trees at Fetuli, Hurso, Tony, Nura Era, Merti, Abadeshka-Jeju, Melkassa, Ethioflora, Shewarobit and Gibe orchards were attacked by several diseases caused by plant pathogens such as *Phytophthora* (primarily on sweet oranges and mandarins), citrus anthracnose (on mandarins) and bacterial canker (on acid limes). During the survey, field symptoms of PLFS disease were observed in twenty-eight citrus orchards (57.14%) in the south, southwest, northwest and central north parts of the country. Symptoms of lichens (fungus and algae that grow together) on leaves and stems of most citrus trees at Bebeka and Ginbo, melanose at Gibe and Lado, sooty mold at Melkassa and Lado, and fruit rots at Melkassa and Tony citrus orchards were recorded during the survey. The problem of nematodes was seen only at Tony farm.

Deformation and scion-rootstock incompatibility were observed at Ethioflora and Dejen (Kurar) citrus orchards. Although there were no soil and tissue analyses, salinity problem was suspected at Ziway orchard. Nutrient imbalance symptoms, mainly deficiency of trace elements were observed in many of the orchards surveyed (65.3%), especially in farms owned by smallholder farmers and by the state (Table 2). At Chagni, nutrient imbalance symptoms were observed on all citrus trees regardless of the species. Serious moisture stress which has caused leaf wilting, fruit cracking and premature fruit drop was also observed in Gota, Nura Era, Abadeshka-Jeju, Jarre, Kurar and Goro citrus orchards.

In Ethiopia, citrus trees have been afflicted due to different diseases incited by many fungi, bacteria, viruses and virus-like organisms. Viruses and virus-like diseases including psorosis, tristeza and greening were reported to be of great economic importance and thought to play a significant role in the decline of citrus plantation in the country (Tesfaye and Habtu, 1985; Lemma, 1994; Mohammed and Getachew, 1995). Citrus canker, caused by a bacterium, is a serious disease of most commercial citrus cultivars and some citrus relatives (Schubert and Miller, 2000). Eshetu and Sijam (2007) reported the occurrence of citrus canker disease on Mexican lime and sour orange in some citrus orchards in Ethiopia. Various fungal pathogens including *Phytophthora*, *Penicillium*, *Colletotrichum* and *Phaeoramularia* species seriously affected citrus trees in Ethiopia (Eshetu, 1999; Yigzaw and Gelelbelu, 2002; Seifu, 2003; Mohammed, 2007; Sisay, 2007). Based on field observations citrus producers had reported 13.6% of citrus trees dieback due to virus problems and over 35% citrus attack by other plant pathogens in the major citrus farms across Ethiopia (Sisay, 2007).

*Phytophthora* species cause the most serious problem and are economically important soil-borne diseases of citrus (Graham and Timmer, 1994). Previous reports in Ethiopia indicated high severity of soil-borne diseases caused by *Phytophthora* species in many citrus orchards (Seifu, 2003; Sisay, 2007). Intrinsic factors such as lack of certified planting material, inappropriate use of cultural practices, and adverse edaphic conditions may increase the rate of infection and the spread of the disease in an orchard (Salerno and Cutuli, 1982). The use of inappropriate irrigation system can result in increased

*Phytophthora* infection and ultimately tree dieback (Caruso and Wilcox, 1990; Oudemans, 1999). Poor rootstock-scion combinations were also attributed to high levels of gummosis and *Phytophthora* root rot (Ippolito *et al.*, 1996). Therefore, the proper use of irrigation methods and selection of disease resistant rootstocks can reduce the risk of infection by soil borne pathogens (Salerno and Cutuli, 1982).

The knowledge and scientific identification of diseases and disorders is very limited in Ethiopia. The use of robust and fast diagnostic techniques for accurate identification is therefore crucial for the development of more effective disease management strategies and to improve the current production practices in the country (Sisay, 2007).

### Insect Pests of Citrus

Insect pests on citrus were found to be equally important to diseases. Based on the information obtained from the respondents and field observations made during the surveys, leafminer (77.6%), red scales (57.1%), citrus thrips (24.5%), cottony cushion scales (16.3%), Mediterranean fruit fly (14.3%), and woolly whitefly (10.2%) were reported as the major insect pests on citrus in the orchards surveyed (Table 2). Attacks by false codling moth, mealy bug, bud mite, orange dog and termites were reported from several citrus orchards. Red scale infestation at Tibila citrus farm was extremely severe; it attacked leaves and fruits, made the twigs dry and led the trees deteriorate. Both red scale and leafminer were the most serious pests attacking citrus trees at Melkassa orchard. At Nura Era, woolly white fly was found the most important pest on citrus trees. Similarly, Sisay (2007) reported more than 50% preharvest fruit damage due to these insect pests of in Ethiopia.

Citrus production in Ethiopia is severely threatened by numerous insect pests. In the past few decades, a large number of insect pests have been identified and documented. Accordingly, the major insect pests of citrus in the country included scales, woolly white fly, Mediterranean fruit fly, false codling moth, citrus thrips, leafminer and fruit flies (Emana *et al.*, 2003; Ferdu *et al.*, 2009). The results of the existing surveys were also consistent with these previous reports.

The start of insect attack and extent of damage in citrus orchards was found to vary depending on tree age and fruit maturity. In the current study, many respondents indicated very high leafminer infestation on leaves of young plants and on newly emerging leaves of older citrus trees. Respondents also reported initial infestation of scale insects and Mediterranean fruit fly attacks during fruit development and ripening stages. In citrus cultivation, regular monitoring of orchard practices from land preparation to fruit maturity and harvesting can provide sufficient information to control infestations of citrus trees (Taylor, 1997).

### Management of Preharvest Diseases and Insect Pests of Citrus

Citrus trees in most of the orchards, especially those owned by smallholder farmers, were not well-managed. The citrus orchard at Bebekka had been neglected for the past several years due to PLFS disease. The disease severely affected the trees. In some other orchards like Guangua weed infestation was very serious. Survey

results showed that no control measures except sanitation in some orchards were practiced against PLFS disease. Citrus orchards at Bikolo, Chagni, Finote Selam, Nura Era and Merti did practice back pruning of dried twigs and branches to rejuvenate new branches. At Erer Gota, the second largest citrus farm next to Nura Era in area coverage, pruning of dead branches and uprooting of dead trees were also practiced. Most medium and large scale citrus plantations were sprayed with pesticides. According to the information obtained from the respondents, some government nurseries such as Bikolo, Chagni and Finote Selam stopped to propagate and distribute citrus planting materials due to PLFS disease.

About 43% of the citrus orchards surveyed apply pesticides as a major means of disease and/or insect pest control (Table 2). The remaining orchards, especially those owned by the smallholder farmers, do not use pesticides. Kocide 101, Mancozeb, Bayleton, Ridomil MZ, Ridomil 5G, and Daconil fungicides, as well as Diazinon, Karate, Selecron, Suprathion, Endosulfan, Ultracide, Confider, White mineral oil, Diazol, Thiodan, Sumithion, Dimethoate and Methidathion insecticides were applied to control diseases and insect pests in the orchards surveyed. Of these pesticides, Kocide 101, Mancozeb and Bayleton fungicides, and Diazinon and Karate insecticides were the most widely applied pesticides for citrus disease and insect pest control, respectively.

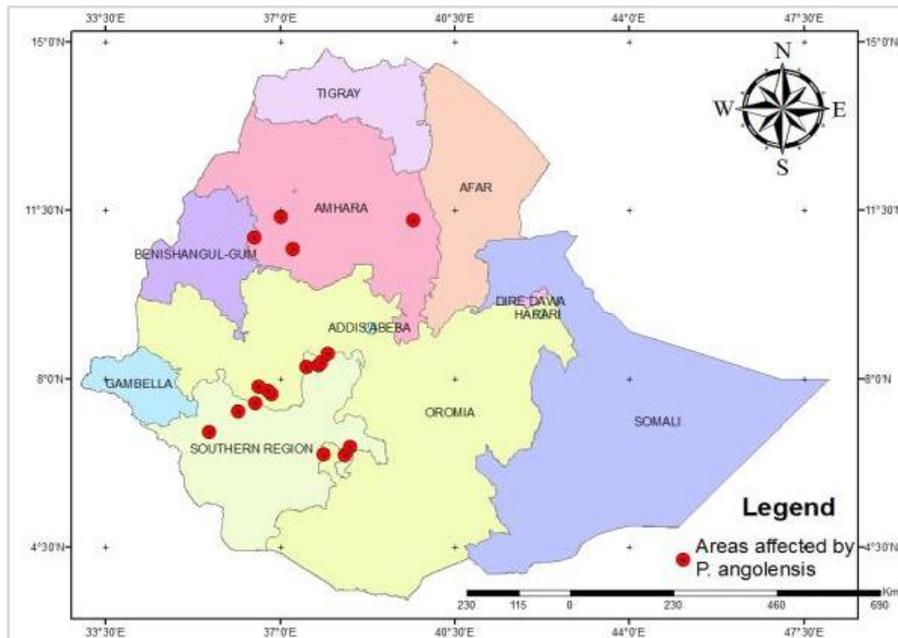
The application of pesticides only during the first observation of the pest may lead to ineffective control and can result in buildup of the inoculum over time and eventually disease outbreak in an area (Fry, 1977). It is required to investigate into alternative natural plant products (Tripathi and Dubey, 2004), microbial antagonists (Droby *et al.*, 1991) and the application of improved sanitary practices (Sierra *et al.*, 1993; Wilson *et al.*, 1995) in order to reduce the risks associated with ineffective application of chemicals and its environmental and health considerations. In Ethiopia, various experiments have been conducted and different pest control methods including cultural practices, bait sprays and attractants, a number of insecticides, parasitoids and predators, and botanicals were recommended for the management and control of major insect pests (Tsedeke 1983, 1991; Ferdu *et al.* 2009). However, most citrus growers in the country do not apply recommended pesticide at a recommended rate and frequency.

### *Pseudocercospora* Leaf and Fruit Spot Disease of Citrus in Ethiopia Prevalence and Distribution

The results of the field surveys indicated the current occurrence and spread of PLFS disease in Ethiopia (Figure 2). The disease was widely spread in the south, southwest, northwest and central north parts of the country. The disease was found prevalent in 63.2% of the citrus orchards surveyed in the major citrus growing administrative zones of the country (Table 3). In south Wello zone, field symptoms of leaf and fruit spot disease were observed only at Jarre. Based on visual observations, no symptoms of PLFS disease were recorded in citrus orchards found in east Gojjam, north Shewa, north Wello, east Shewa, Shinile and Dire Dawa zones. In Jimma zone, the disease was locally known as 'Cholera' due to the devastating nature of the disease.

**Table 2:** Diseases and insect pests recorded, and use of pesticides in citrus orchards surveyed in 2012 and 2013.

Citrus Orchards	Diseases and/or Disorders Observed Based on Field Symptoms	Major Insect Pests Recorded	Chemical Pesticides Use
Bebeka	PLFS, Lichens, Nutrient imbalance, decline	Leafminer, Red scale	No application
Nura Era	Citrus Canker; Sudden death/Tristeza, <i>Phytophthora</i> rot, Dieback, Nutrient imbalance, Water stress	Woolly white fly ("Sukarie"), Med fruit fly, Leafminer, Red scale, Citrus thrips, Cottony cushion scale, Termite	Ultracide, Diazinon, Diazol, White mineral oil
Merti	Anthrachnose, Dieback, Nutrient imbalance	Leafminer, Cottony cushion scale	Ultracide, Diazinon, Diazol, White mineral oil
Abadeshka-Jeju	Anthrachnose, Nutrient imbalance, Water stress	Leafminer, Woolly white fly, Red scale, Citrus thrips	Same as in Merti
Erer	Tristeza, Greening, Nutrient imbalance, Unknown disease	Leafminer, Citrus thrips	Kocide 101, Ridomil MZ, Ridomil 5G, Bayleton, Mancozeb; Selecron, Suprathion, Endosulfan, Karate, Diazinon
Fetuli	Tristeza, Exocortis viroid, <i>Phytophthora</i>	Leafminer	Same as in Erer
Gota	Tristeza, Exocortis viroid, Nutrient imbalance, Water stress	Leafminer, Citrus thrips	Same as in Erer
Hurso	<i>Phytophthora</i> , Tristeza, Greening, Nutritional imbalance	Red scale, Leafminer, Citrus thrips, Bud mite	Mancozeb, Diazinon, Methidathion, Sumithion
Ziway	Decline/Dieback, Nutrient imbalance, Salinity	Cottony cushion scale, Leaf miner, Citrus thrips, Med fruit fly	Karate
Shewarobit	Greening, Dieback, <i>Phytophthora</i> disease	Red scale	Mancozeb, Bayleton
Tibila (Tifhste Genet)	Nutrient imbalance	Red scale, Woolly white fly, Fruit fly, False codling moth, Leafminer, Thrips, Mealy bug, Cottony cushion scale	Diazinon, Confider
Gibe	PLFS, Canker, Dieback, Gummosis, Tristeza, Greening, Melanose, Nutrient imbalance	Leafminer, Scales (red and black), Citrus thrips, Orange dog, Med fruit fly	Ultracide, Karate
Guangua (Dilla area)	PLFS, Nutrient imbalance	Leafminer	No application
Bikolo	PLFS, Nutrient imbalance	Red scale	Kocide
Chagni	PLFS, Nutrient imbalance	Leafminer	Daconil, Kocide
Finote Selam	PLFS	Leafminer, Red scale, Cottony cushion scale	Bayleton
Dejen (Kurar)	Deformation on graft union, Fruit drop, Water stress	Red scale	No application
Harbu (South Wello)	Dieback	Woolly white fly	No application
Melkassa	Dieback, Tristeza, Greening, Citrus Canker, Anthracnose, Fruit rot, Sooty mold, Nutrient imbalance	Red scale, Leafminer, Cottony cushion scale, Citrus thrips, Med fruit fly	Dimethoate, Karate, Diazinon 60
Jarre (Hayk)	Leaf and fruit spot, Water stress	Leafminer, Red scale	No application
Jimma City	PLFS, Nutrient imbalance	Leafminer, Red scale	No application
Tony Farm (Dire Dawa)	Tristeza, Exocortis viroid, Greening <i>Phytophthora</i> / Nematodes, Fruit rot, Nutritional imbalance	Termite, Med fruit fly, Leafminer, Red scale	Methidathion, Sumithion
Ethioflora (Adami Tulu)	<i>Phytophthora</i> , Gummosis, Graft incompatibility, Nutrient imbalance	Leafminer, Med fruit fly	Diazinon, Thiodan
Woldiya	No disease	Red scale	No data
Koka	Decline, Nutrient imbalance	Leafminer	No data
Aleta Wendo	PLFS, Nutrient imbalance	Leafminer	No application
Abeshege (Welkite area)	PLFS, Nutrient imbalance	Leafminer, Red scale	Daconil, Kocide
Kebena	PLFS, Nutrient imbalance	Leafminer	No application
Ginbo	PLFS, Lichens, Greening	Leafminer, Red scale	No application
Lado (Lake Abaya area)	PLFS, Sooty mold, Melanose, Nutrient imbalance	Leafminer, Red scale, Citrus thrips	No application
Senbo	PLFS, Nutrient imbalance	Red scale, Leafminer, Cottony cushion scale	No application
Agaro	PLFS, Nutrient imbalance	Leafminer, Red scales	No application
Yebu	PLFS, Nutrient imbalance	Leafminer, Red scales	No application
Goro (Woliso area)	PLFS, Nutrient imbalance	Leafminer	No application
Harbu Tropical Fruits Propagator Farmers Association	Dieback	Woolly white fly, Leafminer	No data



**Figure 2:** Distribution of *Pseudocercospora* leaf and fruit spot of citrus in Ethiopia

**Table 3:** Prevalence of *Pseudocercospora* leaf and fruit spot disease of citrus observed in the main citrus growing administrative zones of Ethiopia in the 2012 and 2013 surveys

Regional States	Zones	Citrus orchards evaluated	Infected orchards	Prevalence (%)
Amhara	Awi	3	3	100
	West Gojjam	5	5	100
	South Wello	4	1	25
	East Gojjam	3	0	0
	North Shewa	3	0	0
	North Wello	3	0	0
Oromia	Borena	4	4	100
	Jimma City	3	3	100
	Jimma	10	10	100
	Southwest Shewa	3	3	100
	East Shewa	8	0	0
SNNP*	Bench-Maji	3	3	100
	Gurage	5	5	100
	Kaffa	4	4	100
	Sidama	2	2	100
Somali	Shinile	4	0	0
Dire Dawa	Dire Dawa City	1	0	0

SNNP = South Nations, Nationalities and People

**PLFS Disease Incidence and Severity Assessment of Field Symptoms**

The PLFS disease affects the different parts of the plant. In the orchards surveyed, the disease commonly affected leaves and fruits. In addition, symptoms were observed on twigs of sweet oranges and mandarins. During the surveys, citrus growers indicated that the disease begins infection on leaves and then progresses to fruits and twigs. In the field, the disease caused leaf spots, blemish fruits that remain on the tree, premature leaf and fruit drop, and drying of tips of twigs. The characteristic field symptoms of PLFS disease are indicated in Figure 3.

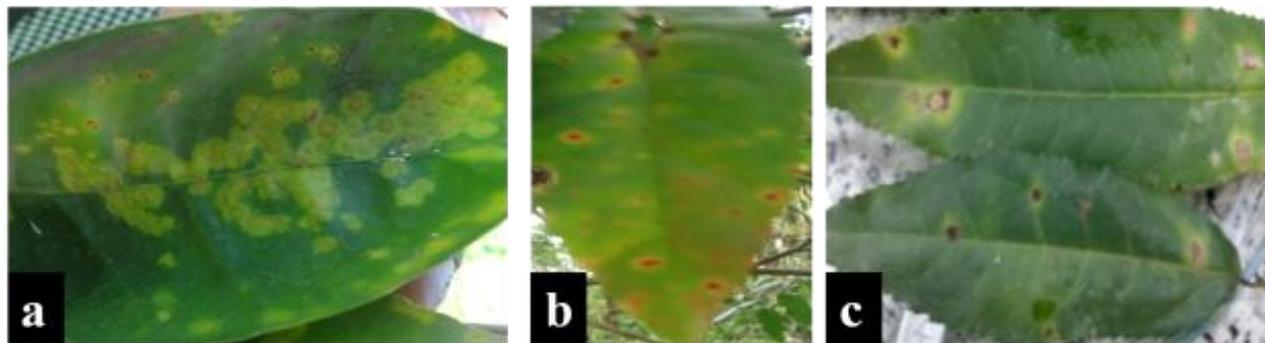
Field symptoms of the spot disease observed on leaves and some fruits of sweet orange at Jarre orchard (Figure 4a) in south Wello zone were somehow different from the symptoms in other orchards. Infected leaf samples were isolated in the laboratory and the causative

agent was culturally and morphologically identified as similar to *P. angolensis*; but it needs further confirmation. Jarre is the only orchard in the central north part of the country where leaf and fruit spot-like disease symptoms were observed. Pretorius (2005) reported similar symptoms in Zimbabwe as concentric ring blotch on citrus leaves caused by grey mite. However, grey mites were not observed at Jarre during the surveys.

At Bebeke, spot-like symptoms that are similar to the PLFS disease were observed on a coffee shade tree (Figure 4b and 4c). Infected leaf samples were isolated in the laboratory and the causative agent was morphologically identified as similar to *P. angolensis*. However, it needs to be confirmed by molecular characterization of the pathogen, and clarified whether the coffee shade tree is an alternate host or the disease infects other plant species outside the genus *Citrus*.



**Figure 3:** Symptoms of *Pseudocercospora* leaf and fruit spot: (a) chlorotic lesions on a leaf; (b) immature fruit with halo, gray colored symptoms on the surface and dead tissue at the center of the lesion; and (c) lesions on small, young branch.



**Figure 4:** Leaf and fruit spot-like symptoms (a) on sweet orange leaf at Jarre orchard, south Wello, and (b, c) on leaves of a coffee shade tree at Bebeka orchard, Bench Maji zone.

#### PLFS Disease Incidence and Severity in the Different Districts

In the districts surveyed, the percentage of PLFS disease incidences on citrus in the various orchards ranged from zero to 100% while its severity ratings varied from one to five (Table 4). The highest mean incidence on leaves was 60.4% at Jimma City followed by 56.1, 53.9 and 50.2% in Aleta Wendo, Abeshege and Kebena, respectively. The maximum incidence (100%) on fruits was recorded in Aleta Wendo. From 50 to 92% average disease incidences on fruits were recorded in Abeshege, Debre Werk, Mana, Kebena, Ginbo, Jimma City and Shebe Senbo. Similarly, the highest severity ratings on leaves (3.8) and fruits (5.0) were recorded in Aleta Wendo (Table 4). Above 2.5 ratings of disease severity on leaves were recorded in Abeshege, Debre Werk and Jabitehnan districts. Higher disease severity ratings on fruits that ranged from 4.0 to 4.6 were recorded in six districts. In Aleta Wendo and several citrus orchards of small-scale farmers in Abeshege and Mana districts, complete damage of fruits was observed. Disease incidence and severity were nil on citrus leaves and fruits in 12 and 13 districts, respectively (Table 4). In the past years, PLFS disease was reported in various areas in the south, southwest, and northwest Ethiopia (Eshetu, 1999; Yigzaw and Gelelbelu, 2002; Kassahun *et al.*, 2006; Mohammed, 2007). The results of the present survey showed that the disease was spread to new areas in the northwest areas such as Bikolo and Bahir Dar, and the north central part like Jarre that were not reported to have the disease in the past. This was a clear indication for the need for continuous monitoring, and putting in place an internal quarantine system to avoid the spread of the disease in to new areas. Disease incidence and severity were higher in areas located in the south and southwest than those in

the northwest and central north Ethiopia. This could be due to the long time disease buildup since its first introduction in the south Ethiopia.

In the current survey, leaves were generally attacked less severely than fruits. Eshetu (1999) reported similar results on sweet orange trees. Disease severity at Aleta Wendo was very high on both citrus leaves and fruits. At Goro, it was observed that only citrus leaves were affected. However, severity on citrus leaves and fruits varied from district to district. Accordingly, mean incidence values on leaves and fruits at district level were 30.7% and 47.7%, while severity ratings were 2.4% and 3.3%, respectively. Eshetu (1999) and Mohammed (2007) also reported the variation of disease severity percentage on leaves and fruits from location to location. Aleta Wendo, Abeshege, Mana, Jimma City, Ginbo, Debre Werk and Kebena were the most seriously affected districts. These districts are located in the south and southwest parts of Ethiopia where the PLFS disease was first introduced in to the country and then spread.

During the survey, farmers in Aleta Wendo stated that symptoms of PLFS disease were first appeared in that area in 1985. Yimenu (1993) reported the first observation of characteristic field symptoms of leaf and fruit spot disease on citrus trees in Aleta Wendo and Dale districts in 1988. According to the farmers, citrus fruit used to be one of their major income sources. They stated that three to four trucks of sweet orange fruits from Aleta Wendo district alone had been supplied to the local markets daily before the PLFS disease affected their citrus plantations. Farmers also indicated that in the past 20 years many citrus trees were abandoned due to PLFS disease, and replaced by other crops like coffee.

**Table 4:** Incidence and severity of *Pseudocercospora* leaf and fruit spot disease in the different districts surveyed in 2012 and 2013 in the main citrus growing regions of Ethiopia

District	Incidence (%)		Severity (1-5 rating scales)	
	Foliage	Fruit	Foliage	Fruit
Aleta Wendo	56.1	100	3.8	5.0
Abeshege	53.9	92.0	2.9	4.6
Debre Werk	28.0	81.5	2.9	4.0
Mana	41.6	72.5	2.2	4.6
Kebena	50.2	72.0	2.0	4.0
Ginbo	34.5	66.0	2.0	4.3
Jimma City	60.4	52.0	4.0	4.6
Shebe Senbo	14.7	50.0	1.5	3.0
Abaya	39.2	38.8	2.4	1.5
Gomma	25.0	31.9	2.4	2.4
Jabitehnan	21.4	29.7	2.7	3.3
Guangua	14.9	29.1	1.9	3.0
Mecha	19.6	28.0	2.0	3.8
Tehuledere	11.8	12.4	1.7	2.0
Sekoru	17.5	6.8	1.8	2.0
Goro	1.6	0.0	2.0	1.0
Adama	0.0	0.0	1.0	1.0
Adami Tulu Jido Combolcha	0.0	0.0	1.0	1.0
Boset	0.0	0.0	1.0	1.0
Dejen	0.0	0.0	1.0	1.0
Dire Dawa City	0.0	0.0	1.0	1.0
Erer	0.0	0.0	1.0	1.0
Guba Lafto	0.0	0.0	1.0	1.0
Jeju	0.0	0.0	1.0	1.0
Kalu	0.0	0.0	1.0	1.0
Kewet	0.0	0.0	1.0	1.0
Lome	0.0	0.0	1.0	1.0
Merti	0.0	0.0	1.0	1.0

#### PLFS Disease Incidence and Severity on the Different Citrus Species

The summary results of the surveys that show the disease incidences on the different citrus species are presented in Table 5. The mean disease incidence was higher on sweet oranges (36.2% on leaves and 63.8% on fruits) followed by mandarins (21.5% on leaves and 29.4% on fruits). Disease incidence was relatively lower on lemons (17.1% on leaves and 18.0% on fruits) and limes (16.3% on leaves but nil on fruits). The disease was not positively identified on sour orange, tangor/tangelo, grapefruit and citron leaves, and on grapefruit and citron fruits. The rate of disease severity followed the same trend as in disease incidence (Table 5). Higher rates of disease severity were recorded on sweet orange (2.6 on leaves and 4.0 on fruits) and mandarin (2.3 on leaves and 3.0 on fruits); but it was less on lemon and lime trees.

Disease incidences on sweet oranges ranged from zero to 76.7% on leaves and from zero to 100% on fruits, while disease severity ratings varied from 1.0 to 3.8 on leaves and from 1.0 to 5.0 on fruits (Table 6). The disease prevailed on all sweet orange fruits sampled at citrus orchards in Aleta Wendo, Ginbo, Senbo and Welkite areas. In these areas, the disease severity ratings were also very high (5.0) and PLFS disease caused nearly complete fruit drop. However, many citrus orchards in 16 surveyed areas did not show any incidence of the disease on both leaves and fruits of sweet orange. Sweet orange fruits were more severely affected by the disease than the leaves. Based on the field observations made, among sweet orange varieties Washington Naval was highly attacked by PLFS while Jaffa and Hamlin were less affected.

Among the locations surveyed, only three of them had PLFS disease incidences on mandarin trees (Table 6). Higher disease incidence and severity were recorded at Finote Selam and Chagni in the northwest Ethiopia. No incidence of PLFS disease was observed on fruits at Gibe.

Disease incidences on lemons and limes occurred at Chagni, Ginbo and Gibe orchards (Table 6). Leaf incidences occurred in all of these orchards; but at Gibe orchard no PLFS disease incidence observed on fruits. Similar trend was happened for disease severity ratings for lemon and lime. Based on the results of the survey, lemons seemed relatively more susceptible to PLFS disease than limes. Lemons and limes at Finote Selam were not infected by the disease. Susceptibility was suggested to vary with different periods of the year and with locations (Kuate, 1998).

During the present surveys, no incidence of PLFS disease was observed on grapefruit at Gota and Melkassa, and on citron at Goro and Melkassa orchards. Symptoms of PLFS disease occurred on leaves of sour orange at Lado (Lake Abaya) and Agaro areas; interestingly it was not observed on fruits. This might be due to the antifungal activities of the essential oils of sour orange fruits which prevented the infection of PLFS disease. Jazet Dongmo *et al.* (2008, 2009) reported that essential oils extracted from *Citrus latifolia* var Tahiti, and *C. aurantifolia* inhibited the growth of *P. angolensis* on artificial medium. Leaf incidence on tangor/tangelo trees was recorded only at Chagni orchard (Table 6).

The results of the surveys showed that citrus species differ in their level of disease resistance. Sweet oranges

and mandarins were more severely affected than tangor/tangelo, lemon and lime. The disease did not affect citron trees at all. Moreover, the disease affected fruits more severely than leaves of the major citrus species. The observed differences in the incidence and severity of PLFS disease among the citrus species, and varieties within a species could be due to the differences in their resistance capabilities. The result was consistent with the findings of various authors (Emechebe, 1981; Kuate, 1998; Seif and Hillocks, 1999; Diallo *et al.*, 2003;

Mohammed, 2007). The variation in resistance between varieties of a species often resides in a physiological or biochemical differences between them (Wutscher, 1997). Sisay (2007) recommended that apart from efficient management practices, farmers should plant more resistant varieties budded on more resistant stocks and adopt a comprehensive integrated approach to disease control involving biological, cultural and chemical methods.

**Table 5:** Average incidence and severity of *Pseudocercospora* leaf and fruit spot disease on different citrus species assessed in 2012 and 2013 in the main citrus growing regions of Ethiopia

Citrus species	Incidence (%)		Severity (1-5 rating scales)	
	Foliage	Fruit	Foliage	Fruit
Sweet orange	36.2	63.8	2.6	4.0
Mandarin	21.5	29.4	2.3	3.0
Lemon	17.1	18.0	2.0	2.0
Lime	16.3	16.7	1.7	2.0
Sour orange	23.3	0.0	2.0	1.0
Tangor/Tangelo	4.0	0.0	2.0	1.0
Grapefruit	0.0	0.0	1.0	1.0
Citron	0.0	0.0	1.0	1.0

**Table 6:** Incidence and severity of *Pseudocercospora* leaf and fruit spot disease on each citrus species in the different locations surveyed in 2012 and 2013 in Ethiopia

Citrus species	Location of Orchards	Incidence (%)		Severity (1-5 rating scales)	
		Foliage	Fruit	Foliage	Fruit
Sweet orange	Aleta Wendo (Omacho Chawa)	56.1	100	3.8	5.0
	Ginbo (Megenagna)	38.0	100	2.0	5.0
	Senbo (Kishe Kosta)	29.5	100	2.0	5.0
	Welkite (Laygnaw Tatessa)	76.7	100	3.0	5.0
	Welkite (Jejeba ena Gasorie)	31.1	84.0	2.8	4.2
	Bebeka	28.0	81.5	2.9	4.0
	Dilla area (Guangua)	44.3	77.5	2.8	2.0
	Yebu (Gube Bosoka)	41.6	72.5	2.2	4.6
	Ginbo (Balewold)	35.9	72.0	2.0	5.0
	Welkite (Aregita)	50.2	72.0	2.0	4.0
	Agaro (Agaro town)	37.5	60.0	2.5	3.0
	Jimma	60.4	52.0	4.0	4.6
	Agaro (Genji Elbu)	31.2	45.0	3.0	3.5
	Chagni	20.0	40.0	2.0	4.0
	Finote Selam	39.5	40.0	4.0	5.0
	Hayk (Jarre)	35.4	37.3	3.0	4.0
	Bikolo	19.6	28.0	2.0	3.0
	Gibe	25.5	27.0	2.0	2.0
	Agaro (Elbu)	18.9	22.5	2.0	2.2
	Goro (Adami Wedessa)	4.9	No fruit	2.0	No fruit
Abadeshka	0.0	0.0	1.0	1.0	
Erer	0.0	0.0	1.0	1.0	
Adami Tulu (Ethioflora)	0.0	0.0	1.0	1.0	
Fetuli	0.0	0.0	1.0	1.0	
Gota	0.0	0.0	1.0	1.0	
Sweet orange	Harbu	0.0	0.0	1.0	1.0
	Melkassa	0.0	0.0	1.0	1.0
	Merti	0.0	0.0	1.0	1.0
	Nura Era	0.0	0.0	1.0	1.0
	Shewarobit	0.0	0.0	1.0	1.0
	Tibila (Tifhste Genet)	0.0	0.0	1.0	1.0
	Dire Dawa	0.0	0.0	1.0	1.0
	Woldiya	0.0	0.0	1.0	1.0
	Ziway	0.0	0.0	1.0	1.0
	Dejen (Kurar)	0.0	0.0	1.0	1.0
Koka	0.0	0.0	1.0	1.0	
Mandarin	Finote Selam	24.6	49.0	3.0	4.0
	Chagni	19.2	39.3	2.0	4.0
	Gibe	20.7	0.0	2.0	1.0
	Abadeshka	0.0	0.0	1.0	1.0

	Gota	0.0	0.0	1.0	1.0
	Harbu	0.0	0.0	1.0	1.0
	Hayk (Jarre)	0.0	0.0	1.0	1.0
	Melkassa	0.0	0.0	1.0	1.0
	Merti	0.0	0.0	1.0	1.0
	Nura Era	0.0	0.0	1.0	1.0
	Tibila (Tifhste Genet)	0.0	0.0	1.0	1.0
	Dire Dawa	0.0	0.0	1.0	1.0
	Woldiya	0.0	0.0	1.0	1.0
	Ziway	0.0	0.0	1.0	1.0
Lemon	Chagni	16.9	36.0	2.0	3.0
	Gibe	17.2	0.0	2.0	1.0
	Finote Selam	0.0	0.0	1.0	1.0
	Hayk (Jarre)	0.0	0.0	1.0	1.0
	Melkassa	0.0	0.0	1.0	1.0
	Dire Dawa	0.0	0.0	1.0	1.0
	Woldiya	0.0	0.0	1.0	1.0
	Ziway	0.0	0.0	1.0	1.0
Lime	Chagni	14.2	30.0	2.0	3.0
	Ginbo (Megenagna)	28.2	20.0	2.0	2.0
	Gibe	6.5	0.0	1.0	1.0
	Finote Selam	0.0	0.0	1.0	1.0
	Goro (Adami Wedessa)	0.0	0.0	1.0	1.0
	Harbu	0.0	0.0	1.0	1.0
	Melkassa	0.0	0.0	1.0	1.0
	Nura Era	0.0	0.0	1.0	1.0
	Senbo (Kishe Kosta)	0.0	0.0	1.0	1.0
Woldiya	0.0	0.0	1.0	1.0	
Grapefruit	Gota	0.0	No fruit	1.0	No fruit
	Melkassa	0.0	0.0	1.0	1.0
Tangor/Tangelo	Chagni	4.0	0.0	1.6	1.0
	Abadeshka	0.0	0.0	1.0	1.0
	Melkassa	0.0	0.0	1.0	1.0
	Merti	0.0	0.0	1.0	1.0
	Nura Era	0.0	0.0	1.0	1.0
Sour Orange	Lake Abaya (Lado)	34.1	0.0	2.0	1.0
	Agaro (Koye Seja)	12.5	0.0	2.0	1.0
Citron	Goro (Adami Wedessa)	0.0	0.0	1.0	1.0
	Melkassa	0.0	0.0	1.0	1.0

### PLFS Disease Severity in Relation to Agro-Climatic Conditions and Citrus Tree Age

Temperature and elevation of the areas considered in the surveys did not contribute much to the incidence and severity of PLFS disease. High plague of PLFS disease of citrus was recorded at all developmental stages of citrus trees. However, citrus orchards that received high rainfalls exhibited high severity of PLFS disease, conversely those areas that did not exhibit the disease had low annual rainfalls (Table 7); indicating the direct relationship between disease incidence and amount of rainfall. Further studies in controlled environment are needed to validate this observation. Districts with PLFS disease incidences had average minimum and maximum temperatures ranging from 12 to 35 °C mean annual rainfalls of 500 to 1750 mm, altitudes that varied from 900 to 2000 m.a.s.l, and citrus trees that aged from 2 to 60 years. On the other hand, those districts free from PLFS disease incidences had temperatures ranging from 10 to 37.5°C, mean annual rainfalls of 316 to 750 mm, altitudes that varied from 1100 to 1760 m.a.s.l, and citrus trees with the age of 3 to 70 years. More severe PLFS disease attacks were recorded in areas with high annual rainfalls like Aleta Wendo, Jimma, Welkite, Yebu, Ginbo and Bebeke. The disease was reported to be more severe during and right after the end of the rainy season, particularly in September and October. These results

were consistent with previous reports (Eshetu, 1999; Yigzaw and Gelebelu, 2002; Mohammed, 2007).

Various authors (Seif and Hillocks, 1993; Eshetu, 1999; Mohammed, 2007) reported that wind-borne conidia of *P. angolensis* infect citrus fruits, leaves and young twigs. Windbreak trees planted around the periphery of the orchards are thought to be the potential sources of the pathogen (Whiteside *et al.*, 1988). Although the use of windbreak trees seems important from agro-ecological point of view and pest trap, field disease control by sanitation and clearing of inoculum source is important (Sierra *et al.*, 1993; Pretorius, 2005).

### Isolation and Identification of the Causative Agent

Isolates grown on PDA medium at 26°C from infected leaf and fruit samples were identified based on cultural and morphological characteristics. They were found identical to *P. angolensis* described by different authors (Kirk, 1986; Kuate, 1998; Eshetu, 1999). The mycelium was mostly creamy white or light grey in culture and compact in density. Conidia were born in branched chains of 2-4, hyaline, cylindrical and some of them had from two to four septa. Pathogenicity tests were also conducted on apparently healthy detached leaves, using Koch's method (Kiraly *et al.*, 1974). The same symptoms were observed on artificially inoculated leaves after a week. The same fungus having the same cultural and morphological characteristics was re-isolated from inoculated leaves.

**Table 7:** *Pseudocercospora* leaf and fruit spot disease severity assessed in relation to the agro-climatic conditions of citrus orchards and tree age in the 2012 and 2013 surveys in Ethiopia

Districts surveyed	Severity (1-5 rating scale)		Average temperature range (°C)	Mean annual rainfall (mm)	Elevation (m)	Tree age (year)
	Foliage	Fruit				
Aleta Wendo	3.8	5.0	12-26	1400	1900	2-60
Jimma City	4.0	4.6	14-30	1150	1780	10
Abeshege	2.9	4.6	10.3-25.9	1244	1830-1860	15-20
Mana	2.2	4.6	11.6-27.1	1640	1630	7-9
Ginbo	2.0	4.3	No data	No data	1440-1500	4-15
Debre Werk	2.9	4.0	15-30	1750	900-1200	22
Kebena	2.0	4.0	10-26	1240	1780	12
Mecha	2.0	3.8	12-21	828	1900	17-28
Jabitehnan	2.7	3.3	14-26	No data	1800	19-36
Guangua	1.9	3.0	15-28	No data	1750	15-19
Shebe Senbo	1.5	3.0	No data	No data	1440	12-30
Gomma	2.4	2.4	No data	No data	1680-2000	6-16
Sekoru	1.8	2.0	15-35	800	1100	3-14
Tehuledere	1.7	2.0	14-28	500	1700	10-25
Abaya	2.4	1.5	18-32	900-1000	1280-1680	10-20
Goro	2.0	1.0	No data	1260	1860	6
Adama	1.0	1.0	13-28	750	1550	7
Adami Tulu	1.0	1.0	13-28	543-600	1600-1680	8-34
Boset	1.0	1.0	10-37.5	316	1100-1205	3-30
Dejen	1.0	1.0	No data	No data	1600	18
Dire Dawa	1.0	1.0	18-28	595	1280	25-60
Erer	1.0	1.0	18-27	425-500	1120-1180	3-70
Guba Lafto	1.0	1.0	No data	800	1760	17
Jeju	1.0	1.0	11-36	383-700	1100-1240	3-28
Kalu	1.0	1.0	No data	No data	1560-1600	3-30
Kewet	1.0	1.0	16-30	425	1320	4-55
Lome	1.0	1.0	No data	No data	1560	16
Merti	1.0	1.0	11-34	383	1100	30

## CONCLUSIONS

*Pseudocercospora* leaf and fruit spot disease is one of the crucial constraints to citrus production in many parts of Ethiopia. The production environment seemed to influence the prevalence of the disease. It has widely spread in most citrus producing areas with high rainfalls; but the disease was not observed in the drier parts of the country. Since the last survey conducted in 2001 and 2002, the disease has spread in to new, PLFS disease free citrus producing areas in the northwest and north central parts of the country. The disease caused severe leaf and fruit damages which render significant defoliation and fruit drop. The severity of the disease varied among different citrus species and areas with different agro-ecologies. Severity increased, in ascending order, for citron, lime, lemon, mandarins, and sweet oranges. Citrus orchards in the south and southwest with high rainfalls were more severely affected by the disease than those in the northwest and central north parts. In some of these areas, complete loss of fruit yield was recorded. Field observations during the surveys indicated that no proper crop management practices were implemented by almost all citrus growers. This has contributed for high disease severity. Results of typical symptoms observed in the field, the macroscopic and microscopic characteristics noticed in the laboratory, and the pathogenicity tests indicated that the causative agent of PLFS disease to be *P. angolensis*. Therefore, due emphasis should be given to the problem and the disease spread has to be ceased.

Citrus producers should be advised to know and maintain the soil nutrient status of their orchards, and to

practice general hygiene and sanitary measures such as removal of infected leaves and fruits, pruning of dead branches and twigs, and clearing of neglected orchards to reduce inoculum source. Domestic quarantine is highly required to limit the spread of the disease to new areas through planting materials and/or fruits. Field application of recommended fungicides such as the mixture of benomyl and chlorothalonil could significantly reduce the damage of the disease. Differences in susceptibility were observed among and within species which necessitate studying the reactions of the available varieties in the country to *P. angolensis*. Thus, selection and use of disease resistant/tolerant scion cultivars may be important. Reliable and quick disease identification and monitoring techniques, and integrated disease management strategies need to be in place in the country.

## ACKNOWLEDGEMENTS

The authors acknowledge all respondents for their unreserved cooperation in providing the necessary information, and for allowing us to record disease intensity data and to collect samples from their orchards. The authors also gratefully acknowledge the Ethiopian Institute of Agricultural Research for financial support, and the National Agricultural Biotechnology Program at the Holetta Agricultural Research Center for allowing the microbial biotechnology laboratory for pathogen isolation, identification and pathogenicity test. The first author would like to thank Dr. Edossa Etissa, National Tropical Fruits Research Project Coordinator, and Dr. Getachew Ayana, Director of Melkassa Agricultural Research Center for facilitating financial matters and vehicles for the field

surveys. The authors are grateful to the Fruits Research Team at Melkassa for their assistances during the field surveys. The authors also thank the Biotechnology Research Team at Holetta for their unreserved support during the laboratory works.

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