OPEN ACCESS

Middle East Research Journal of Biological Sciences
ISSN 2789-7710 (Print & ISSN 2958-2091 (Online)
Frequency: Bi-Monthly
DOI: 10.36348/merjbs.2023.v03i03.009

KUWAIT SCHOLARS PUBLISHER
Website: http://www.kspublisher.com/
Email: office@kspublisher.com

Resistance Evaluation of Gurage Coffee Accessions Against Coffee Berry Disease (*Colletotrichum Kahawae* Waller and Bridge)

Dereje Amare[1*], Gerba Daba[2]
[1]Department of Plant Pathology, Ethiopian Institute of Agricultural Research; Holetta Agricultural Research Center, Ethiopia
[2]Jimma University Colleges of Agriculture and Veterinary Medicine, Ethiopia

Abstract: Coffee Arabica is an important crop in the national economy of Ethiopia. Coffee berry disease (CBD), coffee wilt disease (CLD) and coffee leaf rust (CLR) are the most important coffee diseases in the country. Development of coffee cultivars for different localities having a character of diseases resistant, high yielding and quality coffee is important. Previous research works have little effort to provide varieties that are suitable for Gurage zone southern parts of Ethiopia. Hence, this study was carried out to evaluate Gurage coffee accessions against CBD. Evaluations of Gurage coffee accessions were conducted under field by Attach Berry Test and laboratory by Detach Berry Test methods in Randomized Complete Block Design and Completely Randomized Design, respectively in 2018. The study result indicated that Gurage coffee accessions Gu-18, Gu-1 and Gu-4 had lower CBD infection level in both field and laboratory experiment, which was 5.4, 8.29 and 11.37 and 32.5, 45.0 and 25.8 %, respectively. Those coffee accessions that showed low level of infection are an opportunity for further breading research work and could be the best alternatives to CBD management particularly for the study area. Future research should focus on evaluating the promising Gurage coffee accessions in seedling inoculation test and in multi-location field trials for several years.

Keywords: Coffee Arabica; Coffee berry disease; Disease Resistance; CBD management; Gurage coffee accession.

RESEARCH PAPER

***Corresponding Author:**
Dereje Amare
Department of Plant Pathology, Ethiopian Institute of Agricultural Research; Holetta Agricultural Research Center, Ethiopia

How to cite this paper:
Dereje Amare & Gerba Daba (2023). Resistance Evaluation of Gurage Coffee Accessions Against Coffee Berry Disease (*Colletotrichum Kahawae* Waller and Bridge). *Middle East Res J Biological Sci*, *3*(3): 89-95.

Article History:
| Submit: 21.11.2023 |
| Accepted: 22.12.2023 |
| Published: 29.12.2023 |

1. INTRODUCTION

Coffee is a perennial crop which belongs to the genus *Coffea* in the *Rubiacea* family, and is mostly grown in the tropical and subtropical regions (Berthaud and Charrier, 1988). More than 125 million peoples were earning their income directly or indirectly from Coffee products in worldwide (Lashermes *et al.*, 2012) and is the second most exported commodity after oil (Davis *et al.*, 2012). Major coffee producing sub-Saharan African countries; that they depend for their foreign exchange earnings were; Ethiopia, Uganda, Kenya, Rwanda and Burundi (Phiri *et al.*, 2010). Ethiopia is one of the leading Arabica coffee producers in Africa with a production of 7.65million bags (ICO, 2018). In Ethiopia 15-16 million people's income depends on coffee farming, 10 percent of agricultural production, and about 34 percent of total export earnings over the past decade, and the country is center of origin and genetic diversity of Arabica coffee (Tefera, 2015; Tadesse *et al.*, 2015; ICO, 2018). However, despite the vast area of cultivation, high genetic diversity and importance to the national economy, the productivity of coffee is very low (about 669.6 Kg/ha) in the country (CSA, 2017). These is due to use of unimproved coffee landraces, poor agronomic practices and widespread and prevalence of pests and diseases (Girma *et al.*, 2009).

Coffee berry disease *(Colletotrichum kahawae*), coffee wilt disease (*Gibberella xylarioides*) and coffee leaf rust (*Hemileia vastatrix*) economically important coffee diseases in Ethiopia. Coffee berry disease (CBD) is the most devastating disease in Africa at high altitude. It attacks all stages of the crop from flower to ripe fruits and sometimes leaves, but the highest yield loss occurs when the green berries are infected (Batista *et al.*, 2017). About 24% to 30% yield loss is incurred by CBD it may reach up to 100% in high rainfall, high humidity and high altitude areas (Eshetu and Waller, 2003; Garedew *et al.*, 2017).

Arabica varieties used in crop production and accessions held in germplasm collections were far below the genetic diversity of wild Arabica populations (Labouisse *et al.*, 2008). The wild populations also have high functional diversity in terms of pest and drought tolerance (Girma *et al*, 2005; Taye, 2006). Development of new coffee cultivars for different localities having a

character of diseases resistant, high yielding and possess unique quality profile plays important role for improving coffee productivity and foreign exchange earnings of Ethiopia (Girma *et al.*, 2009). Testing of selecting materials for resistance to CBD in the laboratory and in different locations is very vital (Eshetu, 2000). In Ethiopia CBD resistant cultivars have been identified and commercialized for immediate use, and currently different works have been carried out on the genetic improvement of Arabica coffee. However, this research direction has little effort to provide varieties that are suitable for Gurage zone southern parts of Ethiopia. Currently, Jimma Agricultural Research Center has collected 21 coffee landraces from major coffee producing districts of Gurage zones to evaluate their yield, diseases resistance and quality. Therefore, the present study was carried out on evaluating those Gurage coffee collection for their resistance to Coffee berry disease.

2. MATERIALS AND METHODS

2.1 Description of the Study Area

The attached berry test was conducted in Gera Agricultural Research Sub-center (GARSC) at field condition. The sub center is found in Oromia Regional State in Jimma zone, Ethiopia, located around 7º7'N latitude and 36º0'E longitude and at an altitude of 1900 m a s l. It receive an annual rainfall of 1877.8mm, with mean minimum and maximum temperatures of 10.4ºC and 24.0ºC, respectively (Anteneh and Taye, 2015). The growth room studies were conducted at Jimma Agricultural Research Center (JARC). The center is found at Oromia Regional State in Jimma zone, Ethiopia, located around 07°46'N latitude and 36°47'E longitude coordinate and at an elevation of 1753 m a s l. It receive annual rainfall of 1572mm, with mean minimum and maximum temperatures of 11.6ºC and 26.3ºC, respectively (JARC, 2004).

2.2 Treatments and Experimental Design

Attached berry test was conducted on 17 Gurage coffee accessions in August 2018, which were previously established at GARSC, those score 0-20% CBD visual assessment data in three consecutive years. The established experiment contains 21 Gurage coffee accessions and one field resistant check (74110) were planted with two replications in RCBD design. The ABT was conducted following the methods described by Van der Graaff (1981). Three trees per accessions were selected and divide the selected trees in three strata of branches (top, middle, and bottom) take one middle braches from each stratum for inoculation. Then, record a number of berries in selected branch after removing diseased, wounded, matured and pinhead berries.

In order to obtain the inoculums source, CBD infected green berries with active black lesions were collected from the GARS center. Then, diseased berries were wetted slightly with distilled sterilized water and incubated in a closed plastic box for 24-48 hours which was sufficient time to produce a reasonable sporulation. Then, a conidia suspension was prepared by rinsing the berries in sterilized distilled water and the conidia density was determined by haemocytometer after repeated purifying of suspension through double layers of cheese clothes. The berries in each selected stratum branches were inoculated with a suspension of 2x 10^6 conidia/ml using hand sprayer in late afternoon to avoid excessive heat (Arega 2006; Kamau, 2015). Then, each branch was kept moist and warm over night for 12 hours covered in a plastic 'sleeve'. The plastic sleeve was insulated with paper bag to avoid high temperature. After 21 days of inoculation the numbers of healthy and diseased berries were recorded.

The second method of evaluating Gurage coffee accessions was Detach berry test in green house. Seventeen Gurage coffee accessions (those score 0-20 % CBD in visual assessment) plus 3 cheeks i.e., 741(resistant), 74110 (field resistant and lab moderately resistant) and 370 (susceptible) varieties were used for this test. The experiment was laid out in CRD design in three replications containing 20 berries per replication in plastic boxes that contain misted filter paper. Fifteen weeks old from date of flowering of the expanding coffee berries from those Gurage coffee accessions and the cheeks were collected (Pinard *et al.*, 2012). The berries were picked randomly from bottom, middle and top of the coffee tree in order to have a representative sample. Berries were surfaced sterilized with 5% sodium hypochlorite solution for 2 minutes and rinsed three times with sterile distilled water for 2 minutes each and dried using sterile cotton cloth. The wounded stalk end of the berries was removed with a sterile scalpel to avoid contamination with saprophytic fungi.

Ten days old mycelia colonies culture of EZD isolate, which was more aggressiveness isolate (Amare *et al.*, 2021), was washed by flooding with 10 ml sterilized distilled water, rubbed with sterilized scalpel and then transferred to 50 ml sterilized beaker to harvest conidia. The suspension was stirred with magnetic stirrer for 15 minutes and filtered through double layers of cheese clothes. After repeating the procedure the spore concentration was adjusted to 2 x 10^6conidia/ml and 20µl of conidia suspension was deposited on the berries using a pipette while shaking time to time when drawing the inoculums (Kilambo, 2008; Kamau, 2015). As a control (check) 20µl distilled sterilized water was poured on the berries. Boxes were sealed to provide saturated humid conditions necessary for disease development. Regular opening after every three days was done for 10 minute to allow for aeration of the berries.

The data on infection collected every three days starting from 3rd days post inoculation when CBD symptoms were visible. After 14 days, data on disease intensity (PSI), expressed as pathogenicity level of each isolates were recorded using a scales of 0 to 6 (modified Van der Vossen *et al.*, 1976 adopted from Abdi and

Abdu, 2015) (Table 1). After scoring each coffee berry individually, average infection percentage (AIP) on each isolates across the replicates was calculated as follows:

AIP =∑ [Ir1 + Ir2 + Ir3+ ⋯Irn]/N

Where, I is the sum of disease score; n is the number of replication; Irn is the sum of disease score in replication *n;* N is the total number of berries scored in the replications (Kamau, 2015).

Table 1: Assessment key for evaluation of coffee berry disease severity in *Coffea arabica*

Disease index	Descriptions
0	Healthy green berries without symptoms
1	Black sunken lesions cover < 2% of the green berries surface
2	Black sunken lesions cover 2-5% of the berries surface; approximately 3mm in diameter
3	Black sunken lesions cover 6-10% of the berries surface shows black lesions approximately 5 mm in diameter
4	Black sunken lesions cover 11-50% the berries surface; approximately 7mm in diameter
5	Black sunken lesions cover 51-99% of the berries surface; approximately15 mm in diameter
6	>99% or the whole surface of berries covered with black sunken lesions; mummified berries

Source: Modified Van der Vossen *et al*. 1976

2.3 Data Analysis

All laboratory and field data were summarized, tested for normal distribution using the normality test and subjected to analyses of variance (ANOVA) using SAS program version 9.3 software (SAS, 2011). Before analysis of variance, the field evaluation data (ABT) was transformed with Arcsine transformation.

3. RESULT AND DISCUSSION

The analysis of variance (ANOVA) for attached berry test showed significant difference among the accessions at $p<0.005$ level (Fig. 1). The high CBD severity infection was recorded on accessions Gu-15 (30.07%) which showed highly significant difference for most of Gurage coffee accessions but, there was no significant difference as compare to Gu-19, Gu-10, Gu-6 and Gu-13 accessions. The lowest CBD severity infection was recorded on accession Gu-18 (13.21%) which did not statistically significant different between Gu-7, Gu-12, Gu-1, Gu- 8, Gu-20, Gu-9, Gu-4 and check (74110), but it has significant difference compare to the other accessions (Fig.1). The result of this study clearly indicates that certain Gurage coffee accessions that have been better or comparable CBD resistant to the reference accessions/varieties. Similarly, Tefestewold *et al*., (1995); Bayetta (2001) and Arega (2006) also reported significant differences in seedling percent infection in reaction to CBD for different coffee accessions.

The variation among coffee accessions for the pathogen response can be associated with the genetic makeup of the accessions. The resistance nature of CBD in coffee has horizontal nature (Van Graff 1981; Bayetta, 2001). The host resistance appears to be largely based on the rapid formation of a cork barrier in the pericarp of the developing fruit distal (point of attachment) from the initial infection site that effectively prevents the pathogen from further invading of healthy tissue which totally absent or incomplete in susceptible host plants (Bayetta, 2001; Silva, 2006). CBD resistance of Lyamungu coffee hybrids is partly being contributed by wax surface on green coffee berries (Kilimbo *et al*., 2013). Chen *et al*., (2004) showed the green coffee berries possess inherent antifungal compounds that counteract the infection of coffee by *C. kahawae* strains. In the resistant coffee genotypes, restrict the fungal growth associated with a series of hypersensitive reaction (HR) responses. HR indicates the rapid and efficient plant resistance mechanism that leads to a localized plant cell death in response to invasion by a pathogen and is characterized by a rapid loss of membrane integrity in the infected host cells (Hoglund *et al*., 2005; Singh and Upadhyay, 2013).

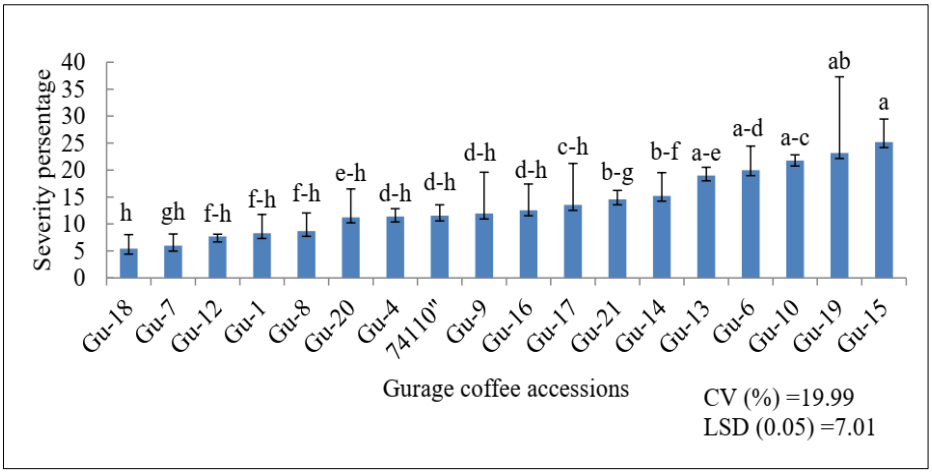

Figure 1: Percent CBD infection of Gurage coffee accessions in attach berry test
Means followed with the same letters are not significantly different at LSD (0.05)

The detach berry test study revealed that there was highly significant difference (p<0.001) among Gurage coffee accessions for reaction of CBD resistant (Fig. 2). Coffee accessions Gu- 4 and Gu-18 showed lower CBD severity infection (25.83 and 32.50%) and statistically highly significant difference as compare to resistant check (741) and other Gurage coffee accessions. The highest CBD severity infections was recorded on coffee accession Gu-7 (88.33%), which was moderately resistant in the field experiment (ABT) and now susceptible in this experiment. The coffee cultivar 74110 was one of the released field CBD resistant cultivars and had moderately susceptible reaction in laboratory (Tefestewold *et al.*, 1995; Bayetta *et al.*, 2001; Kumlachew *et al.*, 2018). Those four Gurage coffee accessions (Gu-7, Gu-19, Gu-16 and Gu-13) showed higher infection percentage more than the well-known susceptible variety (370). The susceptible coffee accessions had their berry surfaces entirely covered by black sunken lesions. On the other hand, those resistance Gurage coffee accessions formed restricted black scab lesions that hindered further penetration of the pathogen into the intercellular parts of the berry (Kamau, 2015) (Fig. 3). Gichuru (1997) findings indicated that scab lesions were the common macroscopic expression of resistance of coffee to CBD. This resistance to CBD is preformed and induced, and it operates at distinct stages of pathogenesis.

In Gurage coffee accessions, the resistant accessions should restricted growth of the pathogen (scab formation). Presence of scab CBD lesions formed in resistant or moderately resistant coffee genotypes suggests the mechanism by which further invasion of the CBD pathogen is blocked (Masaba and van der Vossen, 1982; Chen *et al.*, 2004). On the other hand, scab formation on the green coffee berry surface is due to cork barrier formation and limits fungal hyphal growth inside the plant tissue (Masaba and van der Vossen 1982).

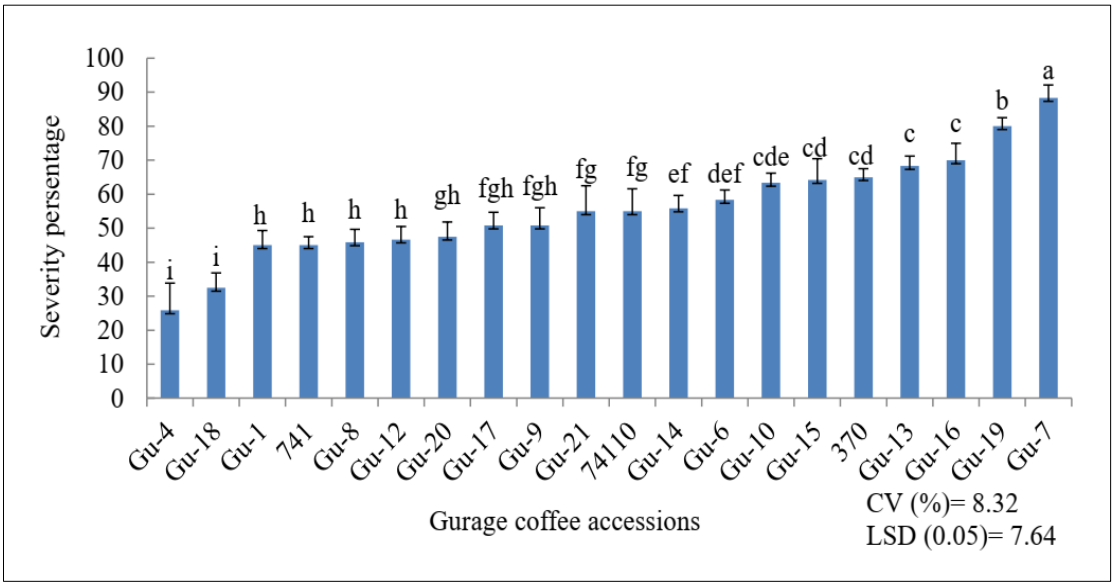

Figure 2: Response of Gurage coffee accessions to *C. kahawae* infection in detached berry test.
Means followed with the same letters are not significantly different at LSD (0.05).

In the resistant variety the restricted hyphal growth was associated with the hypersensitive-like host cell death (HR), and early accumulation of phenolic compounds both in cell walls and in the cytoplasmic contents. These responses were also observed in the susceptible variety, but in a significantly lower percentage of infection sites and did not prevent the fungal growth, as indicated by the appearance of typical anthracnose symptoms and the presence of acervuli (Silva, 2006; Loureiro *et al.*, 2012). Variations in Gurage coffee accessions could be an opportunity for the next better resistant varietal development through breeding. As resistance in perennial crops like coffee is observed and screened during the late stage of development (Van der Vossen *et al.*, 2015), multi-location repeated evaluation of these accessions over time could be the future line of work for pathologists and breeders.

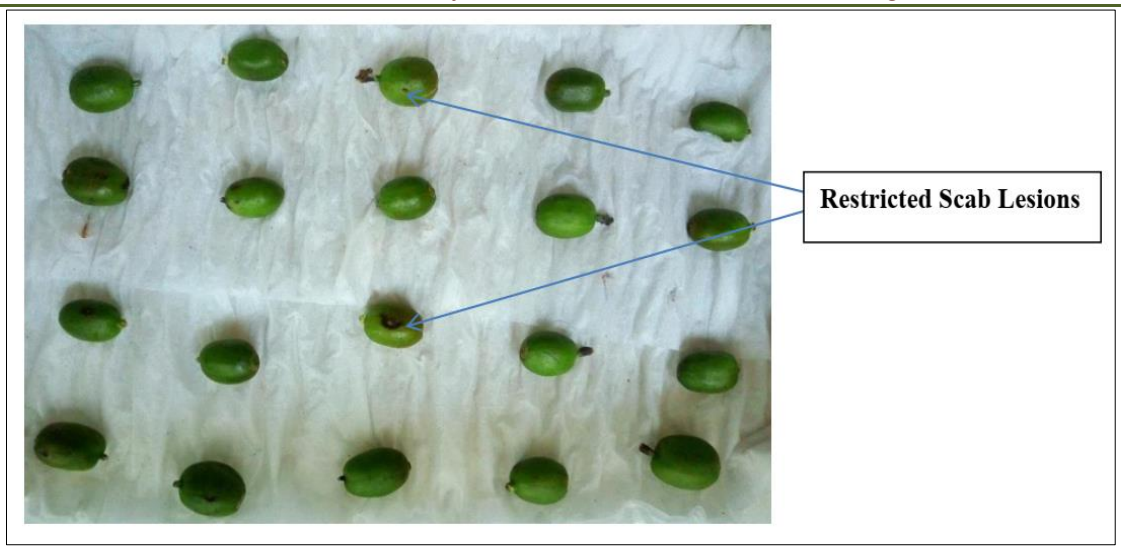

Figure 3: Presence of restricted scab lesions on resistant coffee accessions inoculated with conidia of *C. kahawae* after 21 days

4. SUMMARY AND CONCLUSION

The present study was conducted on evaluation of Gurage coffee accessions resistance to coffee berry disease. Resistance evaluation test under field (ABT) and laboratory (DBT) showed significant variations among of Gurage coffee accessions. The mean percent of berry infection ranged from 13.21% (Gu-18) to 30.07% (Gu-15) in field ABT and 25.83% (Gu-4) to 88.33% (Gu-7) in the laboratory DBT. Gurage coffee accessions Gu-18, Gu-1 and Gu-4 were showed low CBD infection percentage under both ABT and DBT experiments. Results of these experiments confirmed that the variations were mainly due to the existence of difference in genetic makeup of the selected accessions in reaction to CBD. Those coffee accessions that showed low level of infection in CBD resistance evaluation test under field and laboratory conditions are an opportunity for further breading research work and could be the best alternatives to CBD management particularly for the study area. However, these accessions should be evaluated in seedling hypocotyl test and also evaluated under field in multi-location and over years.

Declarations Author Contribution Statement

Dereje Amare and Gerba Daba: Conceived and designed the experiments; performed the experiments, analysis tools or data; wrote the paper.

Funding Statement

This work was supported by Ethiopian Institute of Agricultural Research and Jimma University College of Agriculture and Veterinary Medicine.

Data Availability Statement: The authors do not have permission to share data.

Declaration of Interests' Statement: The authors declare no conflict of interest.

Acknowledgements

We thank Jimma Agricultural Research Center for allowing us to its pathology laboratory and growth room facilities and Gera Agricultural Research sub-center for allowed us field experiment and data collection support.

REFERENCE

- Amare, D., Hailu, B., & Daba, G. (2021). Cultural, morphological and pathogenic variability of Colletotrichum kahawae isolate of Gurage Zone. *Journal of Plant Pathology and Microbiology*, *11*(12), 1-6.
- Anteneh, N., & Taye, K. (2015). Determining suitable shade trees, panting pattern and spacing for Arabica coffee production in south and southwestern Ethiopia. *Journal of Biology, Agriculture and Healthcare*, *5*(17), 9-15.
- Batista, D., Silva, D. N., Vieira, A., Cabral, A., Pires, A. S., Loureiro, A., ... & Varzea, V. (2017). Legitimacy and implications of reducing Colletotrichum kahawae to subspecies in plant pathology. *Frontiers in plant science*, *7*, 2051.
- Bayetta, B. (2001). Arabica coffee breeding for yield and resistance to coffee berry disease (Colletotrichum Kahawae sp. nov) Doctoral Dissertation. *Imperial College at Wye University of London*, 272.
- Berthaud, J., Charrier, A. (1988). Genetic resources of *Coffea*. In: Clarke R J, Macrae R (eds), Coffee: Agronomy, vol. IV, pp.1-42. *Elsevier Applied Science, London*.
- Central statistic Agency (CSA). (2017). *Agricultural sample survey*, Addis Ababa.
- Chen, Z., Nunes, M. A., Silva, M. C., & Rodrigues Jr, C. J. (2004). Appressorium turgor pressure of Colletotrichum kahawae might have a role in coffee cuticle penetration. *Mycologia*, *96*(6), 1199-1208.
- Davis, A. P., Gole, T. W., Baena, S., & Moat, J. (2012). The impact of climate change on indigenous arabica

coffee (Coffea arabica): predicting future trends and identifying priorities. *PloS one*, *7*(11), e47981.

- Derso, E., & Waller, J. M. (2003). Variation among Colletotrichum isolates from diseased coffee berries in Ethiopia. *Crop protection*, *22*(3), 561-565.
- Eshetu, D. (2000). Pre selection method for coffee berry disease (CBD) resistance in Ethiopia. In: proceedings of the workshop on control of coffee berry Disease (CBD) in Ethiopia, *47-57, 13-15 August 1999*, Addis Ababa, Ethiopia.
- Garedew, W., Lemessa, F., & Pinard, F. (2017). Assessment of berry drop due to coffee berry disease and non-CBD factors in Arabica coffee under farmers fields of Southwestern Ethiopia. *Crop protection*, *98*, 276-282. https://doi.org/10.1016/j.cro.
- Gichuru, E. K. (2007). Sensitive response and resistance to bery disease (Colletotrichum kahawae) of two coffee varieties (Coffea arabica and C. canephora): histological comparisons of interactions. *Agronomie Africaine*, *19*(3), 233-240.
- Girma, A., Hindorf, H., Steiner, U., Nirenberg, I., Dehne, W., & Schellander, K. (2005). Genetic diversity in the coffee wilt pathogen (Gibberella xylarioides) populations: differentiation by host specialization and RAPD analysis in Germany. Journal of Plant Diseases and *Protection*, 112(2), 134-145.
- Girma, A., Million, A., Hindorf, H., Arega, Z., Demelash, T., & Chala, J. (2009). Coffee wilt disease in Ethiopia. In: Coffee Wilt Disease, Flood J (ed). CAB International. *Wallingford*, UK, 50-68.
- Höglund, S., Larsson, S., & Wingsle, G. (2005). Both hypersensitive and non-hypersensitive responses are associated with resistance in Salix viminalis against the gall midge Dasineura marginemtorquens. *Journal of Experimental Botany*, *56*(422), 3215-3222.
- ICO (International coffee organization), (2018). Coffee market report in the international trade, *challenges and opportunities facing the sector*, 1-8.
- JARC, (2004). Jimma Agricultural research center annual progress report. 2005, *Jimma, Ethiopia.*
- Kamau, K. (2015). Characterization of coffee genotypes derived from crossing Rume Sudan and SL 28 coffee varieties against coffee berry disease (CBD) causal pathogen (Colletotrichum kahawae) (Doctoral dissertation), 78
- Kilambo, D. L., Guerra-Guimarães, L., Mabagala, R. B., Varzea, V. M. P., Haddad, F., Loureiro, A., & Teri, J. M. (2013). Characterization of Colletotrichum kahawae strains in Tanzania. *International Journal of Microbiology Research*, *5*(2), 382.
- Kilambo, L. (2008). *Virulence of Colletotrichum kahawae strains and their effect on resistant Arabica coffee varieties in Tanzania* (Doctoral dissertation, Sokoine University of Agriculture).
- Kumlachew, A., Girma, A., Fikre, L., & Diriba, M. (2018). Induction of systemic resistance in Arabica coffee (Coffea arabica L.) against coffee berry disease (Colletotrichum kahawae Waller & Bridge) mediated through plant defense activator. *International Journal of Pest Management*, 1-11.
- Labouisse, J. P., Bellachew, B., Kotecha, S., & Bertrand, B. (2008). Current status of coffee (Coffea arabica L.) genetic resources in Ethiopia: implications for conservation. *Genetic Resources and Crop Evolution*, *55*, 1079-1093.
- Lashermes, P., Combes, C., Ansaldi, C., Gichuru, E., & Noir, S. (2012). Analysis of alien introgression in coffee tree (Coffea arabica L.). Molecular breeding, 27(2), 223-232.
- Loureiro, A., Nicole, M. R., Varzea, V., Moncada, P., Bertrand, B., & Silva, M. C. (2012). Coffee resistance to Colletotrichum kahawae is associated with lignification, accumulation of phenols and cell death at infection sites. *Physiological and molecular plant pathology*, *77*(1), 23-32.
- Masaba, D. M., & Van Der Vossen, H. A. H. (1982). Evidence of cork barrier formation as a resistance mechanism to berry disease (Colletotrichum coffeanum) in arabica coffee. *Netherlands Journal of Plant Pathology*, *88*, 19-32.
- Mohammed, A., & Jambo, A. (2015). Importance and characterization of coffee berry disease (Colletotrichum kahawae) in Borena and Guji Zones, Southern Ethiopia. *Journal of Plant Pathology & Microbiology*, *6*(09).
- Phiri, N., Baker, P., Ruther, f. M., Flood, J., Musoli, P., Mmugi, K., Kiambo, D., Aduna, G., Hakiza, G., Pinard, F., & Odour, G. (2010). The regional coffee wilt program, In: Proceedings of 23rd International Scientific Colloquium on Coffee (ASIC) 3rd- 8th October, 2010.Bali, Indonesia, 537-548.
- Pinard, F., Omondi, C. O., & Cilas, C. (2012). Detached berries inoculation for characterization of coffee resistance to coffee berry disease. *Journal of Plant Pathology*, 517-523.
- SAS, S. (2011). STAT 9.3 User‟s guide. Cary, NC: SAS Institute Inc.
- Silva, C. (2006). Coffee resistance to the main diseases: leaf rust and coffee berry disease. Brazilian Journal of Plant Physiology, 18(1), 119 - 147.
- Singh, K., & Upadhyay, S. (2013). The hypersensitive response: a case of cell death induction in plants. International Journal of Engineering, 2, 1-16.
- Tadesse, K., Mekdim, D., & Minten, B. (2015). Coffee Income, Food Security and Diet Diversity of Small holder Coffee Growers in Ethiopia. *EDRI working paper*, 15, 39.
- Taye, K., & Obso, (2006). Eco-physiological Diversity of Wild Arabica Coffee Populations in Ethiopia: *Growth, Water Relations and Hydraulic Characteristics A long a Climatic Gradient, Vol. 46***,** Cuvillier Verlag.
- Tefera, A. (2015). Ethiopia: Coffee Annual Report. GAIN Report Number ET1514 (USDA Foreign Agricultural Service, Ethiopia. *Journal of Plant Pathology Microbes, 6*, 302.

- Tefestewold, B., Omondi, O., & Hindorf, H. (1995). Caffeine content in relation to resistance of coffee Arabica L. to coffee berry disease (*Colletotrichum coffeanum* Noack). *Journal of Plant Diseases and Protection*, *103*, 15-19.
- Van der Graaff, A. (1981). Selection of Arabica coffee types resistant to coffee berry disease in Ethiopia (Doctoral dissertation. In Department of Phytopathology, Agricultural University, Wageningen, *The Netherlands*, 120.
- Van der Vossen, H. A. M., Cook, R. T. A., & Murakaru, G. N. W. (1976). Breeding for resistance to coffee berry disease caused by Colletrotrichum coffeanum Noack (sensu Hindorf) in Coffea arabica LI Methods of preselection for resistance. *Euphytica*, *25*(1), 733-745.
- Van der Vossen, H., Bertrand, B., & Charrier, A. (2015). Next generation variety development for sustainable production of arabica coffee (Coffea arabica L.): a review. *Euphytica*, *204*(2), 243-256. DOI:10.1007/s10681-015-1398-z.
- Zeru, A. (2006). *Diversity of Arabica coffee populations in afromontane rainforests of Ethiopia in relation to Colletotrichum kahawae and Gibberella xylarioides* (Doctoral dissertation, Addis Ababa University).