

Increasing the resilience of coffee production to leaf rust and other diseases in India and four African countries

Coffee Leaf Rust in India and Africa 2008-2013

Final Technical Report – Project number CFC/ICO/40



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Foreword

The purpose of this final technical report is to present the results of the project “Increasing the resilience of coffee production to leaf rust and other diseases in India and four African countries”, which was carried out from 2008 to 2013. The project was developed in response to the worsening Coffee Leaf Rust (CLR) status in the five participating countries of India, Kenya, Rwanda, Uganda and Zimbabwe.

Central to the strategy for managing CLR is the development of resistant varieties, and many of the project’s activities revolved around research, trials, multiplication and distribution of coffee varieties resistant to CLR and other diseases. The pioneering plant breeder Luther Burbank is reputed to have said “*The secret of improved plant breeding, apart from scientific knowledge, is love*”. The project certainly generated and applied a large volume of scientific knowledge. But it was the love and passion for their work and the needs of the rural poor that inspired the many people who worked hard to make this project a success. I salute them all for their commitment and contributions.

The donor, the Common Fund for Commodities (CFC), and the supervising body, the International Coffee Organisation (ICO), are gratefully acknowledged for supporting the project. The collaborating institutions are also recognised for providing co-funding for the project. The Coffee Rust Centre, Portugal (CIFC) did crucial work in analysing CLR races and in training scientists from Africa, for which I thank them sincerely. Special thanks go to Caleb Dengu and Eltha Brown (CFC), and Denis Seudieu (ICO), for their continual support and encouragement.

The management of CLR is an on-going struggle. Recommendations have been provided in this report that highlight areas of focus for continuing research and development of CLR management strategies, as well as for ensuring that the results are of benefit to the rural families whose livelihoods depend on coffee. Decision makers in coffee producing countries are therefore urged to study the findings and recommendations, and act accordingly.

Noah Anthony Phiri

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Principal Contributors

I sincerely thank the following scientists for their major contributions during this project. Many other people were involved in implementing the project, within and without participating institutions, and their work is also gratefully acknowledged.

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Acronyms and Glossary

AGRITEX	Agricultural Technical and Extension Services
ANOVA	Analysis of variance
ARDA	Agriculture and Rural Development Authority (Zimbabwe)
ASIC	Association for Science and Information on Coffee
BAC	Bacterial artificial chromosome
BBC	Bacterial blight of coffee
CABI	CAB International
CBD	Coffee berry disease
CCRI	Central Coffee Research Institute
CFC	Common Fund for Commodities
CGA	Coffee Growers Association
CIFC	Coffee Rust Research Centre
CLR	Coffee Leaf Rust
CRF	Coffee Research Foundation
CRSS	Coffee Research Sub-Station
CWD	Coffee Wilt Disease
DAO	District Agriculture Officer
DAR4D	Department for Agriculture Research for Development
DNA	Deoxyribonucleic acid
EAFCFA	East African Fine Coffees Association
EU	European Union
FAO	Food and Agriculture Organisation
FFS	Farmer Field School
GAP	Good agriculture practice
GBM	Grain Marketing Board
HDT	Hybrido de Timor
ICO	International Coffee Organisation
IP	Intellectual Property
ISSR	Inter-simple sequence repeats
KEPHIS	Kenya Plant Health Inspectorate Services
KIPI	Kenya Industrial Property Institute
MAAIF	Ministry of Agriculture Animal Industry and Fisheries
MOA	Ministry of Agriculture
MSc	Master of Science
NAADS	National Agriculture Advisory Services
NaCORRI	National Coffee Resources Research Institute
NAEB	National Agriculture Export Board
NARO	National Agriculture Research Organisation
NEMA	National Environmental Management Authority
NGO	Non-Governmental Organisation
NUCAFE	National Union of Coffee Agribusiness and Farm Enterprises
P	Probability
PCR	Polymerase chain reaction
PEA	Project Executing Agency
PhD	Doctor of Philosophy

PIA	Project Implementing Agency
QDA	Quantitative Descriptive Analysis
RAB	Rwanda Agriculture Board
RAPD	Random Amplified Polymorphic DNA
RCBD	Randomised Complete Block Design
RFLP	Restriction fragment length polymorphism
RMC	Rwanda Mild Coffee
SCAR	Sequence-characterized amplified regions
SRAP	Sequence-related amplified polymorphism
SSR	Simple Sequence Repeats
UCDA	Uganda Coffee Development Authority
USAID	United States Agency for International Development
USD	United States Dollar
ZCFU	Zimbabwe Commercial Farmers Union
ZCM	Zimbabwe Coffee Mill
ZFC	Zimbabwe Fertilizer Company
ZIMRA	Zimbabwe Revenue Authority

Executive Summary

1. Coffee diseases continue to be a principal production constraint in many localities worldwide, especially for smallholder farmers who have insufficient resources to apply control measures. Among the most serious of these diseases are coffee leaf rust (CLR) and coffee berry disease (CBD). Yield losses attributed to CLR are estimated at 40%, although higher losses are recorded when weather conditions favour the disease.
2. The Common Fund for Commodities (CFC) granted \$2.92m to a 5 year project (2008-2013) to tackle CLR in India, Kenya, Rwanda, Uganda and Zimbabwe. The International Coffee Organisation was the Supervisory Body, and CAB International the Project Executing Agency.
3. The project purpose was to reduce economic and environmental costs of disease management for smallholder coffee farmers by reducing the crop and quality losses caused by the diseases CLR and CBD, especially in regard to smallholder, coffee-based farming systems.
4. The project had five components:
 - a. Rural community responses to CLR and other diseases, and the sourcing and production of coffee genetic material
 - b. Conservation and identification of coffee varieties and disease races
 - c. Trials with new and existing materials
 - d. Scientific management, information systems and dissemination
 - e. Project management and coordination

Component 1

5. Farmers in project areas identified CLR as one of their major constraints. Lack of inputs (fertilizer, pesticides) was also important, along with other diseases including Coffee Berry Disease (CBD), and insects including white coffee stem borer and berry borer.
6. Although most smallholder farmers were able to recognize some of the symptoms of CLR, in Africa many lacked the knowledge needed to manage the disease. Many did little or nothing to control the disease except in India and Kenya where some resistant varieties were available. At the beginning of the project availability of resistant coffee varieties was generally limited.
7. As an entry point for dissemination, and to provide a platform for improving interactions and understanding between different stakeholders, farmer field schools were established in all countries. Farmers took part in exchange visits, open days, and on-farm trials, and reported economic and social benefits from the project.
8. Participating countries identified 31 locally available genotypes for inclusion in varietal trials for screening for resistance to CLR. Selections 5A and 6 were imported to Africa from India, but no African material was introduced to India due to risks of introducing coffee berry disease.

9. India established new seed plots to meet a growing demand for the variety Chandragiri in 2007, and for producing selections SIn 5A, SIn 5B, SIn 6 and SIn9. Over 50 tonnes of seed was produced in India, and nearly 16,000 farmers received seed. Seed gardens and homogeneous seed plots of resistant varieties were set up or improved in Africa, although this was not originally planned. Nearly 3 tonnes of seed was harvested from four gardens in Kenya. Over one million seedlings were produced in nurseries that the project established or renovated in participating countries.
10. Demand for improved CLR and CBD resistant varieties was high in all countries. In Kenya demand greatly exceeded supply following a well-publicised launch of CLR-resistant varieties (Batian 1, 2 and 3).

Component 2

11. In all countries the conservation status of coffee genetic collections was poor. Most are susceptible to CLR leading to severe defoliation and infestations by insect pests. The project rehabilitated, relocated or maintained 56 collections in Zimbabwe, 120 in Uganda, 182 in Rwanda and 250 in India. India produced a monograph on their collections.
12. Conservation strategies for germplasm collections were instituted, including spraying against CLR, managing other pests and diseases, and good agronomic practices. All countries are now conserving their existing germplasm as well as the newly introduced Selections 5A and 6. Kenya published their germplasm conservation strategy.
13. Coffee leaf rust races were collected and determined in all project countries through work at the Coffee Rust Research Centre (CIFC), Portugal. India identified two new races which have not yet been named. Kenya found 6 races not previously recorded there, while Zimbabwe found one.
14. High quality facilities were established at the Central Coffee Research Institute (CCRI) in India for genetic characterisation of germplasm. Marker assisted selection was developed, which is already helping in establishing systems for conserving coffee collections and for improving the cost-effectiveness of coffee breeding programmes. Two SCAR markers were validated and applied, along with 11 genotype specific SRAP markers. Progenies arising from marker assisted selection have already been established in field plots.
15. National research scientists from Africa were trained in marker assisted selection, breeding, pathology, and extension through short courses and attachments at CIFC and CCRI, as well as through post graduate degrees.

Component 3

16. The varieties from India (SI 5A and SI6) were screened for resistance to CLR in Africa. Selection 6 was resistant in all participating countries and Selection 5A was generally resistant but with some individual bushes susceptible, due to the variety being a composite. Selection 6 also has potential field tolerance to Bacterial Blight of Coffee (BBC).

17. SI 5A and SI6 are early maturing and appear high yielding. Both are similar to SL28, a popular tall commercial variety which is susceptible to both CLR and CBD. They have good cup quality in comparison with Ruiru 11 and SL28 in Kenya, and Selection 6 was better in quality than the commercial variety BM 139 in Rwanda.
18. Local varieties were also evaluated, and three CLR resistant high yielding varieties were officially released in Kenya. Catuai x HDT, Colombia, Selection 5B and Chandragiri were all resistant/tolerant to CLR in India. In Uganda varieties NG9257 and Elgon CB were resistant to CLR. Line 13683/35 was resistant to CLR in Zimbabwe, and performed better than the commercial variety Catimor 129.
19. In general the weather conditions were too hot for the development of CBD in Africa during the project, so most countries did not have an opportunity to screen for CBD resistance. Screening was done at CIFC in Portugal and in Rwanda.
20. Biological and pesticidal controls for CLR were tested. In India the microbial *Bacillus brevis* was as effective as the currently recommended fungicide regime of Bordeaux mixture. Cyproconazole was the most effective fungicide for controlling CLR in Rwanda and Zimbabwe, and so was registered for use in Rwanda. Tebuconazole was the most effective fungicide in trials in Uganda. Trials in Kenya showed that fungicide application more than pays for the additional cost of control.

Component 4

21. Farmers and extensionists were targeted through a range of communication activities. Printed materials were produced and disseminated including posters, folders with leaflets, and a coffee recommendation handbook. In India farmers were also reached through short TV programmes aired on ETV. An innovative telephone helpline service for coffee farmers India was piloted in India and has been expanded since the project by the Coffee Board.
22. Face-to-face dissemination and upscaling activities centred on farmer field schools (FFSs). India established FFSs in non-traditional coffee growing areas where farmers had less knowledge. FFS open days and exchange visits were used to promote knowledge sharing between different actors in all countries.
23. The project generated a large amount of scientific knowledge. Scientific papers and posters were published and presented at conferences and workshops. In Kenya a coffee atlas was produced, and in India a monograph on coffee collections was published. Annual scientific symposia were held in association with the African Fine Coffee (AFCA) conferences from 2009 to 2012, and research was presented at the 2010 International Conference on Coffee Science (ASIC).

Component 5

24. Project steering committee meetings were held twice a year in each country to share information, discuss the progress of the project and approve country plans and budgets for the following year. Additional technical and administrative backstopping by the PEA was required over and above what had been planned, including training for implementing partners on financial management to reduce delays in reporting. Progress reports were submitted to CFC and ICO every 6 months, a mid-term review was conducted, and an end-of-project workshop was held in India.

25. The project exit strategy included capacity development of national scientists (not originally planned) that will continue to enhance on-going coffee breeding programmes. Infrastructure was provided or improved, such as nurseries, seed gardens and laboratories that can be used on a continuing basis. Co-financing was provided from participating institutions, and several instances of additional co-financing and support after the end of the project have been observed. Prospects for sustainability of the project outcomes and longer term impacts thus appear good.

Recommendations

26. Varietal resistance to CLR and other diseases is the most effective management strategy, and demand for resistant varieties is high. The improved germplasm multiplication capacity developed during the project needs to be deployed to meet this demand. Opportunities for greater involvement of the private sector should be pursued.

27. Germplasm collections must be continuously maintained according to the defined strategies, and characterised and documented where this has not already been completed. If possible, differentials at CIFC, Portugal need to be imported to Africa, and regular CLR race audits carried out.

28. The new capacity in marker assisted selection must be deployed and further developed. Appropriate coffee research facilities are available in India and Kenya, but in other countries facilities are only available in other institutions, so possibilities for using those facilities for coffee work should be pursued. Marker assisted selection should continue to be used as a basis for pyramiding resistance genes, so that a wide range of CLR resistant varieties can be developed with other traits desired by farmers (such as resistance to other diseases and good yield and quality characteristics).

29. Selection 6 introduced from India to Africa now needs to be multiplied and made available to farmers, along with the other local varieties shown to be resistant to CLR. Further single tree selection should be carried out on Selection 5A in African countries before the variety is released.

30. The microbial *Bacillus brevis* showed potential for control of CLR in India, but further testing is required, and opportunities for commercialisation considered. Scientists in Africa could also screen indigenous microflora as potential biocontrol agents for CLR and CBD.

31. Cyproconazole or tebuconazole can be used for single spray curative treatments, if the necessary safety measures can be taken and Copper can be used for preventative purposes. Work to find lower risk cost-effective fungicides should continue.

32. Communication with farmers should therefore emphasise the use of resistant varieties (including information on where they can be obtained), and good agricultural practices for growing healthy crops. Extension materials and information already prepared should continue to be disseminated through any local channels that are available. The telephone helpline for coffee farmers in India can be extended further, and the approach tested in Africa.

33. Further scientific publications are needed to ensure all the project results are widely available. The coffee atlas, monograph and coffee handbook should be made freely available digitally, not just as hard copies.

I. Coffee Leaf Rust: The Problem and the Project

Coffee exports constitute an important source of foreign exchange earnings for producer countries, and most coffee farmers are small scale growers who depend on coffee for a substantial proportion of their livelihood. This is because coffee is a crop very well-suited to production in a family unit, as well as being appropriate for hillside farms where more demanding cash crops cannot easily be grown.

Coffee diseases were the principal production constraint in many localities at the time the project was formulated, especially for smallholder farmers who have insufficient resources to apply control measures. Two serious diseases of *Coffea arabica* are coffee leaf rust (CLR) and coffee berry disease (CBD). Africa and India were the regions most affected by CLR, and this makes per tonne production costs higher than in other parts of the world. Historically, few inputs are applied to coffee trees by smallholder farmers, resulting in poor plant nutrition, low yields, and fungal and insect damage. Losses attributable to fungal damage, primarily CLR, were estimated to be as high as 40%. In addition, poor crop husbandry and the advanced age of most of the coffee trees further compromises productivity.

Disease resistance offers the most sustainable way of managing CLR and CBD, thus ensuring livelihood security of smallholder farmers. Various research programmes had developed resistant varieties, but most countries in Africa were growing varieties which were susceptible to, or which had had their resistance broken down by CLR and CBD. In the case of India the many CLR virulent races led to many varieties becoming susceptible. The use of varieties susceptible to pests and diseases, combined with inappropriate farming techniques, contributes to high production costs. Full protection against CBD, for example, can reach US\$500/ha/yr. Chemical inputs for control of CLR and CBD were reported make up to 33% of farm inputs where applied, yet if the diseases are not properly controlled, severe losses of 50% or more of a year's production are seen.

The specific objective(s) and expected outputs

The project **purpose** was to reduce economic and environmental costs of disease management for smallholder coffee farmers by reducing the crop and quality losses caused by the diseases CLR and CBD, and resources spent on expensive chemicals, especially in regard to smallholder, coffee-based farming systems.

There were five main outputs planned for the project, one for each component:

- Identification of needs and resources: rural community responses to CLR and other diseases and the sourcing and production of coffee genetic material.
- Conservation and identification of coffee varieties and disease races.

- Trials with new and existing materials under a range of field conditions, on farms and on field station.
- Scientific management, information systems and dissemination
- Project management and coordination

Project Costs and Financing

The total project cost was USD 4,014,313 with a total Common Fund for Commodities (CFC) grant of USD 2,918,720 (of which USD500,000 was contributed by the OPEC Fund). Governments of the participating countries provided counterpart contributions worth USD 1,095,593.

The management and implementation arrangements

Project Executing Agency: CABI Africa and India. CAB International (CABI) is an intergovernmental organisation that provides services worldwide to agriculture, forestry, human health and the management of natural resources. CABI headquarters is in the UK, but there are regional centres in all continents. Address: CABI Africa, P. O. Box 633-00621, Nairobi, Kenya. Email: africa@cabi.org; Website: www.cabi.org

Supervisory body: The International Coffee Organization (ICO), with headquarters located in London, maintains the statistics concerning world coffee production, trade and consumption. Address: International Coffee Organization, 22, Berners Street, London W1T 3 DD, UK. Tel: +44 (0) 20 7612 0600; E-mail: info@ico.org

Project implementing agencies/ project partners

The **Coffee Board of India** is a statutory organisation constituted under the Coffee Act VII of 1942. The Board's main research institute is located at Balehonnur in Chikmagalur District of Karnataka State. The following are the Stations that were involved in the project: Central Coffee Research Institute (CCRI), Coffee Research Station: Balehonnur, Chikmagalur District, Karnataka; Coffee Research Sub-Station, Chettalli, Kodagu District, Karnataka; Regional Coffee Research Station, Thandigudi, Dindigul District, Tamil Nadu; Regional Coffee Research Station, R.V. Nagar, Visakhapatnam District, Andhra Pradesh. Address: Number 1, Dr. B.R. Ambedkar Veedhi, Bangalore, 560 001, India. Tel: + 91 80 2268700; Fax: + 91 80 2255557; Email: director@giabg01.vsnl.net.in

African Partner Institutes

Coffee Research Station, Zimbabwe. The Coffee Research Station is the only research institution in Zimbabwe that is responsible for carrying out investigations to find out the factual information on which improved production of coffee under local conditions can be based. It falls under the Department of Research and Specialist Services, Ministry of Agriculture, Mechanisation and Irrigation Development. Address: Ministry of Agriculture, Mechanisation and Irrigation Development of Zimbabwe,

Department of Research and Specialist Services, Josiah Tongogara Ave./Fifth Street Extension, P.O. Box 8108, Causeway, Harare, Zimbabwe. Tel: +2634704531; Fax: +2634728317. Fax: +263 272 942; Email: dkutywayo@qta.gov.zw

Coffee Research Station (CRF), Kenya. CRF was founded in 1944 and has been actively engaged in coffee research activities in plant improvement, management and protection ever since its inception. Renowned coffee varieties SL 28, SL 34, Ruiru II etc. known for their quality and disease resistance were developed by this institution and released to the growing community of Kenya, but the varieties found their way to other coffee growing countries especially in Africa. Address: Coffee Research Station, Coffee Research Foundation, P. O. Box 4, Ruiru, Kenya. Tel: +254 067 542 39/556 31, +254 067 250 81/250 82; Fax: +254 067 540 48; E-mail: director@crf.co.ke

The National Coffee Resources Research Institute (NaCORRI), Uganda. NaCORRI is a Government institution which was promoted out of the Coffee Research Programme in 1997. Its mandate is to conduct research in priority constraints limiting Arabica and Robusta coffee production in the country. Since its inception, NaCORRI has continued with the priority research projects inherited from the Coffee Research Programme and has initiated a number of new research projects. Address: National Crops Resources Research Institute, National Agricultural Research Organisation, P.O. Box 185, Mukono, Kituza, Uganda. Tel: +256-392-700-725, +256-77-467-631; Email: nacorri@africaonline.co.ug, wagoire1@yahoo.co.uk

Rwanda

Rwanda Agriculture Board (RAB), Rwanda. RAB is a new institute which combined the former research institute, Institute des Sciences Agronomiques du Rwanda (ISAR) and the extension and livestock institutes in 2011. RAB's mandate was established to promote the scientific and technical development, and provision of extension services to farmers of agriculture and livestock in Rwanda. RAB is governed through its board, which is chaired by the Principal Secretary (PS) of the Ministry of Agriculture and Animal Resources. It is accountable to the Ministry of Agriculture and has semi-autonomous status. Address: Rwanda Agriculture Board (RAB), P. O. Box 5016, Kigali, Rwanda. Email: infos@rab.gov.rw

CIFC. The Coffee Rust Research Centre in Portugal has unique facilities for working with coffee diseases, and the location of the CIFC has allowed in-depth research of the CLR and CBD pathogens without the danger of introducing new pathogenic races into coffee growing countries. CIFC was therefore involved in CLR race identification and capacity building of partner institutions in CLR race identification. Address for CIFC: Quinta do Marquês, 2784-505, Oeiras, Portugal. Telephone: + 351 214 544 680/1; Fax: + 351 214 544 689; E-mail: varzea@iict.pt; [ritorvarzea@sapo.pt](mailto: ritorvarzea@sapo.pt)

II. Project components

2.1 Project purpose and indicators

The overall project **purpose** in the logical framework (Appendix 1) was to *reduce economic and environmental costs of disease management for smallholder coffee farmers by reducing the crop and quality losses caused by the diseases CLR and CBD, and resources spent on expensive chemicals, especially in regard to smallholder, coffee-based farming systems.*

The project had 5 components through which the purpose was to be delivered, each component with its own indicators of success. The components are summarised below, and detailed achievements described in Chapter 3.

At the purpose level there were four indicators given.

New technologies and practices are spread outside the project areas

The principle technology developed by the project was coffee varieties resistant to CLR, as well as to CBD. Sections 3.1, 3.2 and 3.3 describe a range of activities and outputs through which this was achieved, including screening for resistance, on-farm trials, establishment of seed production plots and nurseries, and production and distribution of seeds and seedlings. In support of the necessary continual development of new varieties, significant advances were made in understanding the occurrence and distribution of CLR races, identification of genetic markers for use in marker assisted selection, and in building capacity to apply such practices in Africa.

The disease-resistant varieties have been made widely available in the participating countries, with smallholder farmers purchasing seed and/or seedlings. This was achieved through various different communication and dissemination activities targeting farmers within and beyond the project areas. Dissemination activities for promoting uptake of new technologies and practices were part of all project components.

Chemical pesticide sales are reduced while sales of environmentally friendly products increase

A survey of farmers' perceptions and responses to CLR and other diseases indicated that use of pesticides may not have been as widespread as previously assumed. Many farmers in Africa do little or nothing to control CLR, so reducing overall pesticide use was not an appropriate aim. Pesticides were tested for CLR control (Component 4, section 3.3), and of the products tested, the most environmentally friendly (the botanical) was ineffective. A product containing cyproconazole was registered for use in Rwanda, as it was found effective against CLR. Cyproconazole is moderately hazardous according to the WHO pesticide classification based on mammalian toxicity, but from an environmental perspective is lower risk, with relatively low toxicity to birds, fish and bees. However, we expect resistant varieties to become widespread as the main control strategy, reducing the need for fungicides.

Increased volumes of coffee produced by farmers in project area

Farmers in the project areas, particularly those participating in Farmer Field Schools, reported increased production of coffee as a result of what they had learned at the FFS. A study in Kenya found substantial increases in FFS farmers' yields in 3 of 4 districts (Component 1, section 3.1.3). Non-FFS farmers' yields also increased over the same period, but by much lower margins. This may be due to a general increase in yield, but could be as a result of the improved practices adopted by FFS farmers spreading outside the project area. Dissemination activities at FFS included inviting other farmers to open days. As well as increased volumes, farmers also reported increased quality of coffee, and various social benefits.

Disease incidence decreases in project areas

The increase in coffee yields reported from project areas arises from a number of factors including improved management, such as improved soil fertility management and other husbandry practices. This leads to generally healthier plants that are better able to tolerate CLR and other diseases, so we conclude that disease incidence decreased in the project areas. However, a widespread reduction in incidence as a result of the adoption of resistant varieties will take time to occur, once the resistant varieties become more widely grown and their impact on the disease takes effect.

2.2 Project components

The project had five components: (i) Identification of needs and resources: rural community responses to CLR disease and the sourcing and production of coffee genetic materials; (ii) Conservation and identification of coffee varieties and disease races; (iii) Field trials on farm and station; (iv) Scientific management, information systems and communications; and (v) Project management and coordination. The components are summarised here.

2.2.1 Identification of needs and resources: rural community responses to CLR disease and the sourcing and production of coffee genetic material

The objective of the activities under this component was to use best practices in rural extension and action research in order to make the project effective when working with farmers and other stakeholders. Farmers' knowledge and perceptions of CLR and other diseases was ascertained through surveys, highlighting their needs and constraints. Stakeholder analysis was conducted to identify appropriate roles in the coffee innovation system, and Farmer Field Schools (FFS) were utilised as a way of engaging the beneficiaries in the research trials, as well as providing an effective dissemination channel. To increase the supply of recommended planting materials to farmers, nurseries (mother gardens) and seed production sites were established.

2.2.2 Conservation and identification of coffee varieties and disease races

The objective of this component was to source materials for trials, and also to undertake conservation of potentially useful materials in participating countries. Activities improved the conservation of locally collected materials (Kenya and Uganda) and also of those materials collected or introduced in countries during earlier missions and projects.

Countries also carried out identification of coffee leaf rust races, as this knowledge is critical to developing varieties with durable disease resistance. CLR race types for each country were determined, a number of races being found in countries from which they had not previously been reported. As this work needs to be conducted on a continuing basis, scientists from Africa were trained during the race identification work undertaken at the Coffee Rust Research Institute (CIFIC), Portugal.

Facilities were also established at the Central Coffee Research Institute in India for characterisation of germplasm and hybrids in the country. Linked to this was the development of a marker strategy based on purposive samples from which unique DNA markers were isolated, as well as bulk marker profiles using several polymerase chain reaction (PCR) based techniques.

2.2.3 Field trials on farm and station

The objective of this component was to conduct trials to support the development and implementation of cost-effective control strategies. Trials carried out included screening varieties for resistance to CLR and CBD, screening alternative fungicides for efficacy to the diseases, and screening of potential botanicals and biocontrol agents. Varietal trials were designed and planted during the project, so not all had reached full economic yield within the project period. Fungicide, biocontrol and botanical trials were conducted on existing plots of coffee varieties known to be susceptible to coffee leaf rust and coffee berry diseases. Randomised complete block design (RCBD) was used in most countries except Kenya and India where blocks of test varieties were planted in large blocks, with data collected from randomly selected trees within the blocks.

2.2.4 Scientific management, information systems and communications

This component addressed a range of information and communication needs of different stakeholders including researchers, extensionists and farmers. A large number of materials (printed and video) targeting farmers and extensionists were developed, including information on good agriculture practices, resistant varieties, disease management on susceptible coffee varieties, and how to manage coffee in order to increase overall productivity. These materials supported the face-to-face communication at the farmer field schools. An innovative coffee call centre for farmers was also piloted in India, which has expanded since the end of the project.

Scientific information was disseminated through publication in scientific journals and books, as well as through presentations at conferences and symposia. The project hosted annual coffee scientific symposia in association with EAFCA conferences.

2.2.5 Project management and coordination

The project management and coordination component aimed to ensure the project was implemented efficiently, effectively and within budget. This included capacity building for PIAs in administrative and accounting procedures. The Project Executing Agency (PEA) assisted PIAs and the Project Supervising Body, International Coffee Organisation (ICO), with preparation of documentation such as progress reports and the mid-term review.

Project coordination was achieved through annual project planning workshops where work plans and budgets were discussed and agreed. Project implementation was also reviewed at the workshops, discussions held on technical issues, and advice given to project team members as appropriate.

The PEA received funds from CFC, and disbursed them to PIAs against agreed financial reporting procedures. The PEA made consolidated financial reports to CFC. The PEA also ensured annual financial audits were conducted.

III. Project Results Achieved

Results achieved in the project are presented by component. For each component we first provide an overview of the achievements in relation to the indicators of success as in the project logical framework (Appendix I), and then provide details by activity.

3.1 Identification of needs and resources: rural community responses to CLR disease and the sourcing and production of coffee genetic material

The logical framework identified six indicators under this component.

Farmer needs assessments were prepared, the results of which are reported in sections 3.1.1, 3.1.2, and 3.1.3

- Viable rural-based seed/seedling production systems were developed and operationalized in each participating country (sections 3.1.5, 3.1.7 and 3.1.9)
- Seed orchards for mass propagation of high yielding good quality and rust resistant lines were established (sections 3.1.6, 3.1.7 and 3.1.9)
- New networks of coffee sector actors were set up in project areas (sections 3.1.1 and 3.1.3)
- The relative importance to coffee production of CLR, anthracnose & other coffee diseases was clarified and limitations to technology uptake by farmers were identified (section 3.1.2)
- Coffee sector researchers interacted with farmers, extension staff and other actors to generate new approaches, trials and practices responding to needs (sections 3.1.3, 3.1.6, 3.3 and 3.4)

3.1.1 Stakeholder analysis

Workshops with stakeholder representatives were held in each country where a stakeholder analysis was carried out to:

- Identify individuals, groups or organizations that would influence the project
- Determine the roles that stakeholders would play
- Establish stakeholder interests in relation to problems that the project would be seeking to address
- Identify risks that needed to be managed
- Develop strategies to get the most effective support possible for the project and reduce any obstacles to successful implementation

A range of stakeholders were identified in all countries using participatory approaches (Tables 1-4). In all countries farmers, extensionists, institutes of higher learning, players along the coffee value chain, regulators, farm input suppliers and researchers were among the primary stakeholders.

Table 1. List of stakeholders in Rwanda

No.	Broad category	Individual Stakeholders
1	Farmers	Coffee farmers
2	Coffee washing stations	Private people, Cooperatives
3	Coffee cooperatives	Misozi Company, Maraba Coffee, Bufundu Company, COAKAKA
4.	Coffee exporters	RWACOF- Marketing Agents (KCC, KAY CO. Rwanda Mild Coffee (RMC)),
5.	Higher Learning Institutions	National University of Rwanda, Higher Institute of Agriculture and Animal Production (ISAE), FACAGRO, University of Kibungo
6.	Extension (Government of Rwanda [GOR])	Ministry of Agriculture, OCIR CAFÉ, Ministry of commerce, Rwanda Agriculture Development Authority (RADA)
7.	Non-Governmental organizations (NGOs)	SNV, Technoserve, SPREAD (a USAID funded project that improves coffee quality.)
8.	International Organizations	CABI, ICO
9.	Research Institutions	ISAR
10.	Donors	STABEX/EU, USAID, CFC
11.	Certification Programs	UCS, UTZ, CAFÉ PRACTICES
12.	International Buyers	STURBUCKS Regional Office, Japanese Association
13.	Input Suppliers	Agrochemical companies, AGROTECH-SINGETA, Africhem

Table 2. List of stakeholders in Zimbabwe

List of stakeholders for Zimbabwe	
1.	Extensionists: Agricultural Technical and Extension Services (AGRITEX)
2.	Research institutions: Coffee research station, plant protection research institute, Genetic resources institute, Tea Research Foundation, Department of Agricultural Research for Development (DAR4D),
3.	Government Ministries and Government Institutions: Ministry of Agriculture, Ministry of Industry and International Trade, Zimbabwe Revenue Authority (ZIMRA), Agricultural and Rural Development Authority (ARDA), Politicians, Government Departments, Department of Agriculture Regulatory Services
4.	Co-operative societies and Farmer's Unions (representatives of the coffee growers) : Honde Valley Coffee Producers' Co-operative Society, Zimbabwe Commercial Farmers Union (ZCFU), Commercial Farmers Union (CFU), Coffee Growers Association (CGA)
5.	Farmers
6.	Coffee processors and marketers: marketing boards, Zimbabwe Coffee Mill (ZCM), Grain Marketing Board (GMB)
7.	Input suppliers: agrochemical companies, Zimbabwe fertilizer company (ZFC)
8.	Coffee Development Companies: Honde Valley Smallholder Development Company
9.	International Organizations and Donors : International Coffee Organization (ICO), Common Fund for Commodities (CFC), CABI, Foreign Research Institutions
10.	Financial Institutions: banks, funding organizations
11.	Administration/ district authorities/ politicians/ local leaders, etc.

Table 3. List of stakeholders in Uganda

List of stakeholders
1. Farmers
2. Policy making bodies and regulatory bodies [National Agricultural Research Organization (NARO) Council, Uganda Coffee Development Authority (UCDA) council, Top policy management, Ministry of Agriculture Animal Industry and Fisheries (MAAIF)]
3. Extension: (NAADS)
4. Research institutions: NARO, National Crops Resources Research Institute (NaCRRI), Coffee Research Centre (COREC)
5. Government Ministries and Government Institutions: Ministry of Agriculture Animal Industry and Fisheries (MAAIF), Ministry of Finance
6. Co-operative societies and Farmer's Unions (representatives of the coffee growers)
7. Coffee traders: marketing boards, marketers,
8. Environmentalists (NEMA)
9. Coffee consumers
10. Associations (National Union of Coffee Agribusinesses and Farm Enterprises (NUCAFE)
11. Coffee processors (local roasters, star coffees)
12. Input dealers: agrochemical companies
13. International Organizations and Donors : International Coffee Organization (ICO) [supervisory body], Common Fund for Commodities (CFC) [donor], CABI [executing agency], Foreign Research Institutions
14. Funding bodies (Uganda Government, NARO)
15. Local leaders and administrators (district authorities, politicians, etc.)
16. Media
17. Coffee promotion (EAFCA)
18. Credit institutions

Table 4. List of stakeholders in Kenya

Focal Area	Key Stakeholders	Potential Nature of Contribution	Strategy of Engagement
Meru	CRF Substation	Provide trial site Site management	Project to facilitate the officer-in-charge with inputs
	Mukiria Farmers	Farmers mobilization	Involve farmers through the management committee
	MOA-Extension services	Facilitate extension activities	Involve the District Crops officer in the Farmer Field Schools
	Technoserve (NGO)	Currently running a coffee programme in the area	Collaborate
	Meru Central Farmers Union	General Support and Publicity	Keep Union informed through the local co-operative society
	ATC	Possible venue for farmer training sessions	Involve Manager through DAO
Bungoma	Kikai F.C.S Ltd	To provide site for trials Farmer mobilization	Engage Farmers through the Management committee
	MOA- DAO(Bungoma West)	Facilitate/backstop extension activities	Representative to be involved in the Farmer Field Schools
	Coffee Research Foundation - Namwela	Interface with local stakeholders	Involve the farmers in implementation
	Ministry of Cooperative Development	General support	Keep local co-operative officer informed through local co-operative society
Kisii	CRF substation	Provision and management of trial site	Project to facilitate the officer in charge with inputs
	MOA-DAO (Kisii central)	Technical extension support	District Crops officer to be involved in the Farmer Field Schools
	Mobamba F.C.S Ltd	Farmer mobilization	Involve management committee in the FFS

Roles, interests, needs and perceived risks were identified through discussions during the stakeholder analysis, and results are included below for Rwanda, Zimbabwe, Kenya and Uganda for the different stakeholder groups (Table 5-8). Roles included collaboration, information sharing, provision of land, participating in on-farm trials,

training researchers, provision of facilities, mobilising farmers, providing markets for coffee coming from farmers. Analysis of stakeholders in India was similar to the socioeconomic studies in Africa, although emphasis was on the three farmer categories, smallholder, medium and large scale.

Table 5. Stakeholder analysis matrix for Rwanda

Stakeholder	Stakeholder interest (s) in the project	Roles and contribution to project success	Perceived risks
International organizations	(i) Obtaining high production (ii) High quality coffee (iii) Improving livelihoods of small holder farmers	(i) Collaboration (ii) Information sharing (iii) Capacity building	(i) Reluctance of some people towards innovations
Farmers	(i) Acceptance of varieties/ seeds that are resistant to disease, especially coffee leaf rust (CLR) (ii) Obtain cheap means for the control of CLR (iii) Getting inputs at low prices (iv) High coffee output	(i) Provide land, (ii) Provide labour (iii) Participate in on-farm trials	(i) Long drought (ii) Low coffee prices in international markets
Coffee cooperatives	(i) High coffee output (ii) Obtain cheap pesticides for control of CLR	(i). Provide money to farmers for farming (ii). Market the coffee produced	(i). Long drought (ii). Low prices in the international coffee market
NGO (e.g. Technoserve)	Get a sustainable solution to CLR to enable farmers to improve productivity and quality of coffee	Capacity building through farmers training at client cooperatives	Failure of some farmers to receive information
Coffee washing stations	Receive high quality and high quality coffee	Offer training to farmers on good harvesting	Drought
Higher Learning Institutions	(i). Offer training on methods for controlling CLR (ii) prepared thesis, memos and papers from the research project	(i) Avail senior researchers to participate in the project (ii). Supervise students involved in the project (iii) Capacity building	(i). Less funds allocated to these institutions (ii). Lecturers being less committed
Inputs Suppliers	Provision of Products for testing against CLR e.g. systemic products	(i). Technical support (ii). Potential for training (iii). Safe use	(i) Provision of less products for testing (ii). Limited commitment
Extensionists	(i) Availability of information to be disseminated (ii) Availability of seeds resistant to coffee diseases especially Coffee Leaf Rust	(i) Participate in evaluation of all technologies (ii) Scale out the project results (iii) Participate in farmer mobilisation	(i) Less funds allocated to their activities
Coffee exporters	(i) High Coffee production (ii) High quality Coffee (Both coffee attributes resulting from a decrease incidence of diseases especially CLR)	(i) Provision of facilities and capacity for quality testing	(i) Low price in international markets
Research institutions	(i) Collaborate with other institutions/organizations in Research for Development. (ii) Develop technologies for increased farmers' income and improved livelihoods	(i) Participate in implementing the project activities (2) Avail resources for a successful implementation of the project	(i) Project activities may be different from the orientation of Institutions
Donors	(i) Improving livelihoods of poor farmers in southern countries (ii) Information sharing	(i) Provide funds for a successful implementation of the activities (ii) Make follow up so that their money may be used on activities that they have agreed on.	(i) Funds are sometimes stolen or used on activities that are not approved by the donor.
Certification programs	(i) Obtain products without toxic residues	(i) Capacity building in certification	Failure in obtaining

	(ii) Decreased use of chemicals by adopting resistant varieties	matters (ii) Technical support	resistant varieties due for example to a long period of drought.
International buyers	(i) Obtain High Quality coffee (ii) Less use of chemicals and associated toxic residues	(i) Motivate farmers by paying high prices, (ii) Buy all coffee production	(i) Income Decrease being observed in Developed countries

Table 6. Stakeholder analysis matrix for Zimbabwe

Stakeholder	Stakeholder interest(s) in the project	Roles and contribution to project success	Perceived risks
International organizations (CABI, CFC, ICO, etc.)	(i) Increased production, productivity and quality (ii) Improved farmer livelihoods	(i) Funding (ii) Supervision (iii) Execution and control	(i) Failed commitments on counterpart contributions (ii) Inadequate cooperation from collaborators
Extensionists	(i) To help farmers located in the ward/constituency of the extensionists. (ii) Productivity increase (iii) Personal development i.e. training in disease control (iv) Platform for interaction with other stakeholders	(i) Mobilizing and organizing the farmers, data collection (ii) Information dissemination /awareness creation (raising) (iii) Setting up of farmer field schools (group formation). (iv) Farmer training (v) Preparation of project reports and work plans (vi) Project evaluation and feedback	(i) Limited resources e.g. transport and communication. (ii) Protocol (Bureaucracy) (iii) Victims of success whereby good performers are disadvantaged by transfers to different areas
Research Institutions: (i) DAR4D/Coffee Research Stations/ Plant Protection (ii) Researchers	(i) Availability of good germplasm (ii) A comparison of imported germplasm with the local germplasm (iii) Good research trials (iv) Demonstration of results (v) Wide dissemination of information	(i) Research on the disease/ pest resistant varieties (ii) Improve variety productivity (iii) Recommend suitable varieties (iv) Find solutions for control of pests and diseases (v) Recommend import substitutions (vi) Build up germplasm banks for resistant varieties (vii) Research on other alternative and cheap control methods other than chemical (viii) Boost coffee production, quality and marketing	(i) Chances of introducing diseases might arise (ii) Might import infested seeds (iii) Some resistant varieties might not perform well locally leading to low yields e.g. Agaro does well in Malawi but not in Zimbabwe (iv) This might make us neglect our local producing varieties hence leading to loss of germplasm.
Government Institutions	(i) Sound technology (improvement of technology) (ii) High production and quality (iii) Accessible markets (iv) Timely and adequate supply of inputs	(i) research in improved technology (ii) advanced extension (iii) forming marketing authorities (v) consulting fertilizer and seed companies	(i) time consuming in adoption (ii) natural disasters e.g. drought (iii) poor communication
Farmers' co-operatives and Unions	information dissemination and management decrease in coffee losses increased production improved quality profitable marketing disease tolerant varieties	(i) mobilization of the farmers to accept the project (ii) influence political support (iii) Some could contribute some resources to the project e.g. pamphlet generation. (iv) farmer training	(i) may not accept the new technology (ii) May fight for the ownership of the project (ii) political risks (iii) economic risks favouring the status quo e.g. the sale of chemicals

Table 7. Stakeholder analysis matrix for Kenya

Focal area	Key stakeholders	Nature of interest	Strategy of engagement
MERU	District Agriculture Board (DAB)/Provincial Administration	Increased coffee production/ Farmers welfare	To be kept informed
	Political leadership -MPS, Councilors,	Increased coffee production/ farmers' welfare	To be kept informed
BUNGOMA	D.A.C/ Provincial Administration	Increased coffee production/ farmers' welfare	Keep informed
KISII	DAC/Provincial administration/ Ministry of Co-operatives	To ensure farmers interests are cater for	Keep them updated
MACHAKOS	DAC/Provincial Administration	Improved farmers' welfare	To be kept informed
MURANG'A	Provincial Administration	Improved farmers' welfare	To be kept informed
	Ministry of cooperative Development	Improved performance of the cooperatives	Reporting to him

Table 8. Stakeholder analysis for Uganda

Stakeholder	Stakeholder interest(s) in the project	Roles and contribution to project success	Perceived risks
Farmers	Reduced production costs Increased yields Increased income Less environmental hazards Improved health Increased area under production Increased knowledge Reduced child labour Improved livelihood	Offering sites for demonstration Attending sensitization meetings Participating in he practices (planting new varieties) Dissemination of information Have project ownership Multiplication of promising planting materials	Refusal to participate in projects Misunderstanding project role Political instability Theft Weather uncertainties Degree of resistance of improved varieties towards other disease strains Price fluctuation in the global market
Input dealers	Chemical pesticide sales reduced Bio and organic chemical sales increased	Make recommended pesticides available	Campaign against the project
Cooperatives	Increased volume of coffee Improve quality of coffee Increased income Increased dissemination of information on disease resistant varieties of coffee	Strengthen farmer organisations Dissemination of information Supervision of the farmers Monitoring and evaluation of adoption of improved practices Marketing of the product Identification of lead farmers Strengthening marketing according to grade-speciality coffee	Negative attitude and failure to support farmers interested in the project Reduce sustainability of the project in case of no adoption
Coffee traders	Increased coffee volumes Increased quantity Increased sales Increased income Increased livelihood Chemical free products	Source of market for the farmers products Sensitization of the farmers on chemical free products Seek speciality coffee markets	Price manipulation Adulteration of the products Lack of price incentive in relation to quality

3.1.2 Community-based surveys

Socioeconomic surveys were carried out to:

- Understand farmers' perceptions of coffee leaf rust (CLR), coffee berry disease (CBD), and their severity in relation to other production constraints and broader problems such as climate change
- Identify methods used by farmers to control the diseases, including any knowledge of resistant varieties
- Identify needs and resources for increased coffee production, including possibilities for trialling new varieties
- Document expected demand for improved coffee varieties
- Assess options for promoting sustainability, and plan an exit strategy for the project.

The socioeconomic surveys were carried out by CABI and country socioeconomists in the four African countries. A consultant was hired in India to carry out the socioeconomic survey there. Questionnaire surveys, key information interviews and focus group discussions were used.

3.1.2.1 Understand farmers' perceptions of CLR/CBD diseases

Farmer ranking of CLR as a constraint to coffee production

Most farmers were aware of the importance of CLR, and ranked it as the most important constraint to coffee production. In Uganda, farmers ranked CLR as the most important constraint, while aging coffee trees and CBD ranked second and third, respectively (Table 9). In Kenya farmers also ranked CLR as the most important problem, followed by CBD. In Zimbabwe farmers noted that lack of fertilizer and pesticides were the most important constraints, with CLR ranked third.

Thus farmers in all coffee producing areas in Africa recognized the importance of CLR in coffee production, although it is by no means the only serious problem they face. In contrast, over 90% of Indian farmers did not feel CLR caused any economic impact, which could be due to existence of resistant or tolerant varieties. However, the same farmers stated that timely information on management of CLR is important, implying that they indirectly recognise the importance of CLR.

Other biotic constraints to production identified by farmers included a range of insect pests (white coffee stem borer, antestia bug, and berry borer insect pests), and several other diseases (black rot, root rot, coffee trunk canker, berry blight, and Fusarium bark disease). Other less frequently mentioned constraints included coffee theft, transportation bottlenecks, and coffee marketing problems including low prices.

Knowledge on symptoms of CLR

Smallholder farmers were able to recognize some of the CLR symptoms, with leaf symptoms being most easily recognizable. The majority of farmers stated that CLR causes premature shedding of leaves (83%) (Table 10). Interestingly, farmers reported

that the disease is more severe during the dry season (76.3%) and under un-shaded conditions (70.5%). However, farmers felt they needed more information on CLR, which the project subsequently provided.

Trend of disease occurrence and mode of transmission

Some farmers had some knowledge about the biology and epidemiology of the disease. About half of interviewed farmers in Uganda noted that CLR was increasing (48.6%), while in India there was no consensus on whether it is increasing or decreasing. In general many farmers were unaware of CLR trends in surveyed countries. Just below half of the interviewed farmers knew that CLR is transmitted through wind (Table 10). Many farmers did not know of other ways in which CLR is transmitted, which is not surprising.

Varietal susceptibility to CLR on the farm

Farmers growing the susceptible variety SL28 were not able to control CLR. However, in Kenya a number of farmers were aware of the variety Ruiru 11, which is resistant to CLR and CBD. In India, most farmers (94 % in Karnataka, 100 per cent in Kerala and Tamil Nadu) considered use of resistant varieties as the most effective control measure for CLR. The major sources of advice on the use of CLR-resistant varieties included fellow farmers, extension workers and television in all the states and districts of India. In Uganda, farmers were less clear regarding the use of resistant varieties (Table 10). They gave the local variety as least affected by CLR, which is not correct. Although resistant varieties are an economical method for managing CLR, many of the African countries do not have resistant varieties, which is why the farmers are less aware than in India.

Table 9. Constraints in coffee production as recorded in Uganda

Crop/enterprise	Average Ranking at the time of the study
CLR	1
Aging trees	2
CBD	3
Counterfeit inputs and equipment	4
Climatic change	5
Insect pests	6
Declining soil fertility	7
Poor extension services	8

Table 10. Farmers' perception of coffee leaf rust disease as recorded in Uganda

Variable	N	% farmers reporting
Knowledge on symptoms of CLR		
Premature shedding of leaves	147	83.8
Leaves turn to pale yellow/ orange/ rust/ golden colour at the lower surface	125	72.3
White powdery mass on leaves	72	41.6
Drying of coffee tree/bush	25	14.5
Coffee berries fail to ripe	20	11.5
Trend of disease occurrence over the years		
Increasing	84	48.6
Constant	61	35.3
Decreasing	11	6.4
Mode of transmission for CLR		
Wind	45	26.0
Infected plant to non-infected garden	35	20.2
Human and animals activity	15	8.7
Water run off	6	3.5
Field conditions in which CLR is more severe		
Un shaded coffee trees	122	70.5
Unweeded coffee garden	101	58.4
Unpruned coffee	93	53.8
Less fertile parts of the farm	91	52.6
Season of the year when CLR is more severe		
Dry season	132	76.3
Rain/ Wet season	11	6.4
No difference (all seasons throughout the year)	6	2.9
Varieties most affected by CLR on the farm		
Improved variety(mainly SL 14)	89	51.4
Local/ traditional variety	30	17.3
Varieties least affected by CLR on the farm		
local variety	81	46.8
improved variety(mainly SL 14)	7	4.0

N= Total number of observations

3.1.2.2 Identify methods used by farmers to control the diseases

Despite recognising CLR as a serious constraint, most farmers do little or nothing to control it, or other pests and diseases. This could be due to poverty - in India many smallholder farmers (about 10 ha of coffee) were also receiving government rations. However, some farmers in India and Kenya were able to grow varieties tolerant or resistant to CLR and other diseases (eg CBD in Kenya), for example Ruiru 11 in Kenya, and Chandagri in India. The majority who were growing susceptible varieties such as SL28 were not able to control CLR and other diseases. Interestingly, farmers were aware of chemical control (copper oxychloride and *Bordeaux* mixture).

Other methods used by farmers for the control of CLR and other diseases included weeding (81%) and use of fertilizer (21%), increased plant spacing, and use of concoctions in Uganda. However, all this may not work directly in controlling CLR, but weeding and use of fertiliser can make the coffee bush grow more vigorously, thus making it better able to withstand CLR and other diseases. It is not known what concoctions farmers in Uganda apply, and whether they work or not in controlling CLR. In addition, farmers in India (35% to 75% depending on the region) also use replanting as a mitigation strategy for CLR when trees are badly affected. In Zimbabwe a significant number of farmers (55% in Ngarura, 61% in Chiteme and 30% in Samanga-

Dumba) reported that they were not practicing any control against CLR, explaining that their crops produced low yields once affected by CLR, and that there was nothing that could be done about it.

3.1.2.3 Identify needs/resources for increased coffee production

In general there was a limited use of farm inputs, resulting from limited resources among smallholder farmers, which was clearly evidenced by the poor state of coffee bushes in the majority of surveyed countries. Poor quality and unavailability of chemical inputs is a great constraint to coffee production, especially in India. The high cost of the recommended practices was also one of the main constraints to effective management in all countries.

Resistant varieties are the most cost effective, sustainable and environmentally friendly way of controlling CLR, CBD and other diseases. However, availability of CLR and CBD resistant coffee varieties is limited in most countries. Rwanda and Uganda for example have no resistant varieties. Kenya and India have resistant/tolerant varieties, but accessibility is a problem, for example Ruiru 11 in Kenya, and selections 795, 6, and 9, and Chandragiri are resistant/tolerant to CLR in India. In conclusion, the main need for improved production is availability of resistant varieties, environmentally friendly fungicides for the control of CLR and CBD, and other improved coffee production technologies which contribute to general coffee plant health. The technologies need to be made available at the time that farmers need them, and as far as possible, at prices they can afford. In parallel, farmers need information of using the technologies.

3.1.2.4 Assess sustainability and plan exit strategy after the project

Options for sustainability were assessed in each country, and stakeholders agreed that activities started by the project would need to be sustained mainly by the government research and extension institutions leading in-country activities. The project adopted the following exit strategies.

Capacity building

This was carried out by sponsoring members of staff from the identified institutions for PhD, MSc, BSc and attachments to other institutions such as CIFC in Portugal. Capacity was built in specialist techniques that will support coffee breeding programmes in the future, such as marker assisted selection and CLR race characterisation. The members of staff studied subjects related to coffee science, for example plant breeding, plant pathology, and extension, while carrying out research and practical training. See section 3.2.5 for details.

Farmer training activities

The project started by training the farmer field school trainers and facilitators from extension departments, and these skills can be used beyond the life of the project. FFS are known to empower farmers in information seeking behaviours, which means they

are able to apply skills to other problems as they occur. See section 3.1.3 for further details.

Organisation of the project

The project targeted relevant government institutions identified during the stakeholder analysis and brought them on board in order to have a proper takeover/exit strategy at the end of the project. These included government coffee research, extension, coffee regulators/coffee boards/national coffee authorities

Co-financing

The project funding included co-financing from participating institutions. By ensuring institutions invest their own funds, time, infrastructure and other resources into activities during the project, commitment is more likely to continue after the project, as regular budget allocation has already been occurring. This strategy was particularly relevant for activities such as breeding and varietal screening.

Supporting building or renovation of structures

Another element of the exit strategy was the provision of the infrastructure needed for effective use of the enhanced human capacity. Nurseries in all participating countries were renovated so that they can continue producing seedlings of resistant and other commercial varieties beyond the life of the project. In addition, the project supported renovation and equipping of a modern molecular laboratory in India. A well-equipped seed laboratory was also constructed at the Central Coffee Research Institute, India. Seed gardens were developed for old and new varieties thus developing capacity for continued supply of high quality seed for the resistant and other commercial varieties. In cases such as these, recurrent costs are much lower than the initial set-up costs.

3.1.3 Mobilise key stakeholders, including farmers to participate in farm-based activities.

Mobilisation of stakeholders was done at two levels. Workshops were held in all countries, involving the major stakeholder representatives (mostly extensionists, researchers and farmers), at which the project activities under each component were presented and discussed. Country specific plans were drawn up at such meetings. Training of trainers' workshops were held to train Farmer Field School (FFS) facilitators in all participating countries. The second approach concentrated on farmers and facilitators (extensionists) who were mobilised through FFSs and other participatory approaches such as open days at which farmers themselves explained to visitors the different technologies they were 'learning-by-doing'.

Other farmer mobilisation activities included farmer exchange visits between FFSs. In addition, farmers were also mobilised to participate in on-farm varietal trials (screening varieties for resistance to CLR and CBD, and evaluation of fungicides and botanicals). In workshop type meetings with farmers, facilitators helped farmers to agree the person they would like to host the trials. Normally it was a person most people felt free to visit, a

person with coffee to use or land to plant the varietal trials, and a person willing to host other farmers and visitors. All this was done just before starting FFSs, and resulted in getting a buy-in from the main stakeholders and end users of technologies, the farmers.

At FFSs farmers were able to learn by doing. FFSs provided an opportunity for farmers to learn new and existing recommended technologies. According to farmers FFSs provided them an opportunity to increase their knowledge of coffee farming, including in India where most farmers already have good knowledge of coffee farming. They felt that an improvement in their coffee plots was seen after adopting and employing recommended technologies learnt through FFSs. This was clearly expressed during the mid-term review mission of the project in India and Africa.

Table 11. Meetings that were held at FFSs during the Project Period in Kenya

	Bungoma	Kisii	Muran g'a	Machakos	Meru
2009	11	6	11	11	9
2010	8	8	10	10	7
2011	6	6	7	7	5
2012	3	3	3	3	2
Total	28	23	31	31	23

Table 12. Farmer field schools in Rwanda

Name	Numbers		Starting date	Location (District)	Plot size (number of trees)
	male	female			
Abakoraningoga	26	4	23/06/2009	Kirehe	700
Abakundamurimo	27	3	24/7/2009	Rutsiro	176
Duhugukire kawa	21	9	25/8/2009	Nyanza	400
Abakundakawa	23	7	24/10/2009	Nyamasheke	300
Abanozamusaruro	23	7	12/4/2009	Gakenke	300
Abakoranabuhanga	24	6	21/4/2009	Gicumbi	400

3.1.3.1 Exchange visits and open days

Exchange visits were carried out between FFSs to allow farmers to share and learn from each other. One or several FFSs or farmer groups visited another FFS where the hosting members explained to the visitors the practices and technologies they were using and testing. Visitor asked questions and points of clarification, and also shared their own experiences. Open days mainly targeted non-FFS farmers to come and visit the FFS and learn from their fellow farmers. Where possible a guest of honour was invited such as a local chief or even a senior government official (Table 13).

Table 13. Open days held in Kenya

Location	Date Held	Guest of Honour	Subjects covered
Machakos	5/5/2011		Coffee establishment, top working, nutrition, pruning, pest and diseases
Bungoma	5/17/2011	Area Chief	Coffee establishment, top working, nutrition, pruning, pest and diseases
Meru	20/09/2012		Coffee establishment, top working, nutrition, pruning, pest and diseases

In Zimbabwe, three FFS open days and exchange visits were held during the course of the project on a rotational basis, once per year in each of the three participating districts. The open days were held in Chimanimani (2009) with 3 farmer and 3 extensionist representatives from Chipinge and 22 farmers and eight extensionists from Honde Valley attending, together with many other key stakeholders in the coffee industry. The second was in Honde Valley (2010), while the third open day and exchange visit was held in Chipinge (2011). Representatives from the Chimanimani and Mutasa FFSs attended. Stakeholders who attended the occasion included extension department (AGRITEX), representatives from two NGOs (ACTION FAIM and Medicines Du Monde), COTIBA (Coffee Tea and Banana project), Zimbabwe Farmers' Union (ZFU), Coffee Research Institute Staff, local traditional and political leadership (Kraal head and councillor), and a primary school (staff and pupils). Approximately 80 people attended each open day.

Another farmer mobilisation activity was the holding of FFS members' "get-togethers", social events at which a visiting dignitary (such as the CRF Deputy Director) was guest of honour. These meetings were funded by the FFS members themselves, and they helped create external linkages and build social capital.

A range of achievement and impacts were realized from Farmer Field Schools and other mobilization activities. Farmers were able to implement and adopt existing and new recommendations and technologies for coffee such as fertilizer and fungicide application. An example is presented in Table 14 below, which shows a general marked yield increase for FFS farmers. The yield data in Table 14 were obtained from coffee cooperative societies, who keep production figures.

Table 14. Yield benefits among FFS farmers compared to non-FFS farmers in Kenya

Area	Average coffee productivity (kg/tree)		
	Average Yield FFS farmers	Average Yield non-FFS farmers	Average Yield before project (all farmers)
Murang'a	3.54	0.72	0.14
Meru	4.27	0.44	0.35
Kisi	0.23	0.22	0.19
Bungoma	1.49	0.52	0.21

Farmers interviewed by the project team in Kenya identified a range of benefits from FFS (although this was not a structured study):

- Enhanced knowledge of improved technologies for coffee production
- Improved adoption of new technologies
- Increased empowerment of women
- Acquisition of skills on GAP
- Farmers from FFS facilitated in creating new FFSs
- Quality of coffee in FFS members' farms improved. For example in Kenya's Kabati FFS coffee quality improved from class 7 to class 4

3.1.4 Identify locally available genetic material for trial

At the beginning of the project, two hybrids bred in India, Selection (SIn) 5A and SIn 6 from crosses involving diverse sources of resistance were selected for evaluation in trials in India and Africa. In Africa, local genotypes were also identified and included either as local checks or potential sources of resistance. India included variety S.795 at the beginning of the project in 2008; however, they also added a new variety, Chandragiri in the evaluation trials. Kenya included its new crosses which were later officially released as Bati variety after screening during the project. African countries also included their commercial varieties such as SL 28 in Kenya, Bugisu Local in Uganda, BM in Rwanda and Catimor 129 in Zimbabwe. A total of 31 locally available genotypes were used in trials (Table 15).

Table 15. Locally available genetic materials identified for inclusion in trials

No	Variety	Country
1	SL 28	Zimbabwe
2	Yellow Catuai	Zimbabwe
3	Catimor 128	Zimbabwe
4	Catimor 129	Zimbabwe
5	Catimor F6	Zimbabwe
6	K 7	Zimbabwe
7	KP423	Uganda
8	SL28	Uganda
9	NG9257	Uganda
10	SL34	Uganda
11	SL14	Uganda
12	Elgon CB	Uganda
13	SIn.5A	India
14	SIn.5B	India
15	SIn.6	India
16	Chandragiri	India
17	Columbian Catimor	India
18	BBTC Catimor	India
19	Catuai x HDT	India
20	CR27	Kenya
21	CR30	Kenya
22	CR8	Kenya
23	CR22	Kenya
24	CR23	Kenya
25	R11	Kenya
26	SL 28	Kenya
27	BM 139	Rwanda
28	Jackson	Rwanda
29	Harar	Rwanda
30	Selection 5a	Rwanda
31	Selection 6	Rwanda

3.1.5 Manage and expand seed production sites in India for supply of F1 material for trials

In India, after the release of the new variety ‘Chandragiri’ in 2007, a decision was taken to establish new seed plots to meet the growing demand. An action plan was prepared to establish new seed blocks in Research Farms and Technology Evaluation centres to ensure self-sufficiency by 2012. At the beginning of the project in 2008, 6 ha of seed garden were available under nucleus seed plots of the improved varieties SIn.5A, SIn.5B, SIn.6, SIn.9, Chandragiri, in Research Farms and Technology Evaluation Centres located in different coffee growing areas. These nucleus seed plots were maintained and seed of selected varieties was supplied from these for varietal trials both in India and in Africa. A further 19.05 ha of new seed garden was established in addition to maintaining and improving the existing seed gardens. A total of 15,933 farmers benefited from the seed produced in the seed plots (Table 16). Seed from the plots was also used in trials in India, and sent to Africa for research. A total of 25,025 kg of seed were harvested during the project period in India (Table 17).

Table 16. Number of farmers receiving seed in Karnataka and Tamil Nadu

Year	Number of farmers receiving seed		
	Karnataka	Tamil Nadu	Total
2007-2008	1532	389	1921
2008-2009	1708	623	2331
2009-2010	2708	726	3434
2010-2011	2078	696	2774
2011-2012	4663	810	5473
Total	12,689	3,244	15, 933

Table 17. Amount of seed produced from seed gardens in India

Year	Chandragiri (kg)	Other Arabica varieties (kg)	Annual totals for Arabica seed (Kg)
2008-09	655	2,980	3,635
2009-10	1,673	2,264	3,937
2010-11	2,178	1,588	3,766
2011-12	4,439	2,515	6,954
2012-13	4,450	2,283	6,733
Totals	13,395	11,630	25,025

Selections 5A and 6 were used in trials in India and Africa. The seed plots were at Coffee Board’s research centres in all major coffee growing areas, especially in Karnataka and Tamil Nadu regions. Each country in Africa received 5 kg of seed of each selection, although Uganda received 4 kg of Selection 5A after the original seed was lost in transit. In addition, Zimbabwe requested for more seed for planting in seed plots and the field gene bank. In India, seed from seed production sites was also planted in trials in different areas, and on farmers’ fields.

Although this activity was originally planned only for India, some of the African countries, such as Kenya and Zimbabwe, also managed their seed gardens in order to make more seed available to farmers. Zimbabwe managed to produce 89,721 seedlings of 8 coffee varieties (Selection 5A, Selection 6, SL28, Yellow Catuai, K7, Catimor 129, Catimor 128 and Catimor F6) during the project period. Furthermore, homogenous seed plots were developed for existing resistant varieties in Kenya – the strategy involved establishing seed plots next to the trials for varietal resistance. Uganda maintained

homogenous seed plots of the commercial Arabica seed plots only. The contribution to developing mother gardens of Coffee Wilt Disease resistant clones is discussed in Section 3.1.7 below.

An example from Kenya of seed produced from seed gardens managed with the support of the project is presented in Table 18. Although most of the seed gardens were established before the commencement of the project, seed production from the seed gardens established earlier started in 2010 after the release of Batian variety. The project assisted in establishing the Koru seed garden and in maintaining the other gardens. A total of 2,993 kg from 2010 to 2012 were harvested from four seed gardens in Kenya.

Table 18. Seed production in Kenya from 2010 to 2012

Site	Planting date	No of trees	Harvested seeds
CRS	2004	968	834 kg
Mariene	2005	1067	615 kg
Kitale	2004	1024	530 kg
Koru	2009	1256	1014 kg
Kisii	2005	1092	Not harvested*
Total weight of harvested seed			2,993 kg

*Did not harvest seed because of outbreak of Bacterial Blight of Coffee, a bacterial disease

In Rwanda two sites were established and managed for seed production at research stations (Rubona and Ntendezi). Two tonnes of seeds for released coffee varieties were provided to Rwanda Coffee Board (the former OCIR CAFÉ, now the National Agriculture Export Board (NAEB)) for distribution to farmers.

3.1.6 Introduce new material, subject to quarantine and IP provisions

Two Indian coffee hybrids, Selections 5A and 6, were introduced to Kenya, Rwanda, Uganda and Zimbabwe for evaluation against coffee leaf rust and coffee berry diseases. The varieties were included in trials with local varieties including commercial varieties. No new varieties were introduced to India. Participating countries which imported the selections from India used existing quarantine and IP provisions, which included applying for permits to import from India, and complying with the conditions contained within the permits.

The import permits stated how the seed should be treated before sending to the country, such as treating with systemic fungicide. India then applied for a phytosanitary certificate from the Indian authorities which clearly spelt out quantities of seed for each coffee selection, and what was applied to the seed to control diseases. Both documents were packed together with the seed. Upon arrival at the importing country's airport, quarantine authorities went through the seed packages and checked to see if they conformed to the quarantine regulations, and to confirm that what was stated in the permit was adhered to as per the phytosanitary certificate. Some samples of seeds were tested for pathogens.

Initially, there was a proposal for introducing new varieties from collaborating countries in Africa to India. Subsequently, considering the risk involved due to the presence of CBD in all the African countries, India decided not to import new materials from Africa.

3.1.7 Support and develop the nursery production facilities (mother gardens)

Nurseries were developed or rehabilitated in all participating countries. In Rwanda and Zimbabwe new nurseries were established for raising seedlings for the trials and for supplying seedlings to farmers. The nurseries in Africa were also used for raising seedlings for trials, gene banks and seed gardens, and included varieties introduced from India (Selections 5A and 6) and commercial varieties existing in countries.

In Uganda, coffee wilt resistant clones were propagated through tissue culture, and hardened seedlings were made available to private nursery operators to develop mother gardens of the clones. The resistant clones were developed by the previous CFC funded the Regional Coffee Wilt Programme. Mother gardens, once mature, will be a source of cuttings which will be raised in nurseries for distribution to farmers. The project only financially supported renovation of nurseries at government nursery sites, not the private ones.

In Kenya, Coffee Research Foundation (CRF) nurseries were upgraded during the project period with counterpart contribution. The nursery upgrading resulted in increased annual seedling production during the project period (2008 to 2012) at the different sites as follows:

- Ruiru – Increased from 200,000 to 400,000 seedlings per year
- Koru – Increased from 10,000 to 150,000 seedlings per year
- Mariene – Increased from 15,000 to 80,000 seedlings per year
- Namwela – Increased from 0 to 150,000 seedlings per year

Uganda generated 13,000 plantlets of the 7 coffee wilt disease resistant Robusta coffee varieties/clones, which were distributed to 43 nursery operators. India does not raise seedlings for farmers; however, seedlings were raised for on-station, on-farm, and multilocational field trials with new breeding lines. Nursery production sites were therefore developed at Central Coffee Research Institute (CCRI), Coffee Research Sub Stations (CRSSs), Chettalli and Thandigudi with a capacity of 10,000 seedlings per year. A total of 13,500 seedlings were raised for trials until 2010 season. In 2011, 13,250 seedlings of station-bred selections and 5000 seedlings of 34 new breeding lines were raised. Additionally 20,000 seedlings of Chandragiri variety were raised at Chettalli. In 2012 season, 6,000 seedlings of various genotypes covering 15 F₁ progenies, 5 F₂ progenies and 11 parental lines were raised and supplied for trials. In addition, 2,000 seedlings of Chandragiri were raised for planting at Koraput for establishment of a new seed block.

In Zimbabwe, three coffee nurseries, at Coffee Research Institute Chipinge (CoRI), Honde Valley Smallholder Development Company, and Piringani, were supported. One new nursery site was developed at ARDA Katiyo in Honde Valley. A total of 89,721 seedlings of 8 coffee varieties (Selection 5A, Selection 6, SL28, Yellow Catuai, K7, Catimor 129, Catimor 128 and Catimor F6), were produced during the project period. Seedlings were used in trials and for distribution to farmers.

3.1.8 Document expected demand for improved varieties

From the surveys it was apparent from all countries that farmers desire improved varieties for management of CLR, CBD, and other diseases, and for general improved production and productivity. In Zimbabwe farmers were willing to try new coffee varieties that were bred for resistance to CLR and other diseases, although they were wary of the fact that performance of the improved varieties may be affected by challenges in procuring inputs, e.g. fertilizers. In Uganda, demand for improved varieties was high; 86% of interviewed farmers were interested in getting new improved varieties.

In Kenya and India demand for improved varieties was quantified through direct requests for planting materials and seed of improved varieties (Tables 19 and 20).

Table 19. Demand for improved coffee varieties in Zimbabwe, Kenya and India

Season	Zimbabwe	Kenya		India	Comments
	Number of seedlings /equivalent amount of seed (Kg)	Variety	% demand	Amount of seed produced (Kg)	
2009/10	410,000 seedlings or 102.5Kg seed				Documented once for Zimbabwe
2011/12	Demand not documented	Batian	78%	7,500	Over 70% of the seed demand accounts for the new improved Chandragiri variety in India
		Ruiru 11	21%		
		SL 28 (not improved)	1%		
2012/13	Demand not documented	Batian	78%	6,500	65% was for Chandragiri in India
		Ruiru 11	19%		
		SL 28	3%		

Table 20. Demand for improved and traditional varieties in Kenya

Crop Year	Production/ demand/ distribution/ supplied	Improved Varieties (kg)	Traditional Varieties (kg)
2008/09	Production	346,590	170,000
	Demand	401,145	89,818
	Distribution	340,000	89,818
2009/10	Production	315,540	163,600
	Demand	274,397	59,580
	Distribution	274,397	59,580
2010/11	Production	466,180	116,700
	Demand	2,518,585	19,262
	Distribution	460,000	19,262
2011/12	Production	743,474	100,462
	Demand	8,350,047	49,722
	Distribution	743,474	49,722

Demand for seed of improved varieties in Kenya shot up in 2010 and 2011 (Table 20) due to the official release and official launch of Batian varieties which was presided over by the Minister of Agriculture at CRF. The mass media (TV and print), covered the function which resulted in creation of awareness for the improved varieties over the whole country. This suddenly raised the demand for these varieties to millions of kilograms of seed, and CRF could not cope. Although susceptible to CLR and CBD, seed for traditional varieties was still produced as some farmers still demand it due to its high cup quality. Seed gardens of the traditional varieties were established many years ago, and are continuously producing seed. Although production exceeds demand, Table 20 shows that production of traditional varieties is decreasing due to the decreased demand.

3.1.9 Develop homogenous seed production sites for future supply of genotypes for later release, and facilitate the establishment of efficient seed/seedling distribution networks.

This activity included adequate supply of planting material for initial and subsequent field trials. This required planning, quarantine and import of foreign material, and establishment of seed plots for bulk provision of seeds by the end of the project. Initial trials were limited to evaluation of materials present in country and imported (Africa). Development of homogenous sites for future supply of genotypes has been discussed fully under importation of seed; development and management of seed gardens, and improvement or development of nurseries (see sections 3.1.4 to 3.1.5).

However, as a strategy for making seed of improved varieties available, homogenous seed production sites were developed in all areas where varieties were being evaluated. This was a difficult arrangement for some countries since they were not sure which varieties would give good results in trials. In order to overcome this, all varieties being evaluated were planted out in seed gardens.

Seed and seedling distribution networks were already available in participating countries, but the project helped in strengthening them by establishing seed gardens in main coffee growing areas where trials were sited. In Uganda, strengthening of the seed distribution networks included providing CWD resistant Robusta plantlets to the commercial nurseries, in addition to establishing Arabica coffee seed gardens.

3.2 Conservation and identification of coffee varieties and disease races

Four indicators were identified in the logical framework for this component.

- Attributes and conditions of germplasm collections were documented, and material was sampled and conserved as appropriate, as reported in sections 3.2.1 and 3.2.2.

- Field & laboratory facilities were identified, enhanced and utilised in participating institutions & countries (sections 3.2.1, 3.2.2, 3.2.3, 3.2.4 and 3.2.5).
- Genetic/biochemical marker assisted protocols were developed by identifying markers associated with disease resistance, and the skills and knowledge of national scientists were developed through short courses and postgraduate degrees (sections 3.2.3, 3.2.4 and 3.2.5).
- Instruments for sharing coffee germplasm between India and partners in Africa were formulated and used (section 3.2.6).

3.2.1. Identify the conservation status of coffee collections

The conservation status of many coffee collections was found to be poor, and most of the collections needed rehabilitation. In Uganda, the collections had to be propagated and transferred to a safer place due to city expansion.

In Zimbabwe, there was only one field gene bank at Rupinda in Honde Valley. As a result the country had to re-establish field gene banks at Coffee Research Institute, Crake Valley farm, Marimbita farm, Eastern Highlands Estate and Piringani farm. The two Indian hybrids as well as local varieties, SL28, Catimor 129, Catimor F6, K7, Yellow Catuai and Costa Rica were planted and managed in the gene banks during the project period. An inventory of coffee germplasm in Zimbabwe is in Tables 21-23 below.

Table 21. Giant coffee varieties (>3m height) in Zimbabwe

Variety	Details
SL28	<ul style="list-style-type: none"> • Origin: Single tree selection from Tanzania • Source: Tanzania • Year introduced: 1970 • Growth habit: Tall and open • Growing conditions: Tolerant to cool dry conditions • Leaf size: Large • Terminal leaf colour: Copper-coloured • Maturity: Late • Bean size: Large • Yield potential: Very high • Cup quality: Very good, able to produce class 1 • Disease resistance: Susceptible to Coffee Leaf Rust (CLR), Coffee Berry Disease (CBD) & Fusarium Bark Disease (FBD)
K7	<ul style="list-style-type: none"> • Origin: Single tree selection from Kenya • Source: Kenya • Year introduced: 1970 • Growth habit: Tall and open with spreading habit and drooping primaries • Growing conditions: Requires harsher conditions to realise its full bearing potential • Growing conditions: Low altitude, dry conditions • Leaf size: Medium narrow leaves. More vegetative than SL28 Terminal leaf • colour: Bronze-coloured • Maturity: Late • Bean size: Large • Yield potential: Good • Cup quality: Fair • Disease resistance: Tolerant to several races of leaf rust.
Mundo nova	<ul style="list-style-type: none"> • Origin: Natural hybrid between Typica and Bourbon coffee from Brazil • Source: Brazil • Year introduced: 1977

	<ul style="list-style-type: none"> • Growth habit • Drought tolerance • Growing conditions • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Tall, open variety Tolerant 1 000-1600m with an annual rainfall of 1,200-1,800 mm Large Green Medium to late Medium High Fair Susceptible to CLR & CB</p>
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Table 22. Medium size coffee varieties (2–3m height) in Zimbabwe

Variety	Details	
Catimor 128	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Growth habit • Growing conditions • Drought tolerance • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Cross between Hybrid of Timor and Caturra from Colombia Portugal 1982 Semi dwarf, fairly open and rather untidy Requires irrigation Susceptible Large Green Early Large Good Good Resistant to CLR & CBD</p>
Catimor 129	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Growth habit • Growing conditions • Drought tolerance • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Cross between Hybrid of Timor and Caturra from Colombia Portugal 1982 Semi dwarf, fairly open and rather untidy Requires irrigation Susceptible Larger than Catimor 128 Green Early Larger than Catimor 128 Good Good Resistant to CLR & CBD</p>
Ruiru 11	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Growth habit • Growing conditions • Drought tolerance • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Kenya Kenya 1990s Medium sized and fairly open High nutrient and water requirements Tolerant Small Green Early Large High Fair Resistant to CLR & CBD</p>
Yellow Catuai	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Growth habit • Growing conditions • Drought tolerance • Leaf size • Terminal leaf colour • Maturity • Bean size and colour • Yield potential 	<p>Hybrid of Mundo novo and Caturra from Brazil Kenya 1977 Semi-dwarf and fairly open Suitable under harsh conditions Tolerant Small Green Late Small and yellow when ripe High</p>

	<ul style="list-style-type: none"> • Cup quality • Disease resistance 	<p>Fair Susceptible to CLR, CBD & FBD</p>
Red Catuai	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Growth habit • Growing conditions • Drought tolerance • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Hybrid of Mundo novo and Caturra from Brazil Kenya 1977 Semi-dwarf and fairly open Survives under harsh conditions Tolerant Small Green Late Small and red when ripe High Fair Susceptible to CLR, CBD & FBD</p>
Costa Rica 95	<ul style="list-style-type: none"> • Origin • Source • Growth habit • Drought tolerance • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Costa Rica Kenya Semi-dwarf, highly vegetative variety Tolerant Small Bronze Late Small Fair Fair Fairly tolerant to CLR & CBD</p>

Table 23. Dwarf coffee varieties (1.5 – 2m height) in Zimbabwe

Variety	Details	
Caturra	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Growth habit • Drought tolerance • Growing conditions • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Bourbon mutation from Brazil Kenya 1970 Short with a thick core, many secondary branches and dense leaves Tolerant Grows best at 500-1 600m with 2,500-3,500 mm per annum Large leaves with wavy borders similar to Bourbon Green Medium Small beans of lower quality High, decreases with increase in altitude Fair quality, increases with altitude Susceptible to CLR, CBD & FBD</p>
Catimor F6	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Growth habit • Growing conditions • Drought tolerance • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Cross between Hybrid of Timor and Caturra from Colombia Portugal 1982 Dwarf, fairly open Does well in high altitudes and rough conditions Tolerant Small Green Early Small High Fair Tolerant to CLR & CBD</p>
Catimor F4	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Growth habit • Growing conditions • Drought tolerance • Leaf size • Terminal leaf colour 	<p>Cross between Hybrid of Timor and Caturra from Colombia Portugal 1982 Dwarf, fairly open Does well in high altitudes and rough conditions Tolerant Smaller than F6 Green</p>

	<ul style="list-style-type: none"> • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Early Smaller than F6 High Fair Tolerant to CLR & CBD</p>
Icatu	<ul style="list-style-type: none"> • Origin • Source • Year introduced • Reproduction • Growth habit • Growing conditions • Drought tolerance • Leaf size • Terminal leaf colour • Maturity • Bean size • Yield potential • Cup quality • Disease resistance 	<p>Brazil Brazil 1970 Cross between Hybrid of Timor and Caturra Dwarf, fairly open Suitable for rough conditions Tolerant Small Green Early Small Low Fair Susceptible to CLR, CBD & FBD</p>

In Uganda the collections of germplasm needed rehabilitation and transfer. The collections were in dire need of proper agronomic management practices and were under threat because of urbanization (expansion of the City of Kampala). A total of 120 out of 135 Arabica coffee lines in the Kawanda field gene bank were propagated and replanted at Kituza and Bugusege (Table 24). In addition, re-characterisation of the collections started during the project.

Table 24. Germplasm collections transferred from Kawanda to Kituza, Uganda

Germplasm transferred from Kawanda	Origin	Main features
160/26/2	unknown	Unknown
77/6A	unknown	Unknown
Blue mountain	Jamaica	Tall cultivar, Good cup quality, low yielding, fairly susceptible to rust
Bugisu local	Nyasaland/Malawi	Tall cultivar, moderately resistant to leaf rust and CBD, low yielding, good cup quality
Caturra	unknown	Dwarf cultivar, resistant to rust, poor cup quality
Coffea euginoides	unknown	non-coffee species
Cook Island G	Cook Islands	Unknown
DK/1/6	unknown	Unknown
DK2/6	unknown	Unknown
Elgon D/12/11	unknown	Clone, short stature, completely resistant to leaf rust, very high yielding, have good cup and physical bean qualities
Ethiopian 113/18	unknown	Unknown
Ethiopian 35/2 X 134/4/-62	unknown	Unknown
Ethiopian 644/18	unknown	Unknown
Ethiopian 69/11	unknown	Unknown
Hybrid 153	Kawanda crosses	Unknown
Hybrid 24	Kawanda crosses	Unknown
K7/1/3	unknown	Unknown
KA3	unknown	Unknown
KAJ	unknown	Unknown
Kaprweta	unknown	Unknown
KP162	unknown	Tall commercial variety, high yielding, susceptible to rust and CBD
KP228/21/3	unknown	Tall variety, high yielding, very susceptible to CBD and leaf rust, has good cup and physical bean qualities
KP423	unknown	Tall variety, high yielding, very susceptible to CBD and leaf rust, has good cup and physical bean qualities
KP423/11/1	unknown	Unknown
KP423/11/5	unknown	Unknown

Lyamungu B	Lyamungu	Unknown
Lyamungu/C	Lyamungu	Unknown
N197/39/1B	unknown	Unknown
N81/13/5	unknown	Unknown
N5/14/6	unknown	Unknown
NG9257	Papua New Guinea	Dwarf cultivar, high yielding, resistant to rust, susceptible to CBD, has poor cup quality
Ruiru 11	Kenya	Commercial variety in Kenya, resistant to CBD and rust, high yielding but inconsistent cup quality
Rume 12	unknown	Tall cultivar
Rume 14	unknown	Tall cultivar
Rume 17	unknown	Tall cultivar
Rume 4	unknown	Tall cultivar
S12 KAFFA	unknown	Unknown
SL14	unknown	Commercial variety, high yielding, good cup quality, susceptible to rust and CBD
SL28/8/2	unknown	Unknown
SL34	unknown	Tall cultivar, high yielding, good cup quality and susceptible to rust and CBD
SL34/16/3	unknown	Unknown
Turrialba	unknown	Dwarf cultivar, high yielding, resistant to rust, susceptible to CBD, poor cup quality
RE/7 Clone	unknown	Unknown
RG MAGEZI IGNITUS	unknown	Unknown
Castrol 2010 Africano	Colombia	Unknown
Castrol 2011 UCDA	Colombia	Unknown
Indian selection 5	India	Tall cultivar, resistant to rust, has good cup quality
Indian selection 6	India	Tall cultivar, resistant to rust, has good cup quality
KABALE/27 CLONE	Kabale farm	Unknown
Kasanja Group 1	Kasanja farm	Unknown
N135	Kabenge farm	Unknown
Elgon CB (COLOMBIA)	Not known source	Unknown

In Rwanda the germplasm collections were in need of maintenance, and 182 lines were rehabilitated and managed by the project (Table 25).

Table 25. Germplasm collections rehabilitated and managed in Rwanda

Name	Source	Type
Jackson 2/1257	Mulungu (DRC)	Tall
Mibirizi 49 - 1136 BB	Mulungu (DRC)	Tall
Bourbon Mayaguez 139 (BM 139)	Mulungu (DRC)	Tall
Mysore 175	Mulungu (DRC)	Tall
Barbarina	Guatemala	Tall
Harar	Mulungu (DRC)	Tall
Coorg	India	Tall
Amphillo	Mulungu (DRC)	Tall
Harar Dugda Lemita Arussi	Ethiopia	Tall
Irga Ciaffe Sidamo	Ethiopia	Tall
Pop 3303/16	Mulungu (DRC)	Tall
Mizan Taffari Kaffa	Ethiopia	Tall
Bourbon Salvadoreno	Costa Rica	Tall
Bourbon of Kenya	Kenya	Tall
Hybrid Bourbon x Arabica	Rwanda	Tall
Bourbon 72	Mulungu (DRC)	Tall
Locale Bronze 8	Mulungu (DRC)	Tall
Blue Mountain Jamaica 62	Mulungu (DRC)	Tall
Porto – Rico	Columbia	Tall
Eugenioides	Ethiopia	Tall
Kent 508	Kenya	Tall
Kabare 16 - 1112	Mulungu (DRC)	Tall
K 7	Kenya	Tall
SL 14	Kenya	Tall
SL 28	Kenya	Tall
AR 578	Burundi	Tall
Tekisic	Burundi	Tall
LC 370 Bourbon Vermello	Brazil	Tall
Moka Dosidamo Borana	Ethiopia	Tall
Sidamo	Ethiopia	Tall
Ennarea Limu Kaffa	Abyssinie	Tall
Moka d'Aden	Indochina	Tall
Abyssinie 944	Mulungu (DRC)	Tall
Kisenyi 210	Cameroon	Tall
Icatu Vermello RPC2905 C	Brazil	Compact
Java	Cameroon	Tall
Gimma	Ethiopia	Tall
Tannabo Buta Mia Limmu Kaffa	Ethiopia	Tall
Agaro Gimma Kaffa	Ethiopia	Tall
Matinho	Angola	Tall
Sel Kp 423	Angola	Tall
Colombie I	Colombia	Compact
Mundo-Novo	Mulungu (DRC)	Tall
Amarello	Brazil	Compact
Caturra 140* Mibilizi	Rwanda	Compact
San Ramon	Columbia	Compact
Pop 4 (F6)	Portugal	Compact
Caturra 34	Brazil	Compact
Catuai	Mulungu	Compact
Catuai Ameliore	Costa Rica	Compact
Catimor P	Costa Rica	Compact
<i>Coffea liberica</i>	Costa Rica	Tall

Coffea canephora	Uganda	Tall
Las Palmas	Columbia	Tall
Tonkin 123	Indochina	Tall
Guatemala 26	Mulungu (DRC)	Tall
Mulungu 1	Mulungu (DRC)	Tall
Mulungu 32	Mulungu (DRC)	Tall
Porto – Rico	Columbia	Tall
Eugenioides	Ethiopia	Tall
Purpurascens	Ethiopia	Tall

Maragogype	Guatemala	Tall
AR 11C	Burundi	Tall
AR 15	Burundi	Tall
Catuai Amarello	Burundi	Tall
AR 18	Burundi	Tall
AR 54	Burundi	Tall
CIFC 8224	Portugal	Compact
Icatu Vermello RPC2905 C	Brazil	Compact

India had 250 germplasm collections from different countries, collected through different missions including by FAO. As most of the collections had outlived their economic life span, a conservation strategy was planned and the available germplasm collections were retrieved through clonal means and established in an exclusive block at CCRI during the period from 2002 to 2007, before the initiation of the project. However, the project facilitated in improving another field gene bank with 73 core collections established at CRSS, Chettalli. This field gene bank has been consolidated by gap filling using clonal material and also by top grafting of the plants infested with white stem borer. The character profiles of all the 73 germplasm collections have been documented and a monograph has been prepared by incorporating data on vegetative, floral, fruit and bean characters in line with the descriptors for coffee prepared by Bioversity International (IPGRI), Rome. The monograph was published in a book form under the project.

Other constraints were identified during assessment of the status of germplasm collections in relation to CLR and CBD:

- Most collections are susceptible to CLR leading to severe defoliation, and hence being more prone to infestations by insect pests such as stem borers
- This resulted in some of the countries instituting spray regimes against CLR which increased the cost of maintaining the collections
- Rwanda and Uganda did not have characteristics (including disease susceptibility) of collections, and therefore started characterizing them. Rwanda screened materials against CLR and CBD. Examples of CBD (Table 26) and CLR (Table 27) screening results are presented below.

Table 26. Mean infection grade of coffee collections inoculated with CBD in Rwanda

Origin	Name of introduction or cultivar	Mean grade of infection*	Class of resistance**
Ethiopia	Ainamba Babaca Kaffa	8.350	MS
Ethiopia	Babaca Kaffa	9.530	MS
Ethiopia	Debie Sciabile Kaffa	5.547	MR
Ethiopia	Ennarea Limu Kaffa	7.991	MS
Ethiopia	Irgalem Kella Sidamo	7.475	MS
Ethiopia	Wondo Sidamo	8.123	MS
Ethiopia	Harar Dugda Lemita Arussi	4.429	MR
Ethiopia	Teffari Kella Sidamo	8.258	MS
DRC	Bourbon Mayaguez 139*** (BM 139)	3.987	HR
DRC	Bourbon Mayaguez 71*** (BM 71)	5.420	MR
DRC	Harar*	4.893	MR
DRC	Kent 170	4.792	MR
DRC	Kent 198	5.118	MR
DRC	Locale Bronze	3.072	HR
DRC	Locale Bronze 10 – 1721	5.008	MR
DRC	Mibilizi*	7.337	MS

DRC	Population 3303/21*** (Pop 3303/21)	8.235	MS
DRC	Bourbon 72 – 1523	4.307	MR
DRC	Blue Mountain Jamaica	6.268	MR
DRC	Blue Mountain Jamaica 62	5.766	MR
DRC	Jackson 2/1257***	6.332	MR
DRC	Amphillo	9.023	MS

* Mean grade of infection is on a scale of 1-12, resulting from spores inoculated on hypocotyls

**HR= Highly resistant; MR= Moderately resistant; MS= Moderately susceptible; HS= Highly susceptible

*** Commercial cultivars in Rwanda

Table 27. Coffee leaf rust resistance of collections in Rwanda

Origin	Introduction or Cultivar	Score*	Class of resistance**
Burundi	AR 15	7	MS
Burundi	Catuai Amarello	5	MR
Burundi	AR 11 C	5	MR
Portugal	Pop 3/91	5	MR
Portugal	Pop 2/91	3	R
Portugal	CIFC 15706	6	MS
Portugal	Pop 4/91	4	MR
Portugal	Pop 1/91	6	MS
Portugal	CIFC 8224	1	HR
DRC	Jackson 2/1257	8	S
DRC	BM 139	8	S
DRC	Amphillo	4	MR
DRC	Mibilizi 68-00	7	MS
India	Coorg	3	R
India	Devamarchy	0	HR
India	Robarbica	0	HR
Brazil	Icatu Vermello	5	MR
Brazil	Catimor coleçao	4	MR
Brazil	Sarchimor	3	R
Costa Rica	Catimor T8663	1	HR
Rwanda	Caturra 140*Mibilizi	6	MS

*Scores are on a scale of 0-9 after inoculation with CLR

**HR= highly resistant; R= resistant; MR= moderately resistant; MS= moderately susceptible; S= susceptible.

3.2.2. Preparation of an integrated conservation strategy (within wider genetics conservation protocols)

Most countries instituted a conservation strategy of spraying against CLR, weeding, applying fertilisers, and managing pests in order to lengthen the lifespan of germplasm collections. Kenya developed and published a germplasm conservation strategy which is now being used to guide maintenance of germplasm collections. Kenya and Uganda also collected and conserved materials collected from the field. All countries are conserving their existing and newly introduced germplasm (Selections 5A and 6).

Facilities were established at the Central Coffee Research Institute in India for characterisation and conservation of germplasm and hybrids in the country. Linked to this is the development of a marker strategy involving purposive samples from populations to guide work to isolate unique DNA markers, as well as identify bulk marker profiles using the PCR based techniques RAPD, SRAP and ISSR. These established systems for conserving coffee collections and development of methods that can be used to screen them for disease resistance (see section 3.2.3).

3.2.3. Collect CLR isolates for determination of CLR races

3.2.3.1 Determination of CLR races

Resistance to CLR can break down from time to time due to the evolution of new virulent races of the CLR pathogen, *Hemileia vastatrix*. This phenomenon has been more commonly observed in India because of the favourable climatic conditions for CLR build up and selection pressure on the pathogen due to the spread of several rust tolerant cultivars in the field. Resistance to coffee leaf rust is governed by at least nine resistance genes, S_H1 to S_H9, either singly or in combination while the corresponding virulence factors in the pathogen are referred to as v1 to v9. The adaptive capacity of *H. vastatrix* has resulted in the gradual loss of resistance in commercially grown coffee cultivars and all the nine major S_H genes have been overcome by gradual appearance of new physiological races of CLR pathogen. So far, 45 races of rust are known to be distributed in different coffee growing countries. However, the prevalence of each race depends on the spread of corresponding host genotypes. Knowing and monitoring the occurrence of new races is therefore very important so that breeders have an idea of the race diversity and its impact on durability of resistance in the commercially grown resistant cultivars in a country. This information is crucial for formulation of breeding strategies for durable CLR resistance.

Countries carried out collection and identification of coffee leaf rust races. This included scientists from Africa being trained and leaf rust disease samples being sent and/or taken to Portugal for identification at the Coffee Rust Research Institute (CIFC).

Scientists from Kenya, Rwanda, Uganda and Zimbabwe underwent training on identification of coffee leaf rust races at CIFC (Table 28). The training built capacity of staff in determining CLR races in Africa, which will be especially useful once differentials are brought to Africa.

Table 28. Participants trained in coffee leaf rust race characterisation

Name of scientist	Institute
Mr. John Mwangi	Coffee Research Foundation, Ruiru, Kenya
Mr Stanislas Mushimiyimana	Rwanda Agriculture Board (RAB), Kigali, Rwanda
Mr Robert Matovu	Coffee Research Centre, Mukono, Uganda
Mr Neddie Mtetwa	Chipinge Coffee Research Station, Chipinge, Zimbabwe

Staff were trained in methods for determining coffee leaf rust races using differential hosts in June 2010. However, CLR infected leaf samples were collected and sent to CIFC in Portugal in advance of the team's visit. CLR was isolated and inoculated onto differential varieties, which were used to determine CLR races. A number of new races were identified in the four participating African countries and India. The identification of the existing and new races is helping greatly in developing durable resistance by coffee breeders in the participating countries.

India followed two approaches: trapping of the new races by growing differential varieties in the farms; and tracking of the occurrence of any new races on station bred

resistant selections/varieties under cultivation. A set of 35 rust differential varieties and 'A' group plant materials (Sarchimor derivatives, and HDT clones introduced from CIFC resistant to all the known races of rust) was used to trap CLR races. In both methods CLR were isolated, multiplied on susceptible varieties and inoculated on CLR race differential varieties.

Races identified from the different countries under the project are presented in Table 29. Several races were identified not previously reported in the countries, and it is also noted that some countries do not have races occurring in the neighbouring countries. This needs to be taken into account by quarantine institutions when transferring materials from one country to another.

Table 29. Coffee leaf rust races identified through the project

Country	Race identified	Remarks
Zimbabwe	II, XXXIV (previously only race II)	Race XXXIV is virulent to Catimor varieties which are grown in Southern African countries of Malawi, South Africa, Zambia and Zimbabwe
Uganda	I, II, IV, XXX	
Rwanda	II, XV, XLI, XLII XXX	
Kenya	I, II, III, XLI, XLII, XXXVI	The results of this study increased the known CLR races in Kenya from six (6) to twelve (12).
India	I, VIII, XVI, XII, XL, XLI, XVII, XXIII, XXV, XXXVII, XXIV, XXIX, XXXVI, XXXIX	In addition two new races with virulence genes $V_{2,5,6,7,8,9}$ and $V_{2,4,5,6,7,8,9,?}$ were isolated on Sarchimor and HDT clones (832/1 & 832/2) that were re-confirmed as new races at CIFC. Race numbers have not been assigned yet.

Efforts to bring CLR race differential varieties to Africa failed because no courier was willing to carry cuttings of coffee. The differential varieties are supposed to be transported as rooted cuttings, which are quite delicate, so courier companies such as DHL and TNT refused to transport the materials. Efforts to look for alternative ways to bring differentials to Africa were not successful, and it is likely that scientists will have to make trips to Portugal specifically to bring the differentials, although this will be expensive. Unfortunately materials were not ready for transportation when the trainees visited CIFC.

In India, one of the important outcomes of race determination is the interactions of the rust pathogen and Timor hybrids (CIFC 832/1 & CIFC 832/2) used as donors of resistance in CLR resistance breeding programmes worldwide. The clone HDT 832/1, a donor parent in developing Catimor 26 at CIFC, manifested susceptibility, and a new race $V_{2,5,6,7,8,9}$ was isolated from this genotype. Similarly, the clone HDT 832/2, the resistance donor used in developing Sarchimor at CIFC, also showed mild susceptibility, and differentiation of the spore sample resulted in another new race with gene combination $V_{2,4,5,6,7,8,9,?}$. Both these spore samples were sent to CIFC for confirmatory experiments.

The studies revealed that the new race ($V_{2,5,6,7,8,9}$) isolated on HDT 832/1 is able to infect the HDT 832/1 clone, but the new race ($V_{2,4,5,6,7,8,9,?}$) isolated on HDT 832/2 could not infect the HDT 832/2 clone at CIFC. Thus, the resistance spectrum of this clone appears to be more complex. These findings have clearly established that the pathogen

is able to overcome the resistance in the `A` type of plants and the new race has 7 virulence genes out of 9 genes identified so far.

3.2.3.2 Testing varieties for resistance to Coffee berry disease (CBD) in India

Coffee berry disease (CBD) is fortunately absent in India. However, India wanted to understand the resistance to CBD among Indian varieties in order to prepare for a possible outbreak. The project was used as platform for this purpose and two approaches were used: (i) Evaluation of selected Indian varieties against CBD under field conditions in Africa; (ii) Systematic screening of selected coffee genotypes and new breeding lines against different CBD isolates that are different in virulence at CIFC. For the Africa evaluation, two Indian varieties, Sln.5A and Sln.6 were given to all the four participating countries for establishing field trials along with the local check materials. Information on field performance of these two Indian varieties with special reference to agronomical aspects including the resistance against CLR and CBD (where outbreaks occurred) is reported below (see section 3.3.1).

In the second approach, a total of 22 seed coffee samples of different genotypes viz., Sln.11, Sln.5A, S.2790, S.2792, S.2794, S.2800, S.2803 (individual plants) were sent to CIFC for screening against CBD isolates. Additionally, selected plants of two tetraploid inter-specific hybrids S.795 and Sln.11 introgressed with *C. liberica* genome were given to CIFC for screening against two isolates of CBD (Kenya and Cameroon). Results (Figure 1) indicated that Sln.11 manifested relative tolerance (95% mortality against Cameroon isolate, which is more virulent and 41% mortality against Kenya isolate after 22 days of inoculation). Results for S.795 were 99% and 85% mortality when inoculated with Cameroon and Kenya isolates, respectively, after 17 days of incubation, while the control (Caturra) had 100% infection (Figure 2). Cytological studies carried out at CIFC revealed that in Sln11, both fungal isolates presented a lower hyphal length compared to isolates from Catimor 45.

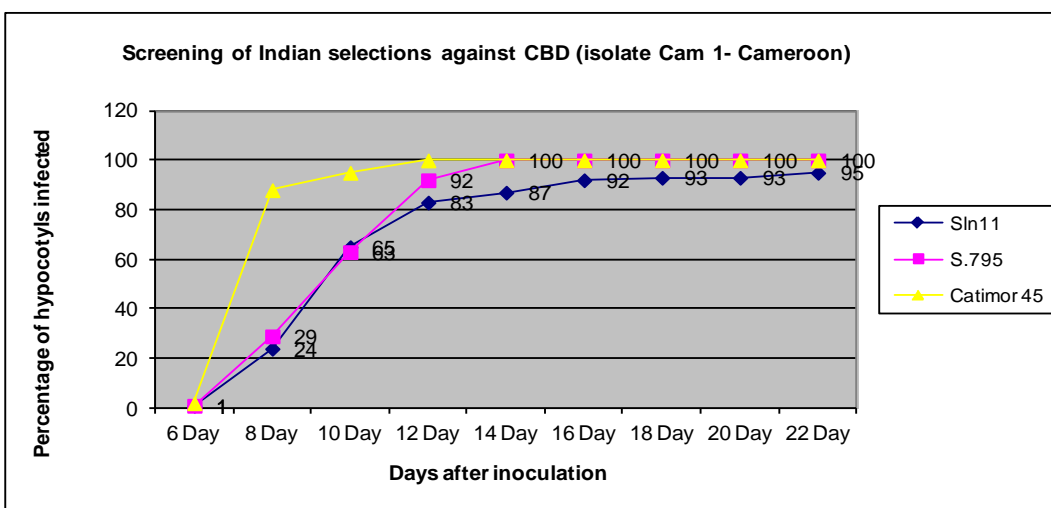


Figure 1. Screening of Indian Selections against CBD (Cameroon isolate)

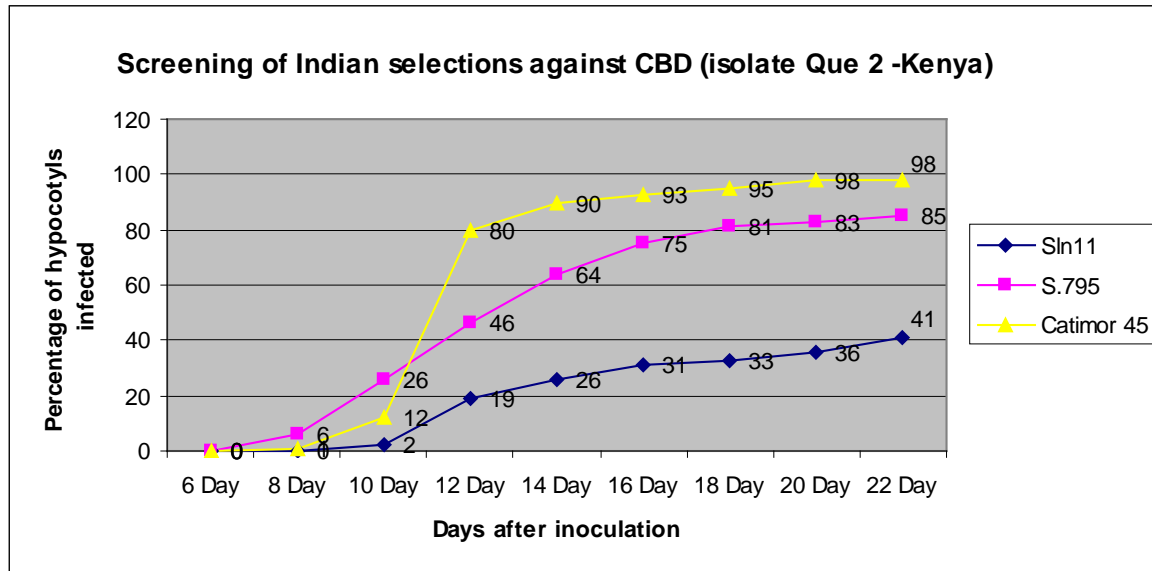


Figure 2. Screening of Indian Selections against CBD (Kenya isolate)

3.2.4 Evaluate coffee hybrids against CLR (and CBD where appropriate) to identify genetic markers associated with disease resistance

This work was mostly done in India and aimed at identifying genetic markers associated with resistance to CLR (and CBD where possible). Studies were carried out under laboratory conditions. Kenya’s capacity to do this work was developed by attaching a Kenyan Coffee Breeder to the Indian laboratory to learn techniques in marker assisted selection.

3.2.4.1 Nursery evaluation of coffee hybrids against CLR races in India

Chandragiri lines were resistant to race XXXIX and $V_{2,5,6,7,8,9}$ in nursery trials on 9-month old seedlings. However, 5 to 7% of seedlings were susceptible to race XXXIX only. Bioassays carried out for presence of the S_{H3} , a CLR resistance gene in selfed progenies of S.795 derived from plants homozygous and heterozygous to the S_{H3} gene, showed segregation in subsequent generations.

This data was used to understand the molecular data generated based on SCAR marker assays. Nursery trials were routinely undertaken to screen the new breeding lines (F_1 progeny) developed by reciprocal crossing of Chandragiri and Sln10 (S_{H3} introgressed line) using marker assisted selection, with an objective of integrating S_{H3} in Chandragiri to achieve broad spectrum (durable) resistance. Bioassays confirmed the presence of the S_{H3} gene in F_1 progeny. Molecular data coupled with bioassays were therefore used for selection of elite (superior) coffee plants.

3.2.4.2 Identification of genetic markers associated with disease resistance

A major challenge in breeding for host resistance in Arabica coffee is the selection of appropriate donor parents and identification of hybrids with alien genome introgression at an early stage of plant growth. Most of the Arabica cultivars are morphologically identical and not easily distinguishable from each other and therefore resistance can only be confirmed through bioassays (screening for resistance). Hence, identification of reliable markers linked to the trait of interest facilitates precise selection of plants and hastens the development of resistant varieties. Molecular markers are more efficient, precise and reliable for discriminating closely related species and cultivars compared to other markers, and are therefore used widely in marker assisted breeding programs.

In this study, application of molecular markers was aimed at:

- Identification of cultivar specific markers for tracking the homogeneity of the seed populations
- Identification and use of markers linked to the leaf rust resistance genes for marker assisted selection and breeding programmes

3.2.4.2.1 Identification of cultivar specific markers

Three types of marker approaches (Random amplified polymorphic DNA (RAPD), Inter-simple sequence repeats (ISSR), and Sequence-related amplified polymorphism (SRAP)) were selected for the purpose. All the three approaches are PCR based. The RAPD marker system generally detects the neutral genetic variation, ISSR markers target the region within the microsatellites repeats, and SRAP markers preferentially detect polymorphism in coding sequences, which are usually conserved among closely related cultivars and species with low mutation rate. Therefore, simultaneous use of different types of molecular markers may be useful in generating the required information.

RAPD/ ISSR/ SRAP Approaches

In order to test the cultivar specific markers with respect to the target varietal selections (SIn.5A, SIn.6, S.795), preliminary screening was undertaken with a total of 100 RAPD primers (decamer primers from Operon Technologies, Almaeda, California). In all, 60 ISSR primers from University of British Columbia were screened against all the Arabica selections including SIn.5A, SIn.6 and the S.795. A total of 425 SRAP primer combinations (12 forward primers and 14 reverse primers) synthesized by Sigma, India were initially used to screen the three Arabica lines (S.795, SIn.5A & SIn.6).

Electrophoresis results

All the three PCRs were repeated at least twice to confirm the reproducibility of each PCR band. Out of the 100 RAPD primers, only five primers were found polymorphic among the three varieties tested. The polymorphic primers were validated on large population of the target selections.

Examples of the gel profiles with RAPD and SRAP assays showing genotype specificity are depicted in Figures 3, 4 and 5.

Key findings were as follows:

- Two RAPD primers (Bot 5 & Bot 65) were specific to Sln.5A while Bot 5 also generated a specific band in variety S.795.
- The ISSR approach is not very promising as there was no polymorphism within the target selections or even among the other Arabica coffee.
- From the preliminary screening using 425 SRAP primers, 42 primer combinations were found polymorphic. These primers were screened on two panels comprising of tall and semi-dwarf Arabica genotypes.
- Of these 42 SRAP primer combinations, 10 primer combinations that showed polymorphism between the three varieties were identified.
- Validation of the 10 SRAP primer combinations revealed that three combinations designated as D11, O24 and S 11 generated cultivar specific profiles with respect to Sln.5A, five combinations (D11, L3, L16, M7 and I10) generated genotype specific profile with respect to Sln.6 and three combinations (M11, M16, P6) generated genotype specific profile with respect to Sln.6.

In conclusion, of the various markers tested, the SRAP markers were found more efficient for detecting polymorphism in Arabica. Hence these markers were routinely used for molecular-genetic analysis. The 11 genotype specific markers were validated on random populations of S.795, Sln.5A, and Sln.6, and were found useful for assessing the homogeneity of seedling progenies. In addition, three SRAP markers specific to the new variety Chandragiri were identified. These markers can distinguish this variety from other semi-dwarf arabica varieties.



Figure 3. Screening of Tall Arabica lines, Sln.5A, S.795 and Sln.6 with RAPD primer BOT 5 showing cultivar specific bands in Sln.5A and S.795. Sample details lanes 1-8 = Sln.5A; 9-17 = Sln.6; 18-24 = S.795; D = Devamachy; K = Kents; M = 1Kb DNA Ladder

RAPD (BOT 5)

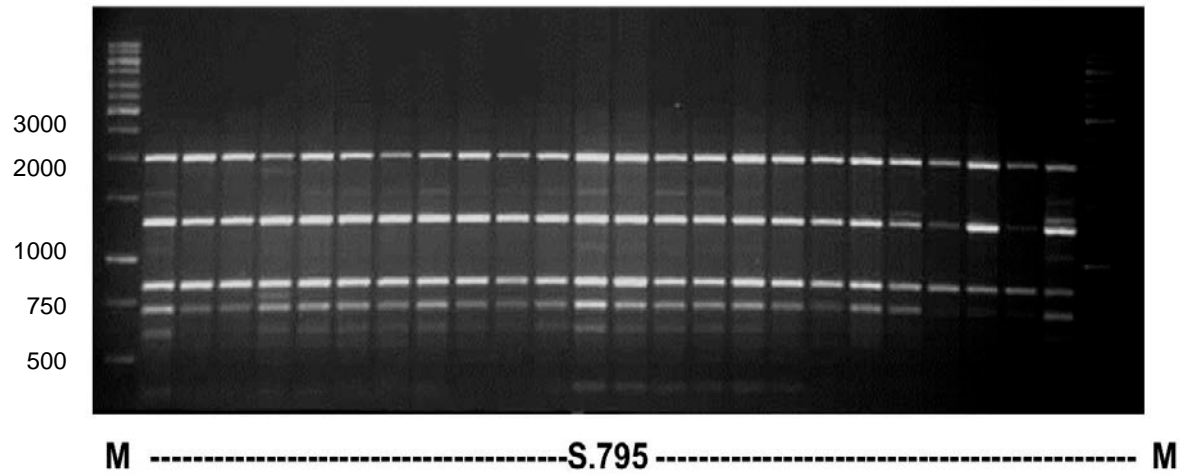


Figure 4. Validation of RAPD primer BOT5 for cultivar specific band (2000bp) in S.795. Sample details, lane 1-8 = Sln.5A; 9-17 = Sln.6; 18-24 = S.795; D = Devamachy; K = Kents; M = 1Kb DNA Ladder

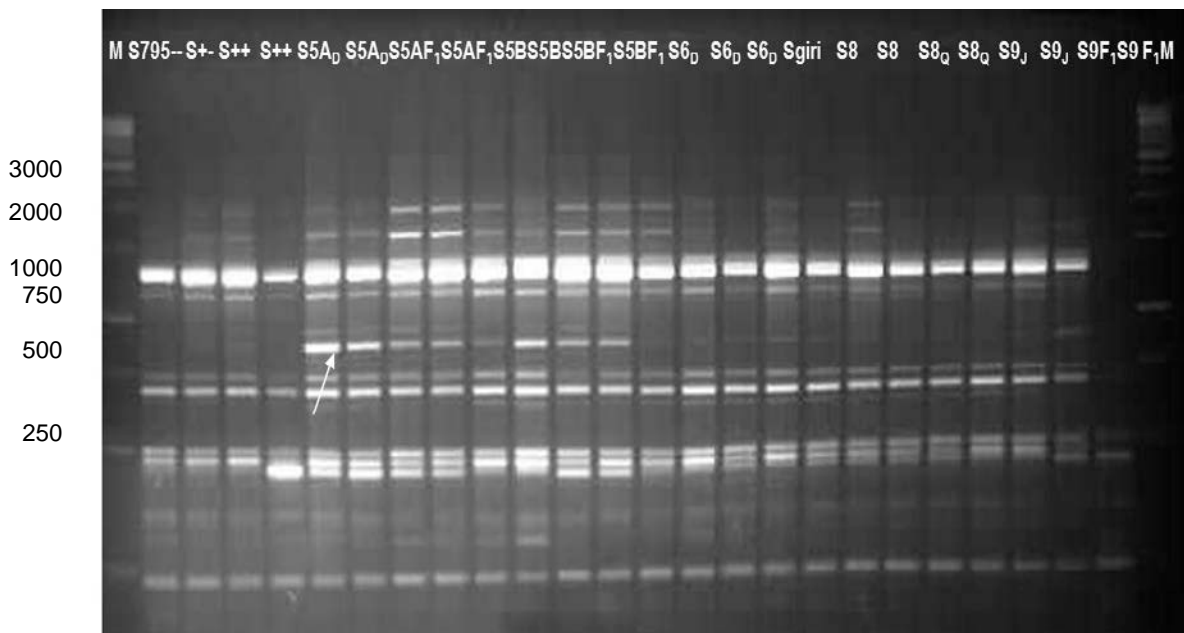


Figure 5. Screening of Tall Arabica lines with SRAP primer O24 showing cultivar specific bands in Sln.5A. Sample details lane 1-4 = S.795; 5-8 = Sln.5A; 9-12 = Sln.5B; 13-16 = Sln.6; 17-20 = Sln.8; 21-24 = Sln.9; M = 1Kb

3.2.4.2.2 Identification and use of markers linked to the CLR resistance genes

Resistance to CLR is reported to be determined by at least nine resistance genes, S_H1 to S_H9, either singly or in combination; the corresponding virulence genes in the pathogen are v1 to v9. Of these resistance genes, S_H1, S_H2, S_H4 and S_H5 were identified in the tetraploid species of *C. arabica* whereas S_H6, S_H7, S_H8 and S_H9, were introgressed to *C. arabica* from the diploid species, *C. canephora*, while S_H3 was introgressed from another diploid species, *C. liberica*.

Prakash *et al.* (2004) reported the first success in identification of amplified fragment length polymorphism (AFLP) markers closely linked to S_H3 gene of resistance. Subsequent success was achieved in developing ten sequence-characterized genetic markers closely associated with the S_H3 CLR resistance gene. This included simple sequence repeats (SSR) markers, sequence-characterized amplified regions (SCAR) markers resulting from the conversion of AFLP markers identified previously, and SCAR markers derived from end-sequences of bacterial artificial chromosome (BAC) clones. The project facilitated the large scale validation of these SCAR markers and routine application of the two SCAR markers closely linked to S_H3 gene in marker assisted selection and breeding programmes.

The SCAR markers linked to S_H3 gene were therefore used for maintenance breeding of S.795 and marker assisted selection for integrating S_H3 gene with resistance genes of Robusta origin (S_H6, S_H7, S_H8, S_H9). The populations of S.795 carry S_H3 resistance gene. In order to rejuvenate this popular and successful variety (grown since 1947), maintenance breeding of S.795 variety has been initiated by systematic monitoring and tracking of the S_H3 gene in the initial populations by using SCAR markers. The S.795 populations established as early as 1950s to 1970s were randomly surveyed and 150 plants manifesting different levels of CLR build up, plant vigour and yield were marked and grouped as vigorous coupled with field tolerance to rust (G1), vigorous and moderately susceptible to rust (G2), less vigorous and susceptible to leaf rust (G3). A total of 90, 40 and 20 plants grouped as G1, G2 and G3, respectively were used for molecular assays, besides the donor parent S.288 for S_H3 gene and Kent Arabica, the susceptible check, for comparison.

Three sequence-characterized DNA markers closely linked to S_H3 gene, Sp-M8-SH3, Sat244, BA-124-12K-f [10] were screened on a small population of S.795. Out of the markers tested, two markers, Sat244 and BA-124-12K-f that gave clear amplification profiles were used for further screening. The sequence of the primers and PCR conditions are presented in Table 30. The PCR assays using specific primer pairs and electrophoresis conditions were followed as described by Combes *et al.* 2000 and Mahe *et al.* 2008.

Table 30. Sequence markers linked to S_H3 gene and PCR conditions used

Marker	Primer sequence (5'-----3')	Amplification product size (bp)	PCR Anneal. Temp. (°C)
Sat244	F: GCATGTGCTTTTTGATGTCGT R: GCATACTAAGGAAATTATCTGACTGCT	300	56
BA-124-12K-f	F: TGATTTTCGCTTGTTGTCGAG R: TGCAGATTGATGGCACGTTA	320	60

Depending on the SCAR marker considered, 2 to 3 different amplified products were observed, but in all cases a marker specific band was found to be present in S.288, the resistant donor parental line and absent in the susceptible parent, Kent arabica. Analysis of the plant population with marker BA-124-12K-f resulted in direct polymorphism with either presence or absence of 320 bp size band (Figure 6). Amplification profiles of different groups of plants revealed that the marker band is amplified in plants grouped under G1 and G2 groups as in case of S.288, the donor parent, while the marker band was found absent in plants belonging to G3 group as in case of Kent arabica, the susceptible check.

In the case of marker Sat 244, an amplification product of 300 to 305 bp size is seen in plants possessing the S_H3 resistance gene, while in plants lacking the S_H3 gene, a band of slightly less size (~ 300 bp) is amplified at the same locus (Figure 7). Further, the marker Sat 244 is more informative as it is possible to distinguish the plants homozygous and heterozygous for S_H3 gene, based on amplification profile. In plants homozygous to S_H3, one intense band is seen while in plants heterozygous to S_H3, two less intense bands are seen (Figure 7). Thus the marker assays proved very effective in identifying plants possessing the S_H3 gene. This was reconfirmed through bioassays that revealed that S_H3-positive plants (both homozygous and heterozygous) were resistant to Race I (v2,5) but susceptible to Race VIII (v2,3,5) while S_H3 negative plants manifested susceptible reaction to both Race I (v2,5) and Race VIII (v2,3,5).

For the second approach of marker assisted gene pyramiding, two F₂ progenies, S.3827 (cross between F₁s of Caturra x S.795 and Caturra x Cioccie) with semi-dwarf phenotype and S.2724 (S.288 x HDT) with tall phenotype formed the study material.

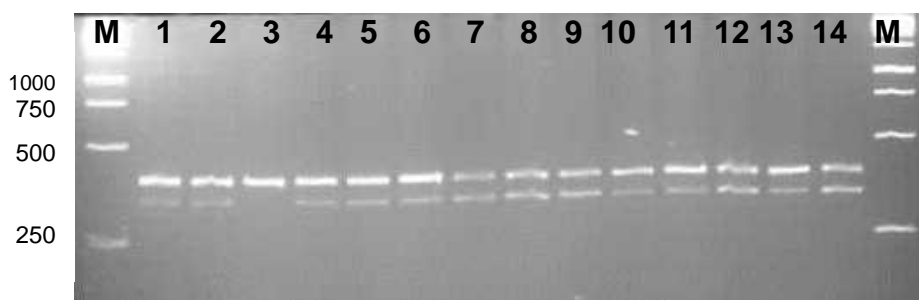


Figure 6. Amplification profile with SCAR marker BA 124 showing presence (samples 1,2,4 to 14) and absence (sample 3) of rust specific SH3 gene, M-1kb.

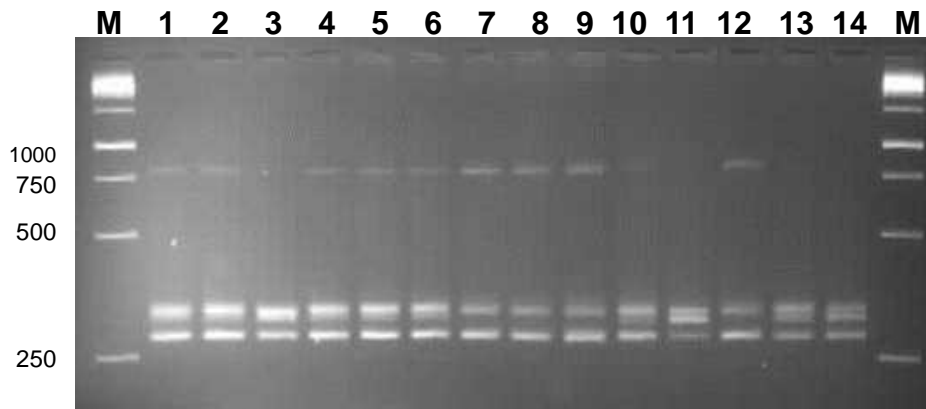


Figure 7. Amplification profile with SCAR marker Sat 244 showing absence (Sample 3) and also presence of rust specific SH3 gene in homozygous (samples 1,2,4 to 10,12) or heterozygous state (samples 11,13,14).

3.2.4.2.3 Marker assisted selection for integrating S_{H3} gene

With an objective of integrating the S_{H3} gene of *C. liberica* origin with the genes of robusta origin (S_{H6} – S_{H9}) for achieving broad spectrum resistance in semi-dwarf genotypes, crosses were effected between Catimor line (derived from crosses between Caturra x HDT - CIFC 1343) and selected Indian cultivars, S.1934 and S.2931. Both S.1934 and S.2931 are the *C. liberica* introgressed lines and were used as donors for the S_{H3} gene, while the Catimor line possesses the genes S_{H6} , S_{H7} , S_{H8} and S_{H9} introgressed from Hybrid de Timor (CIFC 1343). Four F_1 hybrid progenies (S.4814 to S.4817) generated in 1999 and field planted in 2000 at Central Coffee Research Institute were subjected to SCAR assays under the project in order to identify plants homozygous to S_{H3} in F_1 population.

As in the case of S.795 populations, both the markers gave clear amplification profiles that could distinguish the plants for presence or absence of the S_{H3} gene. In the preliminary assays, the analysis of the F_1 hybrid progenies with marker BA-124-12K-f resulted in direct polymorphism with presence and absence of the 320 bp size band (Figure 3). The secondary analysis identified plants homozygous for S_{H3} among the plants that proved positive for S_{H3} in the first screening. Based on individual plant yield, field tolerance to rust and bean quality traits coupled with SCAR marker analysis, elite plants homozygous to S_{H3} were marked and selfed among different progenies.

In conclusion, SCAR marker analysis was used to select and self elite plants with high individual plant yield, field tolerance to CLR, high bean quality traits. The elite plants were homozygous to S_{H3} gene. The selfed progenies raised from individual F_1 plants were established in four different locations for simultaneous evaluation and selection for further exploitation.

3.2.4.2.4 Integration of S_H3 in Sarchimor derivatives

Chandragiri, a Sarchimor derivative, was released for commercial cultivation in India in 2007. The field performance of this variety is very encouraging with high yield potential and field tolerance to rust. The manifestation of high tolerance levels to rust pathogen is primarily due to the rust resistance genes of robusta (S_H6, S_H7, S_H8 and S_H9) introgressed from Timor hybrid (CIFC 832/2). Hence, there is a risk that the races of Catimor on HDT 832/2 may adapt to Chandragiri when the variety is grown on larger areas. Based on these findings, a pre-emptive breeding strategy was initiated in India, to integrate S_H3 into Chandragiri to provide broad spectrum and durable resistance. A cross bred derivative of (Caturra x Ciocee) x (Caturra x S.795), S. 3827, was selected as the S_H3 donor parent. Elite plants homozygous to the S_H3 were selected in S.3827, based on agronomic characters and through marker assisted selection. Reciprocal crosses were made between Chandragiri and S.3827 during 2010 and six progenies were established in the field. SCAR assays of the randomly selected plants of the F₁ progeny revealed the presence of S_H3 gene and studies on field performance are being pursued.

The SCAR markers for S_H3 gene for rust resistance developed by IRD, France were validated and are being used for marker assisted selection and maintenance breeding in S.795 in India. Marker assisted selection using SCAR markers for S_H3 gene has been successfully employed for the first time in coffee breeding towards pyramiding of rust resistance genes, and hybrids have been generated.

3.2.5 Build capacity of country partners in marker assisted selection, breeding, pathology, and extension

The project provided grants for strengthening the marker lab facilities at CCRI in India and a state-of-the-art seed laboratory. The project also provided capacity building of the African scientists in marker assisted selection.

However, the capacity of all participating countries was also strengthened through attachments, short courses (Table 31) and formal training at institutes of higher learning (Table 32).

Table 31. Short courses and attachments

Country	Short course or attachment
India	Students and scientists were trained in marker assisted selection using SCAR markers for S _H 3 gene. Indian students and scientist were attached to the laboratory at CCRI to learn about marker assisted selection in coffee.
Kenya	A Coffee Breeder from Kenya went to India for a short attachment at the CCRI, Coffee Board of India to learn about marker assisted selection in coffee breeding. A coffee scientist went to Portugal (CIFC) to learn about identification of CLR races and resistance to CLR.
Rwanda	One member of staff received training at CIFC in identification of CLR races and resistance to CLR.
Uganda	One scientist received training at CIFC in identification of CLR races.
Zimbabwe	Scientist was attached to the Coffee Research Station in Kenya to learn about marker assisted selection in coffee, which also formed part of his MSc studies. One person received training at the CIFC for CLR race identification.

Table 32. Degree and higher degree training

Country	Number and qualification achieved by staff through sponsorship by the project	Remarks
Kenya	1 PhD in Plant Breeding (Coffee breeding) 1 MSc in Agricultural Extension	Completed successfully Completed successfully
Rwanda	1 MSc in Plant Breeding (Coffee breeding)	Completed successfully
Uganda	1 MSc in Plant Pathology 1 MSc in Plant Breeding	Completed only course work – project research too expensive (molecular based) and could not be completed Completed, thesis submitted.
Zimbabwe	1 MSc in Plant Breeding and Agronomy with University of Zimbabwe 1 BSc in Agriculture Management with the Zimbabwe Open University.	Currently completing his research project To complete in 2014

3.2.6 Address IP and quarantine issues to ensure mutually agreed and safe transfer of genetic material.

With respect to IP and quarantine issues, existing national policies were followed and used in transferring plant materials during the project, as described in section 3.1.6. This also included the transfer of CLR infected leaves which were sent to Portugal. Furthermore, CCRI worked with the Protection of Plant Varieties and Farmers Rights Authority (PPV&FRA), Government of India, to develop Distinctiveness Uniformity and Stability (DUS) criteria for coffee for registering/protecting the Coffee varieties developed by CCRI and to create awareness on IP related issues. CCRI participated in two interactive meetings on these issues and presented the scope and possibilities in coffee.

Countries participating in the project agreed to use existing protocols for exchange of genetic materials. Although India was able to share genetic materials with the African participating countries, India decided not to import materials from Africa in order to avoid the possibility of transferring CBD from Africa to India. Although all countries used the existing IP policies and quarantine regulations in sharing genetic materials, Kenya (Coffee Research Foundation) developed their IP Policy under the legal and professional guidance of Kenya Industrial Property Institute (KIPI) and the Kenya Plant Health Inspectorate Services (KEPHIS). The book is available in hard copy from CRF.

3.3 Field trials on farm and station

There were three indicators for this component in the logical framework.

- Coffee lines combining resistance to CLR with good quality and high yields were selected and popularised among smallholder farmers (section 3.3.1)
- Effective and environmentally friendly chemicals and/or botanicals were identified and methods for incorporation into CLR control were developed and promoted (sections 3.3.2 and 3.3.3).

- Improved disease management options were made available and promoted (sections 3.3.1, 3.3.2, 3.3.3, 3.3.4, and section 3.4).

3.3.1 Trials to screen varieties for resistance to coffee leaf rust and coffee berry diseases

Field trial designs were developed and used in screening coffee germplasm and varieties for resistance to CLR in all participating countries, and for resistance to CBD in in countries where CBD is endemic. The screening included measuring agronomic parameters such as height, number of primaries, and yield. In Africa the two introductions from India, Selection 5A and 6 were evaluated together with the locally available germplasm and commercial varieties. Data were subjected to analysis of variance (ANOVA).

3.3.1.1 Resistance to coffee leaf rust and coffee berry diseases

In India, the new genotype Chandragiri continued to maintain high field tolerance to coffee leaf rust, and only 5 to 7% of the plant population recorded a few pustules. In addition, the Selections 5A and 6, both of which were shared with the four participating African countries, recorded moderate resistance to coffee leaf rust in India. Catuai x HDT, Colombia, Selection 5B, Chandragiri, and Selection 5A, in that order, were all resistant to CLR in India, although as a country they prefer calling them tolerant varieties (Table 33).

Table 33. Coffee leaf rust incidence among six months old varieties screened in India

Genotype	Percentage of leaf rust incidence in different years				Mean of four years
	2009	2010	2011	2012	
Sln.5A	3.92	3.27	3.37	7.95	4.63
Sln.5B	1.64	0.65	2.33	6.02	2.66
Sln.6	2.55	1.94	4.57	5.95	3.75
Chandragiri	4.25	2.25	2.27	5.63	3.60
Columbian Catimor	1.83	2.59	2.91	4.15	2.87
BBTC Catimor	5.05	6.11	7.71	10.45	7.33
Catuai x HDT	0.83	1.69	2.12	3.09	1.93

In Kenya, Ruiru 11, Crosses 8, 22, and 30, and Selections 5A and 6 were resistant to CLR both at Othaya and Kabati where CLR pressure was moderate as recorded in the susceptible variety, SL 28, both sprayed and unsprayed (Figure 8). Ruiru 11, and SL 28 are commercial varieties in Kenya. Crosses 8, 22, 23, 27 and 30 are from a breeding programme which was carried out before the project, but evaluation of the crosses was carried out under the project. In general Selection 6 performed as well as the known resistant variety, Ruiru 11. Although not planned, the varietal trial in Kisii was exposed

to Bacterial blight of coffee (BBC) during the trial period in 2010. The BBC scores indicated that Selection 6 (Robarbica) may also have tolerance to BBC (Figure 9).

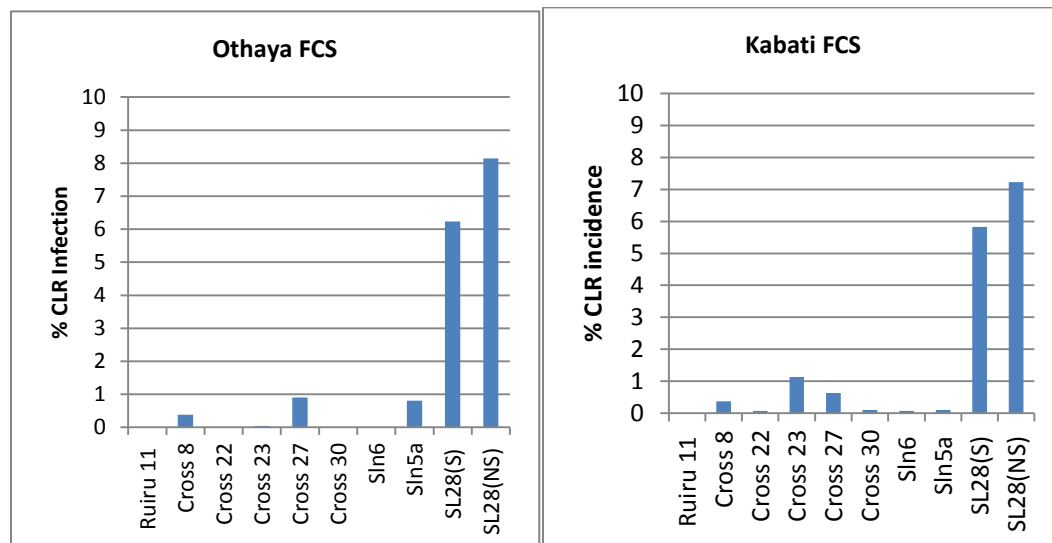


Figure 8. CLR incidence among varieties at two sites in Kenya

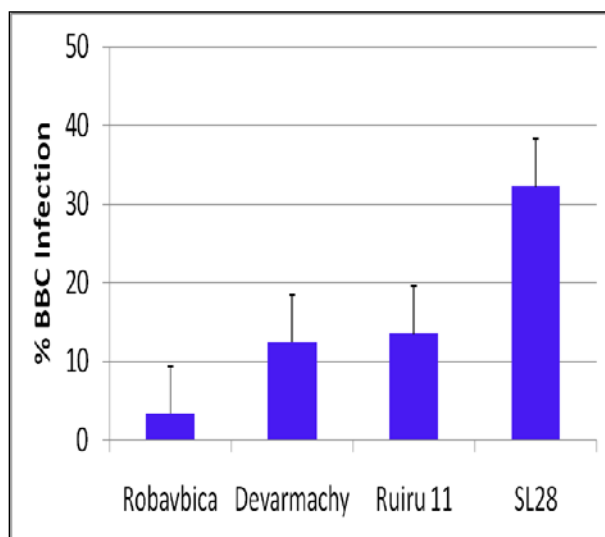


Figure 9. Incidence of Bacterial blight of coffee on 4 selections in Kisii, Kenya

In Uganda four varieties, NG9257, Elgon CB, and the two Indian introductions, Selection 5A and 6, recorded very low CLR incidence and severity scores, thus grouping them into a category of varieties resistant to CLR under both field and laboratory conditions (Table 34). However, it is worth noting that although Selection 5A recorded a relatively high incidence (25% of infected leaf discs under laboratory conditions) the variety recorded a low disease severity score of 2.4, which is within the category of resistance (scale of 0-3). NG9257 is a Catimor variety, while Elgon CB is a locally bred variety.

Table 34. CLR incidence and severity under field and laboratory conditions in Uganda

VARIETY	% CLR Incidence	Mean Severity (Scale: 1- 4)	Reaction Type		% CLR Incidence	Mean Severity (Scale: 0-9)	Reaction Type
Field Assessment				Laboratory Assessment			
KP423	41.4	1.5	MR		15.0	3.5	MR
SL28	55.6	1.8	MR		45.0	5.2	MR
NG9257	0.0	1.0	R		15.0	1.0	R
SL34	45.4	1.6	MR		65.0	5.0	MR
SL14	33.1	1.4	MR		50.0	5.2	MR
Elgon CB	1.86	1.02	R		-	-	-
Selection 5A	0.8	1.0	R		25.0	2.4	R
Selection 6	0.9	1.0	R		10.0	2.0	R
P-Value	<0.001	<0.001			<0.001	<0.001	

Virulence of rust isolates was assessed after 30 days from infection day of leaf discs, using a 0 to 9 disease rating scale. Resistance level for each genotype was determined as follows; discs expressing lesions of type 0 to 3 were considered resistant (R); those expressing lesions of type 4 to 5 were considered moderately resistant (MR); expressing lesions of type 6 to 7 were considered as moderately susceptible (MS) while expressing lesions of type 8 to 9 were considered as Susceptible (S). Disease incidence was determined as percentage of discs infected by CLR for each genotype.

During the time of the highest disease pressure in Uganda (August 2012), the four varieties, NG9257, Elgon CB, Selection 5A and Selection 6 remained resistant (Figure 10). Selections 5A and 6, and NG9257 recorded incidence as low as 0%, while the commercial varieties and controls, KAP423 and SL28 recorded CLR incidence of up to 100% (Figure 9).

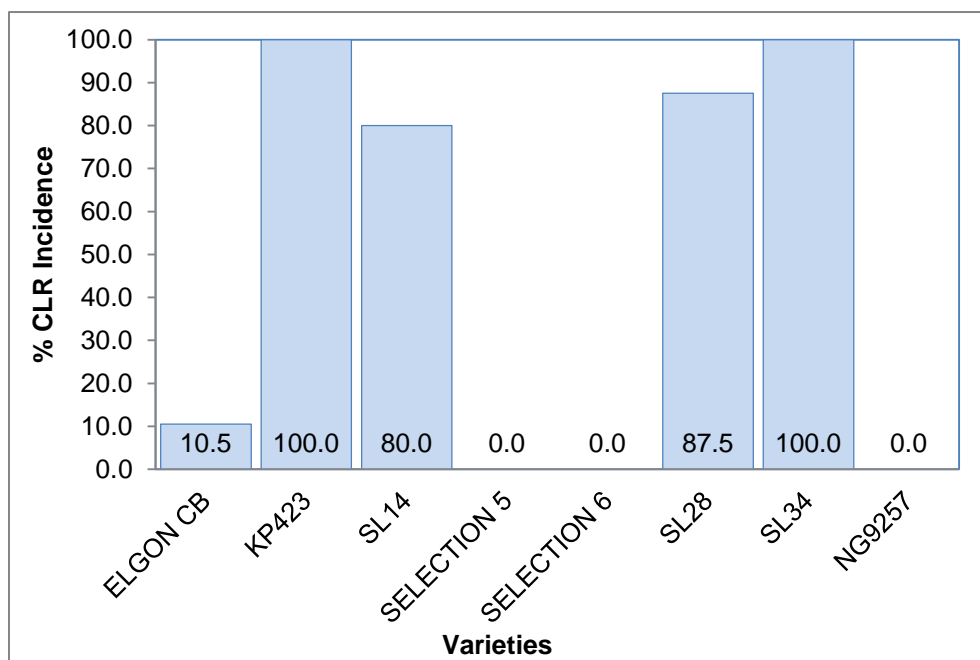


Figure 10. Field incidence of CLR during high disease pressure in Uganda

In Rwanda both Selections 5A and 6 recorded the lowest CLR incidence (almost nothing) (Figure 11) compared to Jackson, Harar, and BM 139, all commercial varieties. As a result the country is going through processes for officially including Selection 6 among the commercial varieties. Results were achieved under field evaluation of Selection 5A, Selection 6, Jackson, Harar, and BM 139. BM 139 was a local check.

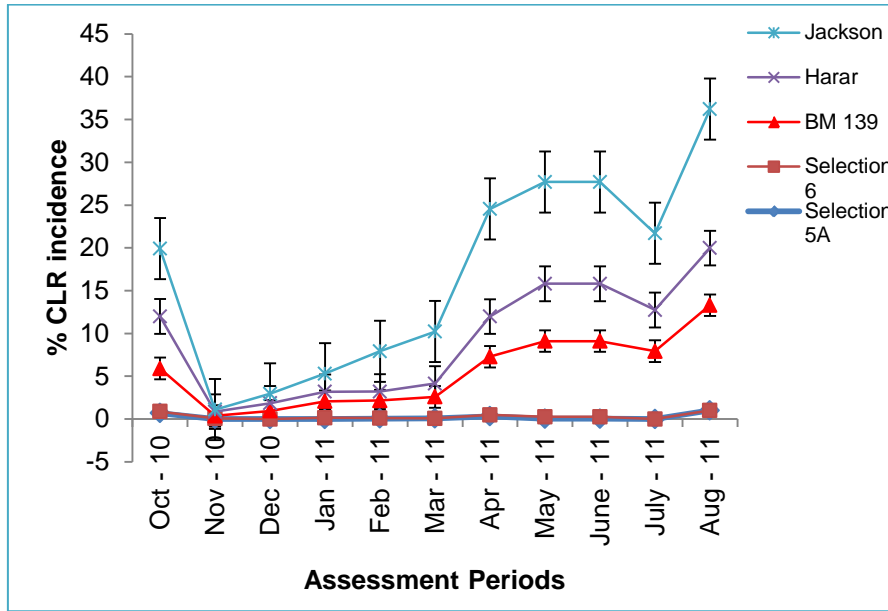


Figure 11. CLR incidence on coffee varieties in Rwanda

Zimbabwe achieved similar results to those in other countries regarding Selection 6. Selection 6 and a coffee line, 13683/35, recorded the lowest CLR incidence (Figure 12). Selection 5A did not perform as well as Selection 6. Selection 5A has been known to segregate, with some individual plants being very resistant while others register a few CLR pustules (spots) on a few leaves. A known resistant variety, Catimor 129, did not perform as well as line 13683/35 and Selection 6. This may confirm the local observations that resistance is breaking down in the variety, which may be attributed to the presence of a new CLR race (see section 3.2.3.1).

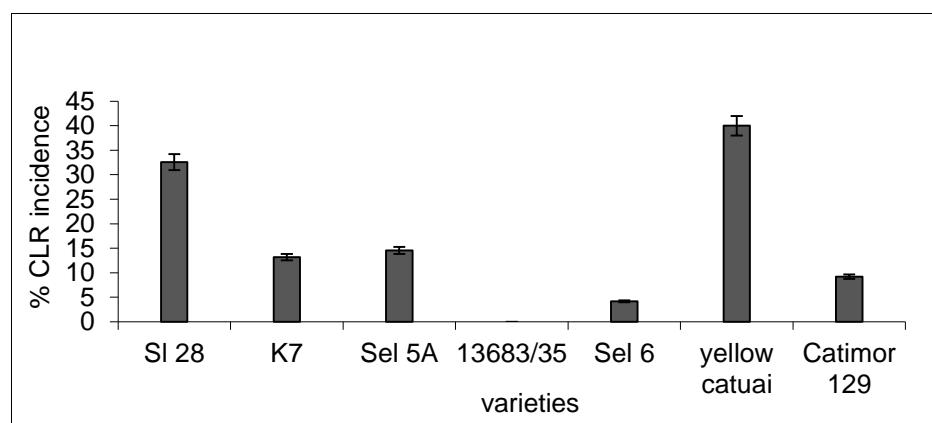


Figure 12. CLR incidence on screened varieties in Zimbabwe

3.3.1.2 Growth parameters of the screened varieties, selections, and lines

Growth parameters which included germination, plant height, stem girth, number of primaries, number of nodes, leaves, and leaf area were assessed. In all countries, growth parameters of the introduced Indian selections, 5A and 6 were not significantly different from those of the tall varieties such as SL28 in Kenya as per combined data from three trial sites (Table 35). However, during earlier stages of growth, the two selections had agronomic parameters between the dwarf varieties such as Ruiru 11 and the tall varieties such as SL28.

Table 35. Growth parameters for some of the evaluated varieties in Kenya

Growth Data 2010 – Machakos, Kisii and Bungoma Combined							
Variety	Height (m)	Nodes	Int. Length ^a	Primaries	%BP ^b	Laterals	Berries
SIn 6	157.87a	31.61a	5.04b	48.24a	66.53a	5.10c	69.17b
SIn 5a	146.01b	29.01bc	5.05b	45.39ab	59.12b	13.07a	45.90c
R11	113.05c	29.50b	3.85c	44.83b	63.01ab	5.84c	104.37a
SL28	151.22ab	28.13c	5.41a	44.13b	65.76a	7.84b	94.76a
LSD _(5%)	6.82	1.28	0.15	2.99	2.24	1.30	16.05

^ainternode length; ^bbearing primaries. Means followed by the same letter are not significantly different at P=0.05.

Assessment of agronomic characteristics was also carried out for the new lines in Kenya, commercial varieties in India, and commercial varieties and the selections in Uganda and Rwanda. The trends are the same in that the new introductions, Selections 5A and 6, are similar to tall varieties such as SL28, NG 9257 and KP 423.

In Zimbabwe, Selections 5A and 6 were significantly ($P \leq 0.001$) taller, had thicker stems, and a larger number of primaries than other varieties such as Catimor or SL28 (Table 36). There were minor differences in the number of flowing primaries, but significant ($P \leq 0.05$) differences in the number of bearing branches (Table 36).

In India, growth data were recorded from 6-months old coffee plants (in the field), hence were very low compared to growth data taken from Africa on the same varieties (Table 37). In general the varieties in India had no significant differences among them, which might have been due to their having similar parentage.

Table 36. Coffee growth parameters and yield after 27 months in the field in Zimbabwe

Variety	Height (cm)	Girth (mm)	Primary branches	Flowering primaries	Bearing branches
SL28	130.4ab	35.3a	45.4ab	28.6	31.1ab
Catimor129	141.9b	36.2a	44.9ab	27.6	30.8ab
Catimor F6	126.4a	34.3a	41.8a	27.7	27.2a
Selection 5A	183.6c	40.8b	48.7bc	28.2	29.9a
Selection 6	210.9d	46.3c	54.1c	33.1	38.8b
p	<.001	<.001	<.001	0.121	0.014
CV%	13.3	14.5	17.5	24.3	11.5

Means followed by the same letter are not significantly different at P=0.05

Table 37. Growth parameters on 6 months old coffee plants at the Central Coffee Research Institute in India

Location/ Variety	Height (cm)	Stem girth (mm)	No of pair of leaves	Tap root length	Shoot weight		Root weight	
					Fresh	Dry	Fresh	Dry
Sln 5A	20.0	3.33	4.32	14.5	6.82	1.57	1.93	0.18
Sln 6	24.3	3.47	4.92	14.8	8.19	1.54	2.35	0.11
S 795	22.2	3.35	4.80	17.1	8.73	1.44	3.10	0.07
Chandragiri	18.5	3.50	4.96	15.7	9.78	2.09	3.57	0.38

In Rwanda, DNA markers were used to assess the genetic diversity between the disease resistant coffee varieties and susceptible cultivars using Random Amplified Polymorphic DNA (RAPD) and Simple Sequence Repeats (SSR) (Figure 13). The analysis of a dendrogram gave the composition of 6 different clusters, but the notable result is that the CLR and CBD resistant varieties, Catimor, HDT, RS, and Selections 5A and 6 were clustered together and separately from susceptible commercial coffee cultivars (Figure 12). This may imply that the two Selections have genes for CBD resistance.

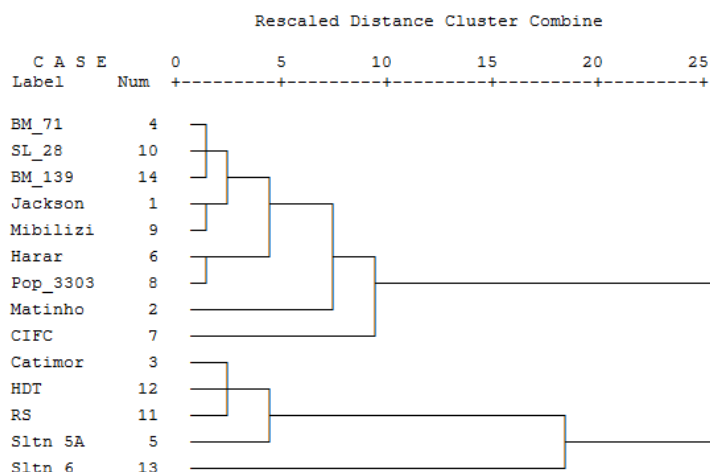


Figure 13. Dendrogram of coffee varieties screened in Rwanda

3.3.1.3 Yield and quality considerations

Although all countries in Africa received Selections 5A and 6, in most countries they did not achieve economic yield during the project, as this takes 3 to 4 years. So apart from in Kenya, yield data from Africa appear relatively low at the time of measurement. In Kenya, the Machakos site provided excellent conditions (including irrigation), and the introduced selections yielded as high as 4.6 tons per ha (Selection 6), which was as high as the commercial variety Ruiru 11 (Table 38). However, the commercial variety SL28 yielded significantly better than Selection 5A, but was not significantly different from Selection 6. The new Kenyan genotypes also yielded very well (above 3 tons per ha) in Machakos and Marience, and yield of most of the lines was not significantly different from the sprayed SL28, the local check (Table 39).

Table 38. Yield Data (kg/ha) 2011 in Kenya

Variety	Kisii 2011 (kg/ha)	Machakos 2011 (kg/ha)
R11	3,666.44a	4,512.09ab
SL28	2,131.55b	6,088.11a
Sln 6	2,716.53ab	4,588.35ab
Sln 5a	1,518.36b	3,433.10b
LSD _(5%)	1,287.45	1,954.37

Means with the same letter are not significantly different at P=0.05

Table 39. Yield of the new coffee lines at five sites in 2011 in Kenya

	Kitale	Koru	Machakos	Mariene	Ruiru
Genotype	Av. Yield (kg/ha)	Av. Yield (kg/ha)	Av. Yield (kg/ha)	Av. Yield (kg/ha)	Av. Yield (kg/ha)
CR27	540.47b	3,853.84abc	4,706.57ab	4,085.63c	1,492.29cd
CR30	508.57b	4,032.60ab	3,516.70bc	5,056.88abc	1,460.33cd
CR8	623.42b	3,336.81cd	3,568.09bc	4,484.79bc	1,060.55d
CR22	784.76b	2,234.24e	3,069.11c	4,236.46bc	1,326.08cd
CR23	1,009.76ab	3,840.81abc	4,957.21a	5,463.75ab	2,049.37b
R11	1,477.14a	4,439.222a	4,057.39abc	6,046.88a	1,232.45cd
SL 28 (Sprayed)	-	3,107.16d	4,402.53ab	-	2,730.57a
SL 28 (Not Sprayed)	995.71ab	3,73.39f	3,772.13abc	4,282.71bc	1,577.04bc
LSD	537.29	733.57	1,290.76	1,340.91	506.76

Means in a column followed by the same letter are not significantly different at $P=0.005$

In Zimbabwe, although still not at full economic production, yield after 27 months in the field was quite high (Table 40). However, Selections 5A and 6 recorded yield which was significantly ($P\leq 0.001$) lower than the commercial varieties, SL28 and Catimor 129 (Table 40). Yield from trials in India was very low (Table 41), which corresponds to agronomic data (section 3.3.1.2), and is due to slow growth of coffee under shade conditions.

Table 40. Yield data in Zimbabwe after 27 months in the field

Variety	Clean coffee yield (kg/ha)
SL28	3,880b
Catimor129	4,007b
Catimor F6	3,986b
Selection 5A	2,455a
Selection 6	2,688a
p	<0.001
CV%	8.5

Means followed by the same letter are not significantly different at $P=0.001$

Table 41. Yield data from India

Variety	Yield (kg/ha)
Sln.6	945.3
S.795	No crop
Chandragiri	674.4

3.3.1.4 Coffee cup quality

Two countries, Kenya and Rwanda, were able to carry out cup quality (sensory) evaluation of the introduced and commercial varieties (Table 42 and 43). The sensory evaluation method which was used is Quantitative Descriptive Analysis (QDA). In Kenya

sensory rankings were highly dependent on the locations where the genotypes were grown, seasons, year of evaluation and spacing. The Indian accessions were comparable to Kenyan SL28 and Ruiru 11 in terms of cup quality and were found to have specialty potential (Table 42).

In Rwanda, results for cup quality testing of two varieties, Selection 6 and BM 139, show that Selection 6 had a slightly better cup quality than the current commercial variety, BM 139 (Table 43). The results are similar to those found in Kenya although there are some differences in the testing system.

Table 42. Cup quality results for four varieties in Kenya

Genotypes	PM	Fragrance	Flavour	After taste	Acidity	Body	Balance	Preference	Total Score
Sln 5a	Dry	7.50	7.44	7.36	7.68	7.46	7.50	7.46	82.43
	Wet	7.75	7.75	7.58	7.75	7.50	7.58	7.71	83.63
Sln 6	Dry	7.50	7.36	7.36	7.50	7.54	7.54	7.36	81.86
	Wet	7.63	7.71	7.75	7.8	7.54	7.63	7.67	83.67
Ruiru 11	Dry	7.36	6.79	7.12	7.50	7.43	7.25	6.86	75.71
	Wet	7.71	7.79	7.58	7.75	7.67	7.63	7.67	83.79
SL28	Dry	7.54	7.46	7.21	7.83	7.50	7.50	7.42	80.79
	Wet	7.67	7.67	7.71	7.79	7.50	7.67	7.75	83.75

Table 43. Cup quality results for two varieties in Rwanda

Site	BM 139 variety		Selection 6 variety	
	Cupping score	Aroma/Flavour	Cupping score	Aroma/Flavour
Rubona	80	Potato taste, fruity, honey, toast, medium acidity and good body	82	Good body and medium acidity
Rutsiro	84	Fruity, citrus acidity, long finish	83	Good body and medium acidity
Gicumbi	84	vanilla, fruity and toast flavour, medium body and acidity with sweet finish	85	Fruity flavour (citrus, strawberry), heavy body balanced with acidity

3.3.1.5 Release of resistant varieties

The following crosses were officially released in Kenya after evaluation under the project:

- Cross 8 (0822) released as Batian 1
- Cross 22 (2222) released as Batian 2
- Cross 30 (3022) released as Batian 3

The three varieties were released in May 2010, and were gazetted on 25th June 2010 (Kenya gazette Vol.CXII-No. 63). Other countries in Africa are still in the process of releasing the varieties which were found resistant.

3.3.2 Screening antifungal botanicals and other bioagents for control of coffee leaf rust

A study was carried out in India to identify anti-fungal botanicals and other bio-agents. Evaluation of botanicals, particularly pawpaw leaf extract, gave poor results in early stages in Africa, except in Rwanda, so it was stopped in the other two countries. However, promising results were achieved in India with antifungal bio-agents, although most of the test materials were not effective against CLR except for *Bacillus brevis* at 1×10^{-7} cfu/ml, which gave the lowest CLR incidence. This was just below that of the most widely used chemical method, Bordeaux mixture at 0.5% (Table 44). However, *Bacillus brevis* needs to be evaluated further for its use in management of CLR in coffee.

Table 44. Effect of different bio control agents and botanicals on CLR incidence on S.795 cultivar at Gundikhan Estate, Balehonnur, India

Treatment Details	% CLR incidence
<i>Pseudomonas fluorescens</i> @ 1×10^{-7} cfu/ml	13.7
<i>Bacillus brevis</i> @ 1×10^{-7} cfu/ml	9.8
<i>Enterobacter intermedium</i> @ 1×10^{-7} cfu/ml	15.1
Neem kernel aqueous extract @ 5%	17.1
Soapnut powder at 5%	23.6
Bordeaux mixture @ 0.5%	8.3
Control (water spray)	23.0
'F' Test	P<0.01
CDF @ 5%	3.75

3.3.3 Screening fungicides for efficacy against coffee leaf rust

Screening of fungicides was carried out in Uganda, Rwanda and Zimbabwe. However, Kenya, which evaluates fungicides routinely before they are registered in the country, opted to assess the economics of applying fungicides. The fungicides tested are shown in Table 45.

Table 45. WHO classification of fungicides tested

Product name	Active ingredient	WHO Classification*	Mammalian LD ₅₀ mg/kg
Alto	Cyproconazole	II	1,020
Bayfidan	Triadimenol	II	900
Bayleton	Triadimefon	II	602
Benlate	Benomyl	U	>10,000
Copper	Copper oxychloride	II	1,440
Daconil, Dacobre	Chlorothalonil	U	>10,000
Kocide	Copper hydroxide	II	1,000
Orius	Tebuconazole	II	1,700

*II=Class 2, Moderately Hazardous; U= Unlikely to present acute hazard in normal use

In Rwanda, Alto (cyproconazole) was the most effective fungicide, with significantly lower CLR incidence and severity (pustules per leaf) than all other fungicides (Figure 14). The fungicide was therefore registered by the National Agriculture Export Board (NAEB) for the control of CLR in Rwanda. The botanical (papaya leaf extract) was ineffective.

In Zimbabwe, Copper (copper oxychloride) and Alto (cyproconazole) gave the best control of CLR with CLR incidence of less than 3 % (Figure 15). Results from Uganda (Figure 16) showed that Orius (tebuconazole) recorded significantly better control of CLR with incidence of 0.3% compared to the control which recorded 22.7% during the screening period. The botanical (papaya leaf extract) was again ineffective. Copper (copper oxychloride) was the second best at 8.77% incidence of CLR. Orius also gave the lowest Area Under Disease Progress Curve (AUDPC) (Figure 16).

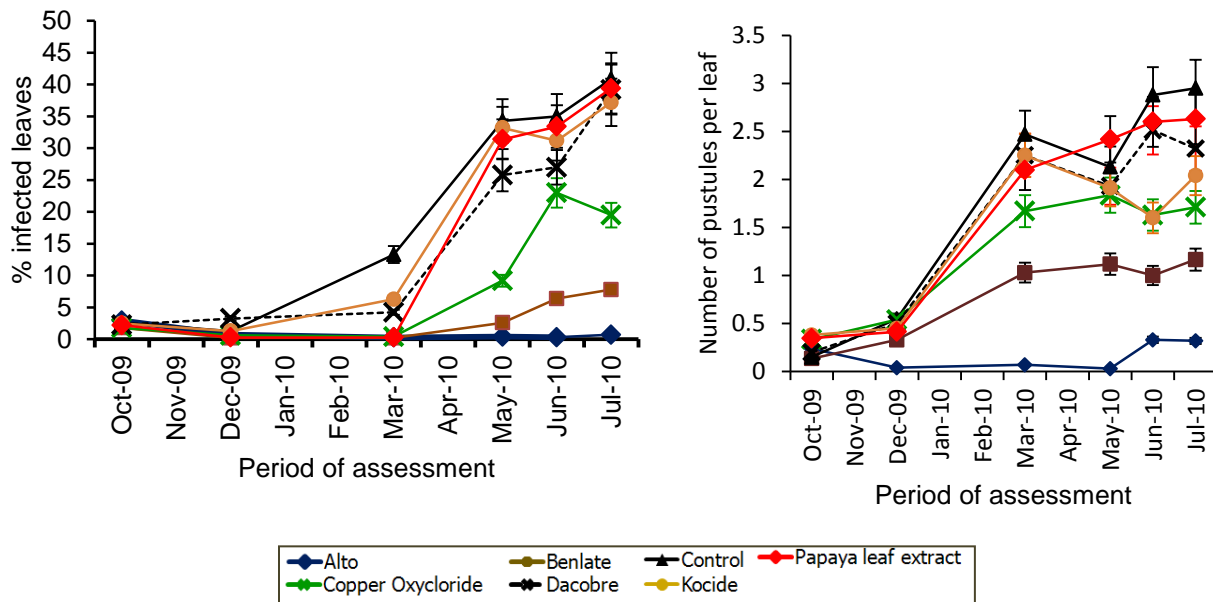


Figure 14. CLR incidence and number of pustules in fungicide trials, Rwanda

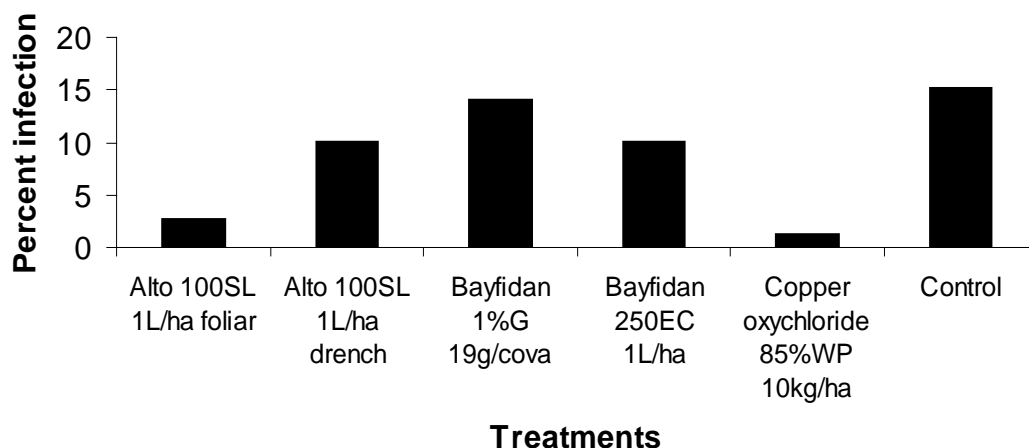


Figure 15. CLR incidence in fungicide trials, Zimbabwe

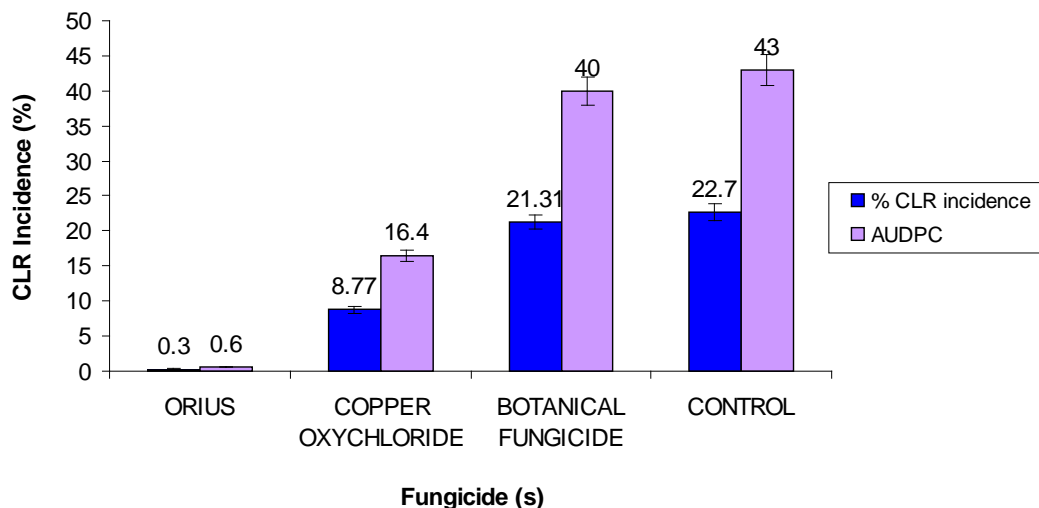


Figure 16. CLR incidence in fungicide trials, Uganda

3.3.4 Studies on epidemiology, adoption of new varieties and economic benefits of using fungicides in Kenya

3.3.4.1 Study of epidemiology of coffee leaf rust in Kenya

Field trials were set up to monitor CLR and CBD under medium and low altitude shade regimes. Twenty two (22) trees were selected along the path of the shadow in the East West direction and six (6) trees in full sun. The study was carried out between 2010 and 2012. Data were recorded on four selected branches for each disease at 2-3 week intervals.

At the start of the CLR infection period, May-June, CLR was higher on the trees at the fringes of the shade and in full sun up to levels of 40% and above, although results for overall effect of shade on CLR were inconclusive (Figure 17). However, when the disease level started to fall mainly due to defoliation, higher levels of CLR were observed on the trees under the shade but the infection did not reach the levels observed on the outer trees earlier in the season. At low rainfall intensity and duration, many water droplets do not reach the coffee trees and spore liberation and dispersal are reduced. This may have resulted in low disease pressure near the shade trees. Effects of shade would be expected to differ with agro-ecological zones and seasonal weather.

Observations from other parts of the country showed that there was higher incidence of CLR in agroecological zone UM3 than in zones UM2 and UM1. In regard to CBD, the disease was very low and when some infections were observed including brown blight during ripening, it exhibited the same pattern as early infection of CLR. It can be deduced that shade slowed down disease progression but maintained the infection when it would be halted or diseased tissues shed off.

It was observed that shading buffered leaf temperature with increasing irradiance. High stomatal conductances were recorded on coffee in full sun during periods of low irradiance since stomatal aperture is greater under shade or on cloudy/rainy days. Shade was also observed to reduce net photosynthetic rate due to insufficient light interception. Coffee leaves exhibit typical shade acclimation features theoretically allowing them to maintain net photosynthetic rates in low light. Limitation of photosynthesis by low light availability has been proposed as one of the main reasons for lower yields of coffee grown in agroforestry systems in optimal coffee production areas. Coffee under shade had higher chlorophyll levels than coffee grown under full sun. Higher chlorophyll content under shade is most likely associated with the larger amounts of nitrogen accumulated in them.

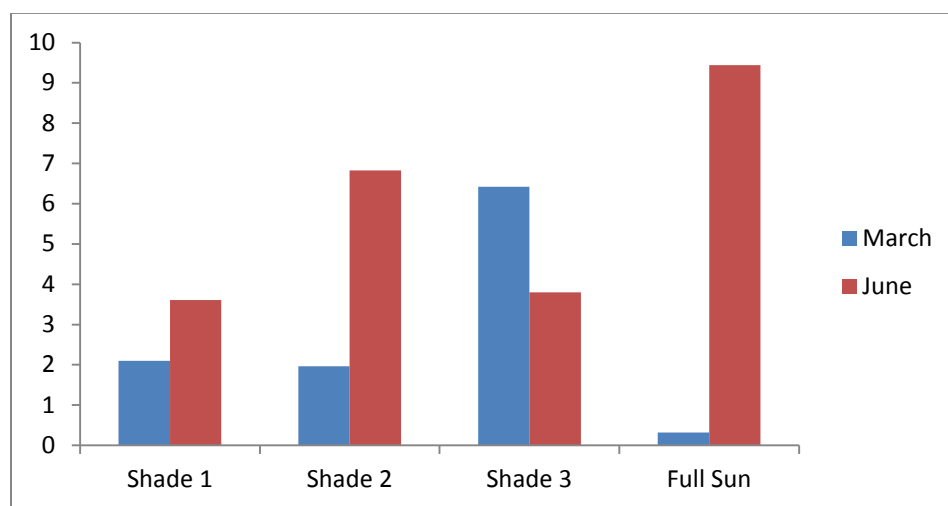


Figure 17. CLR incidence (%) under different shade regimes
Shade 1=25%, Shade 2= 50%, Shade 3=75%

3.3.4.2 Assessment of economic benefits of using fungicides to manage CLR

Field trials were established under different altitude regimes on two sites: Rukera (Medium altitude) and Azania (Low altitude). Coffee leaf rust was severe during the study period with peaks of 40% and above. However, CBD was too low to have a significant impact on yield. There were, however, differences in tree conditions between the treatments with heavy CLR infestation. This also affects the disease progress by altering canopy microclimate and/or availability of substrate. The highest infections were in Daconil (chlorothalonil) treated and unsprayed plots. However, a combination of Copper (copper oxychloride) and Daconil gave the best results. This was also reflected in the analysis of incremental income as indicated in Table 45 below.

Table 46. Yield and net income from fungicide trials in Kenya

		Average yield (kg) per plot	Average total income (Ksh)	% incremental income
A	Copper	28	12,873.28	155
B	Copper + Daconil	30	13,788.29	173
C	Daconil	26	11,971.79	137
D	Bayleton	22	10,123.09	101
E	Control (no spray)	11	5,047.34	-

Thus Copper plus Daconil gave the best incremental income of 173% above the control. Due to low CBD levels, it is not possible to make any valid conclusions regarding its contribution of CBD to the results in this study.

3.4 Scientific management, information systems and communications

The logical framework had 3 indicators for this component. As noted elsewhere, activities in support of communication and dissemination were incorporated in many parts of the project.

- A communication strategy was developed to ensure effective communication between the many project participants (section 3.4.1)
- Many communication activities were implemented with appropriate stakeholders (sections 3.1.3, 3.4.3, 3.4.4)
- Practical recommendations for farmers were prepared and distributed (sections 3.1.3, 3.4.2, 3.4.3 and 3.4.4).

3.4.1 Develop a communication strategy to allow for information sharing within communities and amongst the different actor groups

A communication strategy was developed and used in communicating among project partners. Communication with and among different actor groups is described in the following sections.

3.4.2 Prepare information materials that will be used in communication with farmers and other coffee stakeholders

A range of information materials were produced and disseminated as follows:

- Roll-up Banners (one on coffee leaf rust, one on coffee berry disease) were developed by the PEA and displayed during workshops, and conferences
- Coffee leaf rust and coffee berry disease posters were prepared and disseminated to farmers through FFSs in all participating countries
- A file folder with all the Coffee Research Foundation circulars was prepared and disseminated to farmers through FFS groups in Kenya
- The coffee Atlas was revised as a 2nd edition and made available to farmers and extensionists in Kenya
- The Coffee Recommendation Handbook was reviewed and published in English and Kiswahili.
- The following folders with leaflets were published in local language (Kannada) by CCRI and disseminated to agriculture extensionists and farmers in India:
 - Arabica Coffee Varieties from CCRI
 - Soil Management Practices in Coffee
 - Plant Training and Pruning in Coffee
 - Composting of Coffee wastes
 - Soil Analysis and Liming
 - Nutrition Management in Coffee
 - Management of White Stem Borer
 - Diseases of Coffee- Black Rot
 - Diseases of Coffee- Root Diseases
 - Processing of Coffee wastes
 - Management of Coffee Berry Borer
 - Management of Coffee Leaf Rust
 - Stem Wrapping Method to control white stem borer
 - New Arabica variety `Chandragiri`
- A number of short duration TV modules on coffee cultivation technologies were also prepared jointly by CCRI and ETV Kannada and aired in India
 - Coffee – Pre & Post harvest processing cherry coffee
 - Coffee – Pre & Post harvest processing parchment coffee
 - Coffee Effluent Treatment
 - Arabica Coffee Varieties (Tall varieties)
 - Arabica Coffee Varieties (Semi- dwarf varieties)
 - Robusta Varieties

- Compost
- Coffee Rust Disease
- Soil Sampling (Part-1)
- Soil Sampling (Part-2)
- Coffee (INM) (Part-3)
- Coffee (INM) (Part-4)
- Coffee- Bush training
- Coffee- Pruning
- White Stem Borer management
- Berry Borer Management in Coffee
- Propagation in Coffee - Clones (Part-1)
- Propagation in Coffee - Grafting (Part-2)

As the project included a substantial element of scientific research, various scientific publications, papers and posters were produced and presented at conferences and workshops:

- Bigirimana J., Njoroge K., Muthomi J.W., Gahakwa D., Phiri N.A., Gichuru, E.K., & Walyaro D.J., 2013. Genetic Diversity among Disease Resistant Coffee Varieties and Cultivars in Rwanda Based On RAPD and SSR Markers. *Journal of Renewable Agriculture*, 2013, 1(6): 106-112.
- Bigirimana J., Njoroge K., Muthomi J.W., Gahakwa D., Phiri N.A., 2012. Incidence and severity of coffee leaf rust and other coffee pests and diseases in Rwanda. *African Journal of Agricultural Research* Vol. 7(26), pp. 3847-3852, 10 July, 2012
- Gatarayiha C.M., Mushimiyimana S., Bigirimana J., Phiri N., 2010. Current Status and Management of Coffee Leaf Rust Disease in Rwanda in the “Proceedings of the 23rd International Conference on Coffee Science”, 2010, Bali, Indonesia 3-8 October 2010, Volume 1 and 2. ISBN: 978-1-61839-210-7. Printed from e-media with permission by Curran Associates, Inc. 57 Morehouse Lane, Red Hook, NY 12571, Curran Associates Inc. Proceedings.com
- Gichimu B.M., 2012. Field Screening of Selected *Coffea arabica* L. Genotypes against Coffee Leaf Rust. *Afr. J. Hort. Sci. (June 2012)* 6:82-91
- Gichimu B.M. and Omondi C.O., 2010. Morphological Characterization of Five Newly Developed Lines of Arabica Coffee as Compared to Commercial Cultivars in Kenya. *International Journal of Plant Breeding and Genetics*, 4(4): 238-246.
- Gichimu B.M. and Omondi C.O., 2010. Early Assessment of Growth and Yield Characters of Five Newly Developed Lines of Arabica Coffee in Two Environments and Spacing in Kenya. *Afr. J. Hort. Sci.* 3:98-111.
- Gichimu B.M. and Omondi C.O., 2010. Early Performance of Five Newly Developed Lines of Arabica Coffee under Varying Environment and Spacing in Kenya. *Agriculture and Biology Journal of North America*. 1(1): 32-39.
- Gichimu B.M. and Phiri N.A., 2010. Response of Newly Developed and Introduced Arabica Coffee Genotypes to *Colletotrichum kahawae*, the Coffee Berry Disease Pathogen, in Kenya. *12th Biennial KARI Scientific Conference*

- Gichimu B.M. and Omondi C.O., 2010. Morphological Characterization of Five Newly Developed Lines of Arabica Coffee as Compared to Commercial Cultivars in Kenya. *International Journal of Plant Breeding and Genetics*, 4(4): 238-246.
- Gichimu B.M. and Omondi C.O., 2010. Early Assessment of Growth and Yield Characters of Five Newly Developed Lines of Arabica Coffee in Two Environments and Spacings in Kenya. *African Journal of Horticultural Science (AJHS)*, Vol. 3:98-111.
- Gichimu B.M. and Omondi C.O., 2010. Early Performance of Five Newly Developed Lines of Arabica Coffee under Varying Environment and Spacing in Kenya. *Agriculture and Biology Journal of North America*. 1(1): 32-39.
- Gichuru E.K., Ithiru J.M., Silva M.C., Pereira A.P, Varzea V.M.P., 2012. Restructured sampling plan enables the characterization of more virulence genes of *Hemileia vastatrix* in Kenya. *24th International Conference on Coffee Science, San Jose, Costa Rica*.
- Gichuru E.K., Ithiru J.M., Silva M.C., Pereira A.P, Varzea V.M.P. Additional physiologic races of Coffee Leaf Rust (*Hemileia vastatrix*) identified in Kenya. *Tropical Plant Pathology Journal* (In press)
- Kathurima C.W. and Njoroge E.K., 2012. Effect of Different Shade Regimes on Coffee Quality. *24th International Conference on Coffee Science, San Jose, Costa Rica*.
- Matovu R.J., Kangire A., Phiri N.A., Hakiza G.J., Kagezi G.H. and Musoli P.C., 2013. Ecological factors influencing incidence and severity of Coffee Leaf Rust and Coffee Berry Disease in major Arabica coffee growing districts of Uganda. *Uganda Journal of Agricultural Sciences*, 2013, 14 (1): 87 – 100
- Mayoli R.N. and Gichuru E.K., 2012. Epidemiology of coffee leaf rust: Influence of shade on microclimate and ecophysiology of coffee. *24th International Conference on Coffee Science, San Jose, Costa Rica*.
- Phiri N.A., Gichimu B.M., Gatarayiha C., Kutwayo D., Prakash N., Musoli P., Agwanda C., Kimani M., Musebe R. and Oduor G., 2011. Increasing the resilience of coffee production to Coffee Leaf Rust and other Diseases in India and four other African countries. *Presented on 23rd March 2011 at ICRAF Complex, Nairobi, Kenya*.
- Surya Prakash N., Manoj Kumar M., Padmajvothi, D., Sudhakar S.B., Hanumantha B.Y., Daivasikamani S., Suresh N.R., Soumya P.R., Asha M.B., Madhura M., Divya M.H., Varzea V., Silva M.D., Phiri N., Gichimu B.M., 1910. Evaluation of Coffee Varieties Derived from Diverse Genetic Sources of Resistance for Prospective Exploitation. An International Cooperative Effort in the "Proceedings of the 23rd International Conference on Coffee Science", 2010, Bali, Indonesia 3-8 October 2010, Volume 1 and 2. ISBN: 978-1-61839-210-7. Printed from e-media with permission by Curran Associates, Inc. 57 Morehouse Lane, Red Hook, NY 12571, Curran Associates Inc. Proceedings.com

3.4.3 Carry out communication events for project stakeholders with a variety of objectives

Communication activities for farmers were centred on farmer field schools (FFSs), farmer training and training of extension staff as FFS facilitators, and this has been reported in section 3.1. The training of trainers and backstopping was done by PEA, but implementation and dissemination of information was carried out by all five participating countries. India established FFSs in non-traditional coffee growing areas in order to reach farmers who may not have as much information on coffee growing. Communication events in association with FFS included open days and exchange visits for other FFS and non-FFS farmers.

Communication events for project partners and key stakeholders included steering committee meetings held in participating countries. These were held twice per year to discuss the progress of the project and approve country plans for the following year. The PEA convened annual dissemination workshops (African Coffee Scientific Conferences) from 2009 to 2012 at the international coffee conferences organised by the Eastern and Southern Africa Coffee Association (EAFCA). These were attended by participating countries, but were also open to any interested participants in the EAFCA conferences.

The project team also participated in the 2010 International Conference on Coffee Science (ASIC) in Bali, Indonesia, where posters and papers originating from the project findings were presented (Section 3.4.2)

3.4.4 A website of project activities and outputs

A project website (<http://coffeeleaftrust.ning.com/>) linked to CFC, ICO and partner websites was developed. The ning platform was used so that all project participants could upload materials, rather than having a website controlled by one partner. Non-project participants were given access to the site via a moderator, following an unpleasant incident when the platform was not moderated.

3.4.5 Café Movel: a novel way for sharing information with farmers in India

This activity aimed to enhance information dissemination to farmers in India. The project funded the pilot phase, but the India Coffee Board has since taken over the funding of the service which has been viewed as an important channel for disseminating information to coffee farmers in the country.

Named Café Movel, the service is a helpline which farmers can call to get pre-recorded advice, or to talk to a coffee expert from the Coffee Board. Café Movel was started towards the end of the project in Karnataka Province, but since being taken over by the Coffee Board of India has been extended to other coffee growing areas in Kerala and Tamil Nadu Provinces.

The service includes a help line, answers to frequently asked questions, and alerts. During the pilot period (August to October 2013), a total of 671 calls were made. The service is continuing using the newly launched toll free telephone number (farmers will not pay for calls)

3.4.4.1 Scoping and requirement gathering

Before establishing the service, a participatory needs analysis was conducted during the month of August 2012. Focus group discussions were conducted in 3 major coffee producing zones of Karnataka; Chickmagalur, Hassan and Coorg (Chettali). The objective of the focus groups was to identify existing knowledge gaps, by analysing the key challenges they face throughout the coffee production cycle, how they meet those challenges currently, and what better solutions they would hope or expect to be delivered via the mobile channel. It was noted that some solutions can be delivered through mobile phones whereas others, like demonstration of a technology, are not appropriate. Findings from this activity led to the formulation of messages on different coffee subjects so that farmers were able to listen to areas of interest when they called.

3.4.4.2 Developing Coffee Knowledge Bank

A knowledge bank was developed as a resource for easy access by the coffee experts running the telephone helpline. The primary source of content was the coffee production manual developed by the Coffee Board. Additional information was provided by CABI (e.g. Direct2Farm Knowledge Repository). A research associate was recruited to prepare and compile content from these sources, which was then validated by the respective subject matter specialists from the Coffee Board. The content included information on improved coffee varieties, pest management, fertiliser application, coffee prices, shade management, irrigation, input purchase, bush handling, weed management, harvesting, on-farm processing, pollution management, soil analysis, seed procurement, primary nursery, crop estimates (post-blossom), farm gate, pest and disease surveillance, pest estimate (post-monsoon), water conservation, crop estimate (final), seed procurement and distribution, advisory on management, and advisory on cultivation.

3.4.4.3 Developing the mobile and web applications

The mobile application was developed, and Café Movel services were delivered through a local Karnataka mobile telephone number (+91-9019191000). A brief user manual was prepared in local language and English (Figure 18), so that farmers were aware how to call and get advice or ask questions.

CAFÉ MOVEL: THE MOBILE EXTENSION SERVICE FROM COFFEE BOARD OF INDIA & CABI

What is Café Móvel?

Café Móvel is a Mobile Extension service for the coffee producers of India, brought to you by the Coffee Board of India and CABI. Now you are connected to the Coffee Board, 24x7, through your mobile phone, by just dialing the toll free number 1800-103-1901.

How to Register in Café Móvel?

1. Dial 1800-103-1901 from any mobile phone
2. If you are dialing for the first time, the system will request you to give consent for sending you SMS alert (as per TRA Rule)
3. PRESS 1 two times to give consent
4. After this, the system will ask you a few more questions
5. Please answer by PRESSING the appropriate keys in your phone
6. This information will help us to provide you more customized information

How to use Café Móvel?

1. Dial 1800-103-1901 from any mobile phone
2. Select language: PRESS 1 FOR KANNADA OR PRESS 2 FOR ENGLISH
3. PRESS 3 for Frequently Asked Questions (Advisories on coffee production in south west monsoon area)
4. PRESS 4 for Alerts (Special outlets on weather, market price, coffee schemes etc)
5. PRESS 5 to listen to the Answers to Recorded Queries (answers to the questions that you posted to the Coffee Board experts)
6. PRESS 6 to talk to Experts (you can talk to any experts from the Coffee Board directly)



**Dial
1800-103-1901
For
Café Móvel
Toll Free**

- Listen to advisory from Coffee Board Expert
- Get regular SMS alerts (Weather, Market Price etc.)
- Post any query to Coffee Board and get answer
- Talk to Coffee Board Experts directly; 24x7



**For further assistance; call to 98XXXXXXX*
*Not toll free**

Or Contact
Dr.
Central Coffee Research Institute, Coffee Research Station - 577 117, Chikmagalur Dist.

How to listen to Answers to Recorded Queries?

Café Móvel is like a private discussion forum for you and the Coffee Board experts. You can post a query by recording a note by PRESSING in PAQ Section on In-Headline Section. The recorded query is sent to the expert and when the expert answers your query, you will receive a SMS with a UNIQUE TOKEN ID. The token ID will ensure that discussion between you and the expert is CONFIDENTIAL. Dial 1800-103-1901, select language and PRESS 5 to access the Answer to Your Query. The system will ask the TOKEN ID and after you enter the token ID, it will play the answer to your query.

How to talk to Experts?

Sometimes talking directly to an expert from Coffee Board is all you want. But you may not remember the contact number of the expert. Don't worry, using the Talk to Expert function of Café Móvel, you can talk to any expert, anytime. To access this:

1. Dial 1800-103-1901
2. Select Language
3. PRESS 4 for Talk to Expert
4. The available options are:
 - 1) Insect Pests
 - 2) Diseases
 - 3) Crop Production & Coffee Quality
 - 4) Coffee Varieties
 - 5) Coffee Board Services
 - 6) Weather Information

Once an option is selected, the call will be connected to the respective available expert. In case no expert is available, you will be instructed to record your query. To record query, listen to the BEEP sound and start recording after the BEEP. To end Recording, PRESS 1 a key and system will PLAYBACK the recorded query. If you are satisfied, PRESS 1 or PRESS 0 to RE-RECORD. After PRESS 1, the query is sent to the respective expert and the expert will be notified. You will receive a SMS with a TOKEN ID as soon as your query is answered by the expert. The answer can be listened at the Answers to Recorded Query section of Café Móvel.

Common Questions...

- Which languages are supported in Café Móvel? Kannada & English
- Is Café Móvel available only on Smart Phones? Café Móvel services can be accessed from any phone.
- Can I dial Café Móvel number from outside Karnataka? Yes, you can dial from anywhere in India and it will remain toll free.
- What to do if I do not want to receive SMS from Café Móvel? Send an email to subscriptions@cbi.gov.in with DND written in the subject line.
- What happens if my phone number gets changed after I registered in Café Móvel? You have to register again in Café Móvel with the new number.
- What to do if I want to remove my registered phone number from Café Móvel? Send a mail with subject line "Unsubscribe" to subscriptions@cbi.gov.in

Figure 18. User manual for helping farmers to use Café Móvel in India

3.5 Project management and coordination

This component had one indicator in the logical framework.

- Project outputs were delivered according to the log-frame as reported in sections 3.1 to 3.4. Specific project management and coordination activities are described below, and some of the challenges and lessons learned are listed in section 5.

The PEA had an overall project coordinator, while each participating country had a national project coordinator and a number of team members who managed local activities. The following activities were led by the PEA.

3.5.1 Advise on project operational procedures based on CFC formats

A project launching workshop was held in India in 2008 where representatives of all partners attended. Project objectives and expected outputs were shared with the partner representatives.

Workshops were also held in each country at the beginning of the project to take the country project implementation teams, through project activities they were to implement.

The PEA established administrative and accounting procedures and provided training for local counterparts before the project started in each country – this included opening project accounts or arranging for accounts which could be used for project finances, training accountants in processes for fund utilisation and accounting for project resources.

The PEA also advised PIAs on operational procedures using CFC formats. Advice covered both technical and financial aspects of project implementation. This included a training session for all project accountants from participating countries, that was held in Nairobi. This greatly increased the efficiency of project implementation.

Project staff were given technical backstopping where they were having difficulties in implementing the project activities. Advice was also communicated through emails or meetings, such as annual project planning meetings.

3.5.2 Assist PIAs to prepare necessary documentation

PIAs were assisted during the project period in preparation of budgets, work plans and progress reports, which were reviewed during the annual planning workshops organised by the PEA in collaboration with a hosting country. Reports were submitted to the supervisory body (ICO) and the donor (CFC) every six months throughout the project

Development of work plans and budgets was also done at the end of every project year in preparation for the coming year. The PEA provided templates for both work plans and

budgets and assisted PIAs in their preparation before being discussed in annual planning workshops.

3.5.3 Liaise between project donors and implementers and arrange exchange visits

CABI linked CFC/ICO and PIAs throughout the project period. This involved responding to CFC/ICO request for information from PIAs, and PIAs requesting for funds to be used for an alternative activity. For example, India wanted to use funds budgeted for irrigation on purchasing a project vehicle because they had a functional irrigation system already.

Exchange visits were also arranged for participating scientists. The coffee breeder for Kenya went to India to learn about marker assisted selection, while the coffee breeder from Rwanda went to Kenya for attachment at the Coffee Research Foundation as part of his MSc studies at University of Nairobi. The team from India also visited Coffee Research Station in Kenya and saw how the Indian selections were doing in Africa. One member of staff came to Kenya too for attachment.

3.5.4 Monitor project progress and report on inputs (disbursements), activities undertaken and outputs achieved

Monitoring of progress included the following:

- Monitoring technical progress and implementation of agreed activities. Where the PIA was having problems, CABI staff directly supported the PIAs in implementing activities such as was the case in Rwanda.
- Monitoring financial progress. This included monitoring and assisting PIAs in preparing and submitting financial claims on completed activities. Project accountants were also trained by the CABI accountants where country project accountants were facing problems.
- The PEA coordinated and participated in the midterm review process, and contributed to the midterm review report.

3.5.5 Assist PIAs and collaborators with planning and co-ordination of activities aimed at providing uptake pathways for outputs

Planning and coordination of uptake pathways included the follows:

- PEA personnel supported partners in training trainers for running FFSs, the main uptake pathway for project outputs to farmers
- The PEA also organised workshops/coffee scientific conferences every year in February which were held together with the EAFCA annual coffee conferences
- Progress reports were prepared every six months, in July (six monthly reports) and January (annual reports) each year

3.5.6 Convene an end of project workshop

The PEA, in collaboration with Coffee Board of India, organised and held the end of project workshop from 18 to 22 March 2013 in India. The workshop report is provided in the Appendices.

IV. Dissemination of Project Results

Dissemination of project results was built into the other activities and has been reported above, especially in section 3.1 (dissemination to farmers through FFS) and section 3.4 (dissemination materials and pathways for farmers, extensionists and scientists).

V. Lessons Learned

5.1 Development Lessons

Uganda was hit with two droughts which necessitated replanting of the varietal trials and caused some delay. In retrospect it would have been good to have provided for irrigation at the research stations so that the impact of the droughts would have been reduced.

Capacity building was not included as a main component, but this was found to be important as part of the exit strategy, particularly as some of the project's activities included highly technical methods. Thus the PEA included capacity building activities such as training for partner staff in technical areas, as well as in project management, which was very helpful. A few of those sponsored on higher degrees have still not completed their studies by the end of the project, and this could have been avoided if capacity building activities had been included in the project design.

Another lesson relating to project design is that although there were a number of indicators identified at purpose level in the logical framework, there were few activities that would enable the indicators to be evaluated. Some information for evaluating indicators can be collected during project activities, but in other cases specific studies need to be planned and budgeted for.

5.2 Operational Lessons

The project team was technically sound, and did an excellent job in implementing the project with guidance from the PEA. However, some countries needed more backstopping than others, so in future projects it would be useful to include a reasonable amount of resources for backstopping partners.

On some occasions there were delays in submission of financial reports and claims from the PIAs. This was addressed by inviting project accountants to a training session in Nairobi. This resulted in improvements, but had not been budgeted for when the project was developed.

There were also delays in getting final technical reports from partners in some countries. To avoid this in the future it might be appropriate to include a writeshop for

the final technical report so that all partners come together to do the writing up as a team.

Communication was sometimes a challenge. In some instances partners did not respond to email communication or telephone, necessitating the PEA to visit the country, again adding to backstopping costs. Part of this challenge was due to staff turnover in implementing organisations, with inadequate handover procedures. Such weaknesses cannot easily be managed in a project of this nature, except by ensuring there is adequate provision for the PEA to make regular visits to PIAs.

VI. Conclusions and Recommendations

3.1 Identification of needs and resources: rural community responses to CLR disease and the sourcing and production of coffee genetic material

- A range of stakeholders were identified in all countries following participatory approaches. This resulted in a network of stakeholders who were successfully involved in the project in various roles.
- CLR was identified through socioeconomic surveys as the most important constraint to coffee production for most farmers especially in Africa.
- Lack of inputs e.g. fertilizer and pesticides was also found to be one of the most important constraints especially in Zimbabwe.
- Other diseases, such as CBD, black rot, root rot, berry blight (*Fusarium latelitim*), bark disease (*Fusarium stilboides*), coffee trunk canker, and insect pests such as white coffee stem borer and berry borer were also identified as important constraints.
- Although most smallholder farmers were able to recognize some of the symptoms of CLR, in Africa most were not aware of the trends of CLR incidence, mode of transmission, or field conditions and seasons when CLR is more severe.
- Many farmers in Africa were not aware of varieties resistant or susceptible to CLR, but some farmers especially in India and Kenya had some knowledge.
- Many farmers do little or nothing to control CLR, except in India and to some extent in Kenya, where some farmers use resistant varieties such as Ruiru 11 (in Kenya) and Chandragiri (in India).
- The majority of farmers did not use chemical control due to lack of information, limited financial resources, and untimely availability of fungicides.
- In general there was a limited use of farm inputs, clearly evidenced by the poor state of coffee bushes in surveyed countries.
- At the beginning of the project, availability of resistant coffee varieties was limited in most countries.
- For improved coffee production, there is need for resistant varieties to be made widely available. In addition, environmentally friendly fungicides are required for the control of CLR and CBD, and other improved coffee production technologies which contribute to the general health of coffee plants.
- The project exit strategy included capacity building that was not originally planned. The enhanced capacity will support continuing coffee breeding programmes.
- Co-financing from participating institutions also contributed to sustainability, and several cases of additional co-financing and support after the end of the project have occurred.
- The project also provided infrastructure, such as improved nurseries, renovated laboratories equipped with modern equipment, and seed gardens for improved and old varieties that can be used on a continuing basis.
- We thus conclude that the prospects for sustainability are good.

- Farmer facilitators were trained so that they could facilitate farmer mobilisation through farmer field schools (FFSs) which were run in all countries.
- Through the FFS, farmers were mobilised to participate in and host on-farm trials (varietal and fungicide screening trials). Farmers also visited trials to see the results as they progressed.
- Open days and exchange visits promoted sharing and dissemination of information.
- Farmers were pleased with what they learnt from FFS which they stated increased their knowledge of coffee farming, and led to economic as well as social benefits.
- Participating countries identified 31 locally available genotypes for inclusion in varietal trials for screening for resistance to CLR.
- The four African countries imported Indian Selections 5A and 6 using existing quarantine and IP provisions in Africa and India.
- Due to potential risks of accidentally introducing coffee berry disease to India, no varieties were introduced to India from Africa.
- India established new seed plots to meet the growing demand after the release of the new variety, Chandragiri in 2007. In addition, India established seed blocks for Selections (SIn) 5A, SIn 5B, SIn 6 and SIn9.
- A total of 15,933 farmers received seed in India in addition to seed being used for trials in India and Africa (SIn 5A and 6). 50,050 kg of seed were produced during the project in India.
- In Africa, though not originally planned, seed gardens were set up or maintained in Kenya, Uganda and Zimbabwe. Uganda produced seedlings of the coffee wilt resistant Robusta varieties through tissue culture for developing mother gardens.
- Kenya managed their seed gardens in order to make more seed available to farmers; a total of 2,993 kg of seed was harvested from four gardens. Zimbabwe produced 89,721 seedlings of 8 coffee varieties (Selection 5A, Selection 6, SL28, Yellow Catuai, K7, Catimor 129, Catimor 128 and Catimor F6).
- Kenya, Uganda and Rwanda all developed homogenous seed plots for existing resistant and new varieties.
- Nurseries were renovated or established in all participating countries resulting in the production of over one million seedlings during the project.
- Demand for improved CLR and CBD resistant varieties was high in all countries. In Kenya demand greatly exceeded supply following a well-publicised launch of CLR-resistant Batian varieties.

Recommendations

- Farmers have various constraints, and where possible these should be taken into account in breeding programmes. Selection for resistance to a single disease may often be inadequate for farmers' needs.
- Seed and seedling production facilities and activities set up and supported by the project need ongoing investment to maintain or expand their production of resistant varieties.
- Demand for disease resistant varieties is high, and the public sector organisations responsible for coffee seed and seedling production need to respond to this

demand. Where they cannot meet demand, they should investigate opportunities for involving the private sector, as Uganda has done for clonal, wilt-resistant materials.

- Farmer field schools do not need to run indefinitely, so trained facilitators should be used to start field schools in new areas, especially where resistant varieties or good husbandry practices are not being used.
- Advanced farmers from the field schools can also be facilitated to start FFS on their own (as was done during the project) as a way of disseminating information and technologies.
- Other communication channels that already exist should be used to reach farmers, such as plant clinics. Appropriate information needs to be provided to the channels.

3.2 Conservation and identification of coffee varieties and disease races

- In all countries the conservation status of materials was poor, and most of the collections in field gene banks needed rehabilitation or management.
- Most collections are susceptible to CLR leading to severe defoliation and infestations by insect pests.
- Many collections from within and outside the countries have been rehabilitated or rejuvenated, characterised and documented. Zimbabwe replanted and conserved 56 collections, and characterised 16. Uganda moved 120 collections from the current location to two new sites. Rwanda rehabilitated and maintained 182 germplasm collections in field gene banks. India maintained 250 collections which were established just before the project.
- India produced a monograph of all their collections, incorporating data in line with the descriptors for coffee prepared by Bioversity International (IPGRI), Rome.
- Rwanda's and Uganda's collections were uncharacterized, so characterization has started.
- Countries instituted conservation strategies for germplasm collection, which included spraying against CLR, weeding, applying fertilisers, periodic rejuvenation of collections, gap filling, top working, and managing pests. Kenya published their germplasm conservation strategy.
- All countries are now conserving their existing germplasm as well as the newly introduced Selections 5A and 6.
- Facilities were established at the Central Coffee Research Institute in India for genetic characterisation of germplasm and hybrids in the country.
- India developed a marker strategy which helped in establishing systems for conserving coffee collections and development of methods that can be used to screen them for disease resistance.
- Coffee leaf rust races were determined in all project countries. India identified two new races which have not yet been named. Kenya found 6 races not previously recorded there, while Zimbabwe found one.
- The project validated and applied two SCAR markers closely linked to the S_H3 resistance gene in marker assisted selection and breeding programmes.

- Identification of cultivar specific markers for tracking homogeneity of the varietal populations was carried out. The SRAP markers were found more efficient for detecting polymorphism in Arabica than the other methods tested.
- The SCAR markers linked to S_H3 gene were used for maintenance breeding of S.795 and marker assisted selection for integrating S_H3 gene with resistance genes of Robusta origin (S_H6, S_H7, S_H8, S_H9).
- SCAR marker analysis was used to select and self elite plants (with high individual plant yield, field tolerance to CLR, high bean quality traits)
- The selfed progenies raised from individual F₁ plants were established in four different locations in India for simultaneous evaluation and selection for further exploitation.
- Eleven genotype specific SRAP markers were validated and are useful for assessing the homogeneity of seedling progenies.
- Three SRAP markers specific to Chandragiri were identified which can distinguish this variety from other semi-dwarf Arabicas.
- As a pre-emptive breeding strategy, efforts were made to transfer the S_H3 gene to the Chandragiri variety to provide broad spectrum of resistance.
- Elite plants homozygous to S_H3 were selected in S.3827 and used in reciprocal crosses with Chandragiri. Six progenies were established in the field.
- Marker assisted selection has thus been successfully employed for the first time in coffee breeding in India, leading to further opportunities for pyramiding rust resistance genes.
- The project provided grants for strengthening the marker lab facilities at CCRI in India and a state-of-the-art seed laboratory.
- Capacity of national scientists in Africa was built in the areas of marker assisted selection, breeding, pathology, and extension through short courses and post graduate degrees.
- CCRI worked with the Government of India's Protection of Plant Varieties and Farmers Rights Authority to develop DUS criteria for coffee for registering and protecting the coffee varieties developed by CCRI, and to create awareness on IP related issues.
- All countries used the existing IP policies and quarantine regulations in sharing genetic materials. Kenya (Coffee Research Foundation) further developed their IP Policy under the legal and professional guidance of Kenya Industrial Property Institute (KIPI) and the Kenya Plant Health Inspectorate Services (KEPHIS).

Recommendations

- Rehabilitated, rejuvenated and relocated germplasm collections need to be maintained according to the defined strategies.
- Where characterisation of collections was not completed at the end of the project, this should be done as a priority.
- A way needs to be found to set up differentials in Africa based on the materials at CIFC, Portugal.
- CLR race audits should be carried out periodically in each country on which to base continuing breeding programmes.

- Scientists already trained in marker assisted selection should find ways of utilizing their skills. In Kenya facilities are available at CRF, so scientists from other countries could visit to undertake the work (a student from Rwanda has already done so).
- In some countries facilities are available in other national institutions. Coffee research organisations could investigate the possibilities for using those facilities for coffee work.
- Additional capacity in marker assisted selection should be developed in all the participating countries.
- Marker assisted selection should continue to be used as a basis for pyramiding resistance genes, so that a wide range of CLR resistant varieties can be developed with other traits desired by farmers (such as resistance to other diseases and good yield and quality characteristics).

3.3 Field trials on farm and station

- Varieties from India were screened for resistance to CLR. Selection 6 was resistant in all participating countries.
- Selection 5A was generally resistant but with some individual bushes susceptible, due to the variety being a composite.
- In Kenya Crosses 8, 22, 23, 27, 30, were resistant to CLR and gave high yields. Although CBD pressure was low, earlier studies showed that the varieties are also resistant to CBD.
- Three CLR resistant varieties were officially released in Kenya, named Batian varieties 1,2 and 3.
- Selection 6 also has potential field tolerance to Bacterial Blight of Coffee (BBC).
- Catuai x HDT, Colombia, Selection 5B, Chandragiri, and Selection 5A were all resistant/tolerant to CLR in India.
- In Uganda varieties NG9257 and Elgon CB, as well as Selections 5A and 6, were resistant to CLR.
- Selections 5A and 6 gave consistent CLR resistance in Rwanda.
- Line 13683/35 and Selection 6 were resistant to CLR in Zimbabwe, and were far better than the known resistant commercial variety, Catimor 129.
- Selection 5A and 6 are early maturing and show potential of being high yielding. Both Selections are tall statured similar to SL28, a tall commercial variety in many countries, but which is susceptible to both CLR and CBD.
- Selection 5A and 6 have good cup quality in comparison with Ruiru 11 and SL28 in Kenya, and Selection 6 was better in quality than the commercial variety BM 139 in Rwanda.
- In general the weather conditions were too hot for the development of CBD in Africa during the project, so most countries did not have an opportunity to screen for CBD resistance. Screening was done at CIFC in Portugal and in Rwanda.
- CLR infection did not reach high levels on coffee bushes under shade and this could be attributed to an “umbrella” effect. But coffee under shade had potentially lower yields than the coffee under full sun.

- Among the antifungal botanicals and other bioagents screened in India, *Bacillus brevis* was as good at controlling CLR as the currently recommended fungicide regime of Bordeaux mixture.
- Alto (cyproconazole) was the most effective fungicide for controlling CLR in Rwanda and Zimbabwe. It was therefore registered by the National Agriculture Export Board (NAEB) of Rwanda for use in controlling CLR.
- In Uganda Orius (tebuconazole) was the most effective fungicide for CLR control.
- Studies on fungicides in Kenya showed that use of Copper (Copper oxychloride) plus Daconil (Chlorothalonil) recorded the best yield increase above the control.

Recommendations

- Single tree selection should be carried out on Selection 5A in African countries.
- Selection 6 can be multiplied and grown without further selection; a strategy is required for meeting the expected demand.
- More studies could be carried out with *Bacillus brevis* in India, especially under high disease pressure in order to confirm its effectiveness and safety to non-target organisms. Possible commercialisation paths should be considered.
- Scientists in Africa could also screen indigenous microflora as potential biocontrol agents for CLR and CBD.
- Where a CLR infestation has already occurred, cyproconazole or tebuconazole can be considered as a single curative spray, provided safety measures can be taken. Copper can be used for preventative purposes.
- Work to find lower risk fungicides that are cost effective should continue.

3.4 Scientific management, information systems and communications

- A communication strategy was developed and used in communicating among project partners.
- A range of printed information materials were produced and disseminated for increasing knowledge among farmers and extensionists. The information materials included posters, folders with leaflets, and a coffee recommendation handbook.
- Short duration TV modules on coffee cultivation technologies were prepared and aired in India by ETV, covering a wide range of topics in coffee production and processing.
- Face-to-face dissemination and upscaling activities centred on farmer field schools (FFSs). India established FFSs in non-traditional coffee growing areas where farmers had less knowledge.
- Information and knowledge sharing between different actors took place at open days and exchange visits among farmer field schools.
- Scientific papers and posters from the project research were published and presented at conferences and workshops by the project team members. In Kenya a coffee atlas was produced, and in India a monograph on coffee collections was published.

- Dissemination workshops (African Coffee Scientific Conference) were held annually at Africa Fine Coffee Association (AFCA) conferences from 2009 to 2012.
- The project team presented papers at the 2010 International Conference on Coffee Science (ASIC) in Bali, Indonesia.
- A project website (<http://coffeeleaftrust.ning.com/>) linked to CFC, ICO and partner websites was developed and used by project members and other interested parties to share information from the project.
- Project steering committee meetings were held twice per year in each country to share information, discuss the progress of the project and approve country plans for the following year.
- The project funded a pilot of an innovative helpline service for coffee farmers in India named Café Movel. Services were delivered through a local Karnataka mobile telephone number, supported by a knowledge bank. The Coffee Board of India has taken over Café Movel and has extended it to two other provinces where coffee is grown.

Recommendations

- Communications to farmers should continue to emphasise the use of resistant varieties (including information on where they can be obtained), and good agricultural practices for growing healthy crops.
- Extension materials and information already prepared should be disseminated through whatever local channels are available. For example in Kenya and Rwanda advice on CLR is being disseminated via plant clinics.
- Scientists involved in the project should ensure that all of their findings are published in the scientific literature.
- Publications such as the atlas, monograph and coffee handbook should be made freely available digitally.
- When launching new varieties, high profile events such as the launch of Batians in Kenya should be used.
- The cost effectiveness of Café Movel needs to be monitored, but early signs are that it is worth extending more widely. Similar approaches could be investigated in African countries, perhaps by linking with other initiatives testing the use of mobile phones for agricultural extension.

Appendix I. Logical Framework

<i>Narrative summary</i>	<i>Verifiable indicators</i>	<i>Means of verification</i>	Assumptions
<p><i>Broad goal:</i></p> <p>Contribute to sustainable coffee production and increase producer income and foreign exchange earnings.</p>	<p>Losses due to rust & other diseases decrease & associated returns from coffee production increase.</p> <p>Long-term increase in the frequency/area of disease-free plants in coffee farms in the participant countries</p> <p>Increased foreign exchange from coffee exports in the project countries</p>	<p>Reports & databases from ICO, FAO, Coffee Boards & coffee farmers associations of participating countries.</p> <p>Reports from socio-economic surveys.</p> <p>National reports on foreign exchange earnings from coffee</p>	<p>Coffee remains a valued commodity in the target areas.</p> <p>Market, trade & other production factors do not compromise expected cash gains.</p> <p>Increased returns used to improve livelihoods of farmers.</p> <p>CLR & CBD continue to be strong constraints for coffee production.</p> <p>Other constraints do not overcome benefits from disease control.</p>
<p><i>Project Purpose:</i></p> <p>Reduce economic and environmental costs of disease management for smallholder coffee farmers by reducing the crop and quality losses caused by the diseases CLR and CBD, and resources spent on expensive chemicals, especially in regard to smallholder, coffee-based farming systems.</p>	<p>New technologies and practices are spread outside the project areas</p> <p>Chemical pesticide sales are reduced while sales of environmentally friendly products increase</p> <p>Increased volumes of coffee produced by farmers in project area</p> <p>Disease incidence decreases in project areas</p>	<p>Reports from National extension systems</p> <p>Reports from sales figures of pesticides and environmentally friendly products</p> <p>Survey to score field incidence of disease in the demonstration plots & use of botanicals & bio-agents</p> <p>Reports of socio-economic surveys</p> <p>Media articles/ reports/promotional materials</p> <p>Publications in peer-reviewed journals.</p> <p>Workshop proceedings</p> <p>Reports on demand for new seed & active from commercial nurseries</p>	<p>Socio-political situation remains stable & favourable for wider uptake/adoption of project outputs</p> <p>Disease resistance built in to new materials remains effective against CLR for a long time.</p> <p>Integration of botanicals/ bio-agents & optimised disease control will help small growers in the long run.</p>

Outputs			
<p>1: Identification of needs and resources: rural community responses to coffee disease and the sourcing and production of coffee genetic material</p>	<p>1.0 Farmer needs assessment prepared</p> <p>1.1 Viable rural-based seed/seedling production system developed and operational in each participating country.</p> <p>1.2 Seed orchards for mass propagation of high yielding good quality and rust resistant lines established in each participating country by end of year three.</p> <p>1.3 New networks of coffee sector actors operational in project areas</p> <p>1.4 Relative importance to coffee production of CLR, anthracnose & other coffee diseases clarified & limitations to technology uptake by farmers identified</p> <p>1.5 Coffee sector researchers interacting with farmers, extension staff and other actors to generate new approaches, trials and practices to respond to needs</p>	<p>1.0 Farmer needs assessment report</p> <p>1.1 Project reports</p> <p>1.2 Mid-term review workshops & report</p> <p>1.3 Mid-term review workshops and report</p> <p>1.4 Project technical reports</p> <p>1.5 Country & coffee institute reports</p>	<p>New technologies and practices identified are taken up by farmers</p> <p>Interactions between coffee sector stakeholders contribute to uptake of new technologies and practices</p> <p>Implementing institutes remain sufficiently committed & stable to project outputs lead to project purpose</p> <p>National institutes are able to sustain conservation measures put in place</p> <p>Agreements on protocols to address IPM and quarantine issues are adequate to facilitate plant material transfer between countries</p>

<p>2: Conservation and identification of coffee varieties and disease races</p>	<p>2.1 Germplasm attributes & conditions are understood; material is sampled & conserved as appropriate</p> <p>2.2 Suitable field & laboratory facilities identified & utilised in participating institutions & countries</p> <p>2.3 Genetic/biochemical marker assisted protocol developed, markers associated with disease resistance identified & skills and knowledge of scientist to use the markers upgraded</p> <p>2.4 Instruments for sharing of coffee germplasm between Indian and partners in Africa formulated and endorsed by collaborating countries</p>	<p>2.1 Catalogue of disease resistant varieties</p> <p>2.1/2.2 Strategy paper prepared on Approaches to Conservation of Coffee from the Wild & within Country Germplasm Banks by month 9</p> <p>2.3 Methods description</p> <p>2.3 Researchers use methods for other purposes</p> <p>2.4 Report on any sharing agreements</p>	
<p>3. Field trials on farm and station</p>	<p>3.1 Final selection of lines combining resistance to CLR with good quality and high yields completed by end of year four and selected varieties popularised among smallholder farmers</p> <p>3.2 Effective and environmentally friendly chemicals and/or botanicals identified and methods for incorporation in CLR control developed and promoted by end of year three</p> <p>3.3 Improved disease management options available & promoted by end of year 4</p>	<p>3.1, 3.2, 3.3 Survey of farmer use of new technologies and practices</p> <p>3.2 Cost-benefit analysis of integration of botanical/bio-agent in disease control</p> <p>3.4 Policy brief with recommendations on IP and quarantine issues</p>	
<p>4. Scientific management, information systems and communications</p>	<p>4.1 Communication strategy developed by end of Y1</p> <p>4.2 Communication activities implemented with appropriate stakeholders</p> <p>4.3 Practical recommendations to farmers prepared and distributed</p>	<p>4.1. Project report outlining communication strategy</p> <p>4.1/4.2 Project reports on communication activities, including workshops, meetings, FFS activities etc.</p> <p>4.1/4.2/4.3 Information materials including FFS learning activities, technical briefs and policy briefs videos, demo plots, etc.</p>	
<p>5. Project management and coordination</p>	<p>5 Project delivers outputs according to log-frame or to a modified logframe agreed with donor</p>	<p>5 Annual and final project reports</p>	
<p>Inputs: Activities</p>			

<p><u>1.1:</u> Undertake a stakeholder analysis and plan interactions with most significant stakeholders</p> <p><u>1.2:</u> Carry out community-based surveys assess the impact of coffee leaf rust and other diseases and understand current practices and responses</p> <p><u>1.3:</u> Mobilise key stakeholders, including farmers to participate in farm-based activities and establish linkages with FFS.</p> <p><u>1.4:</u> Identify locally available genetic material for trial, contact and obtain new sources of coffee seed material.</p> <p><u>1.5:</u> Manage and expand seed production sites in India for supply of F1 material for trials, through maintenance of existing seed orchards and establishment of new extensions.</p> <p><u>1.6:</u> Introduce new material, subject to quarantine and IP provisions.</p> <p><u>1.7:</u> Support and develop the nursery production facilities (mother gardens) in Africa and India</p> <p><u>1.8:</u> Document expected demand for improved varieties through solicitation of farmer groups and other stakeholders,</p> <p><u>1.9:</u> Develop homogenous seed production sites for future supply of genotypes and facilitate the establishment of efficient seed/seedling distribution networks.</p> <p><u>1.10:</u> Harvest, process and distribute seed to farmers at selected trial sites.</p>	<p>Component 1: (US\$ 431,382) Identification of needs and resources: rural community responses to CLR disease and the sourcing and production of coffee genetic material</p> <p>Component 2: (US\$ 894,832 total) Conservation and identification of coffee varieties and disease races.</p> <p>Component 3: (US\$ 986,857 total) Field trials, capacity building and establishment of protocols for sharing planting materials</p> <p>Component 4: (US\$ 916,482 total) Scientific management, information systems and communications</p> <p>Component 5: (US\$ 542,850 total) Project management and coordination:</p> <p>Contingency (5%) = US\$ 79,408</p>	<p>Stakeholder analysis</p> <p>Workshop reports</p> <p>Annual project reports</p> <p>Import permits showing exchanges of material between countries</p>	<p>Coffee sector stakeholders are willing to interact and mechanisms can be found that allow each stakeholder to achieve their goals while contributing to those of others.</p> <p>Farmers are willing participants in the process established by the project</p> <p>IPR issues & solutions to differing stakeholder perceptions & policies can be overcome by an agreed IPR protocol</p> <p>Suitable laboratory, screen house & other facilities are available</p> <p>Suitable on-station & on-farm sites can be identified & accessed</p> <p>Seed production areas in India produce hybrid seed to quantity, quality & time specifications</p> <p>India is able to supply material of the original parents to partner African countries</p>
<p><u>2.1:</u> Identify the conservation status of coffee collections and determine the constraints to sustainable conservation.</p> <p><u>2.2:</u> Develop an integrated conservation strategy for existing collections and material secured from the wild in Uganda</p> <p><u>2.3:</u> Survey to establish occurrence of new CLR races, collecting CLR isolates and determining races in Portugal.</p> <p><u>2.4:</u> Nursery evaluation of coffee hybrids against CLR races (and CBD where appropriate) and identification of genetic markers associated with disease resistance</p> <p><u>2.5:</u> Build capacity of country partners in marker assisted selection</p> <p><u>2.6:</u> Address IP and quarantine issues to ensure mutually agreed and safe transfer of genetic material.</p> <p><u>2.7:</u> Protocols to facilitate horizontal exchanges of genetic material</p> <p><u>2.8:</u> A IP communication strategy involving awareness raising and policy influencing activities, as needed.</p>		<p>Report on status of coffee report</p> <p>Report listing CLR races</p> <p>Trainee reports</p> <p>Annual project reports</p>	<p>Security and social conditions permit the implementation of conservation measures.</p> <p>Wild coffees can be found and viable methods found for their <i>in-situ</i> or <i>ex-situ</i> conservation</p> <p>Relevant research capacities utilised & maintained in participating countries</p> <p>No constraints to travel or exchange affect the training programme</p>
<p><u>3.1:</u> Develop field trial design, data</p>		<p>Experimental protocols</p>	<p>Improved disease control</p>

<p>collection and analysis plan</p> <p><u>3.2:</u> Nursery trials to evaluate seedling growth/vigour under a range of ambient conditions.</p> <p><u>3.3:</u> On-farm and on-station trials to evaluate CLR and other disease resistance, agronomic performance, yield and cup quality of hybrids under a range of on-farm and on-station conditions and economic payback time for farmers switching to the new variety.</p> <p><u>3.4:</u> In India, on-station trials to identify anti-fungal botanicals and other bio-agents</p> <p><u>3.5:</u> In Africa, participatory on-farm trials to test alternative fungicides already showing promise</p> <p>3.6 Analyse data obtained from trials</p>		<p>Project reports and papers</p> <p>Workshop reports</p> <p>Material transfer protocols</p>	<p>methods withstand climatic vagaries</p> <p>Farmers are willing to participate in host trials & demonstration plots</p> <p>Other stakeholders contribute to evaluation of trials</p> <p>New disease control measures will be effective in consonance are amenable to small grower lifestyle and capabilities</p> <p>IPR issues are resolvable</p>
<p><u>4.1:</u> Develop a communication strategy to allow for share information amongst the different actor groups</p> <p><u>4.2:</u> Prepare information materials for use in communication strategies with farmers and other coffee stakeholders, including learning activities for FFS groups</p> <p><u>4.3:</u> Carry out communication events for project stakeholders</p> <p><u>4.4:</u> Develop a web based resource of practical knowledge for coffee farming.</p>		<p>Information materials</p> <p>Project papers and reports</p> <p>Report on communication strategy</p> <p>Workshop and other communication event reports</p> <p>Website</p>	<p>Project is able to generate and identify new knowledge that is relevant for the different stakeholders</p>
<p><u>5.1</u> Establish administrative and accounting procedures and train local counterparts</p> <p><u>5.2</u> Advise on operational procedures and initiate consultancies where necessary</p> <p><u>5.3</u> Assist PIs and ICO to prepare necessary documentation, including budgets and work plans.</p> <p><u>5.4</u> Assist organisation of initial meeting of key project partners including PI representatives</p> <p><u>5.5</u> Liaise between project donors and implementers and arrange exchange visits</p> <p><u>5.6</u> Monitor project progress and report on inputs (disbursements), activities undertaken and outputs achieved (to include mid-term impact review and expenditure audits).</p> <p><u>5.7</u> Assist PIAs and collaborators with planning and co-ordination of activities aimed at providing uptake pathways for outputs</p> <p><u>5.8</u> Prepare regular progress reports, mid-term evaluation report, annual accounts, audits and project completion report.</p>		<p>Procedure manual</p> <p>Quarterly financial reports</p> <p>Progress reports, mid-term evaluation report, annual accounts and audits, project completion report</p>	<p>Financing from all sources made on a timely basis in tune with proposed activities & annual work plan, budget etc.</p> <p>Personnel, including external consultants, competent in required skills can be identified & commit to project activities</p> <p>The PEA & collaborative institutions co-ordinate & execute project efficiently.</p> <p>All project participants remain committed to project purpose.</p> <p>Socio-political developments do not prevent effective project implementation</p>

<p>Inputs: Types of resources</p> <p>a. CABI technical, scientific and administrative support; b. Management & technical staff from PIAs;</p> <p>c. Equipment and consumables for use in on-station and on-farm trials d. Farmer groups/associations and participation of coffee sector actors; e. Field sites for implementation of field trials, sharing knowledge and building capacity.</p>			
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Appendix II. Programme for closing workshop



CLOSING WORKSHOP OF THE MULTI-COUNTRY PROJECT 'INCREASING THE RESILIENCE OF COFFEE PRODUCTION TO LEAF RUST AND OTHER DISEASES IN INDIA AND FOUR AFRICAN COUNTRIES (CFC/ICO/40)'

Date: March 19-20, 2013

Venue: GRT Nature Trails Sky Rocca, Yercaud

PROGRAMME		
19th March 2013		
9.30 – 10.00 am	Registration	
10.00 – 11.00 am	Inaugural session	
	Welcome	
	Inauguration	
	Opening Remarks	Chairman, Coffee Board Shri. Jawaid Akhtar , IAS
		Representative from CFC Ms Eltha Brown – CFC
	Representative from ICO Mr. Denis Seudieu - ICO	
	Director, CABI Africa Mr. Morris Akiri, CABI Africa	
11.00 – 11.15 am	Coffee break (Group Photo)	
11.15 am - 01.30 pm	Session I: PROJECT IMPLIMENTATION IN DIFFERENT PARTICIPATING COUNTRIES	
Chair:		
	Overview of the coffee leaf rust project – Presentation of the project outcome - Dr. Noah A. Phiri	
	Presentation of the Report on project activities and accomplishments in India: Dr Nayani Prakash	
	Presentation of the Report on project activities and accomplishments in Kenya: Mr. Bernard Gichimu	
	Presentation of the Report on project activities and accomplishments in Uganda - Dr Pascal Musoli	
01.30 pm – 2.30 pm	Lunch break	

2.30 pm – 3.30 pm	Technical session cont.
	Presentation of the Report on project activities and accomplishments in Rwanda - Dr Celestin Gatarayiha
	Presentation of the Report on project activities and accomplishments in Zimbabwe - Mr. Dumisani Kutwayo
3.30 pm – 4.00 pm	Coffee Break
4.00 pm – 5.30 pm	Discussions on project achievements, short falls, challenges, lessons learnt and significant out come
20th March 2013	
9.00 – 10.30 pm	SESSION 2: FINANCIL REPORTS
Chair:	
	CABI
	India
	Kenya
	Rwanda
	Uganda
	Zimbabwe
10:30-10:45 am	COFFEE BREAK
11.00 – 1.30 pm	Field visits 1. Varietal trial plot at Technology Evaluation Centre, Yercaud. 2. Interaction with FFS groups at Periyakadu
1.30 -2.30 PM	LUNCH BREAK
3.00 – 4.00 PM	Concluding session



Appendix III. Examples of programmes for the African Coffee Scientific Conferences at EAFCA Conferences

6th AFRICAN COFFEE SCIENTIFIC CONFERENCE

10th February 2010 - Makutano Room, the Sarova Whitesands Beach Resort & Spa, Mombasa, Kenya

Theme: Adapting African coffee production systems to changing climate

Workshop Program

08.30 - 09.00	Registration of delegates
09.00 - 09.20	Opening remarks (Morris Akiri , Regional Director, CABI Africa)
09.20 - 10.00	Keynote address - The Changing Climate for Coffee: (Peter Baker)
10.30 – 11.00	Response to Keynote address (Sri. G.V.Krishna Rau)
11.00 - 11.30	Implications of changing climate on world coffee volumes, origins and price (Denis Seudieu , Chief Economist, ICO)
11.30 - 12.00	Mobilising resources for research and development to mitigate climate change impact on coffee (Caleb Dengu , First Project Manager, CFC)
12.00 – 12.15	Plenary discussions
11.15 - 12.00	Coffee Break
12.00 – 12.30	African Robustas getting more “robust”: the Gourmet coffee agenda (Komlan Wegbe)
12.30 – 13.00	Coffee Leaf Rust: Implication of the changing climate on disease management (Noah Phiri)
13.00 – 13.30	Revitalising the competitiveness of African coffees: which way forward? (George Oduor)
13.30 – 14.30	Lunch Break
14.30 – 15.00	Plenary Discussions
15.00 – 16.00	Shaping the coffee research and development agenda in response to a changing climate: What action and by whom? (Open forum)
16.00 – 16.30	Closing remarks (Dennis Rangji)
16.30 – 17.00	Coffee Break and departure



7th AFRICAN COFFEE SCIENTIFIC CONFERENCE 16th February 2011 – Meru Conference Room, the Ngurdoto Mountain Lodge, Arusha, Tanzania

Theme: Empowering coffee smallholder producers for better resilience to the impacts of climate change

Workshop Programme

08.30 - 09.00	Registration of delegates
09.00 - 09.20	Opening remarks (George Oduor , Deputy Director, CABI Africa)
09.20 - 10.00	Keynote address – Increasing competitiveness of African coffee: (Amb. Muchumo)
10.00 - 10.30	Smallholder competitiveness on the world market (Denis Seudieu , Chief Economist, ICO)
10.30 - 11.00	Research and development to increase smallholder coffee productivity (Eltha Brown , First Project Manager, CFC)
11.00 – 11.30	Plenary discussions
11.30 - 12.00	Coffee Break
12.00 – 12.30	Gabon’s Robusta Coffee - a success story in the face of climate change (Jean René Moundounga)
12.30 – 13.00	Making the African Coffee more resilient to Coffee Leaf Rust in line with changing climate: The Kenyan and Rwandan experiences (Bernard Gichimu and Celestin Gatarayiha)
13.00 – 13.30	Revitalizing the competitiveness of African coffees: Key points (George Oduor)
13.30 – 14.30	Lunch Break
14.30-1500	Certification and Verification for Specialty Coffee: The experience of farmers in EAFCO Countries
14.30 – 16.00	Plenary Discussions (Open forum) <ul style="list-style-type: none"> • Emerging issues • Current status with climate change proposal – where we are
16.00 – 16.30	Closing remarks and launching of the Coffee Wilt Book, a product of the CFC funded programme (Amb. Muchumo)
16.30 – 17.00	Coffee Break and departure

Appendix IV. Sample papers published from the project work

1. Effect of different shade regimes on coffee quality in Kenya
2. Incidence and Severity of coffee leaf rust and other coffee pests and diseases in Rwanda – paper published in the African Journal of Agricultural Research
3. Ecological factors influencing incidence and severity of Coffee Leaf Rust and Coffee Berry Disease in major Arabica coffee growing districts of Uganda
4. Genetic Diversity among Disease Resistant Coffee Varieties and Cultivars in Rwanda Based On RAPD and SSR Markers



EFFECT OF DIFFERENT SHADE REGIMES ON COFFEE QUALITY

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Abstract

Current climate change patterns may cause more extreme and variable weather in the future, threatening agricultural productivity in many areas of the world. Use of shade trees in coffee farming systems may offer an effective coping mechanism to coffee in areas that suffer from climate extremes. Field trials were set up in a low altitude zone. A site was selected with a shade tree (*Ficus* spp.) casting shadow over coffee trees from 0 to 6 hours in a day in the morning and afternoon and some trees in full sun. Green coffee grade proportions, sensory and biochemical components were evaluated for the coffees from the different coffee trees. The coffee trees with grade AB at 80% and above were yielded under shade. Likewise, the trees which had the highest proportion of AA grade were under shade. The proportion of grade C was highest in the trees under full sun. The highest total sensory score which is an indicator of the sensory performance was obtained under full sun. No discernible trend was observed in the levels of biochemical components; sucrose, total chlorogenic acids (CGA), oil and trigonelline under shade or in full sun. However, highest caffeine contents were obtained in coffee under shade and lowest levels in coffee under full sun. The results are discussed in relation to possible impact of shade on coffee quality observing that shade/agroforestry is one of the upcoming changes of coffee production systems to mitigate the climate change.

Introduction

Coffee is among the main export crops since its introduction by missionaries in the early 1900s' (Mwangi, 1983) and has remained an important commodity in Kenyan economy. Shade trees in coffee production provide several economic and ecological benefits. Shade trees, especially leguminous species, improve soil fertility, decrease soil erosion and nutrient leaching, reduce excessive solar irradiance and buffer large diurnal variations in air temperature and humidity that are detrimental to coffee physiology (Siles and Vaast, 2002). The crucial consideration for shade in coffee is to identify shade sources with minimal adverse effect on the coffee plant growth, yield and quality but provide all the other beneficial effects (Kimemia, 2004). The current study aimed at investigating the possible impact of shade on coffee grade proportions, cup quality and biochemical composition.

Materials and methods

Site selection

The study was conducted in Azania estate which is in upper midland 3 (UM3) coffee agro-ecological zone at Juja in Kenya. Juja is located at 1° 10' 60S and 37° 7' 0E at an altitude of

1416 meters above sea level. Twenty one trees (21) were selected along the path of the shadow and 8 control trees in full sun.

Processing

Cherry samples were picked separately from each tree and pulped using a disk-pulper during the 2010/11 main coffee season. They were wet processed (pulped, fermented and washed) and dried to final moisture content of 10.5 to 11% using standard recommended procedures (Mburu, 2004). The parchment was finally hulled and graded to seven grades based on size, shape and density. Grade AB was used as a representative grade for cupping and biochemical analysis.

Roasting and sensory evaluation

Roasting of green coffee was done to attain a medium roast using a Probat laboratory roaster within 24 hours of evaluation and coffee allowed to rest for at least eight hours. Samples were weighed out to the predetermined ratio of 8.25g per 150 ml of water. Sensory evaluation procedure described by Lingle (2001) was followed. Fragrance/aroma, flavor, aftertaste, acidity, body, balance and overall were assessed and scored together with three process control parameters (uniformity, clean cup and sweetness) by a panel of seven trained judges on a 10-point scale. All the sensory parameters (including the process control parameters) were added to constitute the total score which is a reflection of the broad quality performance of a particular coffee. This presents the total score as a key characteristic for evaluating the sensory quality performance.

Biochemical analysis

Caffeine, trigonelline and total chlorogenic acids (CGA) were extracted from green coffee powder by refluxing in distilled water. Caffeine, trigonelline and CGA were analysed using HPLC system (KNEUR) equipped with a Supel Co. Discovery column and a diode array detector at three wavelengths, 278nm for caffeine, 266nm for trigonelline and 324nm for CGA. Sucrose was extracted from green coffee powder using the method of Osborne and Voogt (1978) with modifications and analysed using a HPLC system (KNEUR) equipped with a Eurospher 100-5 NH₂ column and a refractive index detector. Caffeine, trigonelline CGA and sucrose were identified by comparing the retention times of standards and their concentrations calculated from peak areas using calibration equations. Crude oil was analysed as outlined in the AOAC (1995).

Results and discussion

The coffee trees with grade AB at 80% and above were yielded under shade (Figure 1). Likewise the trees which had the highest proportion of AA grade were under shade. The proportion of grade C was highest in the trees under full sun. The trees under six hours of shade showed lower total sensory score than the trees under full sun (Figure 2). No discernable trend was observed in the levels of biochemical components; sucrose, total chlorogenic acids (CGA), oil and trigonelline of the coffee from the trees under shade and those in full sun (Figure 3). However, coffee from tree number 15 and 16 each under 6 hours

of shade had the highest caffeine contents 1.44% and 1.38 % (dwb) respectively while tree number 27 under full sun had the lowest amount of caffeine 0.90% (dwb).

Shade resulted in larger coffee beans as reported in previous studies (Muschler, 2001; Youkhana and Idol, 2010) with differing explanations on the contribution of shade to the observations. As with the effect of shade on production, the influence of shade on organoleptic tributes is also controversial (Muschler, 2001, Vaast *et al.*, 2006; Bote and Struik, 2011). In this study no clear gain was observed on the sensory quality parameters due to shade but the contribution of the shade to the increased premium grades (AA and AB) is important since these grades are highly valued in the coffee trade.

ACKNOWLEDGEMENTS

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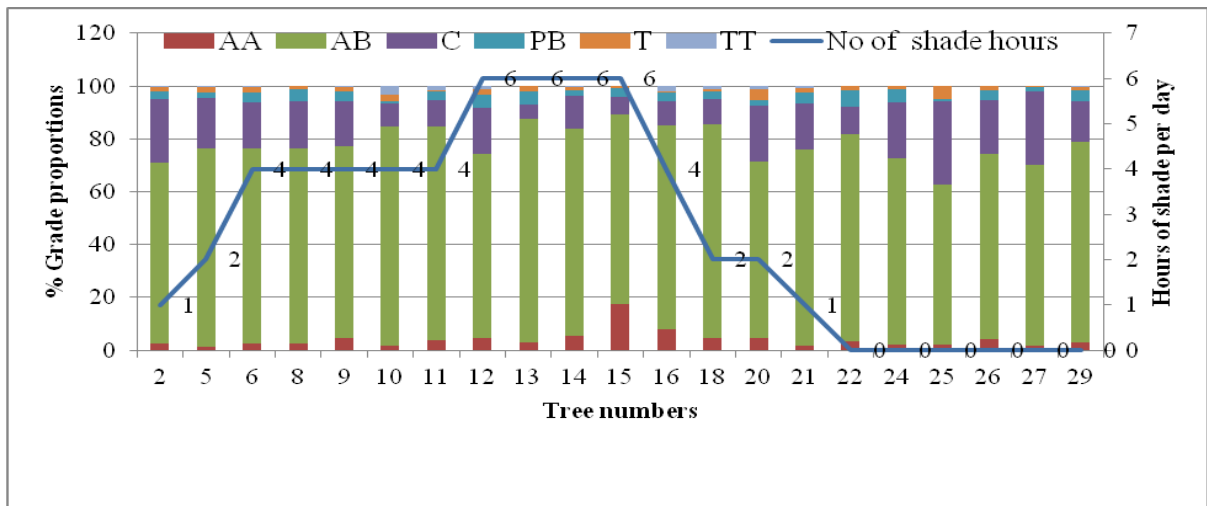


Figure 1: Grades of coffee under different shade regimes and in full sun

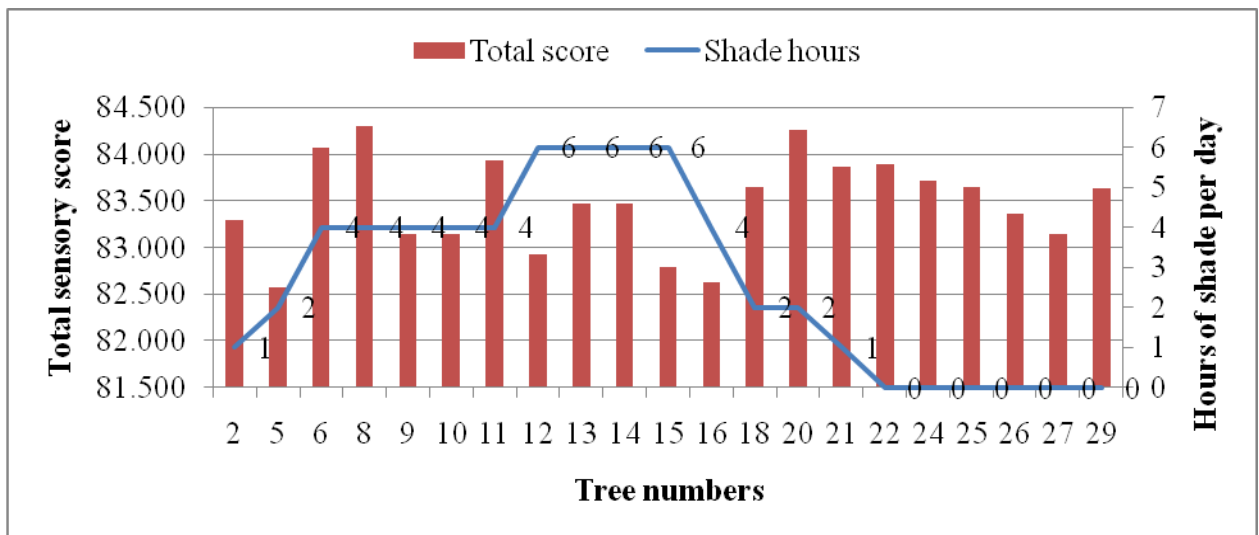


Figure 2: Total sensory score of coffee under different shade regimes and in full sun

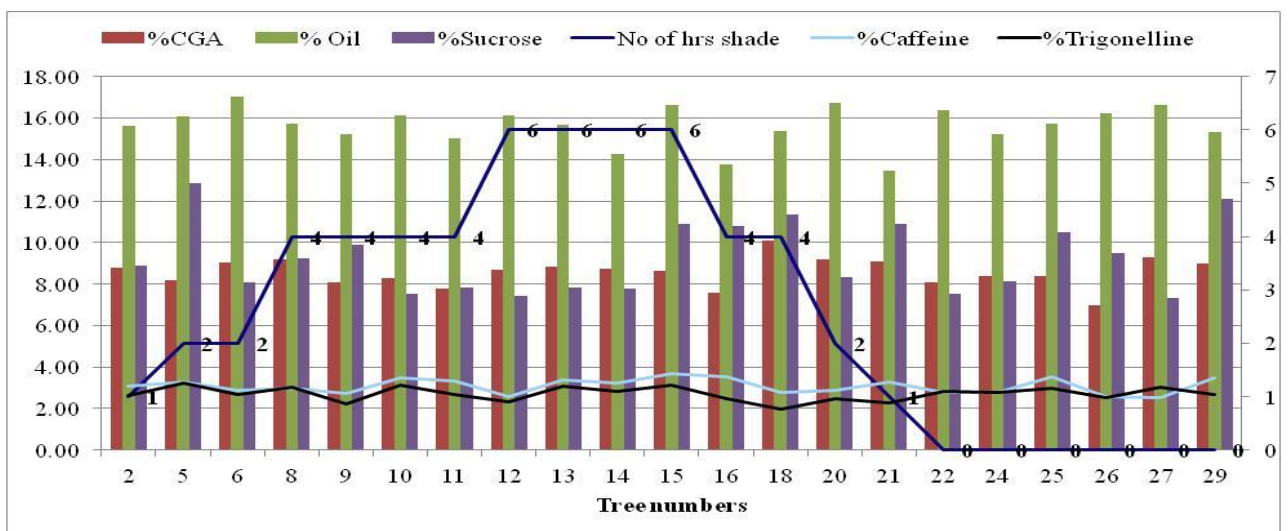


Figure 3: Biochemical components of coffee under different shade regimes and in full sun

Full Length Research Paper

Incidence and severity of coffee leaf rust and other coffee pests and diseases in Rwanda

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Coffee Leaf Rust (CLR) caused by *Hemileia vastatrix* Berk. and Br. has a worldwide distribution and causes enormous yield losses. A survey was conducted in Rwanda to evaluate incidence and severity of CLR and other coffee pests and diseases and to determine how the crop management contributes to CLR severity. A stratified random sample of 307 coffee farms was visited and the prevalence, incidence and severity were recorded. Results showed that all provinces were affected by CLR with the highest severity in Eastern province where the incidence was up to 100%. High altitudes were associated with low disease severity with a negative correlation of - 0.71. Coffee berry disease (CBD) was a minor disease but with a potential to become an epidemic in the future. Coffee leaf miner, Antestia bugs and Coffee stem borer were recorded but were managed below economic injury levels. All commercial cultivars were susceptible to CLR and most management practices such as mulching, pruning and fertilizer application were associated with lower levels of CLR severity except intercropping which resulted in higher disease intensity. Implications of this survey in relation to CLR management in Rwanda are discussed in this study.

Key words: Coffee, diseases, incidence, severity, management, Rwanda.

INTRODUCTION

Coffee is an important export crop and a major foreign exchange earner for Rwanda. It is the second most important agricultural export commodity after tea earning in the country having over 15% of the foreign currency annually (Anonymous, 2009). The crop provides direct employment to a considerable number of workers and it also does well on hillside farms where other more demanding cash crops are not easily grown.

Coffee suffers yield losses from a number of factors including insect pests and diseases. Among these, Coffee leaf rust (CLR) caused by *Hemileia vastatrix* Berk. and Br is the most devastating. Yield losses per year due to CLR range from 30 to 90% depending on the environmental conditions during a given year, especially, if not properly controlled by an intensive program of fungicides

spray (Sera et al, 2005). In Rwanda, the situation is worsened by varieties grown in the country most of which are Bourbon type that is, Jackson, Mibilizi and Bourbon Mayaguez which are mostly susceptible to CLR. An effort was undertaken by the government of Rwanda under the support of the Common Funds for Commodities (CFC) through CAB International to test the Indian hybrids and the most promising coffee germplasm. The intention was to come up with the best varieties, establishment methods and their best growing conditions. However, there was very little background information available on the distribution and severity of CLR in Rwanda and there were no documented records. As part of a project to obtain baseline information for the development of suitable interventions for the management of the disease, a survey was conducted in the coffee growing regions with the following objectives: 1) to evaluate incidence and severity of CLR and other diseases in Rwanda and 2) to determine how various crop management practices contribute to the disease severity.

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Table 1. Distribution of surveyed farms per province.

Provinces	Number of farms
Western	92
Southern	97
Northern	30
Eastern	77
Kigali	11
Total	307

Table 2. Distribution of CLR in surveyed Provinces.

Province	% of farms with CLR	Standard error
North	83.3	6.2
East	97.4	8.3
South	92.2	2.6
West	94.3	2.6
Kigali	94.6	2.8

MATERIALS AND METHODS

Survey methods

A stratified random sample of three hundred and seven coffee farms were selected for a disease survey from a list of coffee growers held by Rwanda Coffee Development Authority (OCIR Café). The number of farms surveyed per province is presented in Table 1. The survey was carried out in Western province, mainly the shores of Lake Kivu, the Northern province covering the border with Uganda to Lake Muhazi, the Southern province from near the city of Kigali to Burundi and in the Eastern province from the central part to the border with Tanzania.

Farms were surveyed from June to August, 2008. For each farm visited, 30 bushes were scored on a diagonal line across the farm for the presence or absence of symptoms of CLR and Coffee berry disease (CBD) caused by *Colletotrichum kahawae*, Waller and Bridge and other pests and diseases. Disease prevalence (percentage of farms where the disease was recorded), incidence (percentage of bushes affected per farm) as well as, severity were recorded. The incidence of CLR and CBD were scored on each of the coffee tree using the following scale: 0 = no disease, 1 = < 10% diseased leaves (CLR) or berries (CBD), 2 = 10 to 30% diseased leaves or berries and 3 = > 30% diseased leaves or berries (Phiri et al., 2001). Severity was recorded as an average number of pustules per leaf for CLR and the average number of lesions per berry for CBD. Other coffee diseases and pests were recorded as either present or absent.

At each farm, a questionnaire was administered and the owner was asked about the age of the bushes, if fertilizer or pesticides were applied and if so, which types were applied, if the plantation was weeded and whether bushes were pruned. This information was collected from each farmer. In addition, it was confirmed by our own observation, particularly, in the case of weeding and pruning. Information was also collected on whether shade trees were used and whether the coffee was intercropped. Altitude in metres above sea level (masl) was taken, using a Garmin Ground Positioning System (GPS) at a central point for each farm surveyed. Data collected on incidence, severity and prevalence were summarized as means for all farms surveyed and standard errors were calculated.

Data were also analyzed for correlations between severity of CLR and altitude. Multiple linear regression analysis was used to determine the relationship between management practices and CLR severity using GenStat Discovery Edition 3 (2007).

RESULTS

Altitude

The altitude of surveyed farms varied considerably with regions surveyed. The highest altitudes were recorded in the Northern province with the average of 1902.6 masl while the lowest altitudes were found in the Eastern province with the mean altitude of 1564.6 masl. The highest altitude point measured was 2045 masl while the lowest point was 1211 masl. Altitude was found to have an implication on the distribution and severity of CLR since the relationship between the disease severity and the altitude was quite apparent during the survey period.

Coffee leaf rust distribution and severity

The survey data showed a wide distribution of CLR in Rwanda (Table 2). All surveyed provinces had the disease ranging from about 80% of surveyed coffee bushes to more than 95%. The highest CLR incidence and severity were recorded in Eastern province while the lowest were observed in the Northern province. The coffee rust incidence, measured as the percentage of diseased leaves per coffee bush, ranged from 0 to 100% in surveyed farms. Results also showed that only 15% of surveyed farms were not diseased. The remaining farms had varying levels of disease incidence (Figure 1).

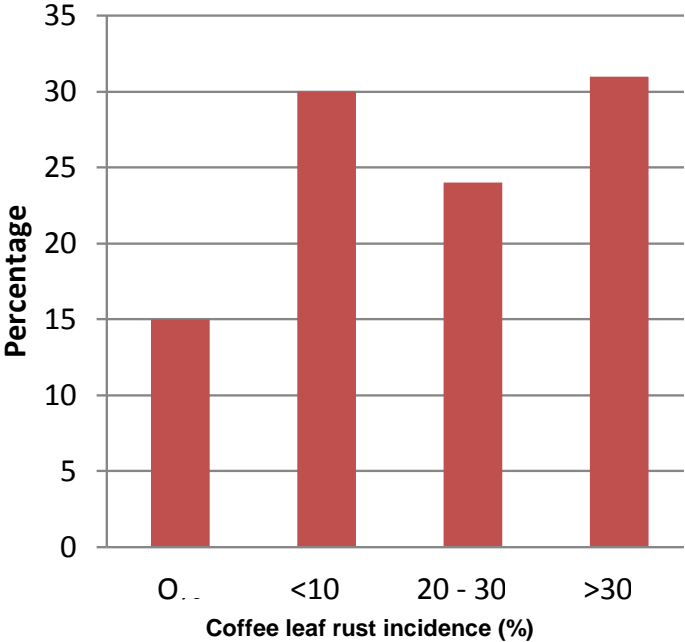


Figure 1. Percentage of farms infected by CLR in Rwanda.

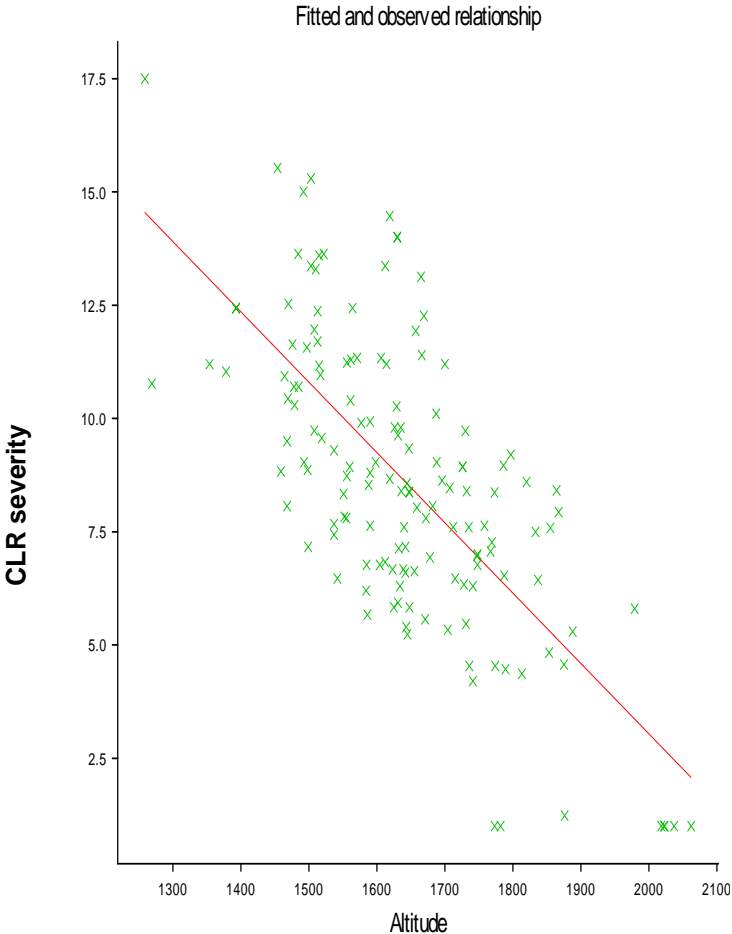


Figure 2. Relationship between CLR severity and altitude.

Table 3. Prevalence of notable coffee pests on surveyed farms.

Province	% of farms with CLM	% of farms with Antestia	% of farms with CSB
West	26.2	0.1	0
North	37.9	6.6	0
East	20.4	0	1.6
South	2.4	2.9	0
Kigali	2.6	0	0

Relationship between coffee leaf rust severity and altitudes

The survey data showed a negative correlation between CLR severity and altitude ($r = -0.71$). The disease severity decreased when altitude increased (Figure 2). As was noticeable during the survey period, farms located at higher altitudes (normally from 1800 masl and above) had less CLR and those at lower altitudes (mainly from 1400 masl and below) were much more diseased. The linear regression equation was found to be:

$$Y = -0.0155 X + 34.093$$

Where Y is the disease severity and X the altitude (Figure 2). Both slope and intercept were highly significant at ($p < 0.001$).

Distribution and severity of coffee berry disease

During the survey period, there was no CBD recorded as it was during the dry season.

Notable pests

Coffee leaf miner (CLM) caused by *Leucoptera coffeella* was recorded in all surveyed provinces (Table 3). The highest percentage of farms infested with CLM was found in the Northern province with more than 35% of surveyed coffee bushes. In the Southern province and Kigali respectively, the number of infested coffee bushes was low (less than 5%). Antestia bug (*Antestiopsis lineaticollis*) was also a major pest recorded in the surveyed area. The highest number of coffee bushes infested by the pest was found in the Northern province with more than 5% of infested farms. In the Southern province and Kigali, there was no Antestia bugs recorded on surveyed farms. Coffee stem borer (CSB) was found in the Eastern province, mainly on old coffee trees where the number of infested coffee bushes was less than 2%. In other surveyed provinces, there was no CSB that was recorded. At national level, only CLM appeared important with the prevalence going up to 22% while it was 2.9 and 0.2% for Antestia and CSB, respectively.

Relationships between management practices and CLR severity

In all surveyed areas, intercropping was found to be practiced to a limited extent that is, less than 10% (Figure 3). Intercrops were sweet potatoes, yams, cassava and bananas. Farms which were intercropped had significantly ($p < 0.001$) higher CLR severity than non-intercropped farms. It was also found that more than 70% of surveyed farms were mulched and the mulching mainly comprised of banana leaves, sorghum and rice straws. The effect of mulch on the severity of CLR was highly significant in surveyed farms at ($P < 0.001$). More than 95% of surveyed farms were weeded and the weeding was done mainly with hoe especially, in non-mulched coffee farms. Weeds were also uprooted by hand in mulched coffee farms. No herbicide was used in surveyed areas. Farms which were weeded had very significantly lower CLR severity than unweeded farms having a value of ($p < 0.001$).

Pruning was found to be practiced at 89.2% in farms where the survey was carried out. Coffee bushes which were pruned had significantly less CLR severity than where bushes were not pruned ($P < 0.001$). Similarly, farms which were fertilized had significantly lower CLR severity than unfertilized farms ($p < 0.001$). Applied fertilizer was either organic (compost made of crop residues and farm yard manure) or inorganic (mainly a combination of nitrogen, phosphorus and potassium to NPK 20 - 10 - 10). Unlike other cropping practices for which management was at a very high level (more than 85%), less than 60% of surveyed farms were fertilized.

The results did not show any relationship between age of coffee trees and CLR severity and coffee trees of different ages suffered heavy CLR infection equally.

Status of commercial cultivars in relation to CLR resistance

There were six varieties of coffee bushes recorded during the survey. These were Bourbon Mayaguez 139 (BM 139), BM 71, Jackson 2/1257, Harar, Pop 3303/21 and Mibilizi. All of these varieties were susceptible to CLR. In all surveyed areas, an average of over two pustules per leaf was observed on all cultivars. The analysis of variance

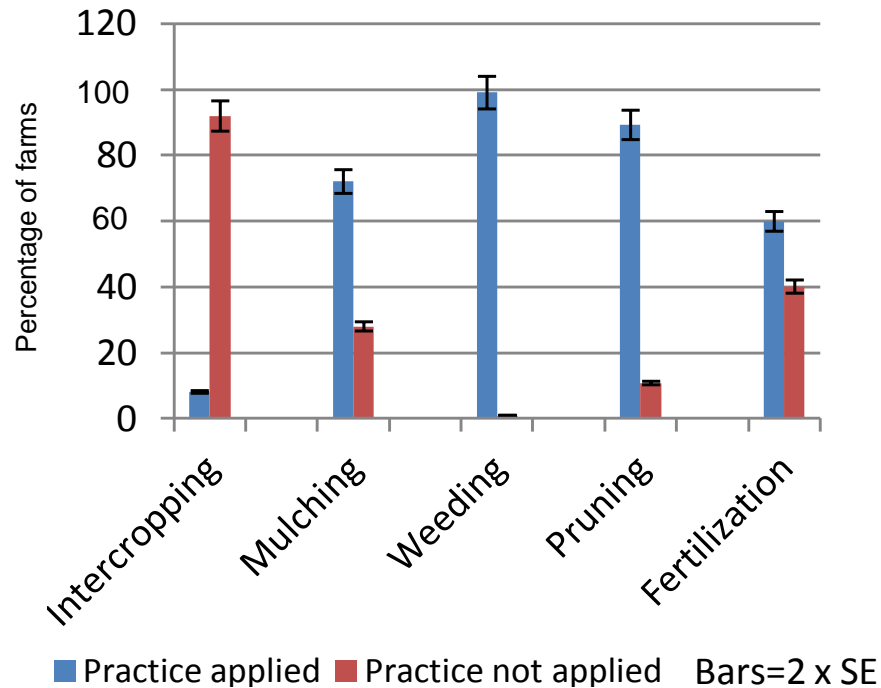


Figure 3. Level of management practices in surveyed farms.

did not reveal any significant difference between the above varieties ($p = 0.08$) in regards to CLR severity and therefore all cultivars exhibited equal susceptibility.

DISCUSSION

Coffee leaf rust

It is apparent from this survey that CLR is widespread throughout the country with the incidence increasing as much as 100% in certain areas. This may partly be explained by climatic conditions prevailing in Rwanda. The alternating wet and dry conditions favor high CLR build up and thus leads to high crop losses (Prakash et al., 2005). On the other hand, this high disease incidence is also compounded by the high susceptibility of commercial cultivars grown in the country. It is worth mentioning that in all provinces and on all varieties, more than two pustules per leaf were recorded.

According to Anonymous (1989), two to three pustules per leaf are required to produce severe leaf defoliation on heavily bearing coffee and can cause die back and biennial bearing, a situation which was already noticeable in certain areas during the survey period. The disease severity has reached such destructive proportions that it is repeatedly causing great concern among both farmers and government. A need therefore, exists to develop CLR resistant varieties with adequate cup quality and local adaptability to coffee growing conditions. Meanwhile,

since copper based fungicides have been effective for the control of coffee rust, the program of timely spraying with such fungicides should also be adopted as a short-term measure.

Coffee berry disease

The survey results did not show any case of CBD infection. One possible reason is because the survey was carried out during the dry season. Griffiths and Waller (1971) found that the occurrence of CBD in Kenya was related to high rainfall and altitude. Though high altitudes (> 1800 masl) were recorded in this survey, it was conducted during the period of very low relative humidity. Water is a requirement for the development, dispersal and germination of *C. kahawae* conidia. It was observed, however, that most of the recommended coffee varieties in Rwanda are uniformly susceptible to CBD (Walyaro, 2010). Though currently a minor disease, it has the potential to flare up into a major epidemic any time. The coffee breeding program should, not only develop varieties that are resistant to CLR, but also to both CLR and CBD in light of the climate changes occurring around the world.

CLR severity and altitudes

The results showed a negative correlation between CLR

severity and altitudes. The negative correlation was also reported by Kushalappa and Eskes (1989) who found that higher altitudes were associated with lower disease severity. Rivera (1984) also observed a lower level of disease intensity at higher altitudes in Guatemala. This finding prioritizes higher lying areas as more deserving of efforts to develop CLR resistant varieties with adaptability to such conditions.

Notable pests

CLM was observed in all surveyed areas. The highest prevalence was recorded in the Northern Province (37%) probably because of conducive climatic conditions of the zone. Oliveira (2006) reported that temperature of around 27°C and nearly saturated relative humidity favoured high pest build up and this closely reflected the environmental conditions of the Northern province during the survey period. Though widespread, CLM has not yet reached economic proportions to necessitate control but its potential to grow into an important pest within a short time remains real considering the levels observed.

Antestia bugs were recorded in the Northern, Western and Southern provinces at relatively low levels (less than 6.6%). This pest was not recorded on coffee bushes in the Eastern province and Kigali. This may be explained by regular pruning since this agricultural practice reduces dense foliage and hence, creates unfavorable conditions for the bugs. In addition, general use of Dursban (Chlorpyrifos) whose wide use was recorded may also be the source of low level of Antestia bugs recorded.

Management practices and CLR severity

Weeding, pruning and mulching are common cultural practices across all provinces unlike the significantly low use of fertilizers. Most farmers were smallholders lacking resources to afford fertilizers. Moreover, coffee faced a stiff competition from the staple crops in regards to organic fertilizer utilization. Results have showed that all cropping practices except intercropping resulted in reduced CLR severity. Both weeding and fertilization increased the plant vigor and made it tolerant to CLR attack. Mulching, in addition to providing organic matter, also adequately conserved soil moisture. Intercrops competed with coffee bushes for water, nutrients and light which resulted in higher CLR severity.

CONCLUSION AND RECOMMENDATIONS

Survey results showed that CLR was a widespread disease in Rwanda. All commercial cultivars grown in the country were susceptible to CLR. Coffee berry disease was a minor disease but with a potential to become a new epidemic. Recorded pests were CLM, Antestia bugs,

and CSB but these were managed below economic injury levels. However, continued monitoring of the pests occurrence was recommended as it is likely to be closely associated with seasonal conditions and the important elements that are likely to be associated with global warming. Most cropping practices were associated with low levels of CLR severity except intercropping which resulted into higher disease intensity. Given the susceptibility of commercial cultivars, there is the need to develop varieties that are resistant to both CLR and CBD.

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Ecological factors influencing incidence and severity of Coffee Leaf Rust and Coffee Berry Disease in major Arabica coffee growing districts of Uganda

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Abstract

Coffee Leaf Rust and Coffee Berry Disease are the most devastating diseases of Arabica coffee in Africa. The importance of coffee in economies of many African countries like Uganda, presents urgent need for cost-effective disease control strategies. In this study, 192 coffee farms were surveyed and their corresponding incidence and severity recorded. Nebbi district had the highest CLR incidence (90.2%) and severity (2.2%) followed by Sironko (67.9% and 1.9%) and least in Kapchorwa (20.4% and 1.3%) respectively. CBD incidence was highest in Kapchorwa (6.0%) followed by Nebbi (1.7%). There was no CBD incidence observed in Sironko. There was a significant ($p < 0.05$) interaction between altitude and disease severity. Thin and medium shade intensity had highest CLR incidence followed by thick and no-shade levels. CLR was highest in farms under mono-shade followed by farms under mixed-shade and least in open-farms. CLR severity was found to be highest at very steep slopes and medium slopes and least on gentle slopes. In conclusion, CLR was present in all surveyed districts while CBD occurred in Kapchorwa and Nebbi districts at intensity levels enough to trigger economic losses.

Key words: Coffee Berry Disease, Coffee Leaf Rust, ecological factors, pathosystem, Uganda

Introduction

Uganda is the second biggest coffee producer and exporter in Africa after Ethiopia (ICO, 2009), contributing an estimated 2.97% of its crop to the world market (ICO, 2011). In the coffee year 2010/2011, Uganda exported 3.15 million 60-kilo bags valued at \$448.89 million, constituting 78.87% Robusta and 21.13% Arabica coffee, (UCDA, 2011). Over 13 million Ugandans derive a substantial proportion of their livelihood from coffee

earnings either directly or indirectly along the value chain (Sayer, 2002).

Two major coffee diseases that have ravaged Arabica coffee production in Uganda are Coffee Leaf Rust (CLR) caused by the biotrophic fungus *Hemileia vastatrix* (Berkeley and Broome) and Coffee Berry Disease (CBD) caused by *Colletotrichum kahawae* (Bridge and waller) (UCDA, 2011). Coffee Leaf Rust is capable of infecting both commercial coffee species; *Coffea arabica* and *Coffea canephora*, although

the former is more susceptible. On the other hand, CBD infects only *C.arabica* at elevations above 1500 meters above sea level (a.s.l) (Hakiza, 1997).

Coffee Leaf Rust causes about 10 to 50% yield loss in farms with susceptible coffee varieties especially if no control measures are undertaken (Van der Vossen, 2001; Silva *et al.*, 2006). In contrast, CBD may cause up to 70 to 80% losses if no control measures are adopted, especially, in years when berry yield is high (Waller, 1985; Silva *et al.*, 2006). Coffee Leaf Rust manifests itself as yellow pustules on the lower surface of leaves turning orange-yellow with powdery masses of urediniospores in later stages. Defoliation of affected plants is a common symptom, which leads to loss of yield and quality of coffee. Coffee Berry Disease infects all stages of the crop from flowers to ripe fruits and seldom leaves. Major losses are observed following infection of green immature berries which then develop dark sunken lesions with sporulation, causing premature dropping and mummification (Silva *et al.*, 2006; Gichimu and Phiri, 2010). Control of CLR by fungicide application with Orius (Tebuconazole) following a bi-weekly spray regime has proved very effective, although copper based fungicides such as Copper oxychloride and Nordox 75% have exhibited moderate potency (Matovu, unpublished data). Unfortunately, fungicide use is fraught with resources constraints since most coffee farmers are smallholders hence making it economically unfeasible. Cultural control measures such as pruning, stumping, de-suckering, fertilizer application and coffee tree spacing, have also shown promise although they cannot stand alone if effective control is to be achieved (Bigirimana *et al.*, 2012).

Host resistance remains the most economically viable option especially for resource constrained farmers (Omondi *et al.*, 2001; Gichuru *et al.*, 2008). This control strategy, however, is challenged by resistance erosion presumably due to an evolutionary response by pathogens to host defense mechanisms through mutation and recombination (Hulbert *et al.*, 2001; Vleeshouwers *et al.*, 2001).

Agrios (2005) and Okori (2004) elucidate the significance of monitoring populations of pathogens and host plants in an evolving environment. Most importantly, such studies should give special emphasis to biotic and abiotic factors in the environment under going strong influence by human activity as a result of disease management. Proper understanding of the interaction of the elements of disease triangle in the pathosystem enables formulation of efficient and cost-effective disease management strategies before disease progress reaches economic injury level. The aim of this study was therefore to determine incidence and severity of Coffee Leaf Rust and Coffee Berry Diseases in major Arabica coffee growing districts of Uganda, and to study the relationship between the observed disease incidences and existing ecological factors.

Materials and methods

Study area

The study was conducted from August to November 2009 in two major Arabica coffee growing regions of Uganda; Mt Elgon region (Kapchorwa and Sironko districts) which borders with Kenya, and West-Nile region (Nebbi district) which borders Democratic Republic of Congo (DRC). The study area in Mt. Elgon lies approximately between latitudes 1° 17'N

and 0° 51'N and longitude 34° 13'E and 34° 25'E at an altitude of 1288-2135M above sea level (van Asten *et al.*, 2011). In Nebbi district, the study area lies between latitudes 2° 14'N and 2° 46'N and longitudes 30° 76'E and 31° 52'E at an altitude of 1450-1800M above sea level. Mt Elgon region receives a mean annual rainfall of more than 1520 mm while West-Nile receives 1100 mm, following a bimodal pattern in both regions. Temperatures at both locations range between 15°C-30°C throughout the year. All soils in Mt Elgon region are derived from volcanic ash and agglomerate. Specifically, Sironko areas generally have dark-brown clays while Kapchorwa has red sandy clay loam soils. On the other hand, the soils in Nebbi are red clay loams derived from amphibolites (Aniku, 2001). A stratified random sampling procedure was adopted where in each district, four sub-counties were randomly selected. From each of the sub-counties, four parishes were chosen and in each of these four villages were selected and subsequently in each village four coffee farms were surveyed. Coffee farmers in each district were contacted and their farms surveyed with the help of district and sub-county agricultural extension officials as guides. The importance of this team was to identify farmers, build trust and provide subsequent follow-ups to the farmers.

Data collection procedures

At each farm, 30 randomly selected trees on a diagonal transect across the farm were assessed for incidence and severity of Coffee Leaf Rust and Coffee Berry Disease. A diagnostic symptom of Coffee Leaf Rust was the presence of yellow-orange pustules on the underside of leaves

while for Coffee Berry Disease was the extensive brown to black sunken lesions on both green and red berries leading to mummification of berries. The sampled trees were then physically counted and tagged from 1-30 with the help of conspicuous coloured labels.

Since both diseases cause observable and distinct symptoms to the coffee plant, a comprehensive and robust approach of quantifying disease was adopted. Disease incidence and severity were selected as variables for data collection. In this case, disease incidence accounted for the proportion of coffee trees diseased out of the 30 trees sampled, while disease severity (intensity) for the relative or absolute area of leaf/berry tissue affected by respective disease. Disease severity was visually estimated with the help of a disease rating scale (1-4) to quantify the extent of infection per tree; 1= No coffee leaf rust or CBD, 2= <10% diseased leaves or berries, 3= 10-30% diseased berries or leaves, 4= >30% diseased leaves or berries (Phiri *et al.*, 2001; Bigirimana *et al.*, 2012). In addition, data on current agronomic practices and environmental properties such as; soil type, shade nature, farm topography and altitude, were collected to determine their influence on coffee disease occurrence. Data on shade types were collected at 3 levels where coffee farms shaded with more than 1 tree species were recorded as mixed tree species, while those with only one tree species were recorded as mono tree species and un-shaded farms in open sun were recorded as no-shade. The nature of shade at each farm was assessed by visual estimates using a rating scale; No shade = 100% light penetration, thin shade = 99% to 70%, Medium shade = 69% to 40% and Thick shade = 39% to 20%. Soil

type was visually assessed by field analysis of soil texture using the 'feeling method'. The nature of topography for each farm was visually assessed based on its degree of inclination; Gentle slope = below 5°, medium slope = 6° to 30°, steep slope = 31° to 50° and very steep slope = 51° to 90°. Altitude in metres above sea level (masl) was recorded, using etrax Ground Positioning System (GPS) at a central point for each farm surveyed.

Data analysis

Completed questionnaires were entered into Microsoft Excel spread sheets where variable codes were assigned. Data were then analysed using Genstat statistical package (13th Edition) and Statistical Package for Social Sciences (SPSS18.0). Analysis of Variance (ANOVA) was used to generate means for disease incidence and severity among discrete independent variables (shade type, shade nature, topography and soil type) were effects were declared significant at 5% level. Least Significant Difference (LSD) was used to determine if there were significant differences among means. Correlation and linear regression analysis were used to test the magnitude and nature of relationships (association) between disease incidence and farm altitude.

Results

Coffee Leaf Rust and Coffee Berry Disease incidence and severity

The survey results indicate a wide distribution of CLR in all major Arabica coffee growing districts of Uganda (Table 1). All surveyed districts had considerable numbers of coffee farms infested with CLR incidence ranging from 20% to over 90%. The highest CLR incidence (90.2%) and severity (2.2%) were observed in Nebbi district while the lowest were observed in Kapchorwa district (20.4 and 1.3%; respectively).

On the other hand, the results reveal a generally low distribution of CBD incidence in surveyed Arabica coffee growing districts. Coffee Berry Disease infested farms were only observed in Kapchorwa and Nebbi districts but not in Sironko district. The highest CBD incidence and severity were observed in Kapchorwa (5.9% and 1.1, respectively) followed by Nebbi district (1.7% and 1.0, respectively).

Relationship between Arabica coffee diseases and ecological factors

Altitude. The results indicate a significant ($P \leq 0.05$) negative relationship between CLR incidence and altitude ($r =$

Table 1. Incidence and severity of Coffee Leaf Rust and Coffee Berry Disease observed in coffee fields sampled in the Arabica coffee growing regions of Uganda

District	Farms analysed	Mean CLR incidence (%)	CLR-Severity	Mean CBD incidence (%)	CBD-severity
Sironko	64	67.9	1.9	0.0	1.0
Kapchorwa	64	20.4	1.3	6.0	1.1
Nebbi	64	90.2	2.2	1.7	1.1
P-value		<0.001	<0.001	0.012	<0.001
LSD		8.25	0.157	4.034	0.058

-0.99). This implies that CLR incidence increases with each unit decrease in farm altitude (Fig. 1). This association is more elaborated in the following resultant linear regression equation: $Y = -0.083X + 182.74$ (where Y = percentage CLR incidence and X = altitude in masl). The equation further predicts that, the probability for a susceptible coffee farm to be infested with CLR is low above 2194 masl while the probability for infection is high below 993 masl.

Coffee Berry Disease incidence and severity on the other hand, shows a significant ($P \leq 0.05$) positive relationship with altitude. CBD incidence therefore increases with each unit increase in farm altitude after 1800 meters above sea level (Fig. 1). The linear regression equation depicting this relationship is: $Y = 0.018X - 26.42$ (where Y = percentage CBD incidence and X = altitude in masl). The equation indicates that the probability for a susceptible coffee farm to be infested with CBD is high at 1800 masl while the probability of infection is low at 1501 masl.

Effect of tree shade on disease incidence and severity

Results indicate 98.5% of coffee farms analysed had been inter-planted with shade trees at various intensities. There was a significant difference in CLR incidence ($P < 0.001$) and severity ($P = 0.003$) among coffee farms situated under different shade intensities. Coffee Leaf Rust incidence was generally highest in farms under thin (71.4%) and medium (61.9%) shade tree cover and lowest in open farms (no shade; 31.4%) and those under thick shade (36.7%).

There was no significant ($P = 0.662$ and $P = 0.943$ respectively) difference in CBD incidence and severity among coffee farms located under the different shade levels. However, similar to CLR, CBD incidence was found more under thin (4.1%) and medium (2.1%) shade levels and lowest under thick (1.3%) and open (1.9%) coffee farms.

Influence of tree shade types to CLR and CBD incidence and severity is presented in Table 3. There was a

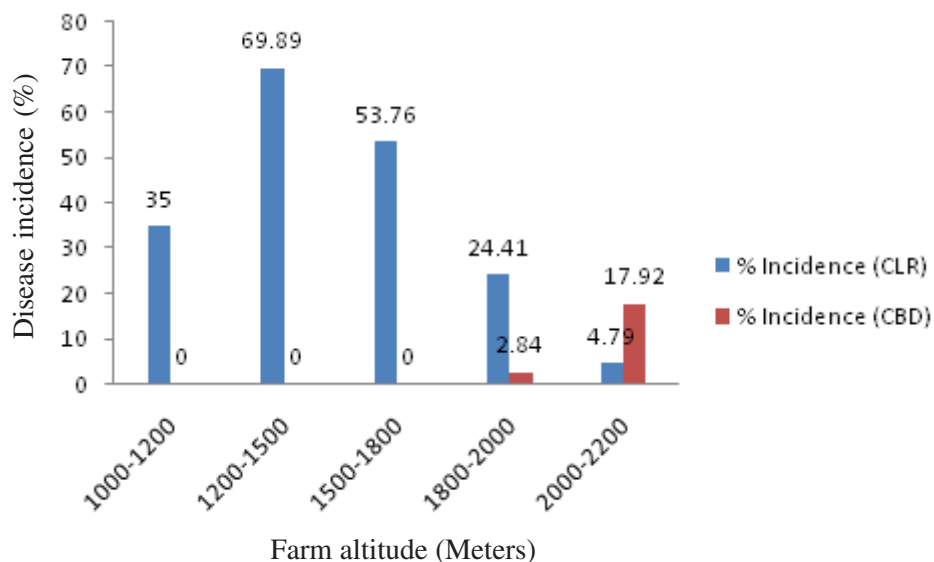


Figure 1. Relationship between altitude and CLR and CBD incidence on coffee farms sampled in the Arabica coffee growing regions of Uganda.

significant ($P=0.018$) difference in CLR incidence among coffee farms located in various tree shade types. Coffee Leaf Rust incidence and severity were generally highest in farms having only one tree species inter-planted (73.0 and 2.0%), mild (55.4 and 1.7%) in farms with more than one tree species and lowest (49.2 and 1.7% respectively) in farms completely without shade. However, the data reveal no relationship between CBD incidence and severity with tree shade types.

Effect of farm soil type

There was no significant difference at $P=0.05$ in incidence of CLR and CBD

between coffee farms situated on clay, sand and loam soil types. Coffee located on clay soils had higher mean incidence of CLR (81.7%) than on loam (59.0%) and sand (0.0%) soil types. In contrast, coffee growing on loam soils had higher mean incidence of CBD (2.6%) than coffee on clay soils (0.0%) and sand soil (0.0%). However, there was a significant ($P=0.037$) difference in mean severity of CLR among coffee farms located on clay, sand and loam soil types. Coffee growing on clay soils had higher CLR mean severity (2.4%) than coffee growing on loam (1.8%) and sand (0.0%) soil types. The data also indicated that 98%

Table 2. Incidence and severity of CLR and CBD as influenced by shade intensity in coffee fields sampled in the Arabica coffee growing regions of Uganda

Shade nature	% Mean incidence (CLR)	Mean severity (CLR)	% mean incidence (CBD)	Mean severity (CBD)	% farms analysed
No shade	31.4	1.8	1.9	1.1	1.5
Thin	71.4	2.0	4.1	1.0	32.5
Medium	61.9	1.9	2.1	1.0	48.4
Thick	36.7	1.5	1.3	1.0	17.6
P-value	<0.001	0.003	0.662	0.943	
LSD	37.41	0.1879	12.469	0.183	

Table 3. Incidence and severity of CLR and CBD as influenced by shade type in coffee farms sampled in the Arabica coffee growing regions of Uganda

Shade type	% Mean incidence (CLR)	Mean severity (CLR)	% mean incidence (CBD)	Mean severity (CBD)	% farms analysed
No shade	49.2	1.7	3.3	1.1	1.5
Mono	73.0	2.0	4.4	1.0	24.6
Mixed	55.4	1.7	2.0	1.0	73.9
P-value	0.018	0.086	0.464	0.794	
LSD	51.47	0.823	16.453	0.2408	

of farms surveyed were located on loam soil type, 2% were located on clay soil and no coffee farm was found located on sand soil.

Effect of farm topography

Table 5 indicates the relationship between farm topography with disease incidence and severity. Majority of coffee farms surveyed (81%) were located on gentle slopes while the rest of farms (less than 19%) existed at medium, steep and very steep topographies. There was a significant ($P < 0.001$) difference between CLR incidence and severity at various farm topographical levels. Coffee Leaf Rust incidence was highest on very steep slopes

(81.1%) followed by medium (79.7%) and steep slopes (64.2%) and least on gentle slope (37.6%). Coffee Leaf Rust mean severity was highest on coffee growing on medium slope (2.1%), followed by steep and very steep (2.0%) and least on gentle slope (1.5%). However, the results show no significant difference at $P < 0.05$ in CBD incidence and severity among all topographical levels surveyed.

Discussion

From the results CLR is present in all surveyed Arabica coffee producing districts of Uganda at varying levels. The high incidence and severity observed is

Table 4. Incidence and severity of Coffee Leaf Rust and Coffee Berry Disease by farm soil type

Soil type	% Mean incidence (CLR)	Mean severity (CLR)	% mean incidence (CBD)	Mean severity (CBD)	% farms analysed
Clay	81.7	2.4	0.0	1.0	4.0
Loam	59.0	1.8	2.6	1.0	188.0
Sand	0.0	0.0	0.0	0.0	0.0
P-value	0.233	0.037	0.661	0.690	
LSD	37.330	0.589	11.770	0.172	

Table 5. Incidence and severity of Coffee Leaf Rust and Coffee Berry Disease by nature of topography

Topography	% Mean incidence (CLR)	Mean severity (CLR)	% mean incidence (CBD)	Mean severity (CBD)	% farms analysed
Gentle	37.6	1.5	2.4	1.0	81.9
Medium	79.7	2.1	6.7	1.1	9.5
Steep	64.2	2.0	6.3	1.1	6.3
Very steep	81.1	2.0	0.0	1.0	2.4
P-value	<0.001	<0.001	0.511	0.207	
LSD	19.38	0.3279	6.93	0.126	

partially attributed to favourable ecological conditions created by optimum temperatures and fairly high precipitation in Uganda (Kabeere and Wulff, 2008). According to reviews by Eastburn *et al.*, (2011), the above environmental factors affect pathogens directly by altering spore germination and hyphal growth rates which as a result affect rate of inoculum production. Studies by Hakiza (1997) and Eskes (1983) relate CLR severity directly to prevailing ecological conditions such as rainfall, temperature, duration of leaf wetness and wind velocity. Similarly, CBD development depends on climatic factors such as rainfall, temperature, and relative humidity (Guyot *et al.*, 2001; MouenBedimo *et al.*, 2010). The high disease incidence is also in part attributed to the presence of predominantly susceptible Arabica coffee varieties to both CLR and CBD notably; Bugisu local, SL14, KP423 and SL28 (Musoli *et al.*, 2001), which were observed throughout the survey. Most coffee farms surveyed had very old trees (30 years and above) which had not been stamped and pruned in a long time (Matovu *et al.* Unpublished data). As a consequence, poor management coupled with prevailing environmental factors could have enhanced susceptibility of coffee to leaf rust and Coffee Berry Disease. Bock (1962) stresses the influence of some agronomic practices such as pruning, weed management and use of soil amendments on CLR development. Hakiza, (1997), MouenBedimo *et al.* (2007) and Bigirimana *et al.* (2012) observed that high level management including:- pruning, mulching, appropriate fertiliser application and good weed control contribute to masking the adverse effect of CLR and CBD epidemic on Arabica and Robusta coffees. These good

management practices increase plant vigour, making them more tolerant to disease attack (Bigirimana *et al.* 2012).

Zambolim *et al.* (2005) concluded that there exists a relationship between CLR incidence and altitude which is more or less opposite to CBD. This observation agrees with findings of this study. This relationship could be due to varying temperatures and moisture conditions as altitude increases. This subsequently translates into successful spore germination and colonisation of CLR and CBD at different altitudes. For instance, in Kenya, CLR is almost not an economic problem at higher altitudes especially if the intensity of rainfall is low (Kushalapa and Eskes, 1989). Similarly, Rivera (1984) and Bigirimana *et al.* (2012) observed a lower level of CLR intensity at higher altitudes in Guatemala and Rwanda respectively. At lower altitudes, CLR may benefit from higher temperatures (Lamoroux *et al.*, 1995). Hindorf and Omondi (2011) stated that CBD occurrence depends mostly on altitude ranges, with high incidences in higher sites with favourable climatic conditions than lower sites. This explains the observation of CBD only in high altitude areas (above 1800 masl) of Kapchorwa and Nebbi districts and not in Sironko district which lies at the foot of Mountain Elgon. These results are in agreement with Mulinge (1971) and Zeru *et al.*, (2009) who observed higher CBD incidence in elevated coffee growing areas than in low areas. Thus, the findings of this study provide vital baseline information which could be used in prioritising research efforts in order to breed resistant varieties with adaptability to such conditions (Bigirimana *et al.* 2012).

The results present a complex relationship between CLR incidence and

severity with shade intensity. Our results are in agreement with López-Bravo *et al.* (2012), who reported that shade effects on coffee rust are often controversial, with two probable pathways antagonising each other. High incidence and severity within thin and medium shade intensities can be explained by presence of optimum micro-climate conditions that favour CLR pathogen infection and colonisation of coffee leaves (Beer *et al.*, 1998). In addition, efficient light penetration under such conditions keeps temperatures well regulated. Temperature is one of the most important environmental factors that determines spore germination and penetration of *Hemileia vastatrix* (Beer *et al.*, 1998). According to Avelino (2010), shade trees tend to reduce berry load on coffee trees, which in turn reduces plant stress and consequently translates into increased plant resistance to CLR. Conversely, the low levels of CLR incidences and severity under thick shade can be explained by the low light penetration under such conditions. This translates into low temperatures which enhance host recognition by the pathogens and eventually successful infection. Shade systems in coffee mainly act on environmental parameters in limiting disease incidence (MouenBedimo *et al.*, 2008). The low CLR incidence in open coffee farms can be attributed to less competition for resources such as soil nutrients, moisture and light which occurs among coffee inter-planted with shade trees. Nutrient stress predisposes coffee to disease infection especially during heavy bearing stages of growth (Agrios, 2005; Waller *et al.*, 2007; McMahon, 2012). Additionally, open coffee farms possess lower humidity levels than shaded farms an environmental factor required during successful penetration of host

tissue by coffee rust. However, our results contrast work by Eskes (1982) who reported a weak correlation between the level of shade and incidence of *Hemileia vastatrix*. In his study, coffee leaf rust caused serious defoliation to both sun grown and shaded coffee. Furthermore, our results show that mean CBD incidence and severity were not significantly different across the various shade intensities. However, the low mean CBD incidence and severity on coffee growing under thick shade cover may be due to reduced rain intensity and subsequently, reduced splash dispersal of *Colletotrichum kahawae* (Ntahimpera *et al.*, 1998; MouenBedimo *et al.*, 2008)

Our data show that the highest CLR incidence was recorded in farms having only one tree species as compared to farms with no shade and mixed tree species. This can in part be attributed to loss of biodiversity in a mono tree species shade system (Avelino, 2010). In addition, a mixed shade type probably possesses numerous ecological benefits to coffee such as nitrogen fixation, manure addition, improvement of soil water retention, reduction in high solar radiation and suppression of weeds. This combination synergistically curtails pathogen incidence. Our results further show that mean CBD incidence was higher on coffee growing under a mono shade tree species system and no shade as compared to that growing under mixed shade tree species. These results are in agreement with MouenBedimo *et al.* (2007) who reported higher CBD infection rate on coffee grown intensively than coffee trees grown in a traditional manner. Mixed cropping with shade plants are cultural practices that limit CBD development (MouenBedimo *et al.*, 2007).

The higher mean incidence in CLR and CBD in clay than in loam soils could in part be attributed to the fact that clay soils are very heavy with excessive water retention and hence are mostly affected by leaching (McCauley *et al.*, 2005 and Waller *et al.*, 2007). This in turn affects the ability to retain soil nutrients (Leslie, 2002) which are indirectly responsible for plant vigour and the state of readiness of plants to defend themselves against pathogenic attack (Agrios, 2005; Stone *et al.*, 2003). This renders coffee trees planted in clay soil very susceptible to CLR. Our results are in agreement with Lamoroux *et al.* (1995) and Medina and Ascanio (1995) who reported that CLR was generally associated with poor soil structure. In contrast, results indicated that loam soils may be better for coffee production owing to their good properties such as increased moisture, air and nutrient retention which enable crop production with increased disease resistance (Stone *et al.*, 2003). Our results agree with findings by Ndo *et al.* (2010). The authors reported lower incidence of *Phaeoramularia* leaf and fruit spot disease of citruson trees growing on volcanic soils known for high fertility.

The lowest CLR and CBD incidence and severity was observed in farms located on gentle slopes. Our results are in line with other researchers who have reported inter-relationships between topography and fungal diseases such as *Colletotrichum coffeanum* (Nutman *et al.*, 1960) and American leaf spot, *Mycenacitricolor* (Avelino *et al.*, 2007). Soils in such topographical locations are characterised by deep fertile soils, less soil erosion and high soil moisture. Although these factors may not directly influence

rust incidence and severity, the crop can benefit from enhanced soil fertility and reduce chances of stress which predisposes host plants to their pathogens (Hakiza, 1997). In contrast, highest disease incidence was observed in farms on very steep, steep and medium slopes. Similarly, Avelino *et al.* (2007) reported that slopes are conducive for epidemics of some fungal diseases such as the American leaf spot, *Mycenacitricolor* and several leaf spots (Malhi and Kutcher, 2004). Such farms are prone to high surface run-off and in extreme cases landslides may occasionally occur for instance in mountain Elgon ranges of Uganda. Farms in such topographical levels possess fragile soil nutrients unless mitigation strategies are put in place. Steep topographies are very synonymous with river valleys, where a micro-climate which favours disease development is created (Nevalainen, 2002).

Conclusions

Coffee Leaf Rust occurs in all surveyed Arabica coffee growing districts of Uganda damaging coffee at substantial levels, enough to cause economic losses. It is evident that CLR is more severe in susceptible farms at low altitude, which is the reverse with CBD. On the other hand, CBD occurs in high altitude areas of Kapchorwa and Nebbi districts, although it can be very destructive even at low altitudes as long as conditions are favourable. All surveyed ecological factors in some way influenced CLR and CBD incidence and severity at a certain threshold level which presents an opportunity for Integrated Pest Management (IPM).

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Genetic Diversity among Disease Resistant Coffee Varieties and Cultivars in Rwanda Based On RAPD and SSR Markers

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Abstract - Understanding the genetic diversity existing in a germplasm collection is important to identify parental combinations with distinct gene sets that can be used in crossing to obtain superior hybrids. This study was carried out to evaluate the genetic diversity in the Rwandan coffee germplasm using both RAPD and SSR markers and to determine any relationship between these two markers. Varieties with resistance to coffee leaf rust and coffee berry disease as well as local cultivars in Rwanda were used for DNA extraction and subjected to PCR amplification. The highest values for genetic distances were obtained between BM 139 and HDT and between Harar and Catimor implying that these varieties were genetically quite distinct. The lowest values for genetic distance were recorded between BM 139 and BM 71 and between Matinho and Rume Sudan implying that these varieties were genetically fairly similar. A Pearson correlation of 55.52% was obtained between the two markers indicating a moderate association between these two analytical procedures. It is recommended that crosses should be made between genetically distant susceptible and resistant varieties to derive hybrids that combine resistance with marked levels of heterosis particularly for yield.

Keywords - DNA Extraction, Heterosis, Genetic Distances, Hybrids

1. Introduction

Coffee is an important agricultural export commodity and accounts for the bulk of the export earnings in more than 50 developing countries of Africa, Asia and Latin America [1]. For Rwanda in particular, it accounts for about 25% of its annual foreign exchange earnings [2]. The major constraints to coffee productivity in Rwanda include prolonged periods of moisture stress, low soil fertility, pests and diseases such as coffee leaf rust (CLR) and coffee berry disease (CBD). CBD for example, can cause up to 80% crop loss if not controlled and conditions are favorable [3]. Preventive control by fungicide sprays can account for 30 – 40% of total production costs and annual economic damage in Africa has been estimated at US\$ 300 - 500 million [4]. CLR, which is the major disease on coffee in Rwanda, is estimated to cause crop losses of up to 40% annually [5]. Cultivation of resistant varieties is the most viable and effective option for disease management.

Rwanda has introduced coffee accessions from various sources including among others, Ethiopia, Democratic Republic of Congo (DRC), Brazil, Uganda and Kenya. These accessions are currently maintained *ex-situ* in germplasm collection plots. The collections include varieties of several

species e.g. *Coffea arabica*, *Coffea canephora* and *Coffea liberica*. Though the commercial coffee varieties grown in Rwanda were selected among these accessions, many others are of no direct commercial value but represent an important source of genetic variation for characteristics such as canopy architecture, pests and disease resistance, yield, quality and other agronomic and industrial traits.

The knowledge of genetic diversity existing in a germplasm collection is useful in the development of new varieties [6]. Determination of genetic diversity helps in identification of parental combinations, in order to use distinct gene sets in crossing to obtain superior hybrids [7]. Determination of genetic diversity among coffee accessions was often based on evaluation of morphometric characters; however, more recently a number of molecular techniques have been used to measure this diversity in coffee. Lashermes et al. [8] used RAPD markers to study the genetic variability and relationships of *Coffea* species and showed consistent relation to the known history and evolution of the *Coffea* species. Aga et al. [9] showed the existence of four different populations for *in-situ* conservation of Ethiopian *Coffea arabica* species

while Masumbuko et al. [10] suggested the widening of the existing genetic base by introducing more accessions in Tanzanian cultivated Arabica coffee.

A combination of different markers may provide more reliable information about genetic diversity compared to the use of a single marker because errors presented by one marker could be minimized using other markers [7, 11]. This study was carried out with the following objectives: (1) to evaluate the genetic diversity in the Rwandan coffee germplasm using RAPD and SSR markers and (2) to determine any relationship between data generated with these two analytical procedures.

Table 1. Description of Coffee Genotypes Used in the Study

Name	Source	Type	CLR and CBD Resistance phenotype
Jackson 2/1257	DRC	Cultivar	susceptible
Matinho	Angola	Semi wild	resistant
Catimor	Costa Rica	Breeding line	resistant
BM 71	DRC	Cultivar	susceptible
Selection 5A	India	Breeding line	resistant
Harar	Ethiopia	Cultivar	susceptible
CIFC 8224	Portugal	Breeding line	resistant
Pop 3303/21	DRC	Cultivar	susceptible
Mibilizi	DRC	Cultivar	susceptible
SL 28	Kenya	Cultivar	susceptible
Rume Sudan	Sudan	Semi wild	resistant
Hibrido de Timor	Kenya	Wild	resistant
Selection 6	India	Breeding line	resistant
BM 139	DRC	Cultivar	CBD resistant but susceptible to CLR

2.2. DNA Extraction

Genomic DNA was extracted from fresh leaves following the method described by Diniz et al. [12] with minor modifications. Five hundred milligrams of fresh leaf material without mid-veins were frozen in liquid nitrogen and ground to powder and 0.35 – 0.4 g of the powder was collected in eppendorf tubes and 800 µl of extraction buffer (400 µl of lysis and 400 µl of extraction buffer) added. The mixture was incubated for 30 min at 62° C and 1 ml of chloroform and isoamyl alcohol (24:1 v/v) was added and centrifuged at 13 000 rpr for 5 min and the supernatant was discarded.

The pellet was then washed with 250 µl of 70 % ethyl alcohol and the mixture was homogenized with gentle inversion and the supernatant was discarded. The pellet was air dried and dissolved in 50 µl of TE (10 mM Tris-HCl, 1 mM EDTA pH 8) and left at 4° C over night to dissolve. The resulting aqueous fraction was incubated with 20 µl of RNAse (10 mg/ml) at 37° C for 30 min and kept at – 20° C for later use. The DNA quality was checked by electrophoresis in 1 % agarose gel at 50 W for 45 min. The DNA was visualized and photographed in UV trans-illuminator chamber after staining

2. Material and Methods

2.1. Plant Material

Fourteen genotypes were used in the study. Their characteristics are described in table 1. Seeds of these varieties were germinated in sterile sand, transplanted into polythene bags and then transferred into a nursery. The leaf samples used for DNA extraction were collected from 8 months old seedlings growing in the nursery.

in Ethidium bromide (2.5 mg/l) for 20 minutes. The DNA quantity was estimated by visually comparing the obtained bands for different varieties with standardized Lambda DNA ladders.

2.3. Determination of Genetic Diversity by RAPD-PCR Analysis

Fifteen primers were screened on all samples to identify the ones which could detect polymorphism among varieties. Nine primers which showed variations were selected for further analyses (Table 2). The DNA amplification was performed on 25 µl reaction mix containing 12 µl of double distilled water, 2.5 µl of the buffer (75 mM Tris – HCl, pH 8.8, 20 mM (NH₄)₂SO₄, 0.01% (v/v) Tween 20), 2.5 µl of MgCl₂ (25mM), 1 µl of the primer (10 mM), 0.2 units of Taq polymerase, 3.8 µl of dNTPs (500 µM) (100 µM each of dATP, dCTP, dGTP and dTTP) and 1 ng per µl of sample DNA. A master mix was prepared for each primer to minimize measurement deviations.

Amplification reactions were carried out in a Techne-thermocycler with one cycle of initial strand separa-

tion at 94°C for 3 minutes followed by 45 cycles of 1 minute at 94°C, 1 minute at 37°C, and 2 minutes at 72°C. The last cycle was followed by an additional extension at 72°C for 10 minutes. The PCR amplification products were separated on 2.3% agarose gel and run in 1 x TE buffer (40 mM Tris acetate pH 8.0, 1 mM EDTA) at 100 volts for 4 hours. Staining was done using ethidium bromide (2.5 mg/l) for 30 minutes and the DNA was visualized on a UV transilluminator chamber and photographed using polaroid film. The gel photographs were evaluated as the presence (1) or absence (0) of the amplified bands.

2.4. Determination of Genetic Diversity by Microsatellite Analysis

Fifteen SSR primer pairs were screened for PCR amplification and only six primers that generated polymorphism were used for SSR analysis. Reaction master mix consisted of 25 µl containing 5 µl of 10 ng/ µl genomic DNA, 2.5 µl of buffer (10X, Promega), 2.5 µl of the MgCl₂ (25 mM, Promega), 7.5 µl of SSR dNTPs (dNTPs stock with a little dATPs), 2.5 µl each of right and left primers, 0.2 units of Taq DNA polymerase and 2.3 µl of double distilled water. The PCR programme consisted of an initial denaturation at 94°C for 2 min, followed by 5 cycles of 45 seconds of denaturation at 94°C, 1 minute primer annealing at 60°C reducing by 1°C every cycle, elongation for 1 minute at 72°C and 30 cycles of 45 seconds of denaturation at 90°C for 45 seconds, primer annealing at 55°C for 1 minute and elongation at 72°C for 1 minute 30 seconds and final extension of 8 minutes at 72°C.

Denaturing polyacrylamide (6%) gel was prepared in 33 cm x 39 cm casting plates separated by 0.35 mm spacers. A plane mould was inserted at the top and the gels were left overnight for use the next day. The gels were assembled in vertical electrophoresis and 1X TE buffer for running was placed into the top reservoirs to cover the inner smaller plates. After ascertaining that there was no leakage, the plane moulds were removed and the gels rinsed with the running buffer.

More running buffer was put into the bottom reservoir and pre-runs were made at 55 W per gel for about 20 min before the samples were loaded. Similar to RAPD analysis, the polyacrylamide gels were interpreted for the presence (1) or absence (0) of the bands. Each of the bands was treated as an independent character.

2.5. Statistical Analysis

Genetic distances (GD) between genotypes were estimated using the method of Van der Peer & De Wachter [13] as follows:

$$GD_{xy} = (N_x + N_y) / (N_x + N_y + N_{xy})$$

Where,

N_x : Number of bands in line X and not in line Y

N_y : Number of bands in line Y and not in line X

N_{xy} : Number of bands in line Y and in line X

The similarity matrices computed for each pair of genotypes were subjected to cluster analysis using the Unweighted Pair – Group Method with Arithmetic Averages (UPGMA) and dendrograms were generated using SPSS (2007) to visualize the genetic similarity between varieties. Data generated using microsatellites and RAPD analyses were also analyzed for correlation using Microsoft Excel 2007.

3. Results

3.1. Polymorphic Information Generated by RAPD Analysis

The nine RAPD primers generated a total of 43 polymorphic bands across 14 coffee varieties. The number of bands per primer varied from two (N-18) to eight (I-7) with an average of 4.8 bands per primer, and the estimated molecular weight was in the range of 180 to 1000 base pairs (Table 2). Of the nine RAPD primers, 60.5 percent of the polymorphic bands were produced by 4 primers (X-20, M-4, I-7 and Y-15) whose frequencies ranged from 0.14 to 0.19.

Table 2. Primers Used in the RAPD Analysis Along with Their Nucleotide Sequences, Number of Polymorphic Bands and Estimated Molecular Size Bands

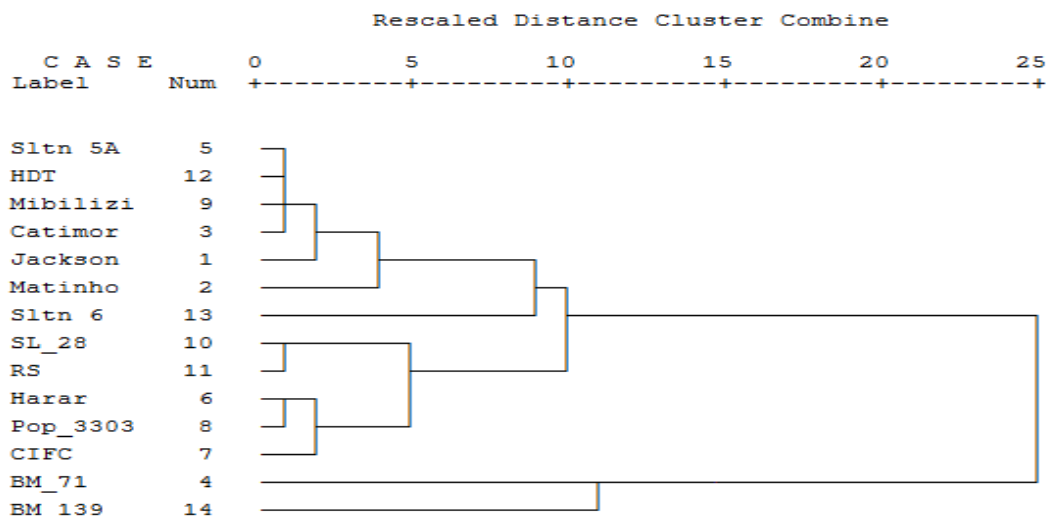
Primer	Sequences 5' to 3'	Number of polymorphic bands	Molecular size range
X-20	CCCAGCTAGA	6	300 to 700 bp
Y-10	CAAACGTGGG	4	200 to 800 bp
M-4	GGCGGTTGTC	6	200 to 700 bp
L-18	ACCACCCACC	3	320 to 600 bp
I-7	CAGCGACAAG	8	180 to 1100 bp
N-18	GGTGAGGTCA	2	350 to 900 bp
J-19	GGACACCACT	5	400 to 950 bp
X-16	CTCTGTTCGG	3	220 to 500 bp
Y-15	AGTCGCCCTT	6	350 to 800 bp
Total		43	
Mean		4.8	
Range		2 to 8	180 to 1100 bp

Table 3. Microsatellite Primers Used, the Number of Alleles Detected Per Primer and Their Estimated Allele Sizes

Primer	Nucleotide sequence	Number of alleles	Estimated allele size
M24	GGCTCGAGATATCTGTTTAG (Forward)	5	450 - 800 bp
M24	TTTAATGGGCATAGGGTCC (Reverse)		
Sat 235	TCGTTCTGTCATTAAATCGTCAA (Forward)	4	320 - 750 bp
Sat 235	GCAAAATCATGAAAATAGTTGGTG (Reverse)		
Sat 172	ACGCAGGTGGTAGAAGAATG (Forward)	3	350 - 600 bp
Sat 172	TCAAAGCAGTAGTAGCGGATG (Reverse)		
Sat 227	TGCTTGGTATCCTCACATCA (Forward)	2	220 - 700 bp
Sat 227	ATCCAATGGAGTGTGTTGCT (Reverse)		
Sat 229	TTCTAAGTTGTTAAACGAGACGCTTA (Forward)	3	180 - 550 bp
Sat 229	TTCCTCCATGCCCATATTG (Reverse)		
Sat 254	ATGTTCTTCGCTTCGCTAAC (Forward)	2	250 - 650 bp
Sat 254	AAGTGTGGGAGTGTCTGCAT (Reverse)		
Total		19	
Mean		3.2	
Range		2 to 5	180 - 800 bp

The similarity matrix revealed values ranging from 0.12 to 0.67 with an average of 0.45 (Table 3). The highest genetic distances were obtained between BM 139 and HDT and between Harar and Catimor implying that these varieties were genetically different. However, the lowest values for genetic

distance were recorded between BM 71 and BM 139 and between Matinho and Rume Sudan implying in turn that these varieties were genetically similar. The generated dendrogram for all analyzed varieties showed five clusters (*figure 1*)

**Figure 1.** Relationships between 14 Arabica Genotypes Generated by Unweighted Pair Group Method Using Arithmetic Averages (UPGMA) Based on RAPD Markers

3.2. Polymorphism Obtained with SSR Markers

The analysis with six SSR markers revealed a total of 19 alleles across the fourteen varieties. The number of alleles per primer ranged from two (Sat 254 and Sat 227) to five (M24) with an average number of 3.2 alleles per used primer. The estimated allele size for all primers ranged from 180 bp to 800

bp (Table 3). Three primers (M24, Sat 235 and Sat 227) generated 57.9% of the total number of alleles with an overall frequency ranging from 0.11 to 0.26. The highest genetic distances were obtained between Mibilizi and Rume Sudan and between BM 139 and Hibrido de Timor indicating that these varieties were genetically distinct. The lowest values were obtained between varieties BM 71 and Mibilizi and BM

139 and BM 71, respectively indicating that these varieties were genetically similar. The similarity matrix showed values ranging from 0.13 to 0.78 with an average of 0.46 (Table 4).

Unlike with the RAPD analysis, CLR and CBD resistant varieties were clustered separately from the commercial coffee cultivars (Figure 2).

Table 4. Matrix of Genetic Distance between 14 Genotypes Based on SSR Markers

	BM 139	Json	Mno	Cmor	BM 71	Sel 5A	Hrar	CIFC	Pop3303	Mzi	SL 28	RS	HDT
Json	0.37												
Mho	0.66	0.48											
Cmor	0.55	0.53	0.58										
BM 71	0.19	0.28	0.55	0.56									
Sel 5A	0.39	0.57	0.67	0.37	0.69								
Harar	0.24	0.34	0.39	0.58	0.33	0.56							
CIFC	0.51	0.43	0.2	0.23	0.53	0.21	0.53						
Pop 3303	0.2	0.26	0.51	0.51	0.17	0.68	0.2	0.57					
Mzi	0.27	0.18	0.65	0.56	0.13	0.69	0.23	0.65	0.35				
SL 28	0.25	0.26	0.37	0.63	0.24	0.66	0.21	0.52	0.38	0.33			
RS	0.6	0.44	0.34	0.23	0.58	0.25	0.23	0.35	0.6	0.78	0.68		
HDT	0.71	0.67	0.55	0.31	0.52	0.37	0.54	0.69	0.53	0.56	0.51	0.45	
Sel 6A	0.68	0.7	0.56	0.32	0.67	0.26	0.63	0.8	0.62	0.7	0.67	0.34	0.3

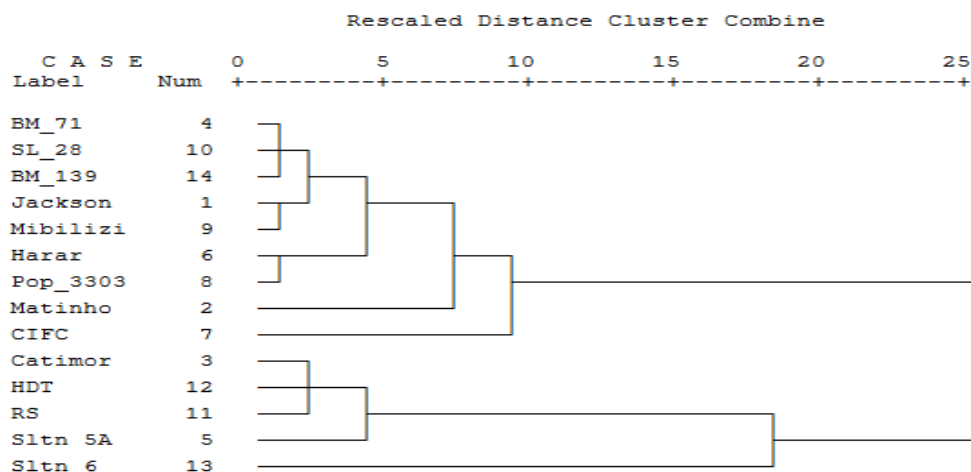


Figure 2. Relationship between 14 Arabica Genotypes Generated by Unweighted Pair Group Method Using Arithmetic Averages (UPGMA) Based on SSR Markers

3.3. Relationship between Genetic Distances Obtained with RAPD and SSR Markers

Estimates of correlation between genetic distances obtained by RAPD and SSR markers were moderate, with magnitudes of 55.52%, indicating that there is a relative pattern of association between results obtained using these two procedures

(Figure 3). The linear regression equation was found to be $Y = 0.8067 X + 0.1144$, where Y was the genetic distance generated based on RAPD markers and X the genetic distance with SSR markers. Both slope and intercept were highly significant ($p < 0.001$).

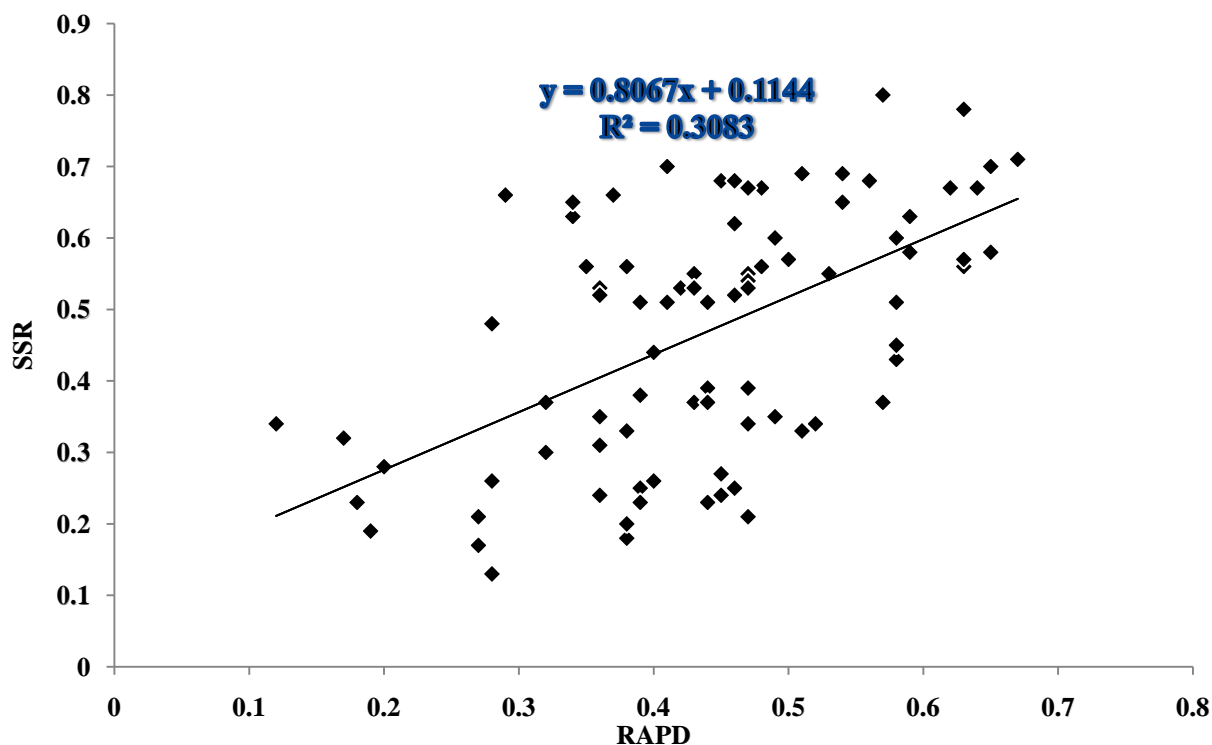


Figure 3. Relationship between Genetic Distances Generated by SSR and RAPD Markers

4. Discussion

The results of the study indicated that the highest values for genetic distance were obtained between Mibilizi and Rume Sudan and between BM 139 and Hibrido de Timor (HDT) for both markers. The genetic dissimilarity between BM 139 and HDT may be explained by their divergent background because BM 139 is a selection among Arabica coffee introductions in Mulungu while HDT is a natural interspecific hybrid between *Coffea arabica* L. and *Coffea canephora* Pierre found in East Timor [14]. While working on different coffee species, Lashermes et al. [8] also observed genetic differences between Arabica coffee and HDT. On the other hand, the genetic differences between Rume Sudan and Mibilizi may be attributed to their genetic genealogy because Mibilizi is a selection from Mulungu whereas Rume Sudan originated in the Boma plateau of Sudan, a region adjacent to the primary centre of genetic diversity situated in the highlands of south west Ethiopia. Leal et al. [7] working on lines with different origins also reported that varieties coming from different locations tended to be genetically different from lines of the same provenance.

While Mibilizi and Rume Sudan showed the highest genetic distance, BM 71 and Mibilizi and BM 139 and BM 71 displayed the lowest values. The genetic similarity between BM 71 and Mibilizi and BM 139 and BM 71 may be explained by their origins as they were selected among introductions from Mulungu. BM 139 and BM 71 were probably developed from the same parents by single tree selections. In addition, these are Arabica coffee varieties which are reported

to have a low genetic diversity [6]. The narrow genetic base in Arabica coffee may also be explained by the high level of homozygosity as *C. arabica* is a self-pollinated species. Analysis by SSR markers grouped local cultivars and varieties that are resistant to CLR and CBD in separate clusters. Similar results are reported by Agwanda et al. [15] using RAPD markers; while Setotaw et al. [16] assessing the genetic diversity of Hibrido de Timor germplasm found that the trees constructed using SSR and RAPD systems were very similar.

In this study, UPGMA analysis of the RAPD and SSR markers showed some similarities. For instance, varieties BM 71 and HDT were placed at the ends of dendrograms. However, some varieties such as Harar and Selection 5A occupied different positions. Maluf et al. [17] evaluating the genetic diversity of cultivated Arabica coffee inbred lines using RAPD, SSR and AFLP markers also observed that some genotypes were not similarly distributed in clusters produced with RAPD and SSR markers. The possible explanation to these dissimilarities may be the fact that molecular markers evaluate different components of DNA variations which can progress in diverse ways (Collard et al. [18]).

Moderate association was obtained between genetic distances based on RAPD and SSR markers, indicating some differences between the two molecular techniques. Similar observations are reported in other studies on maize by Souza et al. [19] and Leal et al. [7] who found correlations of 0.54 and 0.55, respectively, between RAPD and SSR markers.

The study showed genetic diversity between commercial

coffee cultivars in Rwanda and CBD and CLR resistant varieties. The latter category included varieties such as Rume Sudan and Selection 6, Catimor and CIFC 8224. However, despite the poor yield and other inferior agronomic traits, variety Rume Sudan has good resistance to CBD and CLR and has been used along with Catimor in the breeding program for resistance to CBD and CLR in Kenya which led to the development of variety Ruiru 11 [14]. Similarly, concerning disease resistant variety CIFC 8224, Setotaw et al. [16] reported that it originated from natural interspecific hybridization between *C. arabica* and *C. canephora* and has also been used for the development of CBD and CLR resistant varieties all over the world.

The results obtained in this study may be useful in planning breeding procedures for the development of varieties that combine resistance to CLR and CBD and other desirable traits such as high yield and better quality. For example, crosses should be made between genetically distant susceptible and resistant varieties in order to derive hybrids that combine disease resistance with marked levels of heterosis particularly for yield.

5. Conclusions

The knowledge of the genetic diversity is important to identify parental combinations with distinct gene sets that can be used in crossing to obtain superior hybrids. Local cultivars such as BM 139, BM 71 and Mibilizi are genetically quite distinct from CBD and CLR resistant varieties like Rume Sudan, Selection 6 and HDT. Advantage should be taken of this situation to make crosses between these two groups of varieties in order to derive disease resistant hybrids that combine a marked level of heterosis for yield with improved adaptation and quality.

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