INCO : International Scientific Cooperation Projects

Contract number : ICA4-CT-2001-10006

Development of a long term strategy based on genetic resistance and agroecological approaches against Coffee Wilt Disease in Africa.

Fourth intermediate report: covering period 1/11/2004 to 30/04/2005

Acronym: COWIDI

Coordonator principal CIRAD

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Report n° 13/2005
CIRAD/AMIS
Crop Protection Program

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SUMMARY

During the period 1/11/2004 to 31/4/2005, the focus was on survey of wild coffee in primary forests of Uganda and the disease involved, improving the knowledge on genetic diversity of the pathogen with the use of new markers, evaluating influence of the sexual cycle on the diversity of the pathogen, identifying new resistant genotypes and planting resistant genotypes in hot spots disease.

New results

The biomolecular analysis of diversity of the pathogen continued with new markers (MATgenes, RAPD, Tef...) and the results obtained confirms previous results. The markers differentiated the contemporary canephora and arabica strains, from historical strains.

In the absence of genetic diversity detectable with different neutral markers, the MATgene has been identified to be the only marker available for characterization of the progeny in a fertile cross and for a clearer understanding of geographical and historical gene flow. Understanding inheritance and distribution of this marker will enable study of the meiotic recombination thus improve knowledge on the sexual cycle of the pathogen

The trial on the sexual cycle is in progress in UNIKIN with identified mating type in screen house and natural condition (Beni)

The database of isolates has been updated including newly acquired isolates

The host pathogen interaction reveal that historical isolate DSMZ collected on C. excelsa induce symptoms on C. canephora and C. excelsa. This isolate is therefore not specific to *C. excelsa*.

Two surveys of wild *coffea canephora* trees in primary forests (Kibale and Itwara) in Uganda were carry out and samples of fungi involved with wilt symptoms were collected. This is to improve on the knowledge about the genetic diversity of the Ugandan *Coffea canephora* and finf out identified if the wilt is present in primary forest.

This collection of wild coffee will increase the germplasm available for improving resistance to coffee wilt and other agronomic traits (yielding, cup quality...).

Some DRC and Ugandan germplasm are identified resistant to CWD.

New isolates collected in areas newly affected by the disease in the Province de l'Equateur in DRC was included in the evaluation of the aggressiveness.

Priorities for the next 6 months

Study of the fertility between Fusarium xylarioides type A, type C and historical strains.

Comparison of calmoduline sequences.

Identification of fungi involved with wilt on wild coffee in Uganda primaries forests.

Analysis of genetic diversity and evaluation of resistance of wild coffee

Analysis of inheritance of wilt resistance of new hybrids and planting of resistant survivors to artificial inoculations

Hold a short implementation meeting

INCO-DEV COFFEE WILT PROJECT

(Contract no. ICA4-CT-2001-10006)

Development of a long-term strategy based on genetic resistance and agroecological approaches against Coffee Wilt Disease in Africa.

WP1: Pathogen diversity

Biodiversity study of *Fusarium xylarioides*, a phytopathogenic fungus of coffee, taking a microsatellite approach:

The study of the biodiversity of *Fusarium xylarioides* continued by searching for the center of origin of the disease. Recent works by Pascal Musoli showed that a center of origin or diversification of *C. canephora* in Uganda could be in the western primary forest (Kibale forest). The centers of origin or diversification of a botanical species correspond in many cases with the center of origin of the disease where the plant and the pathogenic co-evolved and represent a center of diversity for the pathogen.

During two joint missions of prospections of wild coffee trees in the forests of Kibale national Park in November, 2004 and in the forests of Itwara in February, 2005, a search to identify coffee trees with coffee wilt was carried out. Trees with symptoms similar the wilt were identified. The recognition of the disease in forest is relatively unpredictable because the morphological aspect of coffee trees under strong shade, in these forest are very different from coffee tree plantations.

In Kibale forest, a single tree with symptoms comparable to the symptoms of wilt was taken. In Itwara forest several samplings were made.

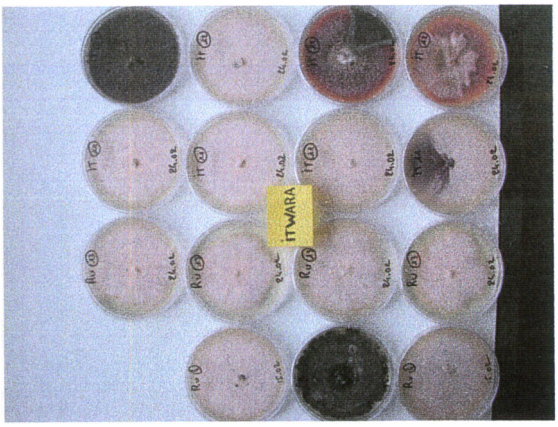
The isolations of fungi from wooden samples were realized and the single spore isolates are now available.

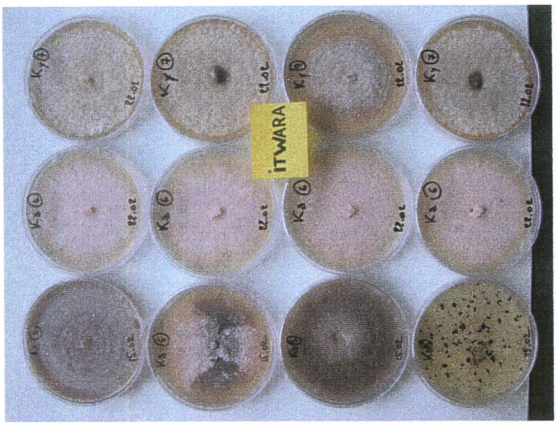
Their morphological analysis and ITS analysis will be carried out.

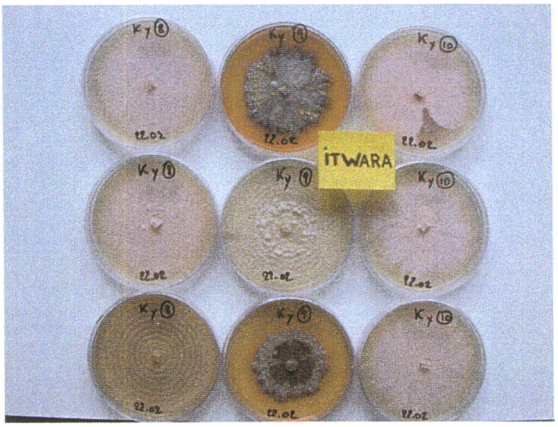
	Lieu	Date	Original strain	Single sporer
Kibale forest	Ngogo Kibuguta	30/11/2004		OUG163
	Ngogo Kibuguta	30/11/2004		OUG164
	Ngogo Kibuguta	30/11/2004		OUG165
	Ngogo Kibuguta	30/11/2004		OUG166
Itwara forest	Rutoona (Block 15)	31/02/2005	OUG167	
	Rutoona (Block 15)	31/02/2005		OUG168
	Kanaaba	01/02/2005		OUG169
	Kanaaba	01/02/2005	OUG170	
	Kanaaba	01/02/2005		OUG171
	Kanaaba	01/02/2005	OUG172	
	Kanaaba	01/02/2005	OUG173	
	Kanaaba	01/02/2005		OUG174
	Kanaaba	01/02/2005	OUG175	
	Kanaaba	01/02/2005	OUG176	
	Kyamuhoro	01/02/2005		OUG177
	Kyamuhoro	01/02/2005	OUG178	
	Kyamuhoro	01/02/2005		OUG179
	Kyamuhoro	01/02/2005	OUG180	
	Itwara	02/02/2005		OUG181
	Itwara	02/02/2005	OUG182	
	Itwara	02/02/2005		OUG183
	Itwara	02/02/2005	OUG184	
	Itwara	02/02/2005		OUG185
	Itwara	02/02/2005	OUG186	

List of strains collected in Kibale and Itwara Forest

Wild coffee canephora with dried leaves similar to C.W.D.







Wild strains collected in Itwara forest

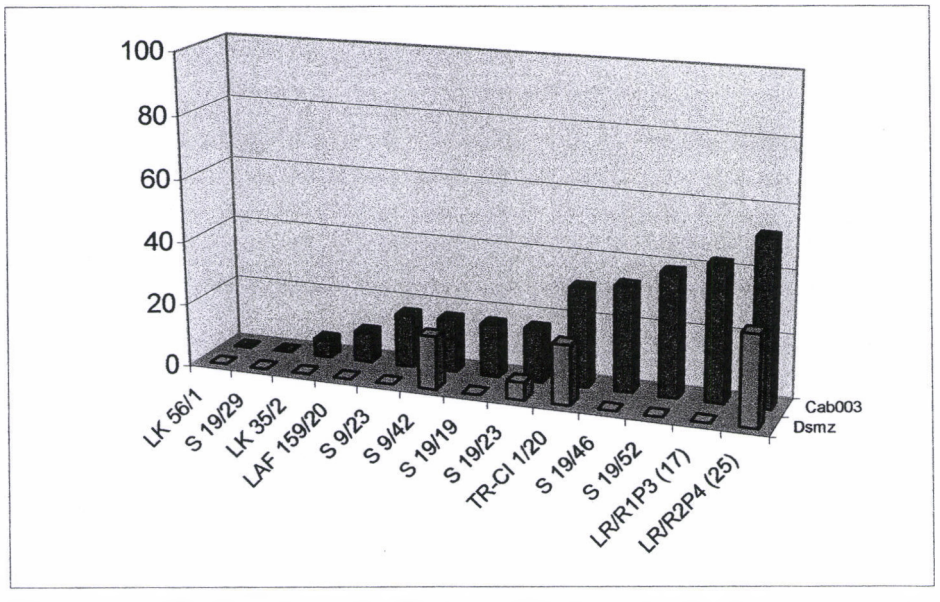
WP2: Host/Pathogen interaction

Task 1: Identification of isolates of *Fusarium xylarioides* representative of genetic diversity and aggressiveness

Host-pathogen interaction was carried out by inoculations on *Coffea canephora* from DRC, in relation to 2 isolates:

- DSMZ collected from *Coffea excelsa* (historical name probably indicating a harvest from *Coffea liberica*, in Cental Africa Republic.

- CAB003, reference strain collected from *C. canephora* in Uganda.



Percentage of infected plants 150 days after inoculation on a set of *C. canephora,* with strains collected from *Coffea canephora* (CAB003) and historical strain *Coffea excelsa* (DSMZ).

Isolate CAB003 induced symptoms on the seedlings of all the populations except population LK56/1 and S 19/29 without symptoms. The percentage of infected plants is 6 to 52%.

Isolate DSMZ induced symptoms on 4 genotypes, susceptible also with CAB003.

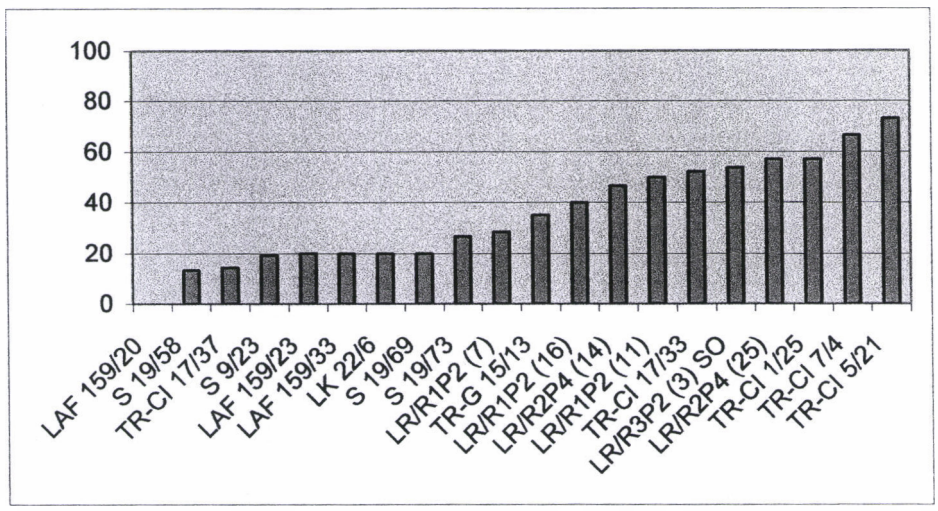
Conclusions

These results indicate that DSMZ collected on *C. excelsa* can induce symptoms and death of *C. canephora* and confirm previous results. This isolate is not specific to coffee trees belonging to *C. excelsa*.

WP3 Breeding for resistance

Task 3: Conduct screening test in both Africa and Europe using isolates with a wide range of aggressiveness.

In this trial, 23 populations belonging to *C. canephora,* from DRC, were tested for their resistance to CAB003 (trial 54).



Ranking of 23 populations of *C. canephora* from DRC inoculated with CAB003, 150 days after inoculation (trial 54)

This result confirm the continuum of aggressiveness of isolate CAB003.

The genotype LAF159/20 did not show wilt symptoms. This inoculation will be repeated in the next trial.

COWIDI-ICA4-2000-10312

Report 1/11/2004-31/04/2005
PARTER II : Université catholique de Louvain (UCL)

OVERALL OBJECTIVE

Identify the mechanisms of variation of the coffee wilt pathogen, *Gibberella xylarioides*, through the sexual and parasexual cycles; and improve the knowledge of the genetic diversity within the pathogen population.

PROGRESS

WP I: Pathogen Diversity

Task I: Collection of both forms of the fungus on various parts of the trees, possibly alternative hosts, in infested regions

Coffea canephora stem samples showing typical coffee wilt symptoms were collected in the Equator Province (DRC) by UNIKIN (September-October 2004). Identification and further analysis of these strains is necessary for completion of the mating type distribution within the DRC and for confirmation that the pathogen present in these regions is identical to the causal agent isolated in the other Congolese, Ugandan and Tanzanian regions.

Task II: Identification, storage, and exchange of isolates

CIRAD has sent strain G3P22 from Ethiopia, and we are awaiting another 4 strains from **CABI** to add to our existing ***G. xylarioides* "Type A"** (isolated from *C. arabica*) collection. Four *Fusarium udum* strains, a close relative of *G. xylarioides* causing pigeonpea wilt in India and Malawi, have equally been ordered from CABI.

Strains NRRL 26064 (*Fusarium sp.*) and NRRL 22540 (*F. udum*), identified as *G. xylarioides* closest relatives based on translation elongation factor 1-∝ (*tef 1*) sequence analysis, were ordered via **K. O'Donnell** for mating type (*MAT*) identification, partial *MAT* gene sequencing, and cross fertility studies with *G. xylarioides*. In order to complete the cross fertility study, thus identifying *G. xylarioides* as a new mating population (MP) within the *G. fujikuroi* species complex (GFC), 18 standard mating population tester strains (MP A-I) have been ordered at the Fungal Genetics Stock Center (**FGSC**).

Task IV : Description of the fungal life cycle, asexual and sexual phases

In the third annual report (1/11/2003 - 1/11/2004 period) the coffee wilt pathogen was described as a **heterothallic** fungus based on *in vitro* crosses and *MAT*-PCR analysis. However, like other heterothallic *Gibberella* species, it is possible that *G. xylarioides* can occasionally self, and that the **partial homothallic nature** hypothesized in the second annual report (1/11/2002 - 1/11/2003 period) is founded. Indeed, a small number of perithecia were formed on a PDA plate of ascospore strain 15/SS06, five ascospores were isolated and stored for further analysis.

Moreover, in one of our last assays, a carrot agar "selfing" of IMI 375908 equally produced the teleomorph. Nine ascospores have been isolated, and parent and progeny will undergo *MAT*-PCR in order to verify the homothallic or heterothallic origin of the progeny.

MAT-1 and *MAT-2 G. xylarioides* "type C" strains (isolated from *C. canephora*) MUCL 44532/MUCL 44536, MUCL 46056/MUCL 46057, and CAB003/OUG008 are being tested to confirm their level of fertility before being sent to FGSC facilities as standard ***G. xylarioides* "Type C" mating population tester strains**. Testers for "Type A" will be established in a similar manner once a larger collection of arabica isolates is available.

In order to ensure ourselves of the heterothallic nature of the resulting perithecia formed in fertile confrontations, the progeny of three crosses (**Table 1**) were analysed by *MAT*-PCR. Both mating types were present in each cross, indicating that the progeny originated from the sexual recombination of both parents and were not of homothallic origin (from one of the parents).

The geographical distribution of *MAT-1* and *MAT-2* isolates will be studied for a clearer understanding of putative geographical and historical gene flows.

Crosses between *G. xylarioides* "type A", "type C", historical isolates, and other closely related *Fusarium* isolates will be carried out in every possible combination. Resulting structures, fertile perithecia as well as infertile fructifications, will be characterized and compared to the literature. Vegetative compatibility tests will also be carried out this summer.

Task V. Evaluation of genetic diversity within *F. xylarioides*

Random amplified polymorphic DNA (**RAPD**) polymerase chain reaction (PCR) was carried out using four decamer primers (A3, A14, A15, and A17) from kit A (Operon Technologies Inc.) that indicated certain levels of interspecies variation in the genus *Fusarium* (unpublished, Munaut). 25µl-PCR reaction mixtures contained 20 ng of template DNA, 1x PCR buffer, 1.5 mM $MgCl_2$, 0.2 mM each dNTP, 0.75µg/ml of each primer and 2.5 U of Taq DNA polymerase. PTC-200 thermocycleur conditions were 7 min at 94°C, followed by 45 cycles of 1.5 min at 94°C, 2 min at 35°C and 3 min at 72°C, followed by a final elongation at 72°C for 7 min (Sreenivasaprasad *et al.*, 1992).

Out of the four 10-mers tested in kit A, all clearly distinguished *F. proliferatum* (MP D), belonging the *G. fujikuroi* species complex (GFC), from *G. xylarioides* isolates (**Figure 1**). Primer A15 failed to differentiate any of the *G. xylarioides* strains, while primers A3 and A14 discriminated three distinct profiles, recent canephora/arabica strains, and group 1 (DSMZ 62457 & ATCC 15664) as well as group 2 (CBS 25852 & CBS 74979) within the historical strains. Primer A17 was the only primer that generated a distinct polymorphism for recent arabica

isolates as well as differentiating recent canephora and the two historical groups. Interestingly, "historical" strain MUCL 14186's profile is identical to recent canephora isolates.

Partial ***MAT* gene** sequencing is completed and has not revealed polymorphism within the *MAT-1* sequence. However, the amplified *MAT-2* fragment using previously described primer sets (Yoshida *et al.*, 1998; Kerényi *et al.*, 1999; Steenkamp *et al.*, 2000) differentiates at least 4 groups. Thirteen out of the 31 (~42%) *G. xylarioides* strains studied are *MAT-1*, with sequences 100% identical to each other regardless of their geographical, historical or host origin. At first glance it could be surprising that ***MAT 1-1*** doesn't differentiate strains, but it should be kept in mind that the *G. fujikuroi MAT 1-1* idiomorph, and supposedly that of *G. xylarioides*, spans more than 4600 bp containing three open reading frames (ORF) of which only ~300 bp (~1/10 of the coding region) in the *MAT 1-1-1* ORF have been sequenced. ***MAT 1-2*** sequences from recent *C. canephora* isolates and historical *C. canephora* and *C. excelsa* strains MUCL 14186 and BBA 62457 were 100% identical to each other regardless of their geographical, historical, or host origin. These strains will be referred to as the "core group" from now on to facilitate understanding. Historical strains CBS 25852 and CBS 74979 differ from each other by a single base pair and differ from the core group by four nucleotides (99% homology). The most distinct sequence is that of the *C. arabica* strains. In total, eight base pairs detach arabica strains from the core group. It should equally be noted that the *MAT 1-2* gene, spanning more than 3800 bp, contains a single ORF of which approximately a third has been sequenced to date. Sequencing of the entire *MAT* idiomorph in these strains should enable the differentiation of eventual "clades" within the actual and historical *G. xylarioides* population, as well as help in the understanding of the functions controlled by the *MAT*-ORFs. Preliminary amplification of the whole *MAT* idiomorph is equally underway.

Translation elongation factor (***tef 1-α***) sequencing is equally completed, allowing the identification of 3 "alleles" within the gene and not only the 2 previously described by Geiser *et al.* (2005). The first group consists of recent canephora, arabica, excelsa strains as well as MUCL 14186, the other two "alleles" being the historical groups identified by RAPD and *MAT* analysis.

Sequencing of the **calmoduline** (CL) gene is underway to see if this 4th marker equally distinguishes strains within the *G. xylarioides* pathogen population.

To date, the only marker available for characterization of the progeny in a fertile cross is *MAT*-PCR. If we are able to induce the sexual cycle between *G. xylarioides* strains originating from different hosts, then RAPD, *tef1*, and possibly CL sequences could enable us to observe how these different markers are inherited and distributed through meiotic recombination.

A first manuscript "*Gibberella xylarioides* from *Coffea canephora*, a new mating population within the *G. fujikuroi* species complex" has been submitted to AEM, and a second manuscript treating of the diversity within the *G. xylarioides* population is in preparation.

		A3			A14		
		Lane	Strains	Access° N°	Lane	Strains	Access° N°
F. proliferatum		2	PLP 11	MUCL 43482	11	PLP 11	MUCL 43482
RECENT	*canephora*	3	PLP 13	CAB 003	12	PLP 13	CAB 003
		4	PLP 14	OUG 008	13	PLP 14	OUG 008
	arabica	5	PLP 38	IMI 204746	14	PLP 38	IMI 204746
HISTORIC	*canephora*	6	PLP 15	MUCL 14186	15	PLP 15	MUCL 14186
	RCA & ?	7	PLP 48	Fus 001	16	PLP 48	Fus 001
		8	PLP 51	Fus 004	17	PLP 51	Fus 004
	IC & Guinea	9	PLP 49	Fus 002	18	PLP 49	Fus 002
		10	PLP 50	Fus 003	19	PLP 50	Fus 003

		A15			A17		
		Lane	Strains	Access° N°	Lane	Strains	Access° N°
F. proliferatum		2	PLP 11	MUCL 43482	11	PLP 11	MUCL 43482
RECENT	*canephora*	3	PLP 13	CAB 003	12	PLP 13	CAB 003
		4	PLP 14	OUG 008	13	PLP 14	OUG 008
	arabica	5	PLP 38	IMI 204746	14	PLP 38	IMI 204746
HISTORIC	*canephora*	6	PLP 15	MUCL 14186	15	PLP 15	MUCL 14186
	RCA & ?	7	PLP 48	Fus 001	16	PLP 48	Fus 001
		8	PLP 51	Fus 004	17	PLP 51	Fus 004
	IC & Guinea	9	PLP 49	Fus 002	18	PLP 49	Fus 002
		10	PLP 50	Fus 003	19	PLP 50	Fus 003

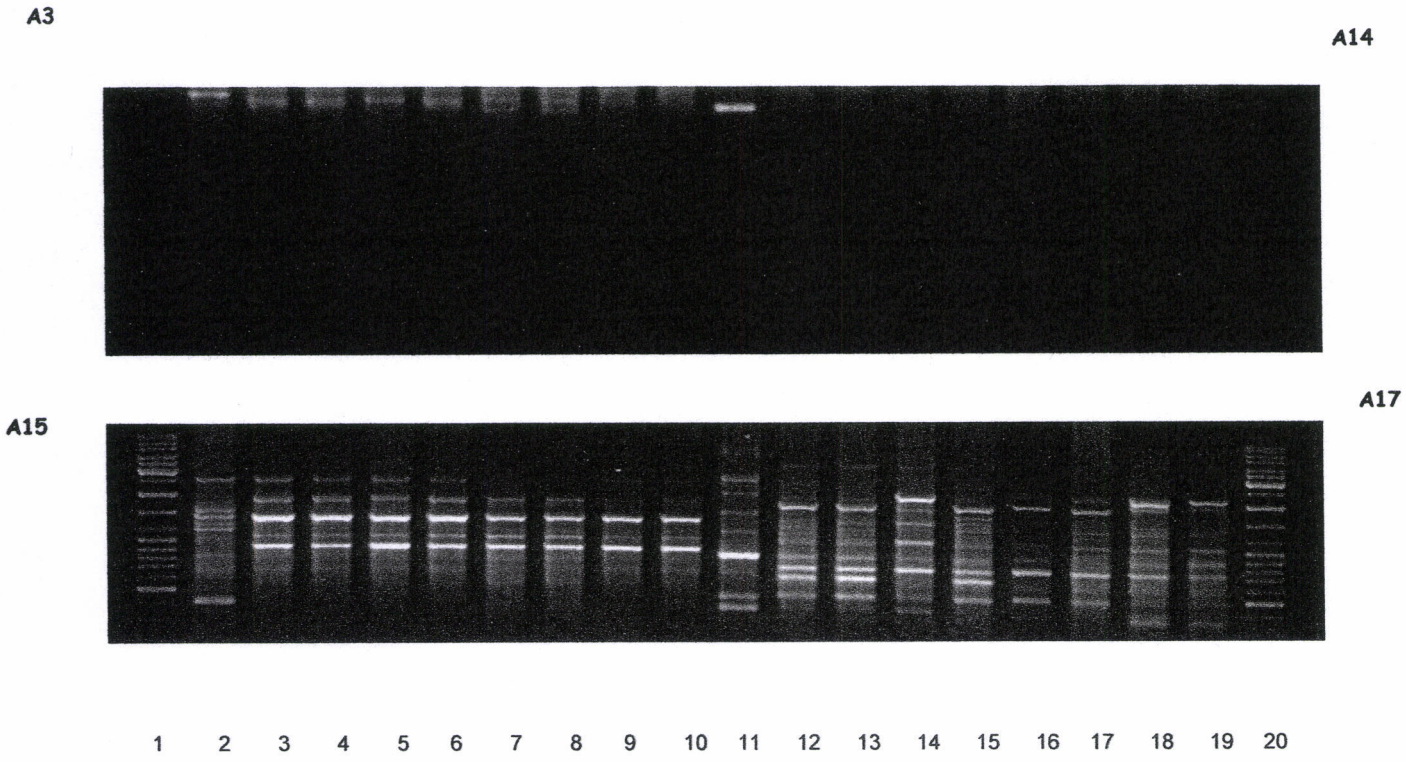


Figure 1: RAPD profiles obtained with primers A3, A14, A15 and A17 for *F. proliferatum* reference strain MUCL 43482 and 8 *G. xylarioides* strains of diverse origin. Lanes 1 and 20 represent gene Ruler DNA ladder Mix (Fermentas)

Table 1: Mating type (*MAT*) PCR identification of ascospore progeny in *G. xylarioides* "type C" crosses

PLP N°	Accession N°	Collector	Date Isol./Rec.	Laboratory cross	Isolate type	MAT PCR	Identification
PLP 55	SR XA/SS01	Lepoint P.	2004	*CAB003a x OUG008*	ascospore	1	*F. xylarioides*
PLP 56	SR XA/SS02	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 57	SR XA/SS03	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 58	SR XA/SS04'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 59	SR XA/SS05	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 60	SR XA/SS06	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 61	SR XA/SS07	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 62	SR XA/SS08	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 63	SR XA/SS09'	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 64	SR XB/SS02	Lepoint P.	2004	*CAB003a x 12B/SS02*	ascospore	1	*F. xylarioides*
PLP 65	SR XB/SS03'	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 66	SR XB/SS04'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 67	SR XB/SS05'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 68	SR XB/SS06	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 69	SR XB/SS07'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 70	SR XB/SS08'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 71	SR XB/SS09	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 72	SR XB/SS10	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 73	SR XC/SS01'	Lepoint P.	2004	*MUCL 14186 x 17B/04a*	ascospore	2	*F. xylarioides*
PLP 74	SR XC/SS02'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 75	SR XC/SS03	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 76	SR XC/SS04'	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 77	SR XC/SS05	Lepoint P.	2004		ascospore	2	*F. xylarioides*
PLP 78	SR XC/SS06	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 79	SR XC/SS07'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 80	SR XC/SS08'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 81	SR XC/SS09'	Lepoint P.	2004		ascospore	1	*F. xylarioides*
PLP 82	SR XC/SS10	Lepoint P.	2004		ascospore	2	*F. xylarioides*

NATIONAL AGRICULTURAL RESEARCH ORGANISATION (NARO)

INTERNATIONAL SCIENTIFIC COOPERATION PROJECT (INCO)

Development of a long-term strategy based on genetic resistance and agro-ecological approaches against Coffee Wilt Disease

Progress report (November 1, 2004 to April 30th 2005)

Coffee Research Institute (CORI), Kituza
P.O. BOX 185
Mukono

UGANDA

WORK PACKAGE (WP) 3: BREEDING FOR RESISTANCE AGAINST COFFEE WILT DISEASE

By

Pascal Musoli

I INTRODUCTION

Coffee wilt disease (CWD) continued to be a major threat to coffee production and productivity in Uganda during the reporting period and variety resistance is still considered the most appropriate option for a cost effective control of the disease. Therefore all breeding activities in the Work Package 3 of this project were given due consideration during the reporting period.

II OBJECTIVES

The objectives of WK3 remained unchanged i.e.

1) Identify sources of resistance against CWD through screen house tests on young seedlings and cuttings and field assessments
2) Assess inheritance of resistance to CWD among robusta coffee in Uganda
3) Evaluate genetic diversity among and between different sources of resistance to CWD in Uganda
4) Define a breeding strategy towards developing varieties with durable resistance to CWD.

III ACTIVITIES AND PROGRESS

The research work is structured along the work package objectives. Activities of the different objectives are however interlinked

A. IDENTIFIFYING SOURCES OF RESISTANCE TO CWD

The anticipated sources of resistance to CWD remained to be local germplasm available in Uganda and germplasm from exotic sources, mainly other African countries with history of having controlled CWD using variety resistance. The local germplasm include:

i) On-station robusta collections and their intraspecific hybrids
ii) Arabica collections and their intraspecific hybrids
iii) Arabusta (interspecific hybrids between robusta and arabica)
iv) On-farm robusta coffee trees surviving in wilt 'hot spots
v) Wild forest robusta coffee from its natural forest habitant.
vi) Exotic/imported germplasm
vii) Other coffee species available in the germplasm collections/fields at CORI and KARI

The search for resistance is based on results of:

a) Screen house tests under uncontrolled room conditions carried out on young rooted cuttings and seedlings
b) Controlled room tests carried out on young rooted cuttings and seedlings

c) Field assessments on young and mature coffee.

Owing to previous results, which proved arabica coffee to be resistant to CWD, screening work at CORI on arabica for resistance to CWD ceased. The most of the effort has concentrated on robusta and arabusta clones. Other coffee species were also not tested during the reporting period. Tests under controlled room conditions are normally carried out by in collaboration with CIRAD but during the reporting period work was only carried out at CORI based on screen house tests and field evaluations.

i) Screen house tests on young rooted cuttings and seedlings of robusta coffee germplasm at CORI/KARI.

During the reporting period

i) Survivor plants (rooted cuttings and seedlings) that have under gone two rounds of inoculation in the screen house were planted out in mother gardens for multiplication
ii) Survivor cuttings and seedlings that had under gone through only one round of inoculation were re-inoculated.
iii) Data collection continued on inoculated seedlings and cuttings

386 individuals out of 3095 (12.5%), belonging to 56 progenies remained healthy looking going through a second round of inoculation in the screen house at CORI. The survivors were planted in a mother garden for multiplications.

Table 1 shows recently re-inoculated rooted cuttings of 96 *C. canephora* clones from the germplasm collection at KARI. These plants are survivors of the first inoculation in the screen house and were re-inoculation under the same condition. After first inoculation, level of infection varied between the clones. 14 clones did not have any of their cuttings dying due to CWD. These clones are likely to be have high level of resistance to CWD, however this will be ascertained from results of the re-inoculation. The re-inoculation was carried out in late March 2005 and by the time of compiling this report it was still early for the inoculated plants to show wilt symptoms and therefore all of them were still looking healthy.

Table 1: Response of rooted cuttings of 96 C. canephora clones from the germplasm collection at KARI to screen house infection at CORI

		Plants inoculated	Plants re-inoculated	Status at 26/4/2005		
	Progeny	29/3/2004	30/3/2005	Sick	Dead	% Infected
1	203/32	9	5	0	0	0
2	1/7	7	7	0	0	0
3	227/59	26	23	0	0	0
4	1/70	18	14	0	0	0
5	254/62	14	12	0	0	0
6	3/20	15	12	0	0	0
7	J15109.4/5	23	22	0	0	0
8	228/57	25	23	0	0	0
9	NGREDOG	42	36	0	0	0
10	$1^{5}/6$	38	35	0	0	0

11	1/48	18	16	0	0	0
12	J74/2/13	9	8	0	0	0
13	13/71	91	82	0	0	0
14	J24/13/5	26	22	0	0	0
15	209/29	11	11	0	0	0
16	14/50	11	11	0	0	0
17	288	110	96	0	0	0
18	261^{s}/2	70	57	0	0	0
19	3/20	33	29	0	0	0
20	254/80	50	47	0	0	0
21	254/80	54	53	0	0	0
22	JB5109.4/3	9	8	0	0	0
23	228/13	11	10	0	0	0
24	258/58	62	58	0	0	0
25	238/29	79	72	0	0	0
26	202/63	47	20	0	0	0
27	1^{s}/2	103	95	0	0	0
28	202/30	181	147	0	0	0
29	266^{s}/11	65	55	0	0	0
30	254/28	63	55	0	0	0
31	1^{s}/3	136	124	0	0	0
32	1/15	76	67	0	0	0
33	223	31	28	0	0	0
34	245/25	89	85	0	0	0
35	J56/20/5	7	5	0	0	0
36	14/70	8	8	0	0	0
37	2/13	6	4	0	0	0
38	J124.9/1	16	13	0	0	0
39	2/57	2	2	0	0	0
40	1/3	3	3	0	0	0
41	JB5109.4/2	4	2	0	0	0
42	261^{s}/21	23	19	0	0	0
43	J56/20/57	8	6	0	0	0
44	22/2	10	9	0	0	0
45	J24/13/5	11	10	0	0	0
46	203/74	9	9	0	0	0
47	286	20	18	0	0	0
48	J56/20/50	3	3	0	0	0
49	234/37	19	13	0	0	0
50	258/28	70	61	0	0	0
51	238/29	28	28	0	0	0
52	NGREDOG/3	8	5	0	0	0
53	258^{s}/3	9	9	0	0	0
54	J24/13/52	7	7	0	0	0
55	J1/14/51	11	7	0	0	0
56	228/63	12	9	0	0	0
57	1/11	7	3	0	0	0
58	267^{s}/6	41	37	0	0	0

62	203/14	7	7	0	0	0
63	227/53	38	36	0	0	0
64	238/29	14	13	0	0	0
65	228/65	4	4	0	0	0
66	2/13	5	3	0	0	0
67	J105203/11	13	12	0	0	0
68	227/54	5	4	0	0	0
69	218/32	2	2	0	0	0
70	203/14	6	5	0	0	0
71	14/60	9	9	0	0	0
72	J24/13/5	15	11	0	0	0
73	2/13	7	4	0	0	0
74	227/56	12	11	0	0	0
75	2/86	31	21	0	0	0
76	JB5109.4/5	33	27	0	0	0
77	1/12	12	11	0	0	0
78	J105203/11	12	8	0	0	0
79	JB5109.4/1	8	7	0	0	0
80	222/65	35	27	0	0	0
81	209/29	9	7	0	0	0
82	227/58	4	3	0	0	0
83	258^{s}/58/3	5	4	0	0	0
84	267/5	3	2	0	0	0
85	207^{s}/15	4	4	0	0	0
86	256/20/6	2	2	0	0	0
87	13/15	2	0	0	0	0
88	J1/14/5	4	3	0	0	0
89	J94/2/13	3	1	0	0	0
90	3/59	2	1	0	0	0
91	J1/14/19	4	2	0	0	0
92	254/80	85	81	0	0	0
93	J24/13/12	1	0	0	0	0
94	Unlabeled 1	47	42	0	0	0
95	Unlabeled 2	19	16	0	0	0
96	Unlabeled 3	5	3	0	0	0

NB: Used inoculum concentration of 200 x $10^{4}ml^{-1}$

Table 2 shows response of 34 open pollinated seedling progenies of *C. canephora* inoculated with CWD in the screen house at CORI. Most of the progenies are either dead or sick with mean percentage infection of 88.4%. Only progeny A/4/13 had percentage infection of 12.5%

Table 3: Response of 34 seedling progenies of C. canephora from the germplasm collection at KARI to screen house infection at CORI

		Plants inoculated	*Status at 28/3/2005*			
	Progeny	*29/09/2004*	*Healthy*	*Sick*	*Dead*	*% Infected*
1	203/14	74	8	4	62	89.2
2	JB51094/3	53	1	2	50	98.1
3	228/57	9	0	0	9	100
4	A/4/13	16	14	2	0	12.5
5	J56/20/	37	0	3	34	100
6	3/54	125	5	6	114	96.0
7	258[S]/58	87	27	8	52	68.9
8	234/37	62	8	8	46	87
9	261[S]/21	250	13	10	227	94.8
10	13/2/1	170	36	13	121	72.9
11	J/24.9/1	94	31	4	59	67
12	3/14	22	1	3	18	95.5
13	228/18	1	0	0	1	100
14	14/70	1	0	0	1	100
15	3/59	2	0	0	2	100
16	J56/20/61	3	0	0	3	100
17	3/62	7	0	0	7	100
18	1/11	16	0	0	16	100
19	J105203/11	37	2	2	33	94.6
20	NGREDOG	27	1	0	26	96.3
21	228/15	27	2	2	23	96.3
22	238/29	93	5	6	82	94.6
22	1/13	141	3	1	137	97.9
23	288	125	4	2	119	96.8
24	R/1/4	66	6	6	54	90.9
25	B/6/2	60	4	4	52	93.3
26	J/1/1	324	80	33	211	75.3
27	C/6/1	197	32	15	150	76.1
28	Q/6/1	188	54	10	123	65.4
29	1[S]/2	100	35	1	64	65
30	B/2/1	145	42	5	98	71
31	B/1/1	136	40	15	81	58.7
32	P/3/6	68	4	2	62	94
33	H/4/1	179	14	10	155	92.2
34	Q/3/4	276	99	4	173	64.1
	Mean % infect.					88.4

NB: Spore concentration used: $225x10^4$ ml^{-1}

ii) **Screen house tests on intraspecific F1 hybrids of controlled crosses of resistant and susceptible robusta coffee clones.**

14 hybrid progenies of crosses between robusta coffee clones with different levels of resistance to CWD were inoculated and data collection initiated (Table 4). Although progenies were small (few individuals for each of the progenies) the preliminary results indicate that the resistance/susceptibility of a progeny is unpredictable irrespective of the resistance status of the parents.

Table 4. Response of 14 F1 progenies of controlled crosses of robusta coffee clones.

		25/10/2004	28/3/2005			
Peogeny	**Cross**	**Inoculated**	**Healthy**	**Sick**	**Dead**	**% infected**
254/80/2 x JB510.4/5/1	R x R	28	12	4	12	57.1
254/80/2 X 202/63/1	R x R	16	4	1	11	75.0
254/80/2 X F.Ruk/1	R x R	49	26	8	15	46.9
JB5109.4/5/1 X F. Ruk/1	R x R	16	6	3	7	62.5
254/80/2 X 258^{S}/24(0)	R x M/R	16	12	1	3	25.0
254/80/2 X 1^{S}/3	R x M/R	13	2	3	8	84.6
F.Ruk/1 x 254/80/2	R x R	12	10	1	1	16.7
1^{s}/2 x 223/32	S x S	26	6	1	19	95.2
254/80/2 x 257/53	R x S	21	18	1	2	14.3
254/80/2 x 1^{s}/2	R x S	9	1	1	7	88.9
254/80/2 x 1s/6	R x M/R	10	5	2	3	50.0
286/1 x 1s/2	R x S	7	2	2	3	71.4
F.Ruk/1 x 202/63/1	R x R	5	5	0	0	0.0
F.Ruk/1 X 1^{S}/3	R x M/R	4	3	1	0	25

iii) Screen house tests of open pollinated arabusta F2 seedlings available le at KARI/CORI

424 individuals out of 587 (72.2%), belonging to 30 F2 open pollinated progenies remained healthy looking after going through two rounds of screen house inoculation. The healthy l0oking plants were planted out in the field for field evaluation.

iv) Screen house tests on plants collected from wilt hotspots

During the reporting period plants collected during the previous reporting period from wilt hotspots in districts of Luwero, Kanungu, Wakiso, Kiboga, Rukungiri, Mubende, Bundibugyo, Mayuge, Bushenyi, Iganga, Mukono and Kyenjojo were raised in the nursery at CORI. Data collection continued on materials inoculated /re-inoculated in previous reporting periods.

Table 5 shows response of 45 open pollinated seedling progenies from *C. canephora* trees in CWD hotspots, that survived first round inoculation, to CWD re-inoculation under screen house conditions at CORI. The materials were collected from districts infected with CWD in different parts of the country. 64.3 mean percentage infection rate of the re-inoculated plants has so been observed. Data collection however is continuing.

Table 5: Response of 45 robusta coffee open pollinated seedling progenies to CWD re inoculation

		Plants inoculated	*Plants re-inoculated*	*Healthy status at 12/10/2004*			*% infected*
	Progeny	**28/02/2004**	*15/09/2004*	*Healthy*	*Sick*	*Dead*	
1	Kanu/kagumira/10	76	53	30	2	21	43.4
2	Kanu/kagumira/1	119	98	61	1	36	37.8
3	Kanu/kagumira/15	21	21	12	2	7	42.9
4	Kanu/kagumira/8	44	33	25	1	7	24.2
5	Kanu/pkabi/1	24	19	12	2	5	36.8

6	Kanu/mwebehire/2	2	2	0	1	1	100
7	Kanu/ndyabagira/2	24	23	17	2	4	26
8	Kanu/kagumira/13	31	21	13	0	8	38
9	Nansubuga M/2	88	79	46	1	32	41.7
10	Kanu/lubinga/3	111	85	62	1	22	27
11	Kanu/tindiwegiwilison/1	19	16	8	0	8	50
12	No label 1	58	52	45	1	6	13.5
13	Haji Kawoya/1	161	130	19	10	101	85.4
14	Haji Kawoya/5	105	97	32	13	52	67
15	Haji kawoya/3	115	75	24	5	46	68
16	Kanu/lubinga/2	113	91	16	12	63	82.4
17	Kanu/lubinga/1	174	149	72	8	69	51.7
18	Kanu/nkumbi w/5	16	8	8	0	0	0
19	Haji kawoya/4	30	23	6	1	16	73.9
20	Kanu/turinamasiko/1	7	5	0	0	5	100
21	Kanu/kagumira/5	60	58	14	7	37	75.9
22	Kanu/kagumira/4	42	37	9	4	24	75.7
23	Kanu/kagumira/9	42	33	13	2	18	60.1
24	Kanu/kagumira/11	138	127	44	20	63	65.4
25	Kanu/africabenard/2	34	28	7	2	19	75
26	No label 2	28	23	12	2	9	47.8
27	Kanu/kanahe/2	63	51	14	7	30	72.5
28	Kanu/mwebehire/1	160	135	50	22	63	62.9
29	Kanu/kahgye p	34	30	2	2	26	93.3
30	Kanu/kagumira/7	95	82	33	6	43	59.8
31	Kanu/kagumira/3	49	37	6	4	27	83.8
32	Kanu/bwengyebukye/3	104	68	5	2	61	92.6
33	Kanu/sausa a	37	26	4	2	20	84.6
34	Kanu/kagumira/12	142	120	30	14	76	75
35	Kanu/kagambagye j /1	68	58	17	9	32	70.7
36	Kanu/kagumira/6	48	39	8	4	27	79.5
37	Kanu/kanahe/5	89	76	9	11	56	88.2
38	Kanu/kagumira/2	83	70	21	11	38	70
39	Kanu/mwebehire/3	-	76	20	20	36	73.7
40	Hajikawoya/2	93	73	13	3	57	82.2
41	Kanu/nkumbi w/4	23	16	4	1	11	75
42	Kanu/tindiwegiwilison/4	22	13	4	1	8	69.2
43	Nansubuga M/3	8	5	0	0	5	100
44	Kanu/nkumbi w/7	4	3	0	1	2	100
45	Walakira md/tr2	5	4	2	1	1	50
	Mean %infection						64.3

Table 6 shows response of rooted cuttings of 112 genotypes collected from CWD hotspots to CWD re-inoculation under screen house conditions at CORI. The re-inoculated cuttings are the survivors of the first round of inoculation. The results reveal that some genotypes are resistant to CWD, particularly genotypes showing 0.0% infection by the time of this data collection. Data collection on the re-inoculated plants will however continue through June 2005.

Table 6: Response of rooted cuttings of robusta coffee from CWD infected farms to CWD infection under screen house conditions

		Plants inoculated	*Plants re-inoculated*	*Healthy status at 12/10/2004*			*% infected*
	Progeny	28/3/2004	15/09/2004	Healthy	Sick	Dead	
80	Buteraba/1	14	9	-	-	-	-
66	Buteraba/3	3	3	3	0	0	0
75	Buteraba/4	12	7	1	1	5	85.7
107	Buteraba/5	1	1	1	0	0	0
58	Buteraba/6	4	1	1	0	0	0
95	Buteraba/8	4	2	1	0	1	50
96	Butereba/9	5	4	2	0	2	50
10	Bwekwaso/6	5	4	3	0	1	25
56	Byekwaso/1	5	2	-	-	-	-
76	Byekwaso/1	17	2	0	0	1	50
88	Byekwaso/2	9	4	0	1	3	100
83	Byekwaso/3	8	5	3	1	1	40
89	Byekwaso/4	9	2	-	-	-	-
77	Byekwaso/8	14	7	1	0	6	85.7
72	Edith/1	23	9	6	0	3	33.3
102	Edith/3	4	1	1	0	0	0
53	Edith/4	10	1	0	1	0	0
84	Hajikawoya/1	7	4	3	1	0	25
67	Hajikawoya/2	23	18	14	3	1	22.2
79	Hajikawoya/4	9	8	6	1	1	25
17	Kanu/africabenard 1	7	7	7	0	0	0
38	Kanu/africabenard 3	3	3	3	0	0	0
41	Kanu/africabenard/2	9	1	0	0	1	100
42	Kanu/bwengyebukye/3	11	11	4	5	2	63.6
24	Kanu/kagambirwe 2	3	2	2	0	0	0
16	Kanu/kagambirwe 3	10	10	9	0	1	10
25	Kanu/kagambirwe/1	10	9	7	2	1	33
3	Kanu/kagumira/1	4	1	0	0	1	100
6	Kanu/kagumira/10	1	1	1	0	0	0
23	Kanu/kagumira/11	6	5	4	1	0	20
12	Kanu/kagumira/12	2	1	0	0	1	50
13	Kanu/kagumira/14	5	5	5	0	0	0
34	Kanu/kagumira/16	3	2	2	0	0	0
37	Kanu/kagumira/17	4	4	3	1	1	50
15	Kanu/kagumira/3	5	4	3	1	0	25
32	Kanu/kagumira/4	9	7	6	1	0	14.3
31	Kanu/kagumira/5	2	2	1	0	1	50
28	Kanu/kagumira/6	9	7	5	1	1	28.6
36	Kanu/kagumira/7	2	1	1	0	0	0
26	Kanu/kagumira/8	9	9	7	1	1	22
9	Kanu/kagumira/9	1	1	0	0	1	100
22	Kanu/kanahe D/2	4	1	1	0	0	0
35	Kanu/kanahe D/3	6	5	5	0	0	0
2	Kanu/kanahe D/4	17	17	15	2	0	11.8

14	Kanu/kanahe D/5	2	2	2	0	0	0
4	Kanu/kanahe D/6	3	3	2	1	0	33.3
39	Kanu/kanahe D/8	10	10	8	0	2	20
43	Kanu/kanahe/7	6	6	2	6	0	100
8	Kanu/mwebehire E/1	1	1	0	1	0	100
18	Kanu/mwebehire E/2	1	1	0	0	1	100
20	Kanu/mwebehire E/3	6	5	4	1	1	50
30	Kanu/ndabangira/5	6	4	3	1	0	25
19	Kanu/ndyabagira/1	3	3	3	0	0	0
29	Kanu/ndyabangira/1	19	7	0	1	6	0
7	Kanu/ndyabangira/2	1	1	1	0	0	0
11	Kanu/ndyabangira/3	4	3	0	0	3	100
33	Kanu/ndyagangira/4	5	4	3	1	0	25
5	Kanu/patricia/	1	1	1	0	0	0
21	Kanu/pkabi/2	5	3	0	0	3	100
27	Kanu/sausa E/	11	11	8	3	0	27.3
40	Kanu/turinamasiko/1	2	1	1	0	0	0
47	Kawoya/5	23	13	3	2	8	76.9
87	Kigoye/10	10	1	0	0	1	100
57	Kigoye/7	7	1	0	0	1	100
93	Kigoye/8	3	2	0	0	2	100
55	Kigoye/9	5	2	0	0	2	100
68	Kiryowa/4	15	14	12	1	1	14.3
98	Kiryowa/6	7	4	3	0	1	25
100	Lubinga A/1	4	1	0	0	1	100
90	Lubinga A/2	6	3	3	0	0	0
105	Lubinga A/3	3	2	1	0	1	50
104	Mayinja/1	4	3	0	1	2	100
52	Mayirane/2	8	6	6	0	0	0
101	Mayirane/3	5	1	-	-	-	-
85	Mayirane/4	6	2	1	0	1	50
59	Mayirane/6	9	8	7	1	0	12.5
71	Mrs walakira/1	22	2	0	0	2	100
94	Mukasa/10	5	5	0	0	5	100
110	Mukasa/19	1	1	1	0	0	0
109	Mukasa/3	-	3	1	0	2	66.7
91	Mukasa/8	4	3	2	0	1	33.3
97	Muza/1	5	4	4	0	0	0
106	Muza/2	4	2	2	0	0	0
49	Muza/3	9	6	2	1	3	66.6
73	Nakazibwe/2	2	2	2	0	0	0
108	Nakazibwe/3	1	1	1	0	0	0
74	Nakazibwe/4	2	1	1	0	0	0
50	Nakazibwe/5	7	7	1	0	2	28.6
51	Namutebi/2	8	4	2	0	2	50
46	Namutebi/3	6	1	0	0	1	100
99	Namutebi/4	4	2	0	0	2	100
103	Namutebi/5	4	3	2	1	0	33.3
45	Namutebi/6	13	7	7	0	0	0

81	Nansubuga M/2	11	10	8	2	0	20
62	Nansubuga M/3	18	16	15	1	0	6.3
44	Nkumbi wilson salongo/1	21	17	15	0	2	11.8
63	Nkumbi wilson salongo/2	16	8	5	2	1	37.5
69	Nkumbi wilson salongo/3	24	20	17	1	2	15
70	Nkumbi wilson salongo/4	15	4	2	0	2	50
60	Nkumbi wilson salongo/5	21	15	8	1	6	46.7
48	Nkumbi wilson salongo/7	24	23	0	0	23	100
111	Not labeled	1-	14	12	1	1	14.3
112	Not labelled	-	1	0	0	1	100
92	Semakula S/2	6	3	0	0	3	100
82	Sempa/1	6	6	5	0	1	16.7
64	Sempa/10	4	2	2	0	0	0
65	Sempa/4	3	1	1	0	0	0
78	Sempa/5	11	10	9	0	1	10
86	Tindiwegiwilison/4	5	3	2	0	1	33.3
54	Tindiwegiwilson/1	21	18	17	0	1	5
61	Tindiwegiwilson/3	9	8	8	0	0	0

Note

- results not available

v) Screen house tests of wild germplasm collected from forests

During the reporting period plant materials of wild robusta coffee were collected from Kibale National Park forest (table 7) and Itwara forest (table 8). Plants collected and planted in the nursery at CORI during the previous reporting period were maintained. The plants are likely to be inoculated during the next reporting period. Owing to poor establishment of plants currently in the nursery, another expedition was conducted to collect more materials from both Kibale and Itwara forests. Summary of newly collected materials is shown in table 9.

Table 7: **Collections of wild robusta coffee from Kibale National Park and planted at Kituza.**

	Site	TREE	Cuttings Planted.	Rooted cuttings	Seeds planted	Seedling available	Volunteer seedlings	Surviving V seedlings
1	Ngogo-Kibuguta A	1	19	4	0	0	62	54
2	Ngogo-Kibuguta A	2	10	0	0	0	30	25
3	Ngogo-Kibuguta A	3	10	4	0	0	6	3
4	Ngogo-Kibuguta B	1	30	5	13	4	13	10
5	Ngogo-Kibuguta B	2	28	9	0	0	8	7
6	Kabwegyemere	1	4	0	20	9	0	0
7	Kabwegyemere	2	13	1	70	20	5	3
8	Mbale-Rwabuhozire	1	10	3	20	13	10	10
9	Mbale-Rwabuhozire	2	20	4	0	0	9	9
10	Mbale-Mahungu	1	10	1	0	0	5	2
11	Mbale-Mahungu	2	5	2	10	1	0	0

12	Mbale-Mahungu	3	3	2	20	0	0	0
13	Mbale-Mahungu	4	30	11	0	0	12	6
14	Kabirizi-Kinyantare	1	18	5	0	0	5	4
15	Kabirizi-Kinyantare	2	12	9	0	0	0	0
	Total		**233**		**159**		**175**	

Table 8 **Collections of wild robusta coffee from Itwara forest and planted at Kituza.**

	Site	TREE	Cuttings Planted.	Rooted cuttings	Seeds planted	Seedling available	Volunteer seedlings	
1	Rutooma comp. 15	1	5	-			3	
2		2	2	-			2	
3		3	23	2			8	
4		4	0	0			5	
5		5	13	0			0	
6		6	5	1			0	
7		7	7	0			0	
8		8	6	0			0	
9	Kanaaba Comp. 14	1	1	0			8	
10		2	36	2			0	
11		3	13	1			0	
12		4	46	2			0	
13		5	13	1			0	
14		6	17	0			0	
15	Kyamuhooro Comp. 11	1	12	0			0	
16		2	15	0			0	
17		3	13	0			0	
18		4	11	0			0	
19		5	30	0			0	
20		6	31	1			21	

Table 9: Plant materials of wild robusta coffee collected from Kibale, Itwara and Kalangala forests in November 2004 and February 2005

Forest	Site	Genotypes with cuttings	Genotypes with berries
Kibale National Park Forest	Ngogo-Kibuguta A	11	5
	Ngogo-Kibuguta B	6	6
	Ngogo-Kissita	0	2
	Mbale Mahungu	18	27
	Kabirizi-Kinyantale	7	3
	Kabwegyemere	8	1
	Sub-total	**50**	**44**
Itwara Forest	Rutooma compartment 15	20	18
	Kanaaba compartment 14	17	7
	Kyamuhuro compartment 11	12	9
	Compartment 19	14	6
	Compartment 18	11	2
	Sub-total	**74**	**42**
Total		**124**	**86**

Vi) Importation and screening exotic germplasm for resistance against CWD

It is still difficult to import materials from other countries for this purpose. Only 69 seedlings of 14 clones received from Ivory Coast through CIRAD in the previous reporting period were planted out in an isolated field at CORI for evaluation.

2) Evaluation for CWD resistance in multi-clonal field trials

During the reporting period, this work centred on the robusta coffee clonal trial at CORI. The trial field was maintained following recommended procedures of maintaining robusta coffee fields. Data collection on yield and CWD incidence continued. Yield data is not presented in this report but results of CWD incidence since 2001 up to April 29, 2005 are shown in table 10. The results reveal a general increase of incidence of the diseases across varieties during the reporting period (April 2005) but varieties Q/3/4 and J/1/1 remained resistant.

Table 10: CWD incidence and severity on 20 clones of *C. canephora* in a field trial at Kituza

	April 2001	April 2002	April 2003	April 2004	April 2005
Clone	**% Incidence**	**% Incidence**	**% Incidence**	**% Incidence**	**% Incidence**
H/4/1	66.7	87.5	87.5	95.8	95.8
P/3/6	75.0	91.7	91.7	91.7	91.7
E/3/6	37.5	75.0	87.5	87.5	87.5
C/1/7	50.0	58.3	75.0	87.5	91.7
B/6/2	58.3	75.0	83.3	87.5	91.7
Q/1/1	70.8	75.0	79.2	79.2	83.4
P/5/1	62.5	66.7	70.8	79.2	87.5
257/53	25.0	45.8	70.8	75.0	83.3
Q/6/1	70.8	75.0	75.0	75.0	75.0
G/3/7	33.3	41.7	58.3	70.8	75.0
B/1/1	29.2	37.5	58.3	66.7	75.0
B/2/1	29.2	45.8	50.0	75.0	75.0
L/2/7	25.0	33.3	45.8	75.0	75.0
R/1/4	33.3	45.8	50.0	70.0	70.0
223/32	16.7	37.5	41.7	45.8	49.9
$1^{S}/2$	4.2	8.3	33.3	50.0	70.9
C/6/1	12.5	16.7	16.7	29.2	50.0
$1^{S}/3$	12.5	12.5	16.7	16.7	37.5
Q/3/4	0	4.2	4.2	4.2	4.2
J/1/1	0	0	0	0	0
	35.6	**46.7**	**54.8**	**63.1**	**68.5**

B: INHERITANCE OF THE CWD RESISTANCE

Activities carried out during the reporting period towards under standing transmission of CWD resistance from parents to progenies include carrying out artificial crosses of CWD resistant and susceptible parents, sowing of available hybrid seed of CWD resistant and susceptible parents and maintenance of available seedlings. Table 11 shows seedlings transplanted from nursery seedbed into polythene pots in the nursery at CORI up to May 2005. More crosses were made to generate more seedlings of each of the progenies for studies on inheritance of CWD resistance and the seeds are yet to be harvested.

Table 11: Number of plants available for each date of transplanting

	J1/1				Q3/4				E3/2				257s/53			
Transplanted	Mar-04	Oct-04	Mar-05	May-05	Mar-04	Oct-04	Mar-05	May-05	Mar-04	Oct-04	Mar-05	May-05	Mar-04	Oct-04	Mar-05	May-05
J1/1					56	101	27	95	23	121	33	98			5	150
Q/3/4	3	129		2			2		78			32	10		1	21
E3/2		22		194		45		83				43				25
257s/3			24			1	29	22				12				

C: EVALUATION OF GENETIC DIVERSITY OF ROBUSTA COFFEE IN UGANDA.

During the reporting period seeds, cuttings and leaves were collected from wild robusta coffee in Kibale and Itwara forests (Table 12). The leaves were submitted to CIRAD in France were their DNA will be extracted and analyzed for diversity using microsetallite markers. The cuttings and seeds were planted in the coffee nursery at CORI for raising to appropriate age before inoculating them with CWD pathogen. Seedlings and rooted cuttings of the previous collection were maintained in the nursery.

Table 12: Plant materials collected from wild coffee in Kibale and Itwara forests

Forest	Collection site	Individual	Leaves	Semi-hard cuttings	Hard cuttings	Seeds
Kibale Forest	Ngogo-Kibuguta A	Tree 1	-	+	-	+
		Tree 2	+	-	-	-
		Tree 3	+	+	-	-
		Tree 4	-	+	-	+
		Tree 5	+	+	-	+
		Tree 6	+	-	-	+
		Tree 7	+	+	-	-
		Tree 8	+	+	-	+
		Tree 9	+	+	-	-
		Tree 10	+	+	-	+
		Tree 11	+	+	-	-
		Tree 12	+	+	-	
		Tree 13	+	+	-	+
		Tree 14	+	+	-	-
		Tree 15	+	+	-	+
	Ngogo-Kibuguta B	Tree 1	-	+	-	+
		Tree 2	-	+	-	-
		Tree 3	+	+	-	+
		Tree 4	+	+	-	+
		Tree 5	+	+	-	-
		Tree 6	+	+	-	+
		Tree 7	+	+	-	+
		Tree 8	+	-	-	+
		Tree 9	+	+	-	-
		Tree 10	+	+	-	+
		Tree 11	+	+	-	+
		Tree 13	+	+	-	-
		Tree 14	+	+	-	-
		Tree 15	+	-	-	+
		Tree 16	+	-	-	+
	Ngogo-Kisita	Tree 1	+	-	-	+
		Tree 2	+	-	-	+
	Mbale Mahungu	Tree 1	-	-	-	+
		Tree 2	-	-	-	+
		Tree 3	-	+	-	+
		Tree 4	-	+	-	+
		Tree 5	+	+	-	+
		Tree 6	+	+	-	+

		Tree 7	+	+	-	+
		Tree 8	+	-	-	+
		Tree 9	+	+	-	+
		Tree 10	+	+	-	+
		Tree 11	+	+	-	+
		Tree 12	+	+	-	+
		Tree 13	+	+	-	+
		Tree 14	+	+	-	+
		Tree 15	+	+	-	+
		Tree 16	+	+	-	+
		Tree 17	+	+	-	+
		Tree 18	+	+	-	+
		Tree 19	+	+	-	+
		Tree 20	+	+	-	+
		Tree 21	+	+	-	+
		Tree 22	+	+	-	+
		Tree 23	+	+	-	+
		Tree 24	+	+	-	-
		Tree 25	+	+	-	+
		Tree 26	+	+	-	+
		Tree 27	+	+	-	+
		Tree 28	+	+	-	+
		Tree 29	+	+	-	+
		Tree 30	+	+	-	+
		Tree 31	+	+	-	+
	Kibirizi-Kinyantale	Tree 1	-	-	-	-
		Tree 2	+	+	-	-
		Tree 3	-	+	-	-
		Tree 4	+	+	-	-
		Tree 5	+	+	-	+
		Tree 6	+	+	-	+
		Tree 7	+	+	-	+
		Tree 8	+	+	-	+
		Tree 9	+	+	-	+
	Kabwegyemere	Tree 1	-	-	-	-
		Tree 2	-	-	-	-
		Tree 3	+	+	-	-
		Tree 4	-	+	-	-
		Tree 5	+	+	-	-
		Tree 6	-	+	-	-
		Tree 7	+	+	-	+
		Tree 8	+	+	-	+
		Tree 9	+	+	-	-
		Tree 10	-	+	-	-
Itwara Forest	Rutooma Compartment 15	Tree 1	+	-	+	+
		Tree 2	+	-	+	+
		Tree 3	+	-	+	+
		Tree 4	+	+	+	+
		Tree 5	+	-	+	+

		Tree 6	+	-	+	+
		Tree 7	-	-	+	-
		Tree 8	+	+	+	+
		Tree 9	+	+	-	-
		Tree 10	+	+	-	+
		Tree 11	+	+	-	+
		Tree 12	+	+	-	+
		Tree 13	+	+	-	+
		Tree 14	+	+	-	-
		Tree 15	+	+	-	+
		Tree 16	+	-	-	+
		Tree 17	+	+	-	+
		Tree 18	+	+	-	+
		Tree 19	+	+	-	+
		Tree 20	+	+	-	+
		Tree 21	+	+	-	+
		Tree 22	+	-	-	-
		Tree 23	+	+	+	-
		Tree 24	+	+	+	+
		Tree 25	+	-	+	-
		Tree 26	+	+	+	-
		Tree 27	+	+	+	-
		Tree 28	+	+	+	-
		Tree 29	+	+	+	+
	Kyamuhoro Compartment 11	Tree 1	-	-	-	-
		Tree 2	-	-	-	-
		Tree 3	+	-	+	-
		Tree 4	+	+	+	-
		Tree 5	+	+	+	+
		Tree 6	-	-	-	-
		Tree 7	+	+	+	+
		Tree 8	+	-	+	+
		Tree 9	+	+	+	+
		Tree 10	+	+	+	+
		Tree 11	+	+	+	+
		Tree 12	+	+	+	+
		Tree 13	+	-	+	+
		Tree 14	+	+	+	-
		Tree 15	+	+	+	+
	Itwara compartment 27	Tree 1	+	-	-	-
	Kanaaba compartment 14	Tree 1	+	+	+	+
		Tree 2	-	-	-	-
		Tree 3	+	-	+	-
		Tree 4	+	+	+	+
		Tree 5	+	+	+	+
		Tree 6	-	+	+	-
		Tree 7	+	-	+	-

		Tree 8	+	-	+	+
		Tree 9	+	+	+	+
		Tree 10	+	+	+	+
		Tree 11	+	+	-	+
		Tree 12	+	+	+	-
		Tree 13	+	+	+	-
		Tree 14	+	+	+	-
		Tree 15	+	+	+	-
		Tree 16	+	+	+	+
		Tree 17	+	+	+	-
		Tree 18	+	-	-	+
		Tree 19	+	+	+	-
		Tree 20	+	+	+	-
		Tree 21	+	+	+	+
	Compartment 19	Tree 1	+	+	+	+
		Tree 2	+	+	+	+
		Tree 3	+	+	+	+
		Tree 4	+	+	+	+
		Tree 5	+	+	+	-
		Tree 6	+	+	+	+
		Tree 7	+	+	+	+
		Tree 8	+	+	+	-
		Tree 9	+	+	+	+
		Tree 10	+	+	+	+
		Tree 11	+	+	+	-
		Tree 12	+	+	+	-
		Tree 13	+	+	+	-
		Tree 14	+	+	+	-
	Compartment 18	Tree 1	+	+	+	-
		Tree 2	+	+	+	+
		Tree 3	+	-	+	-
		Tree 4	+	+	+	+
		Tree 5	+	+	+	+
		Tree 6	+	-	+	-
		Tree 7	+	+	+	-
		Tree 8	+	-	+	-
		Tree 9	+	+	+	-
		Tree 10	+	+	+	-
		Tree 11	+	-	+	-

+ Samples was collected
- No samples

CONCLUSION

Screen house tests CORI has continued to reveal genotypes resistant to CWD among robusta coffee progenies. The field evaluation has shown that resistance level varies according to the genotypes. Preliminary work carried out in the previous period on genetic diversity has so far revealed some level of genetic distinction between the nganda and erecta types of C. canephora in Uganda. The studies also indicated the wild materials to be genetically different from either nganda of erecta types. There is however need to continue and accomplish work on resistance among wild materials. Preliminary work on hybrids of resistant and susceptible parents shows that the inheritance of the resistance may not follow the normal mendelian law. More of this work should be accomplished so as to understand the inheritance of the resistance as a basis for devising an efficient breeding programm.

INCO-DEV WILT PROJECT

Fourth scientific intermediate report: 01/11/2004 to 31/04/2005

WP 1: PATHOGEN DIVERSITY

Task 3: Evaluation of the variability in isolates aggressiveness from Equateur

L'essai d'évaluation d'agressivité des isolats en provenance de la Province de l'Equateur a été installé et les observations sont en cours.

Souches étudiées

A côté de 10 souches en provenance de la province de l'Equateur, les souches historiques de référence de la MUCL et de l'UNIKIN (tableau 1) sont également utilisée dans cet essai.

Tableau 1 : Liste de souches en provenance de la province de l'Equateur en compagnie des souches historiques de référence MUCL et UNIKIN.

N°	Souche	Province d'origine en RDC	Localité d'origine en RDC	Date de récolte par [1]	Date d'isolement par [2]	Identifié par [3]
1	Bandazwa	Equateur	Itimbiri	09/09/2004 K.D.	20/11/2004 T.	T.
2	Bunduki	Equateur	Voisine Oriental	09/09/2004 K.D.	20/11/2004 T.	T.
3	Mangbakapale	Equateur	-	15/09/2004 K.D.	20/11/2004 T.	T.
4	Mindembo Bloc 5	Equateur	Lisala	17/09/2004 K.D.	20/11/2004 T.	T.
5	Moboko	Equateur	Axe Itimbiri	09/09/2004 K.D.	22/11/2004 T.	T.
6	Mongene	Equateur	Itimbiri	10/10/2004 K.D.	22/11/2004 T.	T.
7	Ngwa	Equateur	Itimbiri	10/09/2004 K.D.	22/11/2004 T.	T.
8	Notre-Dame des pauvres	Equateur	Loeka	15/09/2004 K.D.	22/11/2004 T.	T.
9	Yeboka	Equateur	Itimbiri	09/09/2004 K.D.	20/11/2004 T.	T.
10	Zobolia	Equateur	Loeka	15/09/2004 K.D.	20/11/2004 T.	T.
11	MUCL 14186	Orientale	Yangambi	1960 Meyer	Meyer	Meyer
12	MUCL35223	Orientale	Isiro	Nov. 1992 Pochet.	1992 Decock	Decock
13	B10101(2)J ou MUCL 46044	Nord-Kivu	Mutwanga	02/12/02 K.T. D.	02/01/03 T.	T.
14	JE MAMBA	Kinshasa	UNIKIN	13/04/2005 T.	13/04/2005 T.	T.

K.D. : Kalonji et Dibue
K.D.T. : Kalonji, Dibue et Tshilenge

Matériel végétal :

Les plantules issues de la germination des graines récoltées sur le clone L251 à la Station de l'INERA Bongabo (Province de Equateur) ont constitué le matériel végétal pour cet essai. Le clone L251 est choisi pour sa susceptibilité déjà observée dans un essai antérieur (Kalonji, 1975). A

l'inoculation les plantules ont l'âge de 3 mois avec 3 à 4 paires des feuilles. La technique d'inoculation par injection à la seringue (10^6 conidies/ml) est utilisée.

Observés effectuées

Toutes les plantules de chaque traitement, soit 3 x 12 plantules, font l'objet d'observation pour les paramètres végétatifs, les manifestations pathologiques, la production des périthèces ainsi que le réisolement (Postulats de Koch) des pathogènes à la fin de l'essai.

Paramètres végétatifs

- Vigueur (diamètre au collet)
- Hauteur des plants
- Indice de croissance
- Nombre de feuilles formées dans le temps et le rythme de formation

Manifestations pathologiques (symptômes)

Les observations sont enregistrées portent sur l'incidence et sur l'intervalle de temps entre le moment de l'inoculation et l'apparition de différents symptômes. Les symptômes observés sont le flétrissement, le brunissement et le dessèchement des feuilles. La défoliation et le dessèchement de la tige et la mortalité qui en découlent constituent les signes exprimant la sévérité de la maladie.

Task 4: Description of the fungal life cycle, asexual

Essai 1 : Monitoring of périthecia production of *Gibberella xylarioides*

Matériel et méthode

La production des périthèces a été suivie sur différents caféiers ayant fait l'objet des inoculations artificielles au cours de différents essais antérieurs, réalisés au Jardin Expérimental de l'UNIKIN. Le matériel végétal sous observation est constitué d'une part des caféiers adultes âgés de 6 ans et d'autre part de jeunes plantules âgées de moins de 15 moins.

Parmi les premiers un sujet, déjà en production, présente des symptômes de la trachéomycose au stade avancé de la maladie avec une cote 5, équivalant au stade de dessèchement total de la plante (photos 1).

La méthode proposée par Maraite (First intermediate report, pp. 42-45 Report n° 28/2002 CIRAD/AMIS) a été appliquée avec légères modifications portant sur les dimensions du gabarit en fonction des grosseurs des tiges.

Quant aux plantules inoculées à bas âge en pépinière les observations sont faites sur des pieds complètement desséchés. Des sections transversales sont marquées à l'aide d'un stylo à encre indélébile sur la tige pour faire l'objet d'examens sous loupe binoculaire de marque Wild Heerbrugg.

Les données météorologiques de la période concernées sont fournies par la Station climatologique du CREN-K. Elles sont à corréler avec la variation, dans le temps, de la production des périthèces et de l'état de ceux-ci.

Premiers résultats

Les premières observations enregistrées sont présentées dans le tableau 2.

Tableau 2 : Présence et état de maturité des périthèces observés en date du 03 mai 2005 sur des caféiers inoculés artificiellement dans différents essais réalisés au Jardin Expérimental de l'UNIKIN.

Individu observé	Age	Traitement subi	Niveau observé	Abon-dance (Cote)	Etat (Cote)	Remarques
Pied n°39	Sujet adulte (6 ans) en production et portant des fruits	Inoculé en 1999 avec un isolat du *F. xylarioides* conservé dans le bois parasité.	10 cm du collet	0	0	
			20 cm du collet	3	G	
			30 cm du collet	1	G	
			40 cm du collet	0	0	
			50 cm du collet	2	S	
			60 cm du collet	3	G	Présence des macroconidies au microscope
Blessure	Sujet âgé de 18 mois.	Essai méthodes d'inoculation (blessure + fragment de culture)	1 cm sous blessure	2	E	
			1 cm sur blessure	1	G	
			2 cm sur blessure	3	G	
			3 cm sur blessure	0	0	
			4 cm sur blessure	2	S	
			5 cm sur blessure	3	G	
			6 cm sur blessure	3	G	
B10101(2)	Sujet âgé de 18 mois	Essai test d'agressivité	1 cm sur blessure	1	S	

		des isolats de Beni	2 cm sur blessure	1	G	
			3 cm sur blessure	2	G	
			4 cm sur blessure	3	G	
			5 cm sur blessure	3	G	
			6 cm sur blessure	3	G	Périthèces murs éjectant 8 ascospores en chaîne
B20301(3)	Sujet âgé de 18 mois	Essai test d'agressivité des isolats de Beni	1 cm sur blessure	1	S	
			2 cm sur blessure	2	E	
			3 cm sur blessure	3	E	
			4 cm sur blessure	3	S	
			5 cm sur blessure	1	G	
			6 cm sur blessure	0	0	
B30302(2)	Sujet âgé de 18 mois	Essai test d'agressivité des isolats de Beni	1 cm sur blessure	1	E	
			2 cm sur blessure	1	E	
			3 cm sur blessure	0	0	
			4 cm sur blessure	2	S	
			5 cm sur blessure	2	S	
			6 cm sur blessure	3	E	
Témoin	Sujet âgé de 18 mois	Essai test d'agressivité des isolats de Beni	1 cm sur blessure	1	G	Présence des asques non murs
			2 cm sur blessure	0	0	
			3 cm sur blessure	3	S	
			4 cm sur blessure	1	S	
			5 cm sur blessure	0	0	
			6 cm sur blessure	0	0	
Témoin blanco	Sujet âgé de 18 mois	Essai test d'agressivité des isolats de Beni	1 cm sur blessure	1	S	
			2 cm sur blessure	0	0	
			3 cm sur blessure	2	S	
			4 cm sur blessure	3	S	
			5 cm sur blessure	3	S	
			6 cm sur blessure	3	S	

Essai 2 : Etude de fertilité de Mating types *in vivo* en pépinière (UNIKIN) et en conditions naturelles (Beni)

Dans le cadre de l'échange d'isolats entre partenaires Cowidi, les souches MUCL 44532 (Mating type 1) et MUCL 44536 (Mating type 2) ont été fournies à l'UNIKIN par l'UCL (Pascale Lepoint) en vue de confirmer leur fertilité en conditions naturelles de la trachéomycose à Kinshasa et à Beni. Il est prévu que l'analyse de la fertilité des périthèces sera étudiée à l'UCL où les échantillons seront envoyés à la fin de l'essai.

Conduite de l'essai

Les 2 souches sont inoculées au même moment, mais en des endroits différents sur la tige de jeunes caféiers. Le matériel végétal utilisé est constitué de jeunes plantules caféiers issues de la germination des graines récoltées sur un sujet sensible attaqué par la trachéomycose et bien identifié (un caféier Robusta : I1010203/OG) récolté à Beni. La technique d'inoculation a été celle de blessure pratiquée par entaille dans la tige et l'insertion d'un fragment de culture du pathogène sur milieu SNA.

Les objets ont été répartis de la manière suivante :

Objet 1 : Mating type 1 ou 2 inoculé seul à 1 cm en dessous des feuilles cotylédonaires sur le plan de la première paire de vraies feuilles;
Objet 2 : Les souches inoculées à 1 cm en dessous des feuilles cotylédonaires sur le plan de la première paire de vraies feuilles en position face à face.
Objet 3 : les deux souches inoculées en position décalées Mat1/Mat2 à partir du niveau de 1 cm en dessous des feuilles cotylédonaires sur le plan de la première paire de vraies feuilles.
Objet 4 : l'inverse de l'objet 3, c'est-à-dire Mat2/Mat1.

Résultats

Les observations sont en cours sur les mêmes paramètres.

INCO-DEV COFFEE WILT PROJECT
(Contract no. ICA4-CT-2001-10006)

Work Package 1

Progress Report, June 2005

Dr Mike Rutherford
CABI Bioscience (UK)

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Important Note: *It was expected that, under the original contractual arrangements, CABI's inputs to the project would cease at the end of October 2004. However, as a result of a successful application to the EU for an extension to the project CABI has been able to continue its research since that time. However, given that the extension was not funded this research has relied on funds remaining from the original phase of the project and, as such, has been somewhat limited (as reflected in this report). Nevertheless, CABI is pleased to be able to continue to support it's partners in pursuing activities, including secure storage of fusaria and database maintenance, that are fundamental to the ongoing success of the project as a whole.*

1. Project partners

CABI Bioscience (CABI), United Kingdom (leading Work Package 1)
Centre de Cooperation Internationale en Recherche Agronomique pour Le Developpement (CIRAD), France
Coffee Research Institute (CORI), Uganda
Université Catholique de Louvain (UCL), Belgium
University of Kinshasa (UoK), Democratic Republic of Congo

2. Overall objective

To improve knowledge of the coffee wilt pathogen, *Fusarium xylarioides*, with respect to genetic diversity and variation in aggressiveness.

3. Specific activities

Specific activities for which CABI UK Centre (CABI UKC) is responsible for, or involved in, are:

- Establishment of a collection of the anamorph (*F. xylarioides)* and teleomorph (*Gibberella xylarioides)* forms of the CWD pathogen obtained from wilt-affected coffee trees in Uganda, DRC and other regions of Africa as appropriate.
- Confirmation of the identity of isolates to species level based on conventional morphological fungal characters.
- Use of a designated facility to ensure secure, long-term maintenance of representative isolates.
- Investigation of the extent of molecular variability diversity among fusarium isolates
- Establishment and maintenance of baseline data relating to those isolates acquired and held by CABI and its partners on an electronic database.
- Synthesis of results of research activities undertaken by CABI UKC and its partners under WP1.

4. Research progress

4.1. Establishment of a collection of anamorph and teleomorph forms of the CWD pathogen

As reported in January 2005, a comprehensive collection of fusarium isolates has been obtained for use by CABI and it's project partners. More than 250 of these are being held at CABI UKC. Since January only a small number of historial isolates, donated by CIRAD, and several isolates obtained from farm sites in Uganda and provided by CORI, Uganda, were deposited in the collection. All isolates held at CABI UKC are being maintained over the short to medium term on Synthetic Nutrient Agar (SNA) slopes at 5°C. Subcultures continue to be made freely available to project partners with a number of *F. xylarioides* and *F. udum* isolates (the latter from the established GRC) being provided to UCL in May to support ongoing mating studies.

4.2 Confirmation of the identity of isolates to species level based on conventional morphological fungal characters

Drs Mike Rutherford and Paul Cannon have continued to determine or confirm the identity1 of isolates held at CABI based on morphological characters. In the majority of cases it has been possible to identify an isolate to species level although, as found previously, a number of isolates were again found to differ from their original species designations. Where possible, documentation available from the CABI herbarium, the CABI GRC, alternative international collections including ATCC, DSMZ and CBS and other sources has been obtained and closely examined to help clarify discrepancies between this information and the results of morphological and indeed molecular genetic assessments (see Section 4.4 below). Notes to this effect have been included in the database alongside the isolates concerned (see isolates IMI 128389, IMI 127629 and ATCC 15664 for example).

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1 Identifications based on morphological characteristics as described by Booth, C. (1971) *The Genus Fusaium.* Kew, Commonwealth Agricultural Bureaux

4.3 Use of a designated facility to ensure the secure, long-term maintenance of representative isolates.

All of the isolates held at CABI UKC and selected as members of a representative set have now been deposited for long term storage in the GRC at Egham under liquid nitrogen and also in a freeze-dried state. These isolates are considered represenative of the range of *Fusarium* species, geographic origins, host plant species/clones and time of recovery from CWD affected trees in relation to the larger collection acquired (see Table 1). These isolates have now been investigated in depth and information relating to their genetic make-up, pathogenicity, mating type and other traits has and continues to be obtained (see Section 4.5). They now therefore represent an additional and valuable genetic resource that may be accessed by research groups and on which future studies of CWD may be based. In particular, the resource will be of major benefit in the search for resistance to CWD ongoing in Africa and Europe as part of this project and the broader coffee wilt programme.

4.4 Investigation of the extent of molecular variability diversity among fusarium isolates

Analysis of genetic variability within the set of 59 representative fusarium isolates (*F. xylarioides*, *F. solani*, *F. stilboides*, *F. lateritium* and *F. oxysporum*) has continued, with assessment based on the use of ISSR, presumptive mtDNA RFLPs (*Hae*III) and IGS approaches now completed for the entire set. Newly generated data is being analysed using GelCompar. A comprehensive review of the findings of this work and their implications as at the end of October 2004 was provided in the previous report. Findings from the most recent studies continue to strongly support the level of variability and population structure highlighted at that time. A set of fusarium isolates obtained from coffee trees affected by CWD at an on-farm site in Uganda curently being mapped (as part of this project) for the spatial and temporal spread of the disease have also been analysed (as part of a DFID funded project) using a number of the same molecular approaches. No genetic variability was observed within these isolates, again reflecting the lack of variability generally found within isolates obtained from *C. canephora* trees affected by CWD during the recent re-emergence of the disease.

4.5 Establishment and maintenance of baseline data relating to those isolates acquired and held by CABI and its partners on an electronic database.

Input of data to the electronic database has continued during the reporting period, including for newly acquired isolates. The database now holds comprehensive information relating to 284 isolates held at CABI, UCL, CIRAD and their partners in Africa. However, information reflecting the findings of the investigations of genetic variability, pathogenic variability, mating type and other studies remains limited. This information should be provided and input by CABI and it's partners as a priority during the remainder of the project.

4.6 Synthesis of results of research activities undertaken by CABI UKC and its partners under WP1.

An in-depth summary of the findings and implications of the project research was provided in the previous report. New information generated during 2005 generally reinforces the conclusions formed at that time and, as such, no attempt is made here to revise those conclusions. However, the overall situation will be reviewed more fully during a meeting to be held at UCL on 29th June 2005 to allow partners from CIRAD, CABI, UCL and CORI to discuss research progress and to plan future research activities.

Table 1. *Fusarium* isolates obtained from coffee and selected for morphological and genetic characterisation at CABI UKC (i'e' 'representative isolates)

Species[1]	Known isolate accession no.s						Host species	Country of origin	Date isolated or received
F. xylarioides	IMI204746			CAB009			*Coffea sp.*	Ethiopia	1976
F. xylarioides	IMI375907		Gx26				*Coffea arabica*	Ethiopia	1997
F. xylarioides	IMI375908		Gx31	CAB007			*Coffea arabica*	Ethiopia	1997
F. xylarioides	IMI375909		Gx43				*Coffea arabica*	Ethiopia	1997
F. xylarioides	IMI375916		RIGx43	CAB008			*Coffea arabica*	Ethiopia	1997
F. xylarioides	IMI392245	W5106a	IMI369711	CAB013			*Coffea canephora*	DRC	1995
F. xylarioides	IMI392246	W5263c	B	CAB014			*Coffea canephora*	DRC	1996
F. xylarioides	IMI392247	W5267a	C	CAB015			*Coffea canephora*	DRC	1996
F. xylarioides	IMI392248	W5272b	E	CAB016			*Coffea canephora*	DRC	1996
F. xylarioides	IMI392249	W5280b	A	CAB017			*Coffea canephora*	DRC	1996
F. xylarioides	IMI392250	W5432a					*Coffea canephora*	Uganda	1997
F. xylarioides	IMI392251	W5433a		CAB001			*Coffea canephora*	Uganda	1997
F. xylarioides	IMI392252	W5440a					*Coffea canephora*	Uganda	1997
F. xylarioides	IMI392253	W5448a					*Coffea canephora*	Uganda	1997
F. xylarioides	IMI392254	W5543a					*Coffea canephora*	Uganda	1997

F. xylarioides	IMI392255	W5543b					*Coffea canephora*	Uganda	1997
F. xylarioides	IMI392256	W5543c					*Coffea canephora*	Uganda	1997
F. xylarioides	IMI392257	W5543e					*Coffea canephora*	Uganda	1997
F. xylarioides	IMI392258	W5554a					*Coffea canephora*	Uganda	1997
Suspect *F. xylarioides*	IMI392259	H1	S11				*Coffea canephora*	Uganda	
F. xylarioides	IMI392260	R	47				*Coffea canephora*	Uganda	
F. xylarioides	IMI392261	W	24				*Coffea canephora*	Uganda	
F. xylarioides	IMI389563	W7276c		CAB029			*Coffea arabica*	Ethiopia	2002
F. xylarioides	IMI389567	W7279a		CAB033			*Coffea arabica*	Ethiopia	2002
F. xylarioides	IMI389571	W7283a		CAB037			*Coffea arabica*	Ethiopia	2002
F. stilboides	IMI389578	W7277a		CAB011			*Coffea arabica*	Ethiopia	2002
F. lateritium	IMI389579	W7284a		CAB012			*Coffea arabica*	Ethiopia	2002
F. stilboides	IMI389580	W7291a					*Coffea arabica*	Ethiopia	2002
F. oxysporum	IMI389581	W7291b					*Coffea arabica*	Ethiopia	2002
Suspect *F. stilboides*	IMI389582	W7292a					*Coffea arabica*	Ethiopia	2002
Suspect *F. xylarioides*	IMI392263	W 5440a - (16)		CAB 003			*Coffea canephora*	Uganda	11 Oct 2000
F. xylarioides	IMI392264			OUG 008			*Coffea canephora*	Uganda	Aug 1997

Suspect *F. xylarioides*	IMI392265		C12	OUG 151			*Coffea excelsa*	Uganda	Oct-02
F. xylarioides	IMI392681		C12	OUG 152			*Coffea excelsa*	Uganda	Oct-02
F. xylarioides	IMI392682		C13	OUG 154			*Coffea excelsa*	Uganda	Oct-02
Suspect *F. xylarioides*	IMI392266		C13	OUG 155			*Coffea excelsa*	Uganda	Oct-02
Suspect *F. xylarioides*	IMI392267		C14	OUG 157			*Coffea excelsa*	Uganda	Oct-02
F. xylarioides	IMI392268	MUCL 14186		RDC 002			*Coffea canephora*	DRC	1968
Suspect *F. xylarioides*	IMI392269			RDC 004			*Coffea canephora*	DRC	Sep-02
Suspect *F. xylarioides*	IMI392270			RDC 051			*Coffea canephora*	DRC	Sep-02
Suspect *F. xylarioides*	IMI392271		IMU 0101	RDC 068			*Coffea canephora*	DRC	Dec-02
F. xylarioides	IMI392272	MUCL 44508	FYMY access no. = SR 20/10 SCO1						
Suspect *F. xylarioides*	IMI392273	MUCL 44543	FYMY access no. = SR 20/(SS)19				*Coffea canephora*	DRC	12/09/02
F. xylarioides	IMI392274	W7477b					*Coffea canephora*	Tanzania	Feb 2003
F. xylarioides	IMI392275	W7489a					*Coffea canephora*	Tanzania	Feb 2003

Suspect *F. xylarioides*	IMI392276	W7494a					*Coffea canephora*	Tanzania	Feb 2003
F. xylarioides	IMI392277	W7498a					*Coffea canephora*	Tanzania	Feb 2003
F. xylarioides	IMI392278	W7500b					*Coffea canephora*	Tanzania	Feb 2003
Suspect *F. xylarioides*	IMI392279	TZ002						Tanzania	
F. solani	IMI392280	NRLL 25804	CBS 749.79				*Coffea canephora*	Guinea	Mar-03
F. stilboides	IMI392281	NRLL 13277	FRC L101				*Coffea canephora*	Guinea	Mar-03
F. lateritium	IMI392282	NRLL 13275	FRC L84				Coffee berry	Guinea	Mar-03
F. xylarioides	IMI 127629	DSMZ 62457	ATCC 36326	FUS001	4004 (Gordon)	11646 (IMB and Blittersdo rf & Kranz, 1976)	*Coffea excelsa*	French East Africa or Central African Republic	Isolated 1955, received at IMI in 1967
F. xylarioides	IMI392674	NRLL 25486	CBS 25852	FUS 002			*Coffea sp.*	Ivory Coast	1951
F. xylarioides	IMI392675	NRLL 25804	CBS 749.79	FUS 003			*Coffea canephora*	Guinea	1963
F. xylarioides	IMI392676	ATCC1566 4		FUS 004				Unknown	1960s
F. lateritium	IMI392677	DSMZ 62456	ATCC36325	FUS 005			*Coffea excelsa*	Africa	Received IMI 1967, appears to have been

									isolated same year
F. xylarioides	IMI392678	TZ008						Tanzania	2003
F. xylarioides	IMI392679	TZ009						Tanzania	2003
F. xylarioides	IMI392680	G3P22							

1 As confirmed by project scientists or CABI mycologist. Where the identity has not been confirmed, a 'suspect' species based on the designation provided by the donor is given