Banana diversity and the food and income threats of pest and pathogen losses: Priority research areas to deploy diversity to reduce pest and disease losses

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One page summary:
According to the global news, bananas are in crisis for lack of diversity. In the report we mobilize existing information and expertise to provide orientation on the directions of future research investments to strengthen the role of banana diversity in response to the threat of pests and diseases on banana as food security and income among banana-growing countries. Although Cavendish bananas common in supermarkets around the world make up nearly half of world production, between 500-1000 cultivars of bananas divided into six major cultivar groups play an important role in food security and income generation for millions of households. A recent survey of over 500 banana experts worldwide prioritized 6 pests and pathogens among the top ten threats to production. These include black leaf streak (BLS), banana bunchy top disease (BBTD), Fusarium wilt (FW), bacterial wilts (BW), nematodes and weevils. Projected losses from these priority pests and pathogens were estimated as part of a study on research priorities conducted by the research program on roots, tubers and bananas (http://www.rtb.cgiar.org/). Over the next 25 years BBTD, FW and BW were projected to affect 1.5, 1.2 and 2.0 million hectares respectively with further losses as these organisms spread more widely. For BLS, nematodes and weevils which are already found globally, annual losses were estimated to affect 7.5 million hectares. Structured interpretive desk studies of these six major organisms addressed questions about the nature of yield loss, mechanisms for spread and cultivar resistance which provided the basis to rate the applicability of different approaches to reduce yield losses based on the use of Musa diversity. The report identified seven different approaches for the use of Musa diversity to reduce pest and pathogen losses. These were: cultivar substitution based on existing land races, cultivar mixtures, conventional breeding, GMOs, mutation breeding, somaclonal selection and plant microbiome enhancement. The different methods were scored on their applicability to address crop losses due to the six organisms and the status of technology readiness was summarized from proof of concept, advanced prototype to on-farm use. We attempted to assess the role of emerging results from advanced science in likely gains from each method, but this dimension of the study deserves further efforts.

Among conclusions in the evaluation of each method by organism, cultivar substitution offers some applicability although cultivar choice may be limited; especially if a specific market or consumer is targeted. Cultivar mixtures are more applicable for three of the organisms (BLS, nematodes, weevils), less applicable for BW and FW and not at all applicable for BBTD. Conventional breeding already has a base of released cultivars, pre-breeding lines and wild Musa with resistance and new breeding strategies with applicability for BLS, weevils, nematodes and FW. Sources of resistance are less certain for BW and BBTD. GMOs rate relatively high scores tapping on both trans and cis sources of resistance for all six target organisms. Regulatory frameworks and consumer acceptability are major issues. Mutagenesis was generally rated as uncertain with few cases of progress, although cultivars with shorter cycles or improved bunch formation are in use on-farm. The scoring for clonal selection is contradictory with a high score for FW based on successful cultivar development and quite low scores for the other target organisms. Cultivar-specific microbes or cultivars with heightened capacity to recruit microbes were rated as applicable for BLS, FW, nematodes and weevils.

Five priority cultivar deployment initiatives with accompanying methods and types of cultivars to be developed were identified:

- BBTD/food security and local market: 2 cultivars each of AAB plantain, AAA, EAH AAA, AAB dessert using GMOs;
- FW/food security and local market: AAA, EAH AAA, AAB dessert, ABB using conventional breeding, substitution and GMOs;
- BLS/export, large urban and processing market: Cavendish and plantain using conventional breeding, microbiome enhancement and cultivar mixtures;

- FW/export and large urban markets: Cavendish, AAB dessert, other AAA using conventional breeding, microbiome enhancement, clonal selection;
- BW/food security and local markets: ABB, EAH AAA, AAB plantain using GMOs

Research priorities to address these deployment initiatives are proposed:

- understanding molecular and genetic basis of cultivar resistance mechanisms and possible role of plant microbiome coupled with molecular and genetic characterization of diseases and other microbial organisms
- new breeding schemes, marker assistance and the strategies to minimize BSV interference to generate cultivars with multiple resistance with consumer-acceptable fruits with adequate post-harvest characteristics
- applicability of GMO advances in other crops to banana
- viability of shift from trans to cis genesis for key resistance
- applicability of clonal selection to other cultivars and for other pests and pathogens
- high throughput phenotyping for single and multiple resistances for application in conventional breeding, clonal selection, cultivar screening, mutagenesis and GMOs
- modeling epidemiological processes at the field scale and the possible role of cultivar mixtures

Other priority research areas:

- understanding disease spread at the landscape and improved projections of yield loss
- better statistics on banana by cultivar groups, including mapping
- priority research identified in the Global Musa Conservation Strategy, including collecting missions and greater screening and characterization of diversity
- understanding local cultivar diversity in all banana production projects with a past, current and future perspective to understand shifts in cultivar diversity and document the role of minor and rare cultivars in rural livelihoods

Table of contents:

1. Are our bananas threatened by pests and diseases?

In global news cycle the end of the banana gets headlines every couple years. The New Scientist in 2003 cited the director of INIBAP who projected that bananas could be gone in 10 years. In 2006 the threat to wild relatives of the banana was the focus of New Scientist headlines. In 2008 a popular book on the banana and Fusarium wilts again brought headlines to the doomed banana. Fast forward to 2016 and the catastrophe awaiting banana was again reported by different news outlets, primarily spurred by the spread of Fusarium out of Asia and into Africa. The public news cycle has also been paralleled by calls in scientific publications for more research funding to banana with an article in Nature in 2013 and more recently in PLOS in 2016.

At the request of CRP PIM's Flagship 1 cluster on Foresight Modeling, this report was put together based on existing information and expertise to provide orientation on the directions of future research investments to strengthen the role of banana diversity in response to the threat of pests and diseases on banana as food security and income among banana-growing countries. We have three major objectives in the report:

- Explore the interaction between banana diversity and pest and pathogen threats
- Evaluate the applicability of the alternatives to address pest and pathogen losses using Musa diversity
- Highlight priority research areas to improve diversity in bananas directed to the reduction of losses to pests and pathogens

Our work approached was organized in six steps:

1. Compile existing information on banana production and diversity based on existing data
2. Carry out structured interpretive desk studies of the six major banana pests and pathogens addressing questions about the nature of yield loss, the degree of cultivar resistance and the applicability of different approaches to reduce yield losses due to pests and diseases
3. Commission short summaries of 7 different approaches to the use of banana diversity to reduce pest and pathogen losses, contrasting the alternative strategies to use diversity, the steps and time period needed, the role of advanced science and new research developments, the limiting factor most resolvable and examples of success
4. Outside review of desk studies of pest and pathogens, including table on applicability of methods to loss reduction
5. Virtual meetings among key contributors to evaluate consistency and coherence of results and generate summary overviews and priority research areas
6. Complete draft report
7. Further steps being planned for review article

While the reports prepared by participants in the study are heavily referenced, here we have only referenced very specific data in the text. We have also provided references to publications specifically documenting research advances on the use of specific approaches to address pest and disease losses in Tables 11-16. We have also provided references for further reading about pests and pathogens and about the methods for the use of diversity. These references are listed by category at the end of the report. Original reports from participants in the study are also filed for further reference upon request.

2. Global importance of bananas

Most current domesticated bananas are linked back to two Musa species, *acuminata* and *balbisiana*, identified as either A or B. Over time through the process of domestication, these two seed-bearing species have become between 500 to 1000 cultivars which are vegetatively propagated and do not produce viable seed. These are primarily triploids identified with combinations of A and B depending on origin, although numerous diploids are also common in gardens and markets in certain areas. Table 1 provides an overview of primary cultivar groups and their common use, although there are also tetraploids and edible bananas generated from Musa *schizocarpa* and *textilis* referred to S and T genomes which are found in a very small number of cultivars.

Table 1: Uses and distribution of major cultivar groups of bananas

Cultivar group	Use/importance
AAA Cavendish	Primary export banana, also large domestic production in China, India, Brazil, Mexico
Other AAA dessert	Gros Michel previous export banana, diverse cultivars important in national markets in Asia, minor cultivars in Africa and Latin America, novelty red skinned export banana
East African Highland AAA	Food staple and beverage banana in East Africa
AAB plantain	Food staple in Central and West Africa, India and Latin America
Other AAB – South Pacific and dessert	Diverse minor dessert and food staple, although very important in specific countries like Brazil and Peru
ABB	Widely distributed in Asia, Africa and Latin America as minor food staple, although very important in specific countries like Philippines
Diploids AA, AB	Locally important food and dessert bananas in Papua New Guinea, East Africa, India only two-three dessert cultivars found across the continents

Country banana statistics globally do not capture production by cultivar group. FAO crop statistics represent only two groups, banana and plantain, and different countries report their production according to preferences by the national entities reporting the statistics. Burundi reports their national food staple banana as plantain, while Uganda reports the same food staple banana as banana. Mexico reports their national production as plantain (all Musa cultivars in Mexico are "*platano*"). The lack of more detailed data makes efforts to monitor shifts in cultivar make-up difficult. Fortunately the fruits marketing unit at CIRAD generates annual statistics by four cultivar groups (Table 2), dividing production into cooking and dessert with two subgroups in each group. The data show the importance of export or Cavendish banana production in South America, Central America and Asia in terms of volume. The importance of specific groups for food security by region is also noteworthy. In East Africa nearly 80% of production is bananas as daily food staple, while in West Africa plantains also a food staple, make up 70% of production. In South America, Central America and the Caribbean, plantains are also an important source of food, although they represent only 10-30% of regional production.

Table 2: Banana production and export by region and cultivar group

Regions	**Production (t)**	**Export (%)**	**Plantains AAB**	**East African Highland + ABB + others AAB + AAA + AA**	**Sub-group Cavendish AAA**	**Gros Michel & other dessert bananas AA, AAA, AAB, ABB**
East Africa	21 164 244	1,0	966 418	15 785 050	3 519 093	893 683
West and Central Africa	13 616 509	4,3	9 468 569	1 247 796	2 401 702	498 442
North Africa - Middle East	2 307 476	5,3	33	9 067	2 226 494	71 882
Central America	8 333 292	67,1	808 338	62 455	7 390 999	71 500
South America	22 456 387	34,5	5 607 796	388 856	13 049 085	3 410 650
Caribbean	2 698 944	23,8	767 852	665 957	1 096 248	168 887
Asia	61 078 751	5,7	2 113 680	16 406 438	31 098 370	11 460 263
Oceania	1 604 791	0,1	0	1 000	6 525	100
North America	7 625	-	2	17	423 900	27
Europe	423 946	-	1 162	530 706	796 437	276 486
World	133 691 965	16,4	19 733 850	35 097 342	62 008 853	16 851 920

Source: Lescot, (2012)

Production by six cultivar groups was estimated drawing on FAO and Fruitrop statistics as the basis of an exercise to estimate returns to different research options (Table 3). Data for the 25 countries globally which produce more than 1 million tons annually of bananas provide a more detailed profile of the implications of banana for export earnings, national income and food security. Of the 25 countries, nine countries export more than 10% of their production with three countries over 80%. These countries are largely in Latin America and the Caribbean, with only two countries in Africa and one country in Asia. In terms of food consumption, three countries are heavily dependent on bananas as a daily staple food – Uganda, Burundi and Rwanda with annual per capita consumption of banana between 180-270 kg. The loss of bananas as a staple crop in these countries would represent not only a food crisis, but would also result in the conversion of thousands of hectares from a perennial crop to annual cropping with increased soil tillage and erosion. Nine countries from Africa, Latin America and Asia consume between 50-100 kg of banana/year. Table 3 also shows an approximate breakdown between bananas, including plantain, as a food staple and bananas consumed as a fruit, referred to here as dessert bananas.

Table 3: Production, export income and per capita consumption by cultivar type data for 25 countries globally which produce more than 1 million tons annual of bananas. (Data base adapted from Fruitrop and FAOStat for production data from 2011 as part of Pemsl et al, 2014.)

									Consumption per/capita (kg)[1]		
Countries	**Production (t) 2011**	**Cavendish AAA**	**Other AAA, AA**	**EAH AAA**	**AAB Plantain**	**Other AAB**	**ABB**	**Export (%)**	**Total**	**Dessert banana**	**Banana staple**
East Africa											
Burundi	1 855 243	136 564	230 000	1 018 679	170 000	100 000	200 000	0,0	182,6	45,9	136,6
Uganda	9 550 000	241 000	164 000	7 445 000	200 000	500 000	1 000 000	0,3	253,8	23,8	230,0
Kenya	791 570	238 570	80 000	80 000	305 000	8 000	80 000	0,0	17,8	7,4	10,5
Rwanda	2 749 150	120 000	100 000	1 850 000	270 000	150 000	259 150	0,0	233,4	31,4	202,0
Tanzania	2 924 700	100 000	50 000	2 024 000	150 700	300 000	300 000	0,0	59,4	9,1	50,2
West & Central Africa											
Cameroon	2 220 000	500 000	220 000	70 000	1 300 000	0	130 000	13,3	86,5	20,9	65,6
DR Congo	1 566 472	292 472	24 000	100 000	1 045 000	0	105 000	0,2	76,9	15,5	61,4
Côte d'Ivoire	2 111 454	400 000	6 000	0	1 500 000	0	205 454	10,6	26,9	2,2	24,7
Ghana	1 870 000	130 000	10 000	25 000	1 680 000	0	25 000	2,1	69,7	2,9	66,8
Nigeria	2 733 300	263 300	85 000	0	2 258 000	0	127 000	0,0	15,7	2,0	13,7
North Africa											
Egypt	1 028 950	985 949	40 000	0	1	0	3 000	1,2	12,3	12,3	0,0
Central America											
Costa Rica	2 202 000	2 100 000	10 000	0	90 000	0	2 000	91,4	39,0	21,4	17,6
Guatemala	1 737 600	1 500 000	10 000	0	202 600	0	25 000	87,3	19,8	11,4	8,4
Mexico	2 103 360	1 868 360	30 000	0	192 000	3 000	10 000	18,0	15,7	14,1	1,6
South America											
Brazil	6 978 310	3 594 960	200 000	0	453 350	2 700 000	30 000	1,2	34,1	31,7	2,4
Colombia	5 338 390	2 034 340	469 000	60 000	2 650 000	20 000	105 050	30,9	70,5	14,9	55,6
Ecuador	5 867 291	5 200 000	120 000	0	500 000	0	47 291	83,7	48,3	23,8	24,5
Peru	1 450 000	270 000	120 000	0	900 000	160 000	0	6,9	41,4	14,5	26,9
Carribean											
Dominican Rep.	1 085 709	590 000	4 200	0	400 000	45 000	46 509	36,9	72,4	30,0	42,4
Asia											
China	9 848 895	9 042 415	0	0	60	137 995	668 425	0,0	7,2	6,7	0,5
India	31 897 900	6 897 900	10 720 000	0	2 600 000	2 680 000	9 000 000	0,2	25,4	16,2	9,3
Indonesia	5 814 578	2 223 228	1 180 000	0	70 000	41 350	2 300 000	0,1	23,3	13,8	9,5
Philippines	9 101 340	5 000 000	1 300 340	0	1 000	70 000	2 730 000	37,0	72,1	44,4	27,8
Thailand	1 534 900	650 900	124 000	0	60 000	100 000	600 000	2,0	22,5	12,7	9,8
Vietnam	1 481 400	681 400	202 400	0	2 000	0	595 600	0,9	16,3	9,6	6,7

[1] Per capita consumption of national production (imports not included, exports subtracted from total production).

3. Banana diversity – three dimensions

Three dimensions of banana diversity are described in this overview – centers of cultivar diversity, cultivar group diversity of the cultivars commonly used on farm and centers of diversity of wild Musa relatives. A brief introduction here to these three dimensions will later allow us to comment on the threat of pests and pathogens to Musa diversity.

Scientists working on the origins of banana have identified specific geographic zones which for their high diversity of cultivars of a certain genotype have been identified as centers of cultivar diversity (Table 4). These are relatively unique for each cultivar group, although the precise geographic limits are not easy to establish and collection missions are still being organized. A recent mission went to Papua New Guinea, for example, to collect diploids and collected 60 accessions thought to be new. Summing across these sites, upwards of 1000 cultivars of different bananas are found globally, although more conservative estimates suggest upwards of 500. Asia and Oceania are the location of all, but two of these zones. A precise inventory of diversity in each zone is difficult and time consuming to conduct since the same cultivar may have different names in the same zone. Over 60 plantain cultivars have been documented in the Congo basin (Adheka, 2013). For the triploid AAA East African Highland banana over 80 cultivars has been identified (Gold et al 2002), although analysis is still ongoing in both cases.

Table 4: Centers of diversity for cultivars

Centers of diversity of banana	Cultivars
Philippines, Malaysia, Indonesia	AA, AAA, ABB
Papua New Guinea	AA ; AAB other unique cultivars AS, AAS, AAT, ABBT
South Pacific (Polynesia, Melanesia e Micronesia)	AAB
India	AB, ABB, AAB other
West and Central African rainforest centering on the Congo basin and extending into Cameroon	AAB Plantains
East Africa, primarily Uganda, Burundi, Rwanda and parts of Tanzania	EAH AAA
East African coastal region – Tanzania, Comores	AA

Beyond these centers of cultivar diversity, diversity can also be found in the number of cultivars commonly found in backyard gardens, on farm and in markets. Clearly those regions which are centers of origin are high in this scoring, but among banana growing areas which are not centers of origin, some have integrated numerous cultivar groups, while others use primarily only one or two cultivars. Many countries growing bananas in the subtropics, for example, cultivate only one or two cultivars, mostly from the Cavendish group. These include Argentina, Paraguay, South Africa, Zimbabwe, Zambia, Senegal, Sudan, Morocco, Egypt, Iran, Turkey, Yemen, Australia, China and Taiwan. Tropical countries of Latin America and Africa outside of cultivar diversity centers have a much broader set of cultivar groups which are commonly used for dessert and food staples (Table 3).

A third dimension of Musa diversity is the wild relatives which represent the potential source of resistance genes for breeding programs. These are found in disturbed natural vegetation on river banks, forest gaps and secondary growth concentrated in four different regions. Depending on the location, this source of genetic diversity may also offer useful genes as well for abiotic stress like cold temperatures and drought.

Table 5: centers of diversity of wild Musa relatives

Geographic region	**Wild relative group present**
South Pacific (Polynesia, Melanesia e Micronesia)	Fe'i, Eumusa
South-East Asia	Eumusa (A, B, S)
South Vietnam, Peninsular Malaysia, Borneo, Sumatra	Callimusa
Monsoonal areas in mainland southeast Asia (Myanmar and China)	Rhodochlamys

4. Six important banana pests and pathogens

In a survey of over 500 banana experts across the globe carried out in 2013 by the CRP RTB, six of the top 10 yield-limiting factors were specific pests and pathogens (Pemsl et al, 2014). These six pests and pathogens are the focus of our overview here and the analysis in later sections. In addition to the six pests and pathogens, other factors included, in order of ranking: the phytosanitary quality of planting material, water deficit, non-uniform yield potential of planting material and wind.

In the next 14 pages, the six pests and pathogens are described focusing on a common framework which will later allow us to analyze the importance of each for Musa diversity and to assess the seven approaches to the use of diversity in terms of their relevance for each of the six pests and pathogens. The description begins by addressing how yield loss occurs for each pest/pathogen. Here we will see that three of the organisms cause near or complete yield failure mat by mat, while the other three cause yield loss across all mats in small increments, only occasionally resulting in complete yield failure in an individual mat. This is a crucial difference in terms of their threat to Musa diversity both in centers of origin and to cultivar group diversity. For each pest/pathogen we also address differences in cultivar susceptibility, how the organism spreads, the current understanding of the genetic and molecular mechanisms for cultivar resistance and the likely threat to rare cultivar or wild relative diversity and to the diversity of cultivars on farm. The description of each pest/pathogen ends with a summary of management practices. At the end of the section we review the distribution of the pests and pathogens by the 25 leading banana production countries (Table 6) and the centers of diversity (Table 7)

Black leaf streak (BLS)

How does Black Leaf Streak (BLS) threaten yields

The ascomycete *Mycosphaerella fijiensis*, the causal organism of BLS, is a sexual, heterothallic fungus having *Pseudocercospora fijiensis* (M. Morelet) Deighton as the anamorph stage. BLS does not kill the plants, but decreases the photosynthetic area and capacity of leaves, causing a reduction in the quantity and quality of fruit. Specific toxins generated by the disease induce the premature ripening of fruit harvested from infected plants. Pathogen growth occurs from is 12° C to 36°C with an optimum around 27°C. Ascospores germinate when the leaf surface is wet associated with relative humidity higher than 98%. Therefore the disease is more aggressive during extended rainy periods, although newly emerging leaves have less disease if drier conditions prevail.

Cultivar susceptibility

The range of cultivar susceptibility to BLS is wide. Cavendish bananas, plantains and East African Highland bananas, among major market groups globally are highly susceptible. Most of the AAcv cooking-type cultivars originated from Papua New-Guinea are also highly susceptible. The management of BLS is the major challenge to the export banana industry which is based on Cavendish. Organic export Cavendish are grown in dry climates which dramatically reduces BLS pressure. The East African Highland bananas are grown at higher altitudes about 1000 meters above sea level where BLS aggressiveness is also reduced by lower temperatures. Cavendish subgroup (AAA) is highly susceptible. Cultivars resistant to BLS include the partially resistant clones of ABB genomic group like Bluggoe, Pelipita, Saba, Pisang Awak (ABB, *i.e.*, Fougamou), the Mysore subgroup (AAB, i.e. 'Thap Maeo'),the diploid cultivars Pisang Mas, Pisang Madu and others. Among the AAA genomic group, the Ibota sub-group with the reference cultivar 'Yangambi Km5' are highly resistant. Resistance to BLS has been a major focus of breeding programs at CIRAD, Embrapa, FHIA, CARBAP and IITA and in breeding programs in Uganda, India, and Philippines. The varieties used as sources of resistance to BLS are Paka AA, 'Calcutta 4', the diploid banana 'Pisang lilin' and at the lesser extent the triploid 'Yangambi Km5' which pollen fertile.

Does pest or pathogen threaten Musa diversity - rare cultivars and wild relatives

While BLS is a serious disease for commercial plantations and can reduce yields for smallholder, especially when plant nutrition is marginal, it is not a threat to Musa, since mats continue to produce in spite of severe disease levels and recover when drier conditions prevail.

How will the organism cause reduction in diversity of cultivars which are commonly used?

The release of new cultivars or the introduction of more resistant cultivars may lead to a shift in cultivar importance. However, cultivars which have particular uses or functions will be maintained, even if in lower percentages.

Current understanding of genes and other molecular mechanisms related to differences in cultivar resistance

Pending collaborator input

How does BLS spread

The life cycle of BLS starts with leaf infection by either ascospores or conidia. After a period of epiphyllic growth of generally 2–3 days, germ tubes penetrate stomata. In good conditions, the first symptoms appear generally 10–14 days after. The symptoms then gradually evolve from:
Yellowish specks that are less than 1 mm visible only on the underside of the leaf.

1. Red or brown streaks (changing to black) on the underside and later upper side of the leaf.
2. Streaks become longer and larger.
3. Brown elliptical or circular spot on the underside of the leaf and as a black spot on the upper side.
4. The spot is totally black, surrounded by a yellow halo, and has spread to the underside of the leaf blade.
5. The centre of the spot dries out, turns light grey and is surrounded by a well-defined black ring, which is itself surrounded by a bright yellow halo.).

Conidia are produced on young stages of the disease (stage 2–4) and are water dispersed at short distances, even if they are also present in the airspora. Ascospores are produced at the later stage and are wind-dispersed after perithecia burst. Although the epidemiological importance of conidia is still not well understood, it is accepted that ascospores are predominant. The wind dispersion of ascospores is responsible for the infection of new leaves as they unfurl from the center of the leaf whorl. Under conditions of adequate leaf wetness, ascospores germinate and as the leaf ages, the disease on the leaf advances through the progressive stages. When the first necrotic spots appear in stage 4f, new ascospores are produced to be dispersed and result in new infections on newly emerging leaves.

Key management factors

While eradication has been achieved only in northern Australia, the disease has spread from its first appearance in Fiji in 1963. The major focus for BLS management throughout most banana growing areas is on minimizing leaf loss and reducing the potential for the production of ascospores. Key practices are leaf pruning to avoid the development of black necrotic spots which produce ascospores and protection of new leaves with fungicides. In addition, vigorous growth of the plant with adequate fertilization and water ensures that new leaves are emerging at a faster pace than the advance of the disease. The aim of management is to reach harvest with an adequate number of functional leaves to generate market-acceptable fruit quality. At the same time the production of ascospores must be kept to a minimum to avoid the explosion of the disease in the other stems in the mat which will produce the next round of bunches.

Banana bunchy top disease (BBTD)

How does BBTD threaten yields

The causal organism of banana bunchy top disease (BBTD) is the banana bunchy top virus (BBTV Babuvirus). BBTV is a multicomponent isometric virus with at least six circular single stranded DNA (ssDNA) genomes (BBTV DNA-R, -M, -S, -N, C and -U3). BBTV only affects bananas and related Musaceae and other alternative host plants have not yet been confirmed. The virus is transmitted in a *persistent* circulative way by the banana aphid (Pentalonia nigronervosa) which is almost exclusively found on banana. When the infection occurs in adult banana, the main stem may produce a smaller and deformed bunch, but afterwards any later stems will not produce fruit. Plants originating from infected suckers grow poorly, are stunted, and produce no fruit. In a newly infected field the disease progresses mat by mat and yield loss occurs quickly in a field as more and more mats no longer produce. The disease is less common and has lower spread at higher altitude and is not found below 16°C temperature.

Cultivar susceptibility

There are no known cultivars or wild relatives which are completely immune to BBTD. All known germplasm can be infected and serves as host to the disease, although there are differences among genotypes as to the symptomatology and virus titers (virus levels in the host). Cavendish cultivars rapidly express symptoms and experience yield collapse, while AAB plantains are slower to show symptoms. ABB such as Pisang Awak and Saba show fewer symptoms and continue to produce smaller and deformed bunches for a longer time. The search for resistance genes in wild Musa relatives is ongoing. Butuhan (BB) and Khae Phrae (AA) were recently identified as immune to infection. Musa *balbisiana* has also been suggested as source of resistance genes. These initial promising findings are still be verified among virologists.

Does pest or pathogen threaten Musa diversity rare cultivars and wild relatives

The movement of the aphid from mat to mat is particularly problematic in smallholder production or backyard plantings with many different small plots side by side and few management practices aside from harvesting and sucker extraction. In centres of diversity, where within cultivar diversity is still common and rare cultivars are as yet uncollected, the presence and increasing severity of BBTD could lead to the permanent loss of diversity. Since the disease can easily be transmitted into wild bananas as well which are found in natural settings and not subject to management practices, BBTD may also threaten the permanent loss of such wild diversity.

How will the organism cause reduction in diversity of cultivars which are commonly used

The presence of BBTD may also lead to a reduction in the diversity of cultivars which are commonly used. Where the disease has spread unchecked by clean seed based recovery efforts, the only cultivars remaining are more hardy ABB cultivars which continue to produce small and deformed bunches long after the disease is present. This was the case in certain zones of Malawi where Cavendish and plantains had disappeared, while only ABB Pisang Awak remained. In east New Caledonia a reduction of Musa diversity was also registered due to BBTD infection. In zones with sources of BBTD-free planting material, there may be a restricted group of cultivars available with a loss of minor cultivars which have lower demand from farm households, since recovery efforts usually focus on more important market cultivars. Increased planting and promoting of Cavendish types or other very susceptible cultivars with higher market demand can also increase infection pressure on other genotypes.

Current understanding of genes and molecular mechanisms to explain cultivar resistance

Cultivar differences in susceptibility depend on diverse factors, including attractiveness of the cultivar to aphids (studies show quite different aphid levels on different cultivars), ease of infection by aphid, higher or lower accumulation of titre levels in the plant, and expression of symptoms according to titre level. The genetic or molecular control of these characteristics is only beginning to be understood and transgenic strategies for developing crops resistant to this plant virus are being developed as reviewed in Elayabalan et al, 2015.

How does BBTD spread

BBTV is systemic within the banana mat and moves from point of first infection into all plant parts forming after that point, first appearing in leaves as dark green streaks of dot-dash patterns on the lower portion of the leaf's midrib first and on the secondary veins later and then distorting leaf size, color and arrangement (hence the name of the disease). All suckers connected to the infected stems ultimately become infected, even though in initial stages they are asymptomatic. Such suckers are also a means of transmission of the disease. The banana aphid moves BBTV to other mats in the same field in the winged form and even to other fields. The distance beyond which very little movement occurs has been calculated at 100m. Longer distance movement of aphids is very unusual, but occurs assisted by winds. Spread to long distance fields and into new regions and continents occurs through planting material which carries the virus or the vector. One of the most recent outbreaks identified in 2011/12 on the border between Benin and Nigeria is very distant from the border between Gabon and Cameroon where BBTD is more common. The most recent outbreak in South Africa in 2015 may have spread from Mozambique and Zimbabwe. In both cases this major jump occurred probably through infected planting material.

Key management factors

The recovery of banana in areas affected by BBTD involves the elimination of all banana plants in the area to be recovered, including 100m buffer which should remain free of banana, and the maintenance of the area free of banana long enough to ensure no survival of banana aphids. Replanting should be done with BBTV-free planting material while still maintaining the 100m buffer-zone free of banana. Once banana has been replanted, regular inspection of plants ensures early detection of virus symptoms in young plants. Early symptomatic detection may be easier in some varieties than others, as typical early symptoms are variable. Plants with clear symptoms should be rogued ensuring no resprouting occurs and aphids are not dispersed to other plants.

To minimize the spread of BBTD into new zones, there should be zero movement of suckers from areas with BBTD into areas without BBTD. Early detection of infected plants in new areas will depend on ongoing surveillance by public agencies as well as informed rural communities who have been trained to detect symptoms.

Bacterial wilts (BW)

How do Bacterial wilts (BW) threaten yields?

The major bacterial diseases of importance to *Musa* spp. (bananas and plantains) include*:* i) Xanthomonas wilt of banana and enset (XW), caused by *Xanthomonas campestris* pv. *Musacearum* (Xcm); and ii) Moko, Bugtok and banana blood disease, caused by different phylotypes of *Ralstonia solanacearum.*

Both bacteria can cause severe yield loss by affecting the harvestable bunches and sometimes killing ratoon stems within a mat. Both *Xcm* and *Ralstonia solanacearum race 2* (Smith) enter the vascular system of the Musa plant either through open wounds. They are mainly introduced into plants by contaminated tools or insect vectors attracted to sap and nectar in the inflorescence. Infections through soil and water have also been reported to be common for *Ralstonia solanacearum.* Infections in a mat are not completely systemic. External symptoms vary somewhat among the two diseases, but the effect on the plant is similar. Depending on the point of entry, the first symptoms are either yellowing of leaves or a drying rot and blackening of the male bud and/ or the rachis. The fruits ripen unevenly and prematurely. The pulp of infected fingers has orangish-brown stains and is inedible. A yellow-orange streaking of the vascular tissues and discharge of yellow bacterial ooze is revealed when a cross-sectional cut is made on an infected pseudostem.

For Xcm at field or farm level losses can reach up to 100% if control is delayed, although initially bacteria primarily affects productivity of infected stem or bunch, and has only limited movement into other parts of the mat. For Ralstonia, depending on the abundance of the susceptible hosts, losses varying between 36% and 100% have been reported. The highest losses have been reported in the susceptible ABB cultivars such as ‘Saba’ and ‘Pisang Kapok’.

Disease development and spread has been observed to be higher under wet conditions. XW incidence has been observed to decline with increase in altitude, due to the decline in number and activity of insect vectors. Abiotic factors such as nutrient (organic matter and minerals) conditions, soil type, pH, anaerobic conditions, temperature, and water/moisture content are among multiple and complex factors influencing the development of *R. solanacearum*.

Cultivar susceptibility

There are no BW resistant cultivars with artificial inoculation. ABB types ('Pisang Awak, Bluggoe, Saba) with non-persistent male floral bracts and neuter flowers and sweeter sap are more susceptible to insect mediated infections and are the cultivars for which epidemics are commonly found. The persistent floral bracts of ‘Pelipita’ (ABB), ‘Mbwazirume’ (AAA) confer protection from insect vectors, while the lack of developed bracts of Horn plantain (ABB) offer yet another avoidance mechanism. For Cavendish in export plantations, floral buds are removed routinely reducing susceptibility and in areas with a history of outbreaks, all tools used in plantation management are disinfected between each mat.

Do BW threaten diversity of rare cultivars and Musa wild relatives?

In areas of high cultivar diversity of ABBs BW may generate pressure on rare cultivars which are found unattended in backyard gardens and semi-wild. Since simple management including the harvesting of the male bud which is often edible serves to reduce the threat, the threat is not severe and rare uncollected cultivar loss is unlikely.

Will BW lead to a reduction in diversity of cultivars which are commonly used?

The ABB types have been associated with outbreaks of BW at epidemic levels. The disease, if not timely and well managed, can thus selectively eliminate these cultivars from within the affected landscapes. Farmers can also potentially select against these cultivars by growing only cultivars with good level of tolerance thus eliminating them from affected niches. These same ABBs are also susceptible to Foc and the presence of both Foc and BW leads to a dramatic reduction of ABBs among smallholders.

Current understanding of genes and molecular mechanisms to explain cultivar resistance

Currently no edible cultivars resistant to BW are known in the affected zones to use as parents for breeding. Thus, no genes of resistance have been pin-pointed to date. However, resistance towards XW has been observed in *Musa balbisiana* a non-edible wild *Musa* spp. Studies to pin point the mechanism and genes of resistance in *M. balbisiana* have not been conclusive.

How do BW spread?

BW mainly spread within and between fields through insects foraging for sap, nectar and pollen, contaminated farm tools (used for leaf pruning, weeding, harvesting bunches and desuckering) and infected planting materials. Banana weevils and nematodes potentially play a role in local spread of the disease while bats and birds can spread the disease both locally and over long distances. Livestock grazing first on a sick plant and then a healthy plant have been shown to move the bacteria.

The movement of Xcm or *R. solanacearum* within the mat from the one stem to another or into suckers is not frequent. Healthy suckers taken from mats with a diseased bunch stem generally produce a healthy new plant, if care is taken with tool disinfection. Xcm is reported to have limited survival in soil for only a few months, while *R. solanacearum* has been reported to survive in soil and water for variable but longer time periods (1 – 4 years). Thus infections through soil and water are common for *R. solanacearum.*

Key management factors

A set of cultural practices are useful to limit yield losses due to BWs in areas where the disease is present. Removal of male buds immediately after the formation of the last hand using a forked stick ensures that neither cutting tools nor insects will vector the disease. If the disease is already present in the banana stand, tools should be sterilized using fire or 3.5 % sodium hypochlorite between every mat. Removal of diseased stems at first symptom in the bunch or the leaves can also reduce spread.
In export banana plantations where Ralstonia is present, a diseased mat is quarantined up to 5 meters to limit movement in and out of the area of infection and the diseased mat and its neighbors are uprooted and burned on site with a smoldering fire of rice hulls.

Minimum banana-free fallow periods of 6 months are needed on field infested by insect-transmitted infection and up to 12 months for soil-transmitted infection. These periods are shorter for XW than for Moko. The field should then be replanted with clean planting material and cultural practices used as highlighted above.

Fusarium wilts (FW)

How does Fusarium wilt threaten yields?

Fusarium wilt (FW) is caused by the fungus *Fusarium oxysporum f.sp. cubense (Foc).* It provokes total yield loss mat by mat until all mats in a field are affected. Infection occurs through root hairs, root caps and lateral roots. Once the infection is established, the hyphal network grows across intercellular spaces along the junctions of root epidermal cells and once inside the cells, the fungus grows rapidly intercellularly and intracellularly producing microconidia which move into the vascular system. The toxins, tylose and gum produced by the fungus cause parenchymatic companion cell growth. Severe water stress, due to the occlusion of the perforated plates of the xylem vessels, causes the typical symptoms of wilting, eventually leading to the death of the plant. The pathogen invades the parenchyma and sporulates profusely, releasing into the soil conidia and chlamydospores which infect neighboring banana plants, starting new disease cycles. The chlamydospores can remain dormant in the soil for more than 20 years in the absence of banana, but are stimulated to germinate by banana root exudates thus re-initiating the pathogen infection cycle.

Abiotic factors such as temperature, wetness or soil pH can influence the disease intensity. Range temperatures between 23 and 27°C favour pathogen growth and rainy season promotes the spread of spores. Physical and chemical soil characteristics can also influence the disease intensity. High aggregate water-stability, low pH, low clay content, low EC (electrical conductivity) and low levels of soluble Na promote disease infection. Fe availability and Zn deficient conditions increase disease incidence. Ammoniacal nitrogen (NH_4) applications promote disease development.

Cultivar susceptibility

Cultivar susceptibility is based on Foc races, although more recently VCGs (vegetative compatibility groups) have been used to fine tune susceptibility. Based on pathogenicity to different reference varieties in field conditions, Foc has been classified into races. Race 1 affects Gros Michel (AAA), Manzano/Apple/Latundan (Silk, AAB), Pome (AAB), Maqueño (Maia Maoli - Popoulu subgroup, AAB) and Pisang Awak (ABB). Race 2 affects ABB cooking bananas, such as 'Bluggoe' (ABB). Tropical race 4 (TR4) attacks notably Cavendish and a broader list of cultivars which is still being refined. Many cultivars susceptible to races 1 and 2 are also susceptible to TR4. East African Highland bananas also show susceptibility although less than more susceptible cultivars like Cavendish. The AAB plantains appear to be unaffected by TR4, although screening is just beginning. Numerous wild species provide resistance to the different Foc races and include *M. acuminata* subsp. *malaccensis,* subsp. *burmannica*, subsp. *microcarpa* and subsp. *siamea* (to race 1); and *M. acuminata* subsp. *malaccensis,* and subsp. *burmannica*, *M. balbisiana*, *M. basjoo*, *M. itinerans*, *M. nagensium*, *M. ruiliensis*, *M. velutina* and *M. yunnanensis* (to TR4). These have been used to generate pre-breeding and crossing lines by different breeding programs.

Does FOC threaten the diversity of rare cultivars and wild relatives

Fusarium could potentially threaten rare cultivars and wild relatives because of its capacity to kill whole mats and eventually fields of bananas. However, it is mainly present in commercial farms –monoculture fields, and these fields are often constituted by very common varieties. The rarest varieties are often found in the bush or backyard gardens and Fusarium doesn't reach those places as readily. They are usually not cultivated in monoculture conditions – favourable to the spread of Fusarium. Although TR4 has appeared in commercial plantations immediately after forest clearing in Indonesia, little is known about its distribution in undisturbed conditions and only recently has more reliable diagnostic tool been developed to distinguish TR4 from other Foc strains in the soil.

Will Foc lead to a reduction in diversity of cultivars which are commonly used

Varieties susceptible to the different Foc races are found in smallholder production around the world. Once a soil becomes infested, susceptible cultivars cannot easily be grown in the field. To continue in production new clean land must be found, although many farmers opt for a change to resistant cultivars. Foc susceptible cultivars preferred by markets for their flavour are often scarce and higher priced in local markets. These include Latundan (Philippines), Maçã (Brazil), Pisang Rastali (Malaysia), Rasthali (India) which belong to the AAB 'Silk' subgroup; Lady Finger (Australia), Prata (Brazil), Virupakshi (India) which belong to the AAB 'Pome' subgroup and Chuoi Tay (Vietnam), Kayinja (East Africa), Kluai Namwa (Thailand) which belong to the ABB 'Pisang Awak' subgroup. All these cultivars, including Gros Michel (AAA) found globally, will be found with lower frequency in the future as more soils are infested and the market acceptance of resistant cultivars increases.

Current understanding of genes and molecular mechanisms to explain cultivar resistance

Resistance to Foc race 1 in Musa seems conditioned by a single recessive gene designed as panamá disease 1 (pd1). Unfortunately, studies related to gene resistant identification are still far from finding the sequence of the gene or the group of genes responsible for resistance to Fusarium Wilt in bananas.

How does FOC spread?

Once a mat is infected, mycelial growth and microconidia move in xylem and systemically infect all other stems and suckers. Conidia and chlamydospores in the soil infect new banana mats in the field. Spread between fields may occur though infected planting material and infected soil in water, on shoes and nursery substrates. Intercontinental movement can also occur through these same means.

Key management factors

Few practices are available to produce bananas in Foc-infected soils. The substitution of resistant cultivars has been considered the only workable approach. Recently pilot work in Brazil has shown that the use of endophytes, soil amendments, cover crops, non-ammoniacal fertilizers and the avoidance of certain herbicides can reduce infection and extend plantation life, especially when cultivars with partial resistance are planted.

A more important strategy is to avoid infestation of a field. A set of practices is essential: planting material free of Foc, strict supervision of all persons, equipment and machinery entering the field to avoid contaminated soil or water, strict control of any nursery substrate of plants used in the field either from bananas or other crops and careful inspection of surrounding fields from which Foc could enter through soil erosion or water flows.

Weevils

How do weevils threaten yields?

Cosmopolites sordidus Germar, a nocturnally active, free living black weevil, measuring 10-15mm, feeds exclusively on banana. Banana weevils are found between leaf sheaths, in the soil at the base of the mat or associated with crop residues and use fresh and decomposing banana tissues for both food and oviposition. Single eggs are inserted in cavities made by the female's rostrum into the corm and pseudostem base at a rate of 0.5-4 eggs per week. On hatching, the larva feeds on the corm forming tunnels. The complete life cycle, including pupation in the corm, is 6-8 weeks under tropical conditions, with a lifespan of up to 4 years.

The adult weevil does relatively little damage, while the tunnelling larva affects plant growth and yield through damage caused to the rhizome and indirectly the root system, disrupting water and nutrient uptake, and weakening plant anchorage. Yield loss in banana is caused by a combination of reduced bunch weight, plant loss through toppling, snapping or premature death and lengthened crop cycle. This also results in reduced plant size and vigour, and increased susceptibility to other pests and diseases. Weevils are rarely encountered above 1600 masl and egg development does not occur below 12°C.

Cultivar susceptibility

Globally cultivars show great variability in susceptibility. The dessert (AAA and AB) and brewing (AB and ABB) cultivars are considered moderately to highly resistant, while highland cooking banana (AAA-EA) and plantain (AAB) are known to be highly susceptible. Within these susceptible groups more resistant cultivars have been identified such as Karumpoovan and Poozhachendu in India and Kedong kekang in Cameroon for plantain. Some highland banana cultivars like Tereza, Nalukira, Nsowe, Kabula, Nakitembe, and Mbwazirume exhibit intermediate resistance.

Several cultivars from different genome groups have been identified as candidate sources of resistance in breeding programs: triploids include Yangambi Km-5, Pisang Awak, Bluggoe, Sakkali, Senkadali, while diploids include Sannachenkadali, Elacazha, Njalipoovan. Wild species with resistance include Calcutta-4 and M. balbisiana.

Do weevils threaten the diversity of rare cultivars and wild Musa relatives?

Weevils are present across the many areas of high cultivar diversity, but are not a threat to reduce this diversity. Mats may be weakened but are not killed by weevils and clean planting material is relatively easy to obtain on farm.

Will weevils lead to a reduction in diversity of cultivars which are commonly used?

The weevil has been implicated in the decline and disappearance of highland banana from traditional growing zones in East Africa. In response to weevil pressure as well as the spread of BLS, many farmers have shifted from more susceptible highland bananas to more resistant ABB brewing cultivars. This continuous selection may gradually pose a threat to the genetic erosion of some highly susceptible landraces. Similar shifts may occur in other regions, but weevils alone are generally not the major factor.

Current understanding of genes and other molecular mechanisms related to differences in cultivar resistance

Collaborator input still pending

How do weevils spread?

Dissemination of the weevil is through infested plant material and movement of the adults among adjacent old fields to new fields. Active dispersal by crawling is slow and limited, although adult weevils can cover distances of 15 m in 1 day, 35 m in 3 days and 60 m in 5 months within their natural habitat. The beetle is highly susceptible to desiccation and will die within 3 to 10 days on a dry substrate, but can survive considerably longer in moist soil without food. Fields should be fallowed at least 12 months before replanting if weevil infestations were high in the old field.

Key management factors

Management to reduce yield loss due to weevils begins with the use of clean planting material which is not infested with weevil eggs or larvae such as tissue culture plantlets and pared hot water-treated field suckers (at 52-55°C for 15-27 minutes). Suckers from young stands generally have fewer weevils than older fields. Paring alone can also contribute in many cases. All roots and the outer surface of the corm should be removed leaving only white surface with no evidence of tunnels or spots of brown, orange or black. Pesticides have also been used to disinfect suckers. Botanical substances such as neem have been used as weevil control.

The field to be planted should have been free of banana for at least a year. Care can also be taken that nearby banana fields are not a source for weevils moving into the new field. Traps made from cut banana stems can also be used to attract weevils which seek out smells of freshly cut banana tissue. Once a field has been established, a wide range of practices can be deployed to reduce the build-up of weevils. Of high priority is the management of banana stems and corms which have been harvested. Stems should be chopped into pieces to encourage drying and rapid decomposition and corms should be cut as low to soil level as possible and covered with soil. Trapping with sections of freshly cut banana stems can also be used either with hand destruction or chemical or biological control in the trap. Beauveria bassiana can be applied in the traps. Pheromones are available to make trapping more effective. The use of diverse species of predatory ants has also been tested.

Nematodes

How do nematodes threaten yields?

Nematodes are multicellular, vermiform microscopic animals between 0,2 - 1mm long. They can be free-living or parasitic on animals or plants. Plant parasitic nematodes characteristically have a stylet which is used to suck the content of plant cells. The pharynx allows muscular protraction and retraction of the stylet and produces species-specific glandular secretions. Continued puncturing of the cells causes mechanical damage. The glandular secretions induce chemical changes in the plant cells, increasing, for example, the production and solubility of nutrients. On bananas and plantains, plant parasitic nematodes feed on the roots. Depending on the feeding strategy and mobility, plant-parasitic nematodes are called ectoparasites (do not enter plant tissue), migratory endoparasites (mobile nematodes that enter the plant tissue) or sedentary endoparasites (nematodes which cease to be mobile once they have reached a feeding site inside the plant). The most destructive plant parasitic nematodes for bananas and plantains are the migratory endoparasites, or burrowing nematodes, such as *Pratylenchus coffeae* and *Radopholus similis.* Other migratory ecto- and endoparasitic nematodes common to bananas include *Helicotylenchus dihystera, Helicotylenchus multicinctus, Hoplolaimus pararobustus* and *Pratylenchus goodeyi*. Only one sedentary endoparasitic species is commonly found in banana roots, the root knot nematode *Meloidogyne incognito*.

The aboveground symptoms of nematode damage are related to an impaired uptake of nutrients by the plant, resulting in reduced plant growth, lengthening of the growth cycle and reduced bunch weight. The weakened root system can also lead to toppling of the plant before harvest, particularly during strong wind. The cortical damage inflicted by nematodes facilitates invasion by secondary fungal pathogens.

The maxim “where a plant can thrive, a nematode can attack it” is generally applicable. Under favorable conditions in the tropics many species have short life cycles, with several generations possible per season, leading to rapid population build up. Nematode population densities are negatively affected by flooding for a prolonged period of time and by tillage of soil during hot and dry seasons, but are positively affected by high fertility soils. In the absence of banana, nematodes survive on alternate hosts. *R. similis* and *P. coffeae* both have a wide host range, more than 250 species, including citrus, black pepper, coconuts, tea, coffee, ginger and curcuma. They are serious commercial pests on numerous tropical and subtropical crops. *Helicotylenchus* spp. and *Meloidogyne* spp. also have wide host ranges..Temperature preferences of nematodes are species-specific. For example, below 20°C, multiplication rates of *R. similis* are greatly reduced and *P. coffea* optimum life temperature ranges from 25 to 30°C.

Cultivar susceptibility

Although resistant cultivars are a cheap and practical solution for nematode control, only a few banana and plantain landraces show resistance for one or a few pathotypes of a nematode species. The ABB group and many diploids are more resistant which AAA and AAB plantains are more susceptible to nematode damage. Most resistance screening studies focus on the known damaging species *R. similis.* **Resistance sources** have been identified in land races, wild species and new synthetic hybrids from breeding programs. Some of the subspecies of *Musa acuminata* (ssp. *burmanicoides, malaccensis, microcarpa* and *zebrina*) have demonstrated moderate to good resistance to *R. similis* and Fe'i bananas 'Rimini' and 'Menei', some diploids and many accessions of *M. balbisiana* have also proven resistance to *R. similis* in the field. However, resistance to multiple species remains an elusive characteristic. In addition, resistance in different regions to the same species may not be maintained. The cultivar 'Yangambi Km5' (subgroup 'Ibota' AAA) has demonstrated resistance to *R. similis* and *Pratylenchus goodeyi*, although strains of *R. similis* in East Africa have overcome this resistance.

Will nematodes threaten Musa diversity - rare cultivars and wild relatives

Nematodes do not directly threaten the diversity of rare cultivars and wild relatives. While nematodes can build up and reduce production, they do not kill the mat and can be easily managed by simple practices which allow rural communities to continue to exchange planting material. Wild relatives found in disturbed vegetation in natural areas are well protected from the spread of nematodes.

Will nematodes cause reduction in diversity of cultivars which are commonly used

Plant parasitic nematodes may generate shift in the proportions of cultivars if new cultivars are released or introduced which are more resistant. Where high genetic diversity is seen, rural communities may unknowingly select for resistance or tolerance to nematodes by choosing suckers from more vigorous mats leading to some loss of other cultivar characteristics in the process.

Current understanding of genes and molecular mechanisms to explain cultivar resistance

Collaborator input still pending

How do nematodes spread?

Nematodes in the soil infect roots of the first planting material planted in a new field and continue to build up infecting progressively the roots of new suckers and stems formed in the mat from the soil matrix. The use of infested suckers is the most important source of nematode spread from field to field in banana growing regions, although contaminated soil can move through soil erosion into neighboring plots and farm machinery. Nematodes can also be moved in substrate used in nurseries for tissue culture plants or microcorms. All nematode species that affect bananas also survive on alternate host plants, although they cannot survive in the soil in absence of host tissue. Rotations to reduce nematode populations must be carefully planned.

Key management factors

The integrated management of nematodes should incorporate practices to reduce nematode population densities in the soil before establishing a new crop (i.e. crop rotation, fallowing, flooding, solarization, cover- or trap-crops, nematicide application) and practices to reduce nematode population densities on the planting material (i.e. using tissue-cultured plantlets, paring, hot-water or boiling-water treatment, nematicidal dips, endophyte enhancement of tissue-culture plantlets). In addition, once the crop is established practices should focus on building crop capacity to produce well in spite of nematode damage (i.e. mulching, fertilization, propping) and the use of direct control measures with nematicides if the above fail.

Overview of pest and pathogen characteristics related to yield losses and Musa diversity

The six currently most important threats to banana production and productivity vary in several important dimensions.

- Three of the organisms cause total yield loss mat by mat, eventually leading to yield collapse. For bacterial wilts this occurs primarily in the absence of appropriate crop management practices, while for Foc and BBTD, once a field is infected the spread of the disease can be slowed but not easily stopped. Re-organization of production practices and regular replanting may be called for. The other three organisms reduce yield across the field without threatening yield collapse of individual mats. While yield loss can be severe, total collapse is less frequent.
- For two of the organisms (BBTD and bacterial wilts), no immune cultivars are currently known. Certain cultivars have escape mechanisms for bacterial wilt. For the other four organisms sources of resistance in cultivars and in wild relatives are known.
- Clean planting material is important for five of the six organisms. For weevils, nematodes and bacterial wilts practices are available on farm to ensure low risk planting material even with fields with some presence of the organism. For BBTD and Foc, the use of planting material from infected fields represents a high risk, since suckers may have the disease present without visual symptoms. Sources of zero-risk planting material are highly recommended. For BLS, planting material does not play a central role in disease incidence.

Perhaps the most important difference among the six disease and pest organisms is their distribution (Table 6). Weevils, nematodes and BLS are widely distributed, except for a very few remaining pockets where they are not present. For example, Argentina and southern Brazil do not yet have BLS, while parts of Sudan are still free of weevils. However, as Table 6 shows, the three organisms are present in all 25 top producing countries.

However, for the other three diseases, fortunately they are still not global in their spread.

BBTD is found throughout Asia and in 12 countries in Africa. BBTD has recently spread further into Southern and West Africa. The major plantain growing areas of Cameroon, Nigeria, Ghana and Cote d'Ivoire are potentially under threat, while in East Africa BBTD is already present in Burundi, Rwanda and eastern DRC and actively spreading. It is also poised to invade Uganda and Tanzania.

For Foc, Races 1 and 2 are already widespread, while Foc TR4 has been spreading at a steady pace since it was first detected in 1967 in Taiwan. From 1967 to 1978 the disease increased from a single plant to 1500 hectares in spite of rigorous quarantine measures. It is now present in 10 countries in Asia and actively spreading within countries. Three new countries outside of Asia have also reported the disease presence which has generated global concern for the potential for escalating spread and loss in production and infested soils. Jordan and Oman where the disease has been verified are quite small and somewhat isolated from global banana networks raising questions about how the disease reached there, but less concern about a springboard to other major growing areas. However, the disease was also found in a recently established export Cavendish zone in Mozambique which poses a growing threat for spread into the rest of East and Southern Africa. The zone is also linked to other Cavendish growing areas in Latin America and Asia through technical advisors and field staff, raising the possibility of disease transmission through soil on shoes of banana experts. An international alert has been issued to address this issue (http://banana-networks.org/musalac/files/2013/06/RECOMMENDATIONS-for-travellers_Fusarium_09062011_English.pdf).

The bacterial wilts represent a different situation. Bacterial wilts are present in all three continents, but are two different species with the potential of different strains in each species. Xcm in East Africa has spread recently from two initial points in Uganda and eastern Congo to cause widespread losses throughout East and Great Lakes Central Africa. The spread continues into the Congo basin. The distribution of bacterial wilts in Asia is restricted to only three countries, but represents a latent risk of more widespread damage. In Latin America Moko is present in many countries, but not in all areas within each country. Internal quarantine procedures are generally not in place to minimize internal spread.

To highlight the potential threat of pests and pathogens to Musa diversity Table 7 summarizes their presence in the centers of diversity of banana cultivars and of wild Musa relatives. Four of the six organisms are found in all the centers of diversity (weevils, nematodes, BLS, and BBTD), while Foc TR4 and bacterial wilts are only present in certain centers.

The potential losses from these diseases and their threat to Musa diversity will be discussed in the next section.

Table 6: presence of banana diseases by 25 countries

Countries with production >1000000t	**FOC 1-2**	**Foc TR4**	**Nematodes**	**Weevils**	**BLS**	**BBTD**	**Xcm**	**Moko**	**Blood disease and Bugtok**
East Africa									
Burundi	yes	No	Yes	yes	Yes	Yes	yes	no	no
Uganda	yes	No	Yes	yes	Yes	Yes	yes	no	no
Kenya	yes	No	Yes	yes	Yes	Yes	yes	no	no
Rwanda	yes	No	Yes	yes	Yes	Yes	yes	no	no
Tanzania	yes	No	Yes	yes	Yes	Yes	yes	no	no
West and Central Africa									
Cameroon	no?	No	Yes	yes	Yes	Yes	yes	no	no
DR Congo	no?	No	Yes	yes	Yes	Yes	yes	no	no
Côte d'Ivoire	no?	No	Yes	yes	Yes	No	no	no	no
Ghana	no?	No	Yes	yes	Yes	No	no	no	no
Nigeria	no?	No	Yes	yes	Yes	Yes	no	no	no
North Africa									
Egypt	no?	No	Yes	yes	No	Yes	no	no	no
Central America									
Costa Rica	yes	No	Yes	yes	Yes	No	no	yes	no
Guatemala	yes	No	Yes	yes	Yes	No	no	yes	no
Mexico	yes	No	Yes	yes	Yes	No	no	yes	no
South America									
Brazil	yes	No	Yes	yes	Yes	No	no	yes	no
Colombia	yes	No	Yes	yes	Yes	No	no	yes	no

Ecuador	yes	No	Yes	yes	Yes	No	no	yes	no
Peru	yes	No	Yes	yes	Yes	No	no	yes	no
Carribean									
Dominican Republic	yes	No	Yes	yes	Yes	No	no	no	no
Asia									
China	yes	Yes	Yes	yes	Yes	Yes	no	no	no
India	yes	Yes	Yes	yes	Yes	Yes	no	no	no
Indonesia	yes	Yes	Yes	yes	Yes	Yes	no	no	yes
Philippines	yes	Yes	Yes	yes	Yes	Yes	no	no	yes
Thailand	yes	No	Yes	yes	Yes	Yes	no	no	no
Vietnam	yes	Yes	Yes	yes	Yes	Yes	no	no	no

Table 7: Which centers of banana diversity are threatened by diseases?

Centers of diversity of banana	**BBTD**	**Foc TR4**	**weevils**	**nematodes**	**Bacterial wilts**	**BLS**
AAB plantain Congo	Yes	No	Yes	Yes	Yes	Yes
EAH AAA	Yes	No	Yes	Yes	Yes	Yes
India – AB, ABB	Yes	Yes	Yes	Yes	??	Yes
Philippines, Malaysia, Indonesia – AA, AAA, ABB	Yes	Yes	Yes	Yes	Yes	Yes
East African coastal diploids	No	No	Yes	Yes	No	Yes
PNG – Diploids	Yes	No	Yes	Yes	No	Yes
South Pacific – AAB	Yes	No	Yes	Yes	No	Yes
Fe'i (S) Pacific Islands	Yes	No	Yes	Yes	No	Yes
Eumusa (A, B, S) South-east Asia	Yes	No	Yes	Yes	No	Yes
Rhodochlamys – Myanmar and China	Yes/No	Yes/No	Yes	Yes	No	Yes
Australimusa (T) south east Indonesia, southern Philippines to Melanesia	Yes	Yes/No	Yes	Yes	Yes	Yes
Callimusa South Vietnam, Peninsular Malaysia, Borneo, Sumatra	Yes	Yes/No	Yes	Yes	Yes	Yes

5. The threat of pests and pathogens to banana production and diversity

During the period 2012-2015 the CGIAR Consortium Research Program on Roots, Tubers and Bananas carried out a priority assessment exercise of the returns to investment in alternative research lines responding to important constraints identified by banana stakeholders (Pemsl & Staver. 2014). An essential step in the calculation of the economic returns from research investments is an estimation of losses to the spread of disease as well as other opportunities to improve returns to banana growers. The estimation of losses proved to be a serious challenge to the banana research community for four major reasons:

1. While the planting areas of large monocrop bananas are relatively easy to quantify and locate geographically, smallholder production systems often make up a minor land use on the landscape and are distributed in very different ways spatially depending on the production system, the markets and natural resources. The result is that statistics even at the national scale are approximate and the quality and availability of data on within country distribution are highly variable.

2. Banana production systems have highly variable densities of banana from over 2500 mats/ha in annually replanted monocrops to only 300 mats/ha as a secondary crop in perennial agroforestry coffee. While production statistics are somewhat easier to obtain for monocrops, mixed cropping with bananas represents a challenge to crop statistics and for efforts to map banana production areas.
3. The different banana cultivar groups and cultivars with groups have quite different production challenges, including susceptibility to pest and pathogen losses. An estimation of losses requires some detail by cultivars grown.
4. Half of the priority pests and diseases are not yet global and are still spreading. The factors in the rate of spread and vulnerability of different cultivars and production systems are still under study. Often the rate of spread may depend on the degree of connectivity of banana growing areas, although such information is still largely incipient.

The study addressed these four challenges by focusing on national level statistics, although a website to map banana within country by cultivar group and production system has been set up (http://www.crop-mapper.org/banana/). Cross referencing the data bases from Fruitrop (production by cultivar group) and FAOStat (production area, yield/ha) provided an estimate of current production parameters which could be used to estimate both gains in yield as well as recovery from losses or avoided losses from the spread of pests and diseases. Data in Table 3 are taken from this study.

The projected yield losses used in the study are shown in Table 8. For each estimate, different target countries were selected based on banana production and the current spread of the disease.

The spread of BBTD is projected to affect over 8 million rural households producing 1.5 million hectares of bananas in West, Central, Eastern and Southern Africa and Asia by the year 2039. The disease is a threat to Latin America, but was not projected to spread there in the 25 year time span of the study. In the absence of major investments in BBTV-free planting material or immune cultivars, these areas will remain out of production and the disease will continue to spread. The only production system relatively unaffected is the commercial monocrop, either national or export markets, with access to clean planting material.

The spread of Foc TR4 is projected to affect upwards of 6 million households producing 1.2 million hectares on all continents. The disease is currently not in Latin America, but given the global linkages around export banana in technical advisors, containers, planting materials and inputs and in other horticultural activities, losses are also projected for Latin America. Initially production can be shifted to soils which are uninfected which may offset losses. This was not taken into account in the projections, although the shift of production into new areas is well documented in China, for example, where production has shifted from Guangdong and Hainan to Yunnan and into the northern regions of Vietnam, Laos and Myanmar. However, with time the availability of clean soils suitable for banana production will decline. Commercial monocrops will be highly affected. The impact on smallholders will depend on both the cultivar group which they produce and their production system.

For bacterial wilts losses were only projected for Xanthomonas bacterial wilt in East, Great Lakes and Central Africa (Table 8). The disease is already widespread and is spreading from the highlands of Eastern Congo into the lowlands. The disease is projected to affect 10.5 million households cultivating over 2 million hectares with around 60% yield loss. Losses were not projected for the other bacterial wilts in Latin America and Asia which are locally important in specific cultivars, but not to the same extent as Xcm in Africa.

Table 8: Projected losses due to pests and diseases by the end of 25 years

	WCA[1]			**ESA**[2]			**APO**[3]			**LAC**[4]		
	% yield loss	000 ha affected	# households	% yield loss	000 ha affected	# households	% yield loss	000 ha affected	# Households	% yield loss	000 ha affected	# households
BBTD all cultivars	51	612	3 063 323	30	787	3 934 027	27	203	1 012 908	0	0	0
FOC TR4 all cultivars	11	156	778 544	15	273	1 365 749	35	786	3 936 405	3	49	87 357
XBW all cultivars	60	488	2 439 022	59	1850	9 249 199	0	0	0	0	0	0
Weevil Nematodes BLS in plantains[5]	41	1 709	6 749 433	N/A	N/A	N/A	41	1 858	866 667	41	1 514	655 449
Weevils, nematodes, BLS in EAH AAA[6]	39	185	163 889	39	3 408	10376777	0		N/A	0		N/A

[1] Angola, Benin, Cameroon, Central Africa Republic, Congo, DRC, Cote d'Ivoire, Equatorial Guinea, Gabon, Ghana, Liberia, Nigeria

[2] Burundi, Ethiopia, Kenya, Malawi, Mozambique, Rwanda, South Sudan, Tanzania, Uganda, Zambia, Zimbabwe

[3] China, India, Indonesia, Malaysia, Myanmar, Pakistan, PNG, Philippines, Sri Lanka, Thailand, Vietnam

[4] Brazil, Colombia, Costa Rica, Ecuador, Guatemala, Honduras, Mexico, Nicaragua, Panama, Peru, Venezuela

[5] WCA Cote d'Ivoire, Ghana, Cameroon, Nigeria, DRC, Congo, Gabon, APO India, LAC Brazil, Colombia, Costa Rica, Ecuador, Panama, Honduras, Venezuela, Nicaragua. Mexico

[6] WCA Cameroon, ESA Uganda, Rwanda, Tanzania, DRC, Burundi

Nematodes, weevils and BLS are already present globally with yield losses highly variable depending on cultivar and production system. All growers affected by these problems continue to harvest their existing fields and can readily plant new fields in the presence of these problems. This is in sharp contrast to BBTD and Foc which not only collapse production in existing fields, but make difficult or impossible the re-establishment of banana in the same areas. In the priority assessment exercise the calculations of yield gains from new cultivars resistant to these three pest problems provide an approximate estimate of losses for two cultivar groups. For plantains 4 million hectares are affected year after year by this complex of problems with yield losses of 40%, while for East African Highland AAA, the area affected is around 3.5 million hectares with a slightly lower estimated yield loss. This analysis does not address the challenge of BLS in export banana, since the priority assessment exercise was directed at the role of RTB research investments in smallholder agriculture. While both the costs of control and the environmental costs of repeated fungicide applications are high resulting from BLS management, a research alternative directed to this point was not taken into account.

A comparison across these pest and disease problems should be done with caution, since the loss calculations were done to calculate the returns to specific research options. As a general conclusion, losses for all six of the pests and diseases of bananas are substantial and affect millions of households. More specifically, the losses to BBTD and Foc TR4 can be expected to continue to increase as the problem moves into new areas where it is already currently present and into areas where it is currently absent. While the bacterial wilts also can be expected to spread, the yield losses may not reach the levels of the East African Highlands. For weevils, nematodes and BLS, total hectares affected are higher, since all growers suffer some losses, but continue to harvest and replant the crop. For BBTD and Foc TR4, yield collapse is total for those affected. While recovery is possible, this depends on new production systems and the strengthening of off-farm services. In the mean time, more and more households are affected as the organism spreads and more and more lands and communities are affected.

Effects on food security and household income in terms of projections are a combination of the importance of bananas (per capita consumption for the most part as shown in Table 3) and the distribution of the organisms (Table 6). Burundi, Democratic Republic of Congo and Rwanda where bananas are a very important food staple have already been suffering severe losses from two of the three organisms – bacterial wilts and BBTD - with further losses projected. Other counties like Uganda, Tanzania and Malawi may soon have a similar situation as both BBTD and bacterial wilts continue to spread. Foc TR4 is also present in the region in northern Mozambique, but the rate of spread is still to be seen. The spread of BBTD further in Central and West Africa which are still free of the bacterial and Foc wilts will also generate severe dislocation of consumption and land use patterns due to its impact on multiple cultivars. In Asia both BBTD and Foc TR4 are affecting countries like China, Philippines, Indonesia and Vietnam. In all these countries the loss of banana production systems can also be projected to have environmental consequences with a shift to clean tilled annual crops.

The data presented up to this point in the report provide the basis to examine the effect of pests and diseases on banana diversity. While the lack of diversity has been proposed as a reason for the vulnerability of bananas pests and diseases, the presence and spread of pests and disease may also threaten two different dimensions of diversity. First, pests and diseases when present and spreading in the centers of banana cultivar diversity and wild Musa relatives may lead to the permanent disappearance of rare and as yet uncollected cultivars and wild Musa species with a very limited distribution. The summary of the mechanisms for yield loss and spread provide initial information to evaluate whether their presence will lead to the loss of rare cultivar diversity. Only one of the six organisms under study appears to present that risk (Table 9). The banana aphid spreads BBTD from mat to mat leading to complete mat destruction and can prosper in back yard gardens and village fields where rare cultivars in centers of cultivar diversity are likely to be found. These include islands of the South Pacific, the Congo basin and Papua New Guinea. For wild Musa relatives other factors may represent greater threat. Deforestation and loss of natural habitat has been identified as an important factor threatening these species.

Table 9: Pest and disease threats to diversity

Disease	Threat to diversity of rare cultivars and wild Musa relatives	Threat to diversity of commonly used cultivars
BBTD	**Yes:** Congo, South Pacific, PNG, Particularly problematic in smallholder production and transmitted into wild bananas in natural settings. Dangerous in areas where rare cultivars are yet uncollected.	**Yes:** where the disease has spread unchecked by clean seed based recovery efforts, the only cultivars remaining are hardier ABB. In zones with sources of BBTD-free planting material, there may be a restricted group of cultivars available with a loss of minor cultivars which have lower demand.
Foc	**No:** although Foc can kill whole mats, rarest varieties are often found in the bush or backyard gardens and Fusarium doesn't reach those places as readily.	**Yes:** once a soil becomes infested, susceptible cultivars cannot easily be grown in the field and many farmers opt for a change to resistant cultivars. New resistant cultivar introduction may offset cultivar loss.
BW	**No:** although BW may generate pressure on rare cultivars which are found unattended in backyard gardens and semi-wild in areas of high cultivar diversity of ABBs, simple management including the harvesting of the male bud which is often edible serves to reduce the threat.	**Yes:** ABB types have been associated with outbreaks of BW at epidemic levels and can thus selectively eliminate these cultivars if unmanaged. Farmers can also potentially select against susceptible cultivars. The presence of both Foc and BW leads to a dramatic reduction of ABBs among smallholders.
BLS	**No:** mats continue to produce in spite of severe disease levels and recover when drier conditions prevail.	**No:** cultivars which have particular uses or functions will be maintained, even if in lower percentages when resistant varieties are released.
Weevil	**No:** mats may be weakened but are not killed by weevils and clean planting material is relatively easy to obtain on farm	**No:** weevils alone are generally not the major factor in shifting from a more susceptible to a more resistant cultivar.
Nematodes	**No:** mats are not killed and clean planting material is relatively easy to obtain on farm. Wild relatives are well protected.	**No:** may generate some shift in the proportions of cultivars if new cultivars are released or introduced which are more resistant

A second dimension of diversity affected by pests and diseases is the diversity of commonly used cultivars either for market or for home consumption (Table 9). Our preliminary analysis suggests that the pests and diseases which result in some yield loss without threatening mat and field collapse will generally not affect the diversity of commonly used cultivars. However, BBTD, Foc and the bacterial wilts can be responsible for major shifts in cultivars used and loss of diversity.

- If BBTD spreads in the absence of recovery programs only somewhat resistant ABB cultivars will remain producing substandard bunches. Recovery programs focusing on BBTV-free planting material may neglect the full spectrum of cultivars present before BBTD outbreak to focus only on cultivars preferred by the market resulting in a decline in cultivar diversity.
- For Foc races 1 and 2, susceptible cultivars become increasingly scarce as more soils become infected. The introduction of resistant cultivars may compensate production and diversity loss. The full impact of Foc TR4 has not yet played out in areas where the disease is recent. In Taiwan susceptible Cavendish has been replaced by resistant GCT materials. However, Taiwan is not a

country of high cultivar diversity. In the Philippines and Indonesia where cultivar diversity is higher, the relatively slow movement of Foc TR4 out of commercial fields and into smallholder production areas and the buildup of TR4 infected soils merits monitoring.
- The impact of bacterial wilts on the diversity of common grown cultivars appears less severe, although the ABBs which are more susceptible to bacterial wilt have declined in diverse regions in Africa and Latin America.

6. Methods to use diversity to reduce losses to pests and diseases

The losses due to pests and pathogens can be addressed through management practices of different sorts. Direct control measure may include practices to ensure clean planting material, traps, agronomic practices to reduce disease risk and severity, and the use of chemical and biological control practices. Here we will focus on a subset of practice which taps the use Musa diversity. In this section we will briefly describe seven different approaches, while in the following section we will evaluate the applicability of the approaches to the six target pest and pathogens. This will provide us with inputs to sketch out the future priority research areas to promote Musa diversity to address pest and disease losses.

The seven methods considered here are summarized in Table 10. For each approach the table summarizes first how the method uses Musa diversity. Two of the methods make use of cultivar diversity directly either through the substitution of resistant land race cultivars for susceptible cultivars or through the mixture of more susceptible cultivars with more resistant cultivars to reduce losses. Three methods make use of diversity found in both cultivars and wild Musa relatives indirectly by incorporating specific sources of resistance – conventional breeding, cisgenesis and manipulation of the plant microbiome. Mutagenesis generates new diversity within preferred cultivars with radiation, while clonal selection focuses rigorous selection and multiplication of preferred cultivars under conditions in which minor differences in preferred traits can be detected and selected for. Finally, transgenesis which is classed together with cisgenesis as GMOs uses genetic material from outside the Musa group. Table 10 also summarizes the key steps in the process and the role of emerging advanced science in making the method more effective. The different approaches make use of different areas of emerging science from genome sequencing, genetic modification and gene editing, diverse methods to study the plant microbiome and ecological modeling of mixed populations. Several of the methods use high throughput phenotyping to screen large populations for useful traits. Finally Table 10 documents two steps in technology readiness for each method – the existence of advanced prototypes ready for use and applications in use on-farm. In summary, four of the methods have already generated results for use on-farm:
- Additional cultivar substitution following the model used of Cavendish substitution for the Fusarium susceptible Gros Michel
- Cultivar mixtures which are used by smallholders, although uncommonly on more commercial
- Conventional breeding with numerous cultivars in use
- Clonal selection which was used to generate Foc TR 4 resistance in Cavendish cultivars

Other methods have advanced prototypes or partial applications of the approach:
- Trans GMOs are under test or waiting for regulatory validation in Ecuador, Uganda and Australia addressing BLS, Foc and BBTD
- Endophytes applied for control of nematodes, weevils and Foc, although are not yet linked to the development of cultivars which are bred or selected for their specific capacity to recruit a microbial community

- Numerous conventional breeding programs which continue to breed for resistance to BLS, weevils, nematodes and Foc
- Clonal selection procedures in use in numerous sites where Foc resistant GCT clones are planted at a commercial scale.

7. Applicability of methods to use diversity to reduce losses from six pests and diseases

In this section we examine the use of each method to address the losses from the six different organisms under consideration. Seven tables summarize our analysis of the applicability of each of the methods to use diversity. Each method is rated using a 1-4 scoring system as follows:

1) Very low to no applicability to the reduction of losses due to inherent properties of diversity and demands of method
2) Some applicability to reduction of losses, although uncertain chances of success, due to limitations in diversity or method
3) Promising applicability, although limited by availability diversity
4) High applicability with high probability of success based on application of existing principles and available diversity and resulting in wide cultivar success

Following the score, we summarize the reasons for the score. If the score is 1-2, then the reasons focus on why the method does not appear applicable. If the score is 3-4, then the reasons address priority research to move towards workable alternatives. The tables also summarize the status of progress towards on-farm use with references.

For ease of comparison Table 17 at the end of the section presents the scores and the progress for the six pests and diseases and the status of progress. From this summary table we observe that BLS, Foc, nematodes and weevils merited higher scores for more methods with multiple methods offering applicability and for cultivars from multiple groups. BW showed a less limited range of options, while BBTD generally did not have scores above 2 for any method except GMOs (primarily transgenesis).

From a review across the methods we observe:

- Cultivar substitution offers some applicability although cultivar choice may be limited; especially if a specific market or consumer is targeted. Research question pending: Is there as yet uncharacterized useful diversity in centers of cultivar diversity, how extensive is within cultivar variability in land races and what approaches can be tapped to capitalize on this diversity and variability?
- Cultivar mixtures are more applicable for three of the organisms (BLS, nematodes, weevils), less applicable for BW and Foc and not at all applicable for BBTD. Research question pending: what are the mechanisms by which the cultivar mixture ensures higher yields of the more susceptible cultivar and how does that contribute to the overall viability of the cropping system? What percentage of each cultivar and planted in what arrangement is needed for successful mixtures?
- Conventional breeding already has a base of released cultivars, pre-breeding lines and wild Musa with resistance and new breeding strategies with applicability for BLS, weevils, nematodes and Foc. Sources of resistance are less certain for BW and BBTD. Research question pending: will the new breeding schemes, marker assistance and the strategies to minimize BSV interference result in consumer-acceptable fruits with adequate post-harvest characteristics?
- GMOs rate relatively high scores tapping on both trans and cis sources of resistance for all six target organisms. Regulatory frameworks and consumer acceptability are major issues. Research question pending: will the rapidly advancing knowledge across crops find applicability in

bananas? Can the experience gained from relatively quicker advance with transgenes be harnessed into cisgenesis?

- Mutagenesis was generally rated as uncertain with few cases of progress, although cultivars with shorter cycles or improved bunch formation are in use on-farm. Research question pending: Will the random generation of mutations followed by more effective screening lead to the development of useful cultivars more efficiently than GMOs and conventional breeding using advanced bioinformatics?
- The scoring for clonal selection is contradictory with a high score for Foc based on successful cultivar development and quite low scores for the other target organisms. Research question pending: How can selection procedures be developed to accelerate and miniaturize clonal selection processes, since field scale strategies present serious logistical challenges?
- Cultivar-specific microbes or cultivars with heightened capacity to recruit microbes were rated as applicable for BLS, Foc, nematodes and weevils. The identification of effective endophytes is the basis for this scoring, although with pending questions about whether the microbe – cultivar relation is highly specific. Research question pending: Will designer microbes or broad acting microbes be more viable for practical use in banana production?

Table 10: comparative features of different methods

	How does approach use Musa diversity	Key steps in process with approximate time	Areas of emerging advanced science linked to more effective use of diversity	Advanced prototypes under testing	On-farm results in use	Pest and disease applicability
Cultivar Substitution	Cultivars susceptible to a pest or pathogen are replaced by an existing land race cultivar with similar use traits which has resistance	(1) Establish standard screening procedures and reporting platforms (2) Screen current cultivar collections worldwide for resistance characteristics, fruit quality and relevant agronomic traits (3) Develop crowd sourcing tools to identify plants with unique resistance traits within cultivars in centres of diversity (4) Local testing of replacement of a susceptible cultivar with resistant cultivar with similar fruit properties	- Predictive tools based on population genetics to identify potential sources of existing resistance for different cultivars phenotyping procedures to screen for resistance in promising populations. - Use of specific alleles linked to given traits to develop markers that will allow the early detection of the desired trait through molecular screening - application to large collections or within cultivar diversity	dwarf Pisang Awak resistant to Foc R1	Dwarf cultivars for tall cultivars Export: Cavendish for Gros Michel (Foc) Nicaragua: Pelipita for bluggoe (BW/Foc) India: tolerant dwarf Kluai Namwa for Pisang Awak (Foc)	applicable when resistance is found in Musa land races
Cultivar Mixtures	Planting susceptible cultivars among resistant cultivars to slow the spread of the disease and to reduce the yield impact through effect of: barrier, dilution, diversion.	(1) participatory diagnostics of local use of cultivars and pest/pathogen susceptibility (2) household and field surveys to collect information from farmers on crop varietal diversity and disease management practices; (3) studies of pest/pathogen dynamics in resistant and susceptible cultivars (4) field testing of combinations of resistant and susceptible cultivars - pest dynamics and yield (5) participatory trials to develop applied practices building on principles of mixtures	- Use of models to test interactions based on different proportions and arrangements for specific pests and diseases and for combinations of pests and diseases. - Improved understanding of important factors in pest and disease dynamics - improved characterization of resistance mechanisms in different cultivars and their effect on dynamics of mixtures	models for mixed cultivars against nematode	documentation of disease reduction effects of current smallholder mixed cultivar practices for BLS, weevils, nematodes	BLS, weevils, nematodes

Conventional Breeding	Resistance found in wild relatives, diploid cultivars, diploid pre-breeding lines or tetraploids integrated into popular triploid cultivars to generate new triploids with acceptable fruit and agronomic traits and additional resistance	(1) identification of resistance genes and proposed cultivars for improvement (2) development of breeding strategy to be used to generate final triploid (3) hand fertilization of flowers from parents, seed harvest, embryo rescue from collected seeds, screening of resulting plants for resistance (4) field trials to select desirable agronomic and fruit traits (5) multi-locational trials comparing final cultivar candidates with farmers	- Genomics as a basis to understand organization and dynamics of Musa genomes that may affect segregation of breeding lines. - Improved markers to assist crossing strategies - High throughput phenotyping to screen resistance in crosses	all active breeding programs have candidate cultivars under evaluation	FHIAs IITA PITAs CIRAD Cavendish CARBAP IITA/NARO Naritas NARO Nabios Embrapa AAB cultivars	highly applicable for pest and disease resistance
GMO	Direct introduction of genes to create resistance in target cultivar without modification of other characteristics. Cis genesis when genes originate from another species which is sexually compatible (in this case Musa and relatives) and transgenesis from a non-sexually compatible plant or a non-plant organism	(1) development of reliable embryonic cell suspension procedures for target cultivar(2) For cisgenesis, identification of resistance gene from banana source plant which is inserted into susceptible target cultivar cells using AgrobacteriumFor transgenesis, identification of gene for insertion with defense effect on invading disease or pest from non-Musa organism which is inserted into single cells(3) laboratory screening for successfully transformed cells(4) greenhouse testing for appropriate disease or pest resistance(5) field testing for agronomic performance and resistance(6) multi-locational trials to test performance in different environments	- Next generation sequencing (NGS) to boost gene discovery at the whole genome scale that can support trans/cisgenics) - Gene editing technique (e.g. CRISPR/Cas9) to target very specific changes in genes	NARO: plantain and EAH AAA cultivar with weevil and nematode resistanceBBTD resistance: Queensland, HawaiiBW: IITA EAH AAA	none pending regulatory changes permitting release	highly applicable for pest and disease resistance

Mutation Breeding	Mutagenic agents such as radiation and certain chemicals used in tissue culture to induce mutations in existing cultivars at a higher frequency and generate genetic variation from which desirable changes can be selected	(1) At least 2000 shoot tips (or ideally Embryogenic Cell Suspensions -ECS- to avoid chimerism) from target cultivar exposed to dose of gamma radiation to generate initial mutations (2) resulting plants subjected to additional rounds of in vitro multiplication which serve to fix or stabilize initial mutations into complete plant tissue (this step can be avoided by using ECS) (3) Appropriate greenhouse or field evaluation of resulting plants to select for desired trait (4) Evaluation of plants with desired traits for genetic confirmation and agronomic characteristics (5) Multilocation trials to assess performance under different environments	- Next generation sequencing (NGS) more affordable for exploration of genome to trace back the origin of the mutation affecting an observed phenotype. - High throughput phenotyping to screen many, many lines through multiple stages of selection	new project underway, but results pending	Novaria - early flowering Novaria A - Foc tolerance Dwarf Parfitt - Foc tolerance LK-40 dwarfness LT-3 - big bunch size	more commonly used for agronomic traits like earliness, bunch size and shape or dwarfness, but selections for BLS have been made and a project is underway for Foc resistance
Clonal Selection	Based on selection of within cultivar diversity and possible mutations occurring during multiplication, thereby increasing the usefulness, productivity or local adaptability of existing cultivars	(1) Initial selection of plants showing resistance or other exceptional traits under field conditions surrounded by other highly infected or other less productive plants(2) For Foc plants TC multiplied and then screened in densely planted field with severe infestation 500-2,000 propagules per gram of soil. After 3 months, survivors with rhizome section not infected are designated as a somaclonal variant for multiplication for distribution to farmers - new selection of more resistant plants surrounded by infected plants(3) for production traits, selection from new fields planted with material from selected plants to generate new planting material from superior plants	- High throughput phenotyping screening for resistance to identify elite lines	clonal selection ongoing in new locations where GCTs introduced	locally adapted cultivars in Canary IslandsTR4 resistant GCTCVs in widespread use in Taiwan, China and Philippines.	untested applicability in other pests and diseases, although successful in Foc

Cultivar-specific Microbes	cultivar-specific capacity to associate with endophytes to bestow resistance to biotic or abiotic stress thereby harassing diversity in plant microbiome to extend the usefulness of existing diversity	(1) screen banana cultivars and wild relatives for microbial populations (2) laboratory and greenhouse tests for effectiveness of microbes against biotic and abiotic stress for single microbes and in combinations (3) Validate capacity of cultivars to associate with microbes (4) develop efficient inoculation technologies for different production systems	- Metagenomic analysis linked to soil microbiology and plant microbiome - Metatranscriptomics and cDNA amplicons which provide information on activity and function of the communities - Molecular and cellular mechanisms for action by endophytes - Single-strain genomics and transcriptomics to isolate plant-single microbe interactions	endophytes identified for Foc, nematodes, weevils, growth stimulant, but cultivar specificity unexplored	none which are cultivar specific	endophytes identified for Foc, BLS, nematodes, weevils

Table 11: Applicability of methods for reducing losses from BBTD

Method	**BBTD**			
	Score	**Reasons**	**Progress**	**Prototypes and References**
Cultivar Substitution	2	No known immune cultivars, only cultivars slow to show symptoms which survive, but with stagnating sub-optimal yield and often difficult to eradicate in case of renovation with clean planting material.	Field ready, but very little cultivar choice	Saba reported to be least affected by BBTD, although virus infected. Cultivar identification which are slow to show symptoms, although yield declines are common, even if not plant death. Gros Michel (AAA) and Pisang Awak (ABB) (Kumar et al., 2013). 'Kayinja', (ABB), 'FHIA-03' (AABB), 'Prata' (AAB), 'Gisandugu' (ABB), 'Pisang Awak' (ABB), 'Saba' (ABB) and 'Highgate' (AAA, Gros Michel subgroup) (Niyongere et al., 2011).
Conventional Breeding	2	No currently known source of resistance. Possible complex genetic control of virus-vector-host plant to achieve total immunity	Source of resistance not identified	No current breeding for BBTD resistance (most references). Butuhan (BB) and Khae Phrae (AA) found completely immune to BBTV infection. If verified, may serve as a useful source of native banana genes for BBTV resistance (Mware, 2016). Wild balbisiana behaved immune recent study PCAARRD
GMOs	3	Needed: field testing, determination of titre levels long term field tests and application to a greater number of cultivars.	Transgenesis proofs of concept under development	- Successful agrobacterium-mediated transformation of embryogenic banana cell suspensions with constructs that may prevent the replication of BBTV, in lab: - Transformed clones of 'Dwarf Brazilian' (AAB, Pome subgroup) in Hawaii (Borth et al., 2011); - Transformed 'Rasthali' (AAB genome, syn. 'Silk') (Shekhawat et al. 2012); - Transformed Indian hill banana 'Virupakshi' (AAB genome, Pome subgroup) (Elayabalan et al., 2013). - 20 putatively resistant lines currently on field experiment in Hawaii (Kumar et al., 2013). - Cavendish types in Malawi (Queensland University)
Mutation Breeding	1	mutation stability in doubt	Initial proof of concept under development	Mutation in tissue-cultured banana cv 'Lakatan' plantlets was carried out in Philippines with Cobalt 60 gamma irradiation. 29 lines were found highly resistant to BBTV (Damasco et al., 2005).
Clonal or mass selection	1	No evidence for within cultivar variability. Gradual improvement not likely to lead to cultivar with zero virus titre and no infectivity	Initial proof of concept under development	Resistant somaclonal variant of banana cv. Lakatan field tested in Philippines: five out of the 1354 plantlets evaluated (0.36%) showed varying degree of resistance to BBTV, while yield and fruit characteristics remained comparable with the tissue culture control lines. One selected SV line ('SV 6-30') showed stability of BBTV resistance after field evaluation of the first, second and third generation plants (Damasco et al., 2005).

Cultivars mixtures	1	No known cultivar immunity and radius of movement of aphids limits mixture complementarity. Rate of decline of susceptible cultivar not greatly extended. Mixture becomes monoculture of resistant cultivar	No clear mechanism for effectiveness	High resistance of Saba, although not immunity, may have potential. Extremely low rate of transmission from Saba to interplanted susceptible cultivars would be key to possible success, although viable market for Saba also important for on-farm success
Cultivar-specific Microbes	1	Mechanism of action by endophytes likely to improve resistance but not bestow immunity	Initial proof of concept under development	Bio-formulations of mixtures of the rhizobacterial isolate Pseudomonas fluorescens (Pf1) and endophytic Bacillus spp. (EPB22) found to reduce the incidence of BBTV under green-house (80%) and field conditions (52%). Yield of inoculated plants also higher (53.33%) compared to the control plants (Harisha et al., 2009), although validation of these promising results not reported

Table 12: Applicability of methods for reducing losses from BLS

Method	**BLS**			
	Score	**Reasons**	**Progress**	**Prototypes and References**
Cultivar Substitution	3	Market acceptability. Field performance	Field ready, but not wide cultivar choice	Resistant varieties available in the existing germplasm. Some have been field tested and distributed. Example: large-scale replacement of plantains by BLS resistant ABB cooking banana 'Burro Cemsa' in Cuba (Lescot et al., 2009).
Conventional Breeding	4	Market acceptability. Resistance to other pests and diseases (ex. BSV for AAB). Unapplicable for some cultivars (no fertility)	Numerous hybrids on farm and in market involving processed products like chips and flour	IITA hybrids developed in Cameroon 'PITA 21', 'PITA 23' and 'FHIA 25' (triploid) and 'PITA 14', 'PITA 17', 'BITA 3' and 'FHIA 23' (tetraploid) were selected with input from farmers and exhibited higher levels of resistance to BLS, shorter growth cycle and higher bunch yield in comparison to the landraces (Njukwe et al., 2010). In Côte d'Ivoire tested BLS resistance in hybrids: 'FHIA-21' and 'BITA-3' can be distributed to farmers while 'FHIA-25' can be used as female parent in a breeding scheme (Kobenan et al., 2009). In Guadeloupe and Martinique a CIRAD 925 dessert banana hybrid under evaluation for export (Promusa 2016). In Cameroon, CARBAP and CIRAD plantain-like triploid hybrid under evaluation in field (Promusa 2016).
GMOs	3	Market acceptability. Resistance genes. Instability of the resistance in the few GMO obtained	Proof of concept, greenhouse	Potential of rice chitinase genes to enhance resistance against M. *fijiensis* in banana, lab demonstrated Kovács and Sági (2013). Transformation of Cavendish for BLS awaiting approval for testing: (Santos, 2014)
Mutation Breeding	2	Mutation stability. What genes to target	Proof of concept, greenhouse	13 clones selected for further evaluation due to BLS tolerance in in vitro mutation breeding in Costa Rica (Valerin et al., 1995) IAEA selected few clones (GN35-I to GN35-VIII) with gamma rays technique, which appear tolerant to the toxin Juglone (main toxic component of Mycosphaerella fijiensis), field tests pending (Roux et al., 2002).

Clonal or mass selection	1	Little documented within cultivar variability, although growers suggest certain variability	Little within cultivars variability as basis for selection	No documentation available
Cultivars mixtures	3	Market acceptability of more resistant cultivar, minimum % of resistant cultivar for useful disease reduction	Effect documented	Increased diversity of crop varieties, measured by number of varieties (richness) and their evenness of distribution, corresponded to a decrease in the average damage levels across sites and to a reduction of variance of disease damage in Uganda (Mulumba et al., 2012). Testimony from Bouganville, PNG, where fields with diploids only are more infected than fields where diploids are mixed with triploids. Experiments conducted in Nigeria with BLS resistant PITA cultivars mixed with local plantains (not referenced)
Cultivar-specific Microbes	3	More screening in different locations.	Endophytes with potential identified, although not cultivar specific	The strain RD_MAAMIA_05 delayed the appearance of the first leaf with necrotic symptoms (Marcano et al., 2016) endophyte collection for BLS reported in Colombia, but not verified

Table 13: Applicability of methods for reducing losses from FOC

Method	**FOC**			
	Score	**Reasons**	**Progress**	**Prototypes and References**
Cultivar Substitution	3	Characterization of TR4 resistance cultivars still incipient with potential for useful cultivars or lines	In use, but TR4 screening incomplete	Cavendish substituted for R1-susceptible Gros Michel for world trade production. Pelipita or Saba substitute for R2-susceptible Bluggoe dwarf Pisang Awak substitute for R-1 susceptible normal Pisang Awak
Conventional Breeding	3	Active breeding programs screening TR4 in breeding lines – major challenge is fruit quality for export or to meet local consumer preferences	In use	Replacement of "Prata Anã", susceptible to Foc race 1 with the variety "BRS Platina" and the replacement of "Maçã" variety (Apple), also susceptible to Foc race 1 with "BRS Princesa". Both varieties developed by EMBRAPA and successfully in use in Brazil (da Silva et al., 2008; EMBRAPA, 2016). FHIA hybrids resistant to Foc race 1 and TR4: FHIA-01, FHIA-18, FHIA-21 and FHIA- 25. FHIA-18 and FHIA-21 have been tested by Smith et al. (2014) along with SH-3640.10 and they resulted very productive in the Australian sub-tropics. FHIA-01 was released in Australia, but wasn't a success on the commercial market (Whiley, 1996). Many sources of resistance identified for use in breeding
GMOs	3	Numerous programs for TR4 resistant cultivars in China. Consumer acceptance challenges for marketing of GM bananas.	Green house	To date, experiments with GM banana plants have only been tested in vitro, plant growth chamber or green house. Results from Foc race 1 field are not yet available (Subramaniam et al., 2006; Paul et al., 2011; Ghag et al., 2012), while there are not TR4 field tests (Hu et al., 2013; Mahdavi et al., 2013; Yip et al., 2011). Active research programs in China and field trials in Australia
Mutation Breeding	2	Decreasing interest in mutation breeding. No clear results have been obtained in banana.	Initial proof of concept under development	Currently, Till (2016) are working on the development of a mutant population of approximately 10,000 lines by treatment of shoot tips with gamma irradiation and modifying sequencing techniques to recover large genomic indels caused by the treatment with the aim of obtaining banana plants resistant to Foc TR4. Australia dwarf Cavendish (ref)
Clonal or mass selection	3	Dynamic of inoculum in soil to determine need for replanting and use of other practices to reduce Foc disease pressure; Criteria for on-going selection for resistance; Applicability for other cultivars affected by Foc not widely tested	Cultivars in use with on-going clonal selection	Somaclonal mutants Giant Cavendish Tissue Culture Variants (GCTCV) clones are currently being tested in fields infested with Foc TR4 (Hwang & Ko, 2004). Commercial somaclones in use in Southeast Asia (Peng et al, 2013), although only partially resistant (replacement after 2 or 3 cycles) and other unfavourable agronomical traits (Ploetz, 2015, Hwang & Ko, 2004). commercial use and expansion of area under production of GCTs in Philippines and testing globally in monocrop Cavendish areas with TR4
Cultivars mixtures	2	Susceptible cultivars continue to be produced in smallholder mixed cropping, but mechanism	Proof of concept pending	Potential to use resistant cultivars as barrier crops along field borders to reduce risk of field to field spread or to be used into the field mixed with susceptible varieties to decrease the inoculum level in the soil Li et al. (2011).

		not clear. Escape due to planting in clean soil, slower buildup of inoculum or real suppressive effect of cultivar mixtures		
Cultivar-specific Microbes	3	Understanding of effectivity of current endophytes - specific to cultivars or even specific lines within cultivars. Delivery and durability of endophyte effect related practices for greater effectiveness	Endophytes with potential identified, although not cultivar specific	Populations of *Actinomycetes* (Peng et al., 1999), nonpathogenic F. *oxysporum* strains (Forsyth et al., 2006), *Chthonomonas, Pseudomonas* and *Tumebacillus* genera (Shen et al., 2015), *Bacillus, Rhizobium, Bhargavaea, Pseudolabrys,* and *Sinorhizobium* (Xue et al., 2015) linked to soils suppressive to Foc. The consecutive compost application in banana to suppress Foc changes the rhizosphere microbiome and effectively suppress Foc in fields infested with Foc TR4 in China. No current evidence of specificity of microbiome of banana plants to certain cultivars or lines and its role in Foc control.

Table 14: Applicability of methods for reducing losses from BW

Method	**BW**			
	Score	**Reasons**	**Progress**	**Prototypes and References**
Cultivar Substitution	3	Although no resistance to artificial inoculation, diverse escape mechanisms including retained flower bracts which greatly reduce disease dynamics. More thorough screening of escape mechanisms proposed for increased cultivar substitution	In use, although limited cultivar choice	ABB cultivar ‘Pelipita’ has filled the niche left by bluggoe which is susceptible to Moko. Both cultivars are highly resistant to most pests and diseases, although bluggoe is susceptible to Moko as well as Foc R2
Conventional Breeding	2	No current conventional breeding programs for bacterial wilts with no clear source of resistance yet identified	No resistance for use in conventional breeding	For XW disease, only resistance has been observed in Musa balbisiana a non-edible wild Musa spp. (Ssekiwoko et al., 2014; 2015). Resistance and tolerance towards Moko has been reported in some cultivars such as ‘Pelipita’ with retained flower bracts. Identification of Musa balbisiana genitors devoid of infectious endogenous Banana streak virus sequences may offer way forward.
GMOs	3	Trans GMOs for Xcm pending insertion into more market-preferred cultivars Potential to insert genes of resistance in the non-edible wild Musa balbisiana to the edible cultivars. Both regulatory	Proof of concept cultivar validated in field	Trans GMO breeding for XW resistance has been successfully achieved using genes from green pepper (Tripathi et al., 2010; 2014).

		procedures and market acceptability need to be addressed. Similarity of Moko, Bugtok and blood disease suggest applicability of cis- and trans-genes.		
Mutation Breeding	2	Basis for prediction of probability of success unexplored. Decline in use of approach for breeding	No programs breeding for bacterial wilts	No reports of mutation breeding against banana bacterial pathogens.
Clonal or mass selection	1	Within cultivar variability not studied for traits which may confer escape from infection - bract retention	Not under exploration	No known programs looking at improvement for bacterial wilts
Cultivars mixtures	2	Lack of resistance to BW, although potential to use cultivars with escape mechanisms. Proximity of resistant and susceptible cultivars may increase risk of tool mediated infections. For Moko which is more persistent in soils, mixtures unlikely to be effective.	Not under exploration	No known programs looking at improvement for bacterial wilts
Cultivar-specific Microbes	2	Studies to determine Xcm-suppressive microbiome are just beginning with no exploration of cultivar specificity	Initial screening of organisms and development of techniques	Three species of endophytic bacteria (Burkholderia spp., Herbaspirillum spp., and Enterobacter spp.) isolated from banana plants in XW endemic zones in Uganda have been found to suppress Xcm in in-vitro assays (Were, 2016).

Table 15: Applicability of methods for reducing losses from nematodes

Method	**Nematodes**			
	Score	**Reasons**	**Progress**	**Prototypes and References**
Cultivar Substitution	3	The replacement of susceptible cultivars by resistant or tolerant cultivars provides a sustainable solution for nematode related damage and yield losses.	In use, although limited cultivar choice	Nematode resistance one of several traits which contribute to cultivar uptake, but not determinant to change cultivar use. Speijer and Bosch (1996) and Kikulwe et al. (2007) report cultivar shifts in the Kagera region of Tanzania related to increased pest and disease pressure. Common use of Yangambi Km5 in East Africa.
Conventional Breeding	4	Multiple sources of resistance often associated with other disease resistance, although not a primary focus of breeding	Numerous cultivars on farm	Cultivar resistance to nematodes often increased in hybrids bred for BLS or Foc which have been major focus of breeding programs - FHIA, Embrapa, IITA, Cirad and Carbap hybrids. Several hybrids obtained through conventional breeding techniques show resistance to R. similis, including

		programs		diploid AA and tetraploid hybrids. However, only few have been tested in the field (Dochez, 2005). The resistance of 'Pisang Jari Buaya' to R. similis incorporated into the widely used diploid parent plant from the FHIA program, 'SH-3142' (AA genome; Pinochet and Rowe, 1979). 'Yangambi Km 5' partially resistant to both *R. similis* and *P. coffeae,* used as parent by Carbap for elite triploid hybrid resistant to nematodes (Promusa 2016)
GMOs	4	Resistance genes from many other crops (eg. maize, potato, tomato, beet, rice etc) show promise for GM bananas This approach is suitable for all cultivar groups.	Proof of concept developed with transfer to market cultivars pending regulatory approval	Two resistance mechanisms were harnessed to create transgenic plantains (Tripathi et al., 2015): a small protein inhibitor (cystatin) inhibits the nematode digestive enzyme cysteine proteinase (Atkinson et al., 2004; Roderick et al., 2012) and a synthetic peptide disrupts nematode chemoreception (Liu et al., 2005; Winter et al., 2002). Tripathi et al (2015) developed transgenes inserting maize-derived cystatin stacked with two synthetic peptides, or each separately (cystatin or peptide) into the plantain cultivar 'Gonja manjaya'. Several transgenic lines showed resistance against H. *multicinctus* and R. *similis* in the field, and the four best lines pending test to assess traits stability with plans to test against *Meloidogyne* spp and *Pratylenchus* spp.
Mutation Breeding	2	There are few reports on the success of mutation breeding for nematode control and it is not suitable for AAB.	No improvement programs using this approach	Kumar et al. (2012) identified two resistant mutants (from a Silk AAB and a Cavendish AAA) and one moderately resistant mutant (from a Cavendish AAA). Higher quantities of phenol, tannin, lignin identified in the roots of the mutant lines showing resistance
Clonal or mass selection	1	Clonal selection not studied to address nematode control. within cultivar variability to nematodes has not been studied	Not tested yet	no known reports of clonal selection being used to specifically improve resistance or tolerance to nematodes in the literature.
Cultivars mixtures	3	Field studies to test mixture percentages and arrangements for effective control	Well known	Quénéhervé et al., (2011) reported shifts in nematode community composition and population densities when cultivars with mixed susceptibility for R. *similis* and P. *coffeae* were grown together.
Cultivar-specific Microbes	3	Endophytic inoculation has delivered promising results, with reports of higher effectiveness for multi-species inoculations. Soils may also contain high antagonistic potential ("suppressive soils"). Cultivar specific endophytes or cultivar capacity to recruit endophytes not yet	endophytes identified and validated in field, although not cultivar specific	Mycorrhiza-induced resistance has been observed against R. similis in banana, due in part to mycorrhizal root exudates at the pre-infectional stage (i.e. attraction and penetration) of R. similis infection (Vos et al., 2012). Such mutualistic endophytes from roots inhabit the same niche as plant-parasitic nematodes (Sikora et al., 2008). Suppressive soils have been identified in commercial banana plantations (zum Felde et al., 2005), where antagonistic micro-organisms were isolated, including endophytic fungi and bacteria which demonstrate nematode-antagonistic activity (eg. Carñizares Monteros, 2003; Meneses Hernández, 2003; zum Felde, 2002). Multi-species endophytic inoculations

		studied		increase nematode control over individual fungal applications (zum Felde et al., 2006), with additional antagonists, fungal or bacterial, providing an additive protective effect (Mendoza and Sikora, 2009).

Table 16: Applicability of methods for reducing losses from weevils

Method	**Weevil**			
	Score	**Reasons**	**Progress**	**Prototypes and References**
Cultivar Substitution	3	Most resistant cultivars are not cooking types	In use, although limited cultivar choice	Many cultivars such as Yangambi- Km5 (AAA), Cavendish (AAA), Gros Michel (AAA), Kayinja (ABB), Ndiizi (AAB), and Kisubi (AB) appeared to be moderately to highly resistant (Kiggundu et al., 2003). (Highland bananas and plantains are considered highly susceptible to the weevil (Ortiz et al., 1995; Kiggundu et al., 2003).
Conventional Breeding	4	Multiple sources of resistance often associated with other disease resistance (FHIA3, Km5, Calcutta-4, and Bluggoe), although not a primary focus of breeding programs	Numerous cultivars on farm	New hybrids Development of with demonstrated resistance against weevils e.g. FHIAs, M9 & NABIOs. Yangambi Km5 has been used as parent by Carbap and one elite triploid hybrid has been obtained with tolerance to black weevil (Promusa 2016).
GMOs	4	Biosafety and biosecurity capacity building; Absence of enabling law to regulate biotechnology activities	Proof of concept developed with transfer to market cultivars pending regulatory approval	Resistance genes have been discovered: Cry from the Bacillus thurungiensis and Cystatin from Carica papaya. These have been successfully transformed into 'Sukali ndiizi' (Namuddu et al., 2013). Screen house evaluation of EAHB cv Nakitembe transgenics lines has shown promising enhanced efficacy against weevil (Tazuba et al Unpublished). Sequencing of banana weevil transcriptome has been done and RNA silencing exploited to suppress mortality in banana weevil larvae (Ocimati et al. 2015). Transgenic plants have been generated from embryogenic cell suspensions of different cultivars like; Gonja, Sukali ndizi, Cavendish, hybrids (M9 and NABIOs)
Mutation Breeding	2	Limited studies and reduced interest in mutation breeding.	No current programs	There are no known reports of mutation breeding being used to specifically improve resistance or tolerance to weevils in the literature.
Clonal or mass selection	1	Within cultivar variability for weevil resistance not characterized	Not tested yet	There are no known reports of clonal selection being used to specifically improve resistance or tolerance to weevils in the literature.
Cultivars mixtures	3	Weevil radius of movement greater than distance commonly used in banana stands, perhaps useful with very low percentage of susceptible cultivar with	Smallholder practice with potential weevil effects	Increased diversity of crop varieties, measured by number of varieties (richness) and their evenness of distribution, corresponded to a decrease in the average damage levels across sites and to a reduction of variance of disease damage (Mulumba et al., 2012). Mixing resistant cultivars (Km5, Pisang awak,) with susceptible cultivars (plantain and EAHBs) may suppress

		high proportion of resistant cultivar, but marketing challenges		weevil build up for plant growth and yield losses.
Cultivar-specific Microbes	3	Need for further research on molecular understanding of the plant-microbiome interaction and its impact on plant health and productivity.	Endophytes identified and validated in field, although not cultivar specific	Entomopathogenic fungi, *M. bassiana* offers great potential to control weevil (Kaaya *et al*., 1993; Koppenhofer *et al*., 1993; Feng *et al*., 1994; Nakinga 1994, 2000; Gold *et al*., 2003; Paparu *et al*., 2006; Thompson *et al*., 2006; Akello *et al*., 2008). An improved M. *bassiana* delivery system with different formulations and amendments exists.

Table 17: comparative scores and progress

Method	**BBTD**		**BLS**		**FOC**		**BW**		**Nematodes**		**Weevil**	
	Score	**Progress**	**Score**	**Progress**	**Score**	**Progress**	**Score**	**Progress**	**Score**	**Progress**	**Score**	**Progress**
Cultivar Substitution	2	field ready, but very little cultivar choice	3	field ready, but not wide cultivar choice	3	in use, but for TR4 screening incomplete	3	in use, although limited cultivar choice	3	in use, although limited cultivar choice	3	in use, although limited cultivar choice
Conventional Breeding	2	source of resistance not identified	4	numerous cultivars on farm	3	in use	2	no resistance for use in conventional breeding	4	numerous cultivars on farm	4	numerous cultivars on farm
GMOs	3	Transgenic proofs of concept under development	3	proof of concept, greenhouse	3	green house	3	proof of concept cultivar validated in field	4	proof of concept developed with transfer to market cultivars pending regulatory approval	4	proof of concept developed with transfer to market cultivars pending regulatory approval
Mutation Breeding	1	initial proof of concept under development	2	proof of concept, greenhouse	2	initial proof of concept under development	2	no programs breeding for bacterial wilts	2	no improvement programs using this approach	2	no current programs
Clonal or mass selection	1	initial proof of concept under development	1	little within cultivars variability as basis for selection	3	cultivars in use with on-going clonal selection	1	not under exploration	1	not tested yet	1	not tested yet
Cultivars mixtures	1	no clear mechanism for effectiveness	3	effect documented	2	proof of concept pending	2	not under exploration	3	well known	3	smallholder practice with potential weevil effects
Cultivar-specific Microbes	1	initial proof of concept under development	3	endophytes with potential identified, although not cultivar specific	3	endophytes with potential identified, although not cultivar specific	2	initial screening of organisms and development of techniques	3	endophytes identified and validated in field, although not cultivar specific	3	endophytes identified and validated in field, although not cultivar specific

8. Priority research areas to deploy diversity to reduce pest and disease losses

To conclude this preliminary analysis of the use of diversity to reduce pest and disease losses, we first identify the likely methods to be used for each pest and pathogen. Here we have included other practices with a crucial role in loss reduction. Then we propose the major cultivar development initiatives to respond to the different pest and disease threats. Finally we identify research areas by theme and further steps in the analysis.

Our analysis suggests that certain methods for the use of diversity are more appropriate for specific pests and diseases. Table 18 makes a distinction between alternatives for food security and local markets versus export and large national commercial markets. Three major differences between these two categories are the potential to use GMOs, the flexibility for cultivar substitution and the service support for cultivar specific microbes or microbe specific cultivars.

- Conventional wisdom suggests that GMOs may find greater applicability for local food security. This has not yet proven to be so for bananas. GMO bananas are in the pipeline in Uganda, Malawi and Ecuador. In Table 18 we still assume that GMOs will be approved on a country by country basis. This in itself represents a major challenge. A still bigger step will be consumer and certification acceptance of gene-edited cisgenetic vegetatively-propagated banana in export markets depending on price pressure and environmental concerns.
- Criteria for cultivar substitution for food security may be less strict than for export markets, but material needs to be made available for testing. While specialized niche and exotic food items are also increasingly promoted for export, volumes and areas remain very small.
- Currently endophytes are provided as an input and as such demand an infrastructure to ensure purity and storage conditions. The form this method will take on-farm in the future is uncertain, but would appear to be more applicable for market-oriented growers with input service access. Cultivars with the capacity to recruit microbes for plant defense from the soil is still a distant goal (example: soybean with more promiscuous inoculation rather than highly specialized).

While four of our target pests and pathogens can be addressed by diverse methods to harness diversity, BBTD stands out for the lack of methods (Table 18). Advanced prototypes of transgenic GMOs are under testing to address BBTD, but may not find applicability in export plantations where clean planting material and rigorous roguing and replanting have minimized BBTD losses. For smallholders this type of GMO may be applicable, although the clean planting material approach may still need attention for banana recovery in such systems. The clean seed approach may be the only means to maintain minor cultivars which do not attract the attention of GMO labs. Sanitation and cultural practices will also play a role for smallholder production for BWs as well as nematodes and weevils, although new cultivars from different methods will also play a role.

Based on our analysis we also asked ourselves how many cultivars and from which cultivar groups should be targeted over the next 15 years to address pest and pathogen losses and threats. As Table 19 shows, new cultivars need to incorporate a bundle of pest and disease resistances, not just the main target pest or pathogen. The first four lines of the table summarize the major breeding challenges.

- To address banana recovery in BBTD-affected areas of Asia and Africa with high cultivar diversity will require at least 2 cultivars each from four different cultivar groups. The only method with advanced prototypes for BBTD is GMO through transgenesis. Ideally these cultivars would also have Foc TR4 resistance as well as nematode and weevil resistance, but rural communities in the Congo basin and other areas severely affected by BBTD may find highly acceptable their current cultivars just with BBTD immunity.

- Pesticides are a central part of BLS management in Cavendish and plantain production. BLS resistant varieties would reduce production and environmental costs. Additional dimensions of resistance are necessary to ensure the value of the cultivars as part of current production systems. Conventional breeding and targeted microbial associations appear to be most promising. New cultivars with acceptable fruit quality and post-harvest characteristics would also enable cultivar mixtures in which high value susceptible cultivars are produced within a larger matrix of resistant plants.
- The challenges to reduce Foc losses depend on the production system. For smallholders who do not manage replanting and other practices, very high resistance or even immunity is desirable which may be addressed with conventional breeding, GMOs and cultivar substitution. For export and national monocrop, immunity is still very useful, but not essential with an opening for clonal selection and other methods in which cultivar resistance is increased without achieving immunity. Other cropping systems practices including microbial based management may contribute to successful cropping of bananas in Foc TR4 infected soils.
- For bacterial wilts the lack of genetic sources of resistance leaves transgenesis as the principal current option. Since sources of resistance to Xcm are still unidentified, the use of transgenesis on new cultivars achieved through conventional breeding may add value to cultivars for smallholder situations. This approach could be used both in land races as well as new cultivars which have other useful traits.
- We observe in Table 19 that nematode and weevil resistance may be achieved based on the prebreeding lines for higher priority problems like BLS or Foc. To date they have seldom been the principal target of breeding programs. While extremely high levels of resistance may not be achieved through this approach, improved resistance can be complemented with other practices.

Table 18: More promising methods to reduce pest and pathogen losses for each organism, including other critical practices when diversity may not offer alternatives (*in italics*)

Disease	Food security and local markets	Export and commercial monoculture
BBTD	Transgenic GMOs *Clean planting material for replanting following banana-free period and buffer*	*Clean planting material for replanting following banana-free period and buffer*
BLS	Conventional breeding, GMOs	Conventional breeding, cultivar specific microbes, cultivar mixtures
Foc	Conventional breeding, GMOs, cultivar substitution	Clonal selection, conventional breeding, cultivar specific microbes
BWs	GMOs, cultivar substitution *Sanitation and cultural practices*	*Sanitation and cultural practices*
Nematodes	*Clean planting material, agronomic practices* Cultivar substitution, Conventional breeding, GMOs	Conventional breeding, cultivar specific microbes
Weevil	*Clean planting material, agronomic practices* Cultivar substitution, Conventional breeding, GMOs	Conventional breeding, cultivar specific microbes

Priority research areas to address Table 19 include:

- understanding molecular and genetic basis of cultivar resistance mechanisms and possible role of plant microbiome coupled with molecular and genetic characterization of diseases and other microbial organisms
- new breeding schemes, marker assistance and the strategies to minimize BSV interference to generate cultivars with multiple resistance with consumer-acceptable fruits with adequate post-harvest characteristics

- applicability of GMO advances in other crops to banana
- viability of shift from trans to cis genesis for key resistance
- applicability of clonal selection to other cultivars and for other pests and pathogens
- high throughput phenotyping for single and multiple resistances for application in conventional breeding, clonal selection, cultivar screening, mutagenesis and GMOs
- modeling epidemiological processes at the field scale and the possible role of cultivar mixtures

Other priority research areas

- understanding disease spread at the landscape and improved projections of yield loss
- better statistics on banana by cultivar groups, including mapping
- priority research identified in the Global Musa Conservation Strategy
- understanding local cultivar diversity in all banana production projects with a past, current and future perspective to understand shifts in cultivar diversity and document the role of minor and rare cultivars in rural livelihoods

Table 19: Measures of success for use of diversity in next 25 years

Pest/Pathogen Market	**Cultivar group**	**How many new cultivars**	**Other resistance**	**Method to use diversity**
BBTD: smallholder food security and local market	AAB plantain, AAA, EAH AAA, AAB dessert	2 cultivars each	Nematodes, weevils, Foc	Transgenic GMOs
BLS: export, national	Cavendish export Plantain national	3 Cavendish with different abiotic stress responses 2 plantain for high and low input systems	Foc, nematodes, weevils	Conventional breeding, cultivar specific microbe, cultivar mixtures
Foc: smallholder food security and local market	AAA, EAH AAA, AAB dessert, ABB	2 cultivars each with taste and cooking quality differences for low input systems	Nematodes, weevils	Conventional breeding, substitution, GMOs
Foc: export and national market	Cavendish, other AAA and AAB dessert types	3 of each type with different abiotic stress responses	Nematodes, weevils	Clonal selection, conventional breeding, cultivar specific microbe
BWs: Smallholder for food security and local market	ABB, EAH AAA, AAB plantain	2 cultivars each with taste and cooking quality differences for low input systems	Foc, nematodes, weevils	Conventional + GMOs
Nematodes Weevils	Not a stand alone goal for diversity use			

Next steps to consolidate the proposals in this report:

- more complete review of current status and projections of emerging advanced science to the use of diversity (genetic and molecular understanding of Musa, pests and diseases and microbiome, ecological modeling of epidemiological processes and yield response, high throughput phenotyping and others)
- costs for cultivar development strategies in Table 19

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