

COMMENTARY

African Cassava Whitefly, *Bemisia tabaci*, Resistance in African and South American Cassava Genotypes

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Abstract

The whitefly, *Bemisia tabaci*, is a major pest of cassava, particularly in Africa where it is responsible both for the transmission of plant viruses and, increasingly, for direct damage due to feeding by high populations. To date, there have been no practical solutions to combat this emerging problem, due to the inability of the subsistence farmers that grow cassava to afford expensive inputs such as insecticides. A programme of research was carried out linking institutes in Africa, the UK and South America, to identify possible resistance sources in cassava to the whitefly, *Bemisia tabaci*. The South American genotype MEcu 72 and several Ugandan cassava landraces including Ofumba Chai, Nabwire 1 and Mercury showed good levels of resistance to *B. tabaci*. Field and screen-house experiments showed that all of the improved, high-yielding cassava mosaic disease (CMD) resistant cassava genotypes assessed were highly susceptible to *B. tabaci* and supported high populations of all life stages. These data support the hypothesis that the continuing high populations of cassava *B. tabaci* in Uganda are due, in part, to the widespread adoption of CMD-resistant cassava varieties during the CMD pandemic. They also show that the whitefly, *Aleurotrachelus socialis*, resistance present in the South American cassava genotypes could have broader applicability in the Old World.

Key words: whitefly resistance, cassava, direct pest, Africa, South America

INTRODUCTION

Many whitefly species are important pests of cassava in South America, India and Africa. They represent significant constraints to cassava production, both as direct pests and plant-virus vectors. One of the most damaging of these species is the whitefly, *Bemisia tabaci* (Gennadius), which is also the vector of the cassava mosaic begomoviruses (CMVs) that cause cassava mosaic disease (CMD) (Fishpool and Burban 1994; Colvin *et al.* 2004, 2006).

A pandemic of unusually severe CMD, which first arose in Uganda in the late 1980s (Otim-Nape *et al.* 1994) has now spread to the main cassava-growing regions of nine African countries (Legg *et al.* 2006). One of its characteristics is the high populations of *B. tabaci* associated with its spread (Legg and Ogwal 1998; Otim-Nape *et al.* 2000; Colvin *et al.* 2004, 2006). Prior to the advent of the pandemic, African *B. tabaci* populations on cassava were consistently low, even at the most favourable time of year for survival and development (Fishpool and Burban 1994; Colvin *et al.* 2004). The high populations have persisted in pan-

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demic affected areas and, in addition to the yield losses caused by CMV transmission, they have now caused *B. tabaci* to become a direct cassava pest in its own right. Yield losses of more than 50%, for instance, have been recorded in the worst affected cassava varieties (Legg *et al.* 2004; Omongo *et al.* 2004), some of which are the most preferred amongst farmers, due to their good tuber quality and resistance to CMD.

An additional factor contributing to the continuing presence of large cassava *B. tabaci* populations is that several of the most popular CMD-resistant varieties, which were introduced during the pandemic to combat CMD, are highly suitable hosts for *B. tabaci* and appear to have little whitefly resistance (Omongo 2003). This undesirable trait adversely affects their effectiveness for CMD control, because adult *B. tabaci* are highly mobile and so these populations contribute to a heightened CMV inoculum pressure and the rapid spread of CMVs to any susceptible varieties grown in the vicinity (Omongo 2003). They therefore perpetuate the superabundance of *B. tabaci*, increasing the risk of new pandemics (Alicai *et al.* 2007) and the evolution of resistance-breaking viruses (Seal *et al.* 2006).

The direct feeding damage by whiteflies on cassava appears as chlorotic mottling and twisting or curling, particularly on upper leaves (Bellotti and Arias 2001; Legg *et al.* 2004). If large populations develop early in the life of the crop, plant vigour and tuber size are reduced and general plant stunting occurs. Another effect of the large whitefly populations is the production of honey dew, which falls onto the lower leaves. This is subsequently colonised by black sooty mould (Schuster *et al.* 1996; Bellotti *et al.* 1999; Bellotti and Arias 2001; Legg *et al.* 2004), which reduces the ability of the leaves to photosynthesise and contributes to yield losses.

The occurrence of black sooty mould on the upper surface of the lower leaves on cassava plants is now commonplace in Uganda and this recent phenomenon is referred to by farmers as “black mosaic”. This name suggests that the farmers are aware of the reduced yields that result, because “mosaic” is a term closely associated with the CMD pandemic that devastated cassava production in the country, causing famine and suffering to the people whose livelihood depended on the crop (Thresh *et al.* 1994; Otim-Nape *et al.* 2000). While a solution to CMD has been found through the develop-

ment and deployment of CMD-resistant varieties, very limited research has been carried out to date to address the emerging problem of whitefly as a direct pest of cassava in Africa.

Differences in the acceptability of cassava varieties to whiteflies have been recognized for some time (Nair and Daniel 1983; Maruthi *et al.* 2001) and high levels of resistance to the South American whitefly species, *Aleurotrachelus socialis* (Bondar), have been identified in neotropical cassava genotypes (Gomez and Bellotti 2003; Arias *et al.* 2004) and to biotype-B of *B. tabaci* (Burbano 2003), now known as the Middle East-Asia Minor 1 (MEAM1) mtCOI phylogenetic group (Dinsdale *et al.* 2010; De Barro *et al.* 2011; Liu *et al.* 2012).

In the area of Uganda where the field work was carried out and where the population of *B. tabaci* was collected for the laboratory-based work in the UK, two mitochondrial cytochrome oxidase subunit one gene (mtCOI) haplotypes have been identified previously on cassava (Legg *et al.* 2002; Colvin *et al.* 2006). At the time of this study, however, only one mtCOI genetic group was present on cassava (Sseruwagi *et al.* 2005; Colvin *et al.* 2006), which is currently named Sub-Saharan Africa 1 (Dinsdale *et al.* 2010; De Barro *et al.* 2011; Liu *et al.* 2012).

Cassava is grown by the poorest of subsistence farmers in Africa who have few, if any, additional resources to invest in their cassava crops. An ‘embedded technology’, such as whitefly resistance would have the extremely important additional advantage of restoring the efficacy of cultural disease control practices, such as improved phytosanitation and the rouging of virus-diseased plants. Whitefly resistance, therefore, offers a low-cost, sustainable solution with the best chances of widespread adoption by subsistence farmers (Bellotti *et al.* 1999; Bellotti and Arias 2001). The research described here represents initial research to assess the South American whitefly-resistant germplasm from the neotropics against African cassava *B. tabaci*. Several different experimental designs were necessary to achieve this due to the susceptibility of the South American cassava genotypes to African cassava mosaic viruses. The paper also presents data on the relative resistances of various cassava landraces, with the aim of identifying potentially useful genotypes for further resistance breeding and ultimately the development

of effective management technologies able to control the super-abundant whitefly populations now present on cassava in many parts of Africa.

RESULTS

B. tabaci populations on CMD-resistant cassava genotypes in the uniform yield trial

There was a significant difference in *B. tabaci* adult populations between the cassava genotypes ($F=3.0$, $df=9$, $P=0.02$) evaluated in the Uniform Yield Trial at Namulonge. The *B. tabaci* adult population was lowest on genotype MM96/4271 followed by MM96/0116, with average mean counts of adults per top 5 leaves of 12.9 ± 1.6 and 18.8 ± 1.3 , respectively (Table 1). The number of adults was highest on genotype MM96/4235, which had 67.9 ± 20.7 adults per top 5 leaves. Numbers of *B. tabaci* eggs and nymphs also differed significantly between the cassava genotypes ($F=3.1$, $df=9$, $P=0.01$). The lowest and the highest mean egg and nymph numbers per top 5 leaves were recorded on genotypes MM96/4271 (143.9 ± 38.5) and MM96/4235 (605.3 ± 170.5). There was a highly significant explanatory relationship ($y=8.8x(\pm SE1.02)+29(\pm 32.7)$, $P<0.001$) between the adult populations and the eggs and nymphs recorded on these genotypes (Fig. 1). There were no obvious correlations, however, between plant traits such as “bitter or sweet”, “leaf width/area”, “leaf colour” and resistance to *B. tabaci*.

B. tabaci populations on local landraces

Differential varietal reactions to *B. tabaci* were apparent amongst the cassava landraces (ANOVA, $P<0.001$). The *B. tabaci* adult populations were the lowest on the landraces Ofumba Chai, Nabwire 1 and Mercury, with means of less than 20 adults per top 5 leaves. Adult populations were significantly greater on Sporoza, Egabu, Kagita 3 and Duma 1 (Fig. 2).

B. tabaci infestation on CMD-resistant genotypes and local landraces in the screen-house

There were significant differences between the geno-

Table 1 Mean numbers of *B. tabaci* adult, eggs and nymphs on 5 top leaves of CMD-resistant cassava genotypes in a Uniform Yield Trial (UYT) in Uganda

Cassava genotype	Mean adults (\pm SEM)	Mean eggs and nymphs (\pm SEM)
MM96/4235	67.9 \pm 20.7	605.3 \pm 170.5
MM96/0102	30.6 \pm 6.3	319.0 \pm 13.4
MM96/0469	29.8 \pm 9.2	377.9 \pm 106.6
MM96/0686	29.3 \pm 4.1	314.9 \pm 38.7
MM96/4799	28.6 \pm 1.3	276.3 \pm 66.6
Nase 10 (95/NA-2-00063)	23.9 \pm 6.4	243.7 \pm 79.5
Nase 12	23.6 \pm 3.8	174.0 \pm 32.1
MM96/0232	23.4 \pm 5.2	179.0 \pm 40.3
MM96/0116	18.8 \pm 1.3	189.3 \pm 30.5
MM96/4271	12.9 \pm 1.6	143.9 \pm 38.5

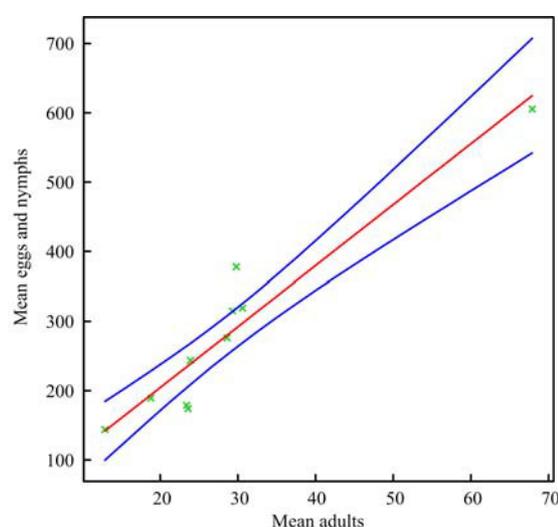


Fig. 1 Fitted and observed relationship with 95% confidence limits between mean adult *B. tabaci* numbers and mean juvenile stages on CMD-resistant cassava genotypes.

types ($F=4.1$, $df=18$, $P<0.001$) in the mean numbers of *B. tabaci* recorded during the trial. Generally, *B. tabaci* adult number was lower on the local landraces than on the CMD-resistant genotypes TMS I92/0067, Nase 3, Nase 9 and Nase 12 (Table 2). *B. tabaci* populations were low (<20 adults per top 5 leaves) on 8 of the 15 local landraces evaluated with the lowest populations recorded on Njule Red, Soya, Bao and Njule White. Overall, mean *B. tabaci* adult number was the highest on CMD-resistant genotype TMSI92/0067 (39.2 ± 4.4 adults per top 5 leaves) and the greatest population on the local landraces was recorded again on Duma 1 (37.4 ± 7.2 adults per top 5 leaves) (Table 2).

The *B. tabaci* egg and nymph populations on the

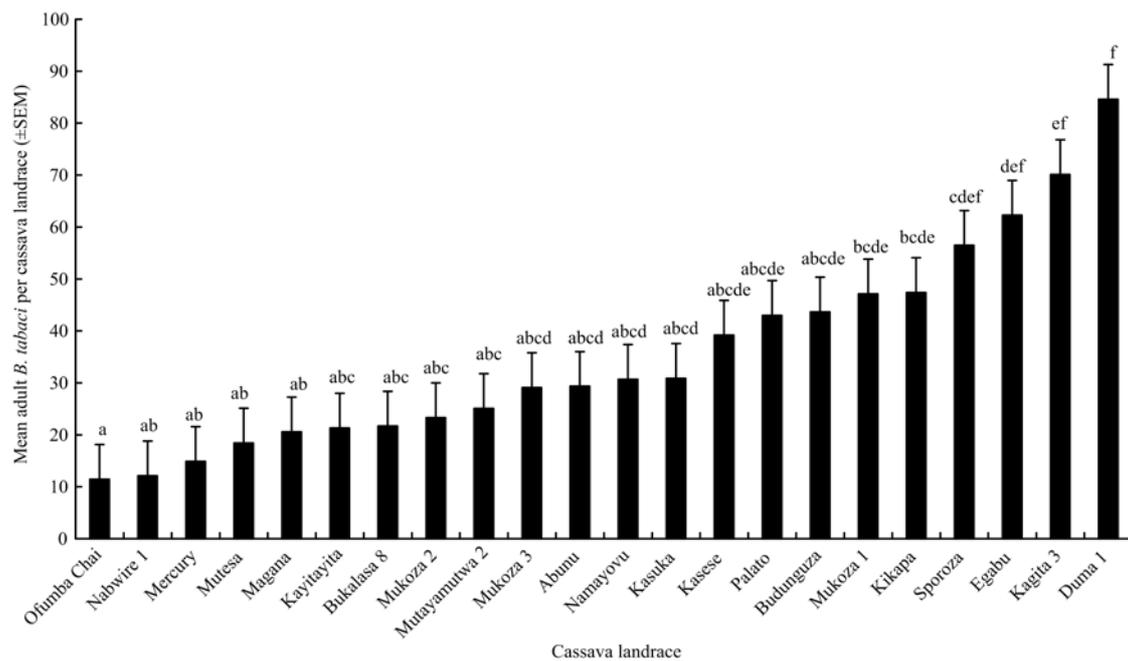


Fig. 2 Variation in the mean populations of *B. tabaci* present on 22 cassava local land races collected from Uganda and grown at NaCRRI. Means annotated with the same letter are not significantly different (ANOVA plus Tukey test at $P < 0.005$).

Table 2 Mean *B. tabaci* populations (±SEM), feeding damage and sooty mould severity on cassava cultivars under free-choice colonization

Cultivar	Adult no./top 5 leaves ¹⁾	37 DAP egg & nymph no./top 5 leaves	83 DAP damage (scale 1-5)	83 DAP Sooty mould (scale 1-5)
I92/0067	39.2±4.4	1393.8±160.6	3.8	4.8
Duma 1	37.4±7.2	1161.6±210.0	4.2	5.0
Nase 3 (TMS 30572)	37.1±8.3	912.2±229.3	3.9	4.7
Bukalasa 8	32.9±3.6	575.7±87.9	3.6	4.2
Nase 9 (30555-17)	30.8±5.1	1176.0±234.2	3.5	4.4
Nase 12	30.3±4.8	550.4±75.8	4.2	4.6
Aladu	27.1±9.0	1308.8±382.8	3.4	3.9
Magana	25.7±6.9	624.6±158.2	3.8	4.3
Mutesa	23.3±5.0	632.1±73.3	3.1	3.4
Mercury	21.3±6.2	746.1±185.3	3.3	3.3
Egabu	21.2±6.4	971.0±187.7	4.0	4.6
Senyonjo	16.4±3.5	563.1±35.0	2.9	3.3
Nylon	15.9±2.8	581.0±99.4	3.8	4.4
Namayovu	14.5±3.0	795.8±341.0	3.6	4.3
Ebwanatereka	12.9±2.9	436.2±107.2	3.3	3.6
Njule White	12.7±2.9	535.2±110.4	2.4	3.0
Bao	11.4±2.6	431.4±103.9	3.0	3.9
Soya	11.3±2.4	457.4±86.6	3.3	3.8
Njule Red	5.4±1.7	228.6±98.0	2.1	2.6

¹⁾ means of 15 consecutive daily records: 28-43 DAP (day after planting).

plants showed significant differences ($F=3.1$, $df=18$, $P<0.001$) amongst the genotypes. Generally there were more eggs and nymphs on the CMD-resistant genotypes compared to the local landraces (Table 2). Mean egg and nymph numbers on the CMD-resistant genotype Nase12 ((550.4±75.8)/top 5 leaves), and the local landraces Aladu ((1 308.8±382.8)/top 5 leaves) and

Duma 1 ((1 161.6±210.0)/top 5 leaves) deviated from the general trend. The highest egg and nymph numbers were recorded on I92/0067 with means of (1 393.8±160.6)/top 5 leaves and the lowest were on Njule Red with means of 228.6±98.0 (Table 2).

Feeding by whitefly adults and nymphs induced a characteristic yellow-to-green mottled appearance and

twisting and curling, particularly on the apical leaves. High populations also caused the development of sooty moulds on the upper surfaces of lower leaves. Physical feeding damage and high sooty mould coverage were recorded on all the CMD-resistant genotypes (Table 2). High feeding damage and sooty mould coverage were also recorded on the local landraces Duma 1, Egabu, Magana, and Namayovu. The lowest feeding damage and sooty mould severity were recorded on local landraces Njule Red, Njule White, and Senyonjo (Table 2), which was associated with their resistances to *B. tabaci*.

Population growth of African cassava *B. tabaci* on South American cassava genotypes

18 d after the confinement of the African cassava *B. tabaci* onto the cassava genotypes, there were no significant differences between the populations and it was evident that all of the genotypes could be colonized. The variety with the greatest level of resistance, however, was MEcu 72 (Table 3). 38 d after the African cassava *B. tabaci* were released, MEcu 72 still showed the highest level of resistance and the difference was now significant (Table 3).

Table 3 The total populations of African *B. tabaci*, 18 and 38 d after 10 male and 10 female adults were confined on each plant

Cassava genotype	Number of plants	18 d mean eggs & nymphs (\pm SEM) ¹⁾	38 d mean eggs, nymphs & adults (\pm SEM) ²⁾
Col 1468	10	458.3 \pm 40.7	2 514.5 \pm 257
Col 2063	15	439.8 \pm 27.7	2 010.5 \pm 129
CG 489-34	13	511.6 \pm 30.4	2 797.2 \pm 285
MEcu 72	12	391.0 \pm 22.3	1 639.3 \pm 89.1

¹⁾ ANOVA $P=0.053$. All initial adults had died at the time of assessment.

²⁾ ANOVA $P<0.001$. Variances varied and so a Tukey test of multiple comparisons could not be carried out.

B. tabaci populations on South American genotypes in Uganda under choice conditions

The population of *B. tabaci* adults varied significantly ($F=3.96$, $df=4$, $P=0.009$) between the 5 cassava genotypes (Fig. 3). MEcu 72 and Colombian showed the lowest and highest preference by *B. tabaci*, respectively.

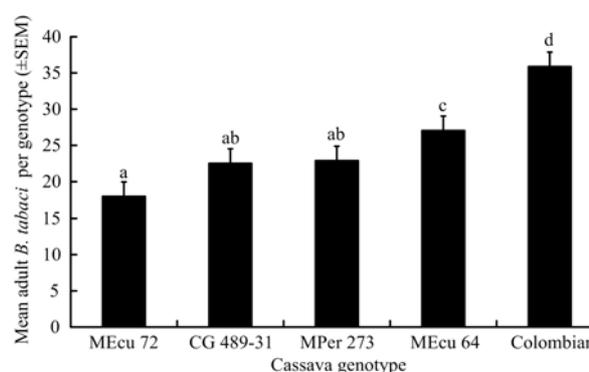


Fig. 3 The settling and feeding preference of African cassava *B. tabaci* on South American cassava genotypes. Means annotated with the same letter are not significantly different (ANOVA followed by a Tukey test at $P<0.005$).

DISCUSSION

This study addressed the pressing need to identify durable resistance sources to African cassava *B. tabaci*. This search is complicated by the presence of CMVs in Africa and the high susceptibility of South American cassava genotypes to them. Virus infection in the South American genotypes would have complicated the interpretation of the experimental results, because cassava infected with *East African cassava mosaic virus* (EACMV-UG), for example, has been shown to cause the *B. tabaci* populations on them to increase significantly more rapidly than on equivalent healthy plants (Colvin *et al.* 2006).

Significant differences in the level of colonization

and population growth by *B. tabaci* on cassava genotypes were apparent under field, screen-house and insectary conditions. This variability demonstrates that sources of resistance to African cassava *B. tabaci* exist both within cassava germplasm currently available in Africa and from additional genotypes from the neotropics. This is important because this genetic diversity can be exploited by farmers and agricultural research scientists to increase crop productivity and as a

means of avoiding the most damaging effects of pests and pathogens (Thresh *et al.* 1994).

The UYT confirmed that improved CMD-resistant genotypes are highly susceptible to *B. tabaci*. The number of *B. tabaci* adults on cultivar MM96/4235, which was the worst affected in the field was about 5 times higher compared to the populations on MM96/4271, which was the least colonized by *B. tabaci* (see Table 1). Variation in susceptibility to *B. tabaci* was also evident amongst the local landraces and Duma 1 had consistently high *B. tabaci* populations, whereas Njule Red supported the lowest *B. tabaci* populations.

Cassava genotype leaf area affected the severity score for sooty mould and so, for instance, the egg and nymph number on variety Nase 12 was much lower than that on variety Aladu, but the feeding damage and sooty mould severity on was higher. Nase 12 has much broader leaves than Aladu and so we presume that much more of the honey dew was caught on the upper surface of Nase 12's leaves.

B. tabaci egg and nymph populations were highly related to the adult populations, providing additional evidence of variability in resistance. The results from the screen-house experiment where CMD-resistant cultivars and landraces were tested together showed that all the CMD-resistant cultivars, except Nase 12, had very high numbers of eggs and nymphs (>900 per top 5 leaves). The high suitability for oviposition makes the cultivars prone to serious physical damage due to feeding by all of the life-stages of *B. tabaci*. In contrast, the relatively low numbers of eggs and nymphs (<500 per top 5 leaves) on Njule Red, Bao, Tereka, and Soya indicated that these landraces are less preferred for oviposition and the damage on them, consequently, was lower. These data strongly support the hypothesis that the current high populations of *B. tabaci* seen in many parts of Uganda are, in part, due to the now widespread adoption and use of CMD-resistant cassava genotypes, and why "black mosaic" is also associated with them.

As the level of physical damage due to whitefly feeding and the severity of sooty mould on Njule Red, Njule White and Senyonjo showed the lowest mean scores for damage (2.1) and sooty mould (2.6), this suggests that these landraces could be used in breeding programmes to introduce their whitefly resistance into the CMD-resistant genotypes.

Several cultivars with high levels of resistance to South American whitefly species have been identified

previously (Bellotti and Arias 2001). The most prominent resistant cultivars include MEcu 72, MEcu 64 and hybrids CG 489-31 and CG 489-34. This study's data suggest that MEcu 72 shows the most promise against African cassava *B. tabaci*. In the experiments where whitefly population growth was assessed on the different cassava genotypes in the insectary, at NRI, MEcu 72 was clearly the most resistant to African *B. tabaci*, as it had the fewest eggs and nymphs present on it 18 d after colony initiation. After a further 18 d, this difference was still apparent and had increased and become highly significant. These results were consistent with those from South America, where *A. socialis* oviposition has been reported to be greater and the development period shorter on the susceptible cultivar Colombian than on the resistant cultivar CG 489-31 (Bellotti and Arias 2001). The latter genotype was created from a cross involving the whitefly-resistant MEcu 72 as one of the parents.

When African cassava *B. tabaci* were given a choice of which cassava genotype to settle on, MEcu was the least preferred. This suggests strongly that MEcu 72 may possess plant defence compounds active against African cassava *B. tabaci*, and/or its phloem sap composition may be less suitable for *B. tabaci* growth and reproduction.

The current study only involved one African *B. tabaci* phylogenetic group (putative species), Sub-Saharan Africa 1, yet other putative *B. tabaci* species in Africa have been recorded to colonise cassava (Maruthi *et al.* 2001, 2004; Sseruwagi *et al.* 2005; Dinsdale *et al.* 2010). It remains to be discovered, therefore, whether or not the resistance identified here is also effective against the other cassava-colonising, African *B. tabaci* putative species.

FUTURE RESEARCH

In Uganda, numerous requests to deal with the new 'black mosaic' have been received from most districts and the lack of reference to the whiteflies themselves demonstrates the dearth of knowledge about this emerging problem. As the CMD pandemic continues to spread throughout Sub-Saharan Africa and is tackled with the introduction of the same CMD-resistant varieties, it is probable that the demand for a solution to this emerg-

ing problem will be echoed elsewhere in Africa. The whitefly problem is also a threat to the development of cassava into a more important and industrial crop, with increasing importance to the Ugandan economy. The other major risk is that the high populations may increase other virus problems like *Cassava brown streak virus* (CBSV) and also increase the risk that the current resistance to CMD will be broken. Concerted efforts should therefore be made to develop cassava varieties that have combined resistance to CMD, cassava brown streak disease and the different cassava colonising *B. tabaci* putative species. The Ugandan landraces and South American genotypes identified in this study, which show the strongest whitefly resistance could provide the parental genotypes required for future breeding programmes.

CONCLUSION

This study describes the serious and ongoing pest and disease problems in Uganda, resulting from super-abundant populations of the African cassava whitefly, *B. tabaci*. New resistance sources in cassava to cassava *B. tabaci*, are reported here. The South American genotype MEcu 72 and several Ugandan cassava landraces including Ofumba Chai, Nabwire 1 and Mercury showed the most promising levels of resistance. The experimental data also show for the first time that the whitefly, *Aleurotrachelus socialis*, resistance present in some South American cassava genotypes could also have broader applicability in the Old World.

MATERIALS AND METHODS

B. tabaci populations on CMD-resistant cassava genotypes in Uganda

Ten cassava genotypes (Table 1) were planted in May 2003 in the Uniform Yield Trial (UYT) at the National Crops Resources Research Institute (NaCRRI), Uganda, and were evaluated for resistance to *B. tabaci*. The experiment had a randomised complete block (RCB) design with each genotype planted in 30 m² plots (5 m×6 m) and replicated four times. The distance between the plots was 2 m and the experiment was conducted under open-field conditions and natural *B. tabaci* infestation. *B. tabaci* adult, egg and nymph numbers were counted on 5 top open leaves per

plant from 20 randomly selected plants per plot. Previous work demonstrated this technique to be suitable for monitoring adult populations in the field (Fishpool *et al.* 1995; Colvin *et al.* 2004) and also found that the *B. tabaci* population at NaCRRI during this period of research was the phylogenetic group now termed Sub-Saharan Africa 1 (Colvin *et al.* 2006). Monthly population counts were started at three months after planting (MAP) and terminated at seven MAP. Mean values were calculated for adult, egg and nymph counts and the data were subjected to repeated measurements analysis of variance (ANOVA) using GenStat (2008) to test for statistically significant differences in the population with respect to the genotypes.

B. tabaci populations on Ugandan local cassava landraces

Twenty-two cassava landraces (genotypes selected and grown by farmers) were collected within Uganda and planted at National Crops Resources Research Institute (NaCRRI) for characterization under field conditions. Cuttings obtained from plants free of cassava mosaic virus symptoms of each variety were planted in single row plots (10 plants/row) in two replicates. While the assessment of agronomic traits was the main aim of the trial, data on the major pests and diseases occurring on the germplasm collection were also recorded. To identify resistance to *B. tabaci*, adult numbers were counted on the 5 top leaves of a randomly selected sub-set of 10 plants per landrace, starting at 82 d after planting (DAP) and, thereafter, every 30 d for four consecutive periods. The data sets were summarised for all the plants per variety and mean values from the two replicates were obtained. Means were analysed by analyses of variance followed by a Tukey test for multiple comparisons (GenStat 2008).

Screenhouse evaluation of cassava local landraces for *B. tabaci* resistance

Stem cuttings from three to 5 mon-old CMD symptomless plants of local landraces were collected from different parts of Uganda in September 2004 and planted directly in the earth floor of a screen-house at NaCRRI. The selection of landraces included seven of those used in the field assessment of the landraces to provide comparative data, e. g., Duma 1 and Mercury, which were *B. tabaci* susceptible and tolerant, respectively (the above section). The experimental set-up comprised single-row plots of three plants per landrace, spaced at 50 cm between plants and 65 cm between rows. The plots were replicated three times giving a total of nine plants per landrace. Four CMD resistant varieties were included in the trial as *B. tabaci* susceptible (TMS I92/0067, Nase 3 (TMS 30572) and Nase 12 (MM95/0414)) and resistant/tolerant (Nase 9 [30555-17]) checks.

At 21 DAP, 10–15 thousand *B. tabaci* adults of both sexes were released in the experimental screenhouse. The whiteflies were collected from the middle of a multiplication block of improved CMD-resistant cassava variety Nase 10 (non-symptomatic) and so were assumed to be non-viruliferous. The released insects were then left for a period of 7 d. Thereafter, adult numbers on the top 5 leaves of every plant were counted. The count was repeated after a further 7 d. No additional counts were made to avoid the inclusion of newly-emerged adults in the data. Number of eggs and nymphs were counted on the plants at 37 DAP.

The established plants were then left exposed to the introduced whiteflies for 3 mon and the *B. tabaci* population was allowed to develop within the screen-house. This was done to enable an assessment of physical damage to the cassava plants by the colonizing whitefly. The level of damage due to feeding and the severity of sooty mould on the plants were scored on a scale of 1–5 (feeding damage: 1=no damage and 5=severe damage; sooty mould severity: 1=no sooty mould and 5=more than 75% of the plant covered by sooty mould) at 83 DAP to assess susceptibility to whitefly.

The resistance of South American cassava genotypes to African cassava *B. tabaci* under 'no-choice' conditions

Four cassava genotypes, Col 1468, MCol 2063, CG 489-34, and MEcu 72 with various degrees of resistance to South American whitefly species (Bellotti *et al.* 1999) were sent by the International Centre for Tropical Agriculture (CIAT) to the Natural Resources Institute (NRI), UK. The genotypes were received in glass tubes as tissue cultured plants (*in vitro* propagated). These were further sub-cultured and multiplied successfully by using the combined *in vitro* propagation and glasshouse protocols established at NRI.

B. tabaci (phylogenetic group Sub-Saharan Africa 1) from Namulonge (Uganda) were collected and established on the *B. tabaci* susceptible cassava genotype "Colombian" in the insectary at NRI. They were maintained for two generations prior to being used for experimentation under the following environmental conditions, (27±0.5)°C, 70% r.h. and LD 12:12 h.

Cassava plantlets, with 5–8 expanded leaves, of each genotype were enclosed separately in individual, insect-proof containers. Ten male and ten female, 2-d-old, adult *B. tabaci* were released onto each plant using a pooter (Watkins & Doncaster, UK, with a large entry aperture of 0.5 cm diameter and perspex 'holding' tube internal diameter of 3.2 cm). Each 'treatment' consisted of 3 to 5 plants, replicated three times. 18 d after release, adults were removed from the plants and the numbers of eggs and nymphs per plant were recorded. After data collection,

adults were released back on to the plants and the populations were left to increase for a further 18 d. Mortality of adults during this procedure was negligible, due to minimal handling and a maximum period of 15 min spent in the pooter. At this point, the number of eggs, nymphs and adults per plant were recorded for a second time.

The resistance of South American cassava genotypes to African cassava *B. tabaci* under 'choice' conditions

The initial screen at NRI (no-choice experiment) showed that only MEcu 72 showed good resistance to Sub-Saharan Africa 1 *B. tabaci*. We therefore obtained additional South American cassava genotypes with resistance to South American whitefly species for testing in Africa under the more natural 'choice' conditions. Five genotypes, MEcu 64, CG 489-31, MPer 273, MEcu 72, and "Colombian" were sent as tissue cultured plantlets by NRI in November 2005 to Uganda. Established plants of similar sizes were selected from the hardened-off material and tested twice in the screenhouse to establish their susceptibility to *B. tabaci*. The experiment involved a RCB design with three replications. The screen-house conditions were necessary, because the South American genotypes were susceptible to CMVs. Each replicate contained three plants per cultivar and the plants were randomly assigned to each block. The spacing between plants was 50 cm and a space of 75 cm separated the blocks. Five to seven thousand *B. tabaci* adults of both sexes were collected from the field between 08:00 and 09:00 h and released immediately on to the plants in the experimental screen-house. The whiteflies were collected from the middle of a multiplication block of improved CMD-resistant, symptomless cassava variety Nase 10 (95/NA-2-00063). Symptomless leaves of this variety give a negative result when tested for geminiviruses by PCR and so these whitefly were assumed to be non-viruliferous.

Numbers of whitefly adults were initially recorded on all of the leaves of every plant. The first record was taken at 17:00 h on the day the whitefly adults were released onto the plants (i.e., 8 h after release). Adult counts on three subsequent days were carried out between 08:00 and 09:00 h.

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References

- Alicai T, Omongo C A, Maruthi M N, Hillocks R J, Baguma Y, Kawuki R, Bua A, Otim-Nape G W, Colvin J. 2007. Cassava brown streak disease re-emerges in Uganda. *Plant Disease*, **91**, 24-29.
- Arias B V, Bellotti A C, Vargas H L B. 2004. Nataima-31, a cassava (*Manihot esculenta*) variety resistant to the whitefly, *Aleurotrachelus socialis*. In: *Sixth International Scientific Meeting of the Cassava Biotechnology Network*, 8-14 March, 2004, CIAT, Cali, Colombia. p. 66.
- De Barro P J, Liu S S, Boykin L M, Dinsdale A B. 2011. *Bemisia tabaci*: a statement of species status. *Annual Review of Entomology*, **56**, 1-19.
- Bellotti A C, Arias B. 2001. Host plant resistance to whiteflies with emphasis on cassava as a case study. *Crop Protection*, **20**, 813-823.
- Bellotti A C, Smith L, Lapointe S L. 1999. Recent advances in cassava pest management. *Annual Review of Entomology*, **44**, 343-370.
- Burbano M. 2003. Studies on the biology and development of biotype B of *Bemisia tabaci* on cassava, *Manihot esculenta*, and the wild species, *Manihot carthaginensis*. In: *CIAT Annual Report 2003, Project IP3: Improving Cassava for the Developing World*.
- Colvin J, Omongo C A, Govindappa M R, Stevenson P C, Maruthi M N, Gibson G, Seal S E, Muniyappa V. 2006. Host-plant viral infection effects on arthropod-vector population growth, development and behaviour: management and epidemiological implications. *Advances in Virus Research*, **67**, 419-451.
- Colvin J, Omongo C A, Maruthi M N, Otim-Nape G W, Thresh J M. 2004. Dual begomovirus infection and high *Bemisia tabaci* populations: Two factors driving the spread of cassava mosaic disease pandemic. *Plant Pathology*, **53**, 577-584.
- Dinsdale A, Cook L, Riginos C, Buckley Y, De Barro P J. 2010. Refined global analysis of *Bemisia tabaci* (Gennadius) (Hemiptera: Sternorrhyncha: Aleyrodoidea) mitochondrial CO1 to identify species level genetic boundaries. *Annals of Entomological Society of America*, **103**, 196-208.
- Fishpool L D C, Burban C. 1994. *Bemisia tabaci*: the whitefly vector of African cassava mosaic virus. *Tropical Science*, **34**, 55-72.
- Fishpool L D C, Fauquet C, Fargette D, Thouvenel J C, Burban C, Colvin J. 1995. The phenology of *Bemisia tabaci* (Homoptera: Aleyrodidae) populations on cassava in southern Côte d'Ivoire. *Bulletin of Entomological Research*, **85**, 197-207.
- GenStat Release 11 VSN International Ltd. 2008. *Reference Manual, Part 3 Procedure Library PL19*. Hemel Hempstead, UK.
- Gomez M J, Bellotti A C. 2003. Studies on whitefly (*Aleurotrachelus socialis*) resistance mechanisms in selected cassava genotypes. In: *CIAT Annual Report 2003. Project IP3. Improving Cassava for the Developing World*.
- Legg J P, Ogwal S. 1998. Changes in the incidence of African cassava mosaic geminivirus and the abundance of its whitefly vector along the south-north transects in Uganda. *Journal of Applied Entomology*, **122**, 167-178.
- Legg J P, Sseruwagi P, Brown J. 2004. *Bemisia* whiteflies cause physical damage and yield loss to cassava in Africa. In: *Sixth International Scientific Meeting of the Cassava Biotechnology Network*, 8-14 March, 2004, CIAT, Cali, Colombia. p. 78.
- Legg J P, Owor B, Sseruwagi P, Ndunguru J. 2006. Cassava mosaic virus disease in east and central Africa: Epidemiology and management of a regional pandemic. *Advances in Virus Research*, **67**, 255-418.
- Legg J P, French R, Rogan D, Okao-Okuja G, Brown J K. 2002. A distinct *Bemisia tabaci* (Gennadius) (Hemiptera: Sternorrhyncha: Aleyrodidae) genotype cluster is associated with the epidemic of severe cassava mosaic disease in Uganda. *Molecular Ecology*, **11**, 1219-1229.
- Liu S S, Colvin J, De Barro P J. 2012. Species concepts as applied to the whitefly *Bemisia tabaci* systematics: how many species are there? *Journal of Integrative Agriculture*, **11**, 176-186.
- Maruthi M N, Colvin J, Seal S E. 2001. Mating incompatibility, life-history traits and RAPD-PCR variation in *Bemisia tabaci* associated with the cassava mosaic disease pandemic in East Africa. *Entomolgia Experimentalis et Applicata*, **99**, 13-23.
- Maruthi M N, Colvin J, Thwaites R M, Banks G K, Gibson G, Seal S E. 2004. Reproductive incompatibility and cytochrome oxidase I gene sequence variability amongst host-adapted and geographically separate *Bemisia tabaci* populations. *Systematic Entomology*, **29**, 560-568.
- Nair N G, Daniel R S. 1983. Preference of *Bemisia tabaci* Gen. to cassava varieties and their reaction to cassava mosaic disease. *Journal of Root Crops*, **9**, 45-49.
- Omongo C A. 2003. Cassava whitefly, *Bemisia tabaci*, behaviour and ecology in relation to the spread of cassava mosaic epidemic in Uganda. Ph D thesis, University of Greenwich, UK.
- Omongo C A, Colvin J, Sserubombwe W, Alicai T, Baguma Y, Bua A, Legg J P, Gibson R W. 2004. Host-plant resistance to African *Bemisia tabaci* in local landraces and improved cassava mosaic disease resistant genotypes in Uganda. In: *Sixth International Scientific Meeting of the Cassava Biotechnology Network*. 8-14 March 2004, CIAT, Cali, Colombia. p. 84.
- Otim-Nape G W, Bua A, Baguma Y. 1994. Accelerating the transfer of improved crop production technologies: controlling African cassava virus disease in Uganda. *African Crop Science Journal*, **2**, 479-495.
- Otim-Nape G W, Bua A, Thresh J M, Baguma Y, Ogwal S, Semakula G N, Acola G, Byabakama B, Colvin J, Cooter R J, et al. 2000. *Cassava Mosaic Virus Disease in*

- Uganda: The Current Pandemic and Approaches to Control*. University of Greenwich, PSTC28, UK.
- Schuster D J, Stansly P A, Polston J E. 1996. Expression of plant damage by *Bemisia*. In: Gerling D, Mayer R T, ed., *Bemisia 1995: Taxonomy, Biology, Damage, Control and Management*. Intercept, Andover, UK. pp. 153-165.
- Seal S E, Jeger M J, van den Bosch F. 2006. Begomovirus evolution and disease management. *Advances in Virus Research*, **67**, 297-316.
- Sseruwagi P, Legg J P, Maruthi M N, Colvin J, Rey M E C, Brown J K. 2005. Genetic diversity of *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) populations and the presence of the B biotype and a non-B biotype that can induce silverleaf symptoms in squash, in Uganda. *Annals of Applied Biology*, **147**, 253-265.
- Thresh J M, Otim-Nape G W, Jennings D L. 1994. Exploiting resistance to African cassava mosaic virus. *Aspects of Applied Biology*, **39**, 51-60.

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