## **CBSD** leaf and root symptoms

# Disparity between leaf and root symptoms and crop losses associated with cassava brown streak disease in four countries in eastern Africa.

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## Abstract

Cassava brown streak disease is endemic to the coastal regions of East Africa and from around 2004 the disease resurged and became epidemic in the Great Lakes Region, where it continues to spread. In both these areas CBSD leaf symptoms occur at high incidences. However, it is the associated symptom of root rot (necrosis) in the starch-bearing tissues that renders the root unfit for human consumption. Because the extent of root necrosis is not known until the crop is harvested and surveys require destructive sampling, root symptoms are much less frequently assessed than are the above-ground symptoms on the leaves and stems. Surveys were undertaken in selected villages in Tanzania, Kenya, Uganda and Malawi to assess the incidence of CBSD leaf symptoms and the incidence and severity of root symptoms, in order to estimate the impact of the disease on household food security and on cassava processing. CBSD leaf symptoms were recorded at high incidences [40 - 90% in individual fields] in all fields visited throughout East Africa but root necrosis incidence was lower than would be expected from the high incidence of leaf symptoms. Severe root necrosis at high incidence was found only on a few varieties, usually grown to a limited extent. It appears that varieties that are prone to root necrosis are being abandoned in favour of those with a lower propensity to develop root necrosis after infection by the virus.

## Introduction

For almost 100 years cassava brown streak disease (CBSD) has been known to occur widely in the coastal areas of east Africa from Kenya down to the Zambezi River in Mozambique (Hillocks and Jennings 2003). The cassava brown streak viruses (CBSVs) are spread through planting of infected cuttings and by the main insect vector, the whitefly (*Bemisia tabaci*) (Maruthi et al. 2005). The disease began to spread in Uganda around 2004 (Alicai et al. 2007) and since then, the epidemic has spread around the Great Lakes Region to affect Eastern Uganda, Western Kenya, the Lake Zone of Tanzania, Rwanda, Burundi and the DRC (Patil et al. 2015). The 'syndrome' of CBSD includes symptoms on the leaves stem and roots but it is the symptom of root necrosis which is most damaging and can cause total crop loss in individual plants. Infection by the virus may cause a decrease in root weight, the extent of which depends on cassava variety. The main source of crop loss is the symptom of root necrosis that makes the root partly or completely unusable for human consumption. Root symptoms are often not thoroughly assessed due to the need for destructive sampling of large numbers of plants.

As cassava becomes increasingly used as a source of value added products and income for smallholders, CBSD poses a potential constraint to root supply for domestic and commercial processing. The activities described were conducted to assess the impact of CBSD on food security and income generation. This was done by surveys in parts of four East African countries where high incidences of CBSD leaf symptoms have been reported.

#### **Material and Methods**

Three villages were chosen for the survey in each of Uganda and Tanzania [2 locations -Lake Zone and Southern Zone] but in Malawi, only 2 villages were included. In Kenya 22 farms were surveyed in Migori County where homesteads tend not to form discrete villages. The target villages were located in areas known to have a high incidence of CBSD. Ten cassava fields per village were surveyed and these were chosen because the households were members of existing farming groups or chosen by extension officers or village heads. Disease incidence for CBSD and a second virus disease, cassava mosaic disease (CMD) based on foliar symptoms, was assessed in May/June 2013 on 30 plants per field (first 30 plants encountered on a diagonal transect across the field, excluding field margins] and variety names were also recorded. Plants showing leaf symptoms of CBSD were marked with coloured tags or paint. Leaf symptoms are clearest during the wet period when foliage is green and there is less damage from green mite. Root symptoms begin to appear around six months after planting and it is better to wait until at least eight months after planting before sampling for root necrosis. Therefore, CBSD root necrosis severity was assessed in July/August in the same fields where the leaf symptoms had been recorded, by harvesting 15 plants per field. The harvested plants were those which had been marked as showing leaf

symptoms or were still showing above ground symptoms at the time of uprooting. Fifteen plants per field or 150 per village, was the maximum number that could be realistically uprooted and assessed within one day. After uprooting the 15 plants, total root weight was recorded for each plant, the roots were then cut longitudinally, up to a maximum of six roots per plant and examined for signs of root necrosis. Root necrosis was scored on a 1 - 5 scale for each of the roots (Fig 1) and score 1 was included in calculating mean severity. Over 1500 plants were uprooted during the course of the survey.

In order to estimate the impact of losses due to CBSD in each village, the proportion of each variety grown was noted and it was assumed that a CBSD root necrosis score of three or above represented a total loss for both household consumption and for processing. Crop losses were estimated based on the frequency of occurrence of each variety in the village and proportion of roots with root necrosis score greater than three. Previous research has shown that roots with a necrosis score >3 are unsuitable for processing and usually rejected by processors (Abayomi et al. 2015). Losses per village were calculated from the percentage of each variety showing a score of >3 multiplied by the proportion of the total cassava crop in the village represented by that variety.

For comparison and more relevant to household food security when necrotic parts of CBSDaffected roots may be cut out before cooking, a second method of loss estimate was used, whereby the percentage of roots showing necrosis grades 3, 4 and 5 was calculated for each farm surveyed and a mean for each grade obtained for the village. The estimate for loss was obtained by multiplying the mean % of each grade by 25%, 35% and 58% which represents the average proportion of root tissue lost during cutting out the necrotic areas for CBSD necrosis grades 3, 4 and 5 respectively (Abayomi et al. 2015).

#### Results

In general across the four countries the results were similar but with important differences in each country. The mean incidence of CBSD leaf symptoms was high in each country, ranging from 45% in Uganda to 67% in the Lake Zone of Tanzania. The incidences of CMD were lower than for CBSD, ranging from only 3% in the Lake Zone of Tanzania to 48% in Migori County of western Kenya (Table 1). In contrast to the results for leaf symptoms, the incidence of CBSD root necrosis was low. Even in the varieties that were more prone to root necrosis, the mean root necrosis score was low due to the high proportion of symptomless roots. The association between CBSD leaf incidence and root necrosis score was tested with Spearmans' Rank Correlation Test but there was no correlation (results were non-significant for all sites so data not presented), due to a number of varieties showing high leaf incidence but little root necrosis. Differences in root necrosis severity between the varieties became more apparent if the proportion of roots with a severity score of three or above was used as the measure.

#### Uganda

A single variety, 'Migyera' (TMS 30754) was dominant in all three villages in Uganda but particularly at Nandere and Kakuja, where it was more than 90% of the crop in the surveyed fields. With the exception of 'Bibina' at Natoto Village, the other varieties were seen in small numbers in the fields or at only one farm. Migyera showed a very low incidence of CBSD leaf symptoms and little root necrosis (Table 1). The same was true of the variety TME204 on which root necrosis did not reach severity score three. There were three local varieties that showed very high incidences of leaf symptoms and significant levels of root necrosis, especially 'Namuyangu' and 'Bibina'. CMD was present at high incidences on Migyera in some fields, reaching 57% in one village. 'Bibina' had a high incidence (75%) and 'Kabaratekere' had a moderately high incidence (38%), while it was only about 10% in the other two varieties (Table 1).

Crop loss due to CBSD (assuming root necrosiss core >3 represents a total loss) was low at Nandere (1.9%) and Kajuka (3.3%) villages, due to the high proportion of Migyera planted in those villages (Table 2). At Natoto Village the crop loss was much higher at 13.9%, due to significant planting of the susceptible variety, Bibina.

#### Tanzania Lake Zone

In Sengerema District of Tanzania, there was more diversity of cassava varieties than in Busogo District of Uganda. 'Mkombozi' is an improved variety regarded as having some resistance to both CBSD and CMD. The others in the survey were described to us with local names but some may have been introduced improved varieties. The incidence of CBSD leaf symptoms was high, reaching 90% in some fields. Mkombozi showed the lowest incidence but leaf symptoms in the others ranged from 41 - 94%. The highest incidences occurred in the varieties 'Ngarabuto' and 'Lufunganya'. CMD incidence was very low in the area surveyed (Sengerema District) and none of the main varieties (except Ngarabuto at 13%) had a mean incidence greater than 5% (Table 1).

Severe CBSD root necrosis was rare and the majority of roots were disease-free, resulting in low scores for mean severity of root necrosis. The highest necrosis severity occurred in the varieties 'Busanagulwa' and 'Lufaili'. The varieties 'Malamogonzobe' and Ngarabuto showed a high incidence of CBSD leaf symptoms but low root necrosis scores. Four varieties had significant levels of root necrosis >3, including Mkombozi (Table 2). Crop loss was only 3.1% at Kijuka village and 3.7% at Nyamezeze but it was higher (13.5%) at Nyampande, due the growing of two varieties that are prone to root necrosis (Table 2).

#### Tanzania Southern Zone

CBSD leaf symptoms were widespread in all three villages surveyed and CBSD incidence was highest in Hiyari village reaching over 50%, while the incidence of CMD was low in all three villages, ranging from 7 - 23% (Table 1). CBSD incidence was highest in the varieties 'Vincent' at Mkunwa village, in 'Lihipukalyene' at Maranje village and in 'Nachinyaya' at Hiyari village. No varieties were completely free of leaf symptoms.

Root necrosis was most severe in the varieties Lihipukalyene and Mtukane and least in Nachinyaya (Table 1).

CMD incidence was generally low to moderate and above 20% only in the varieties Nakuchima and Lihipukalyene. The lowest incidence of CMD was found in the variety Vincent (Table 1).

Crop loss in individual varieties ranged from less than 1% in Nachinyaya to over 4% in Kigoma Red and Mtukane (Table 2). Total crop loss to CBSD, assuming that a score of three or above represented total crop loss, was 11% at Maranje and below 10% at both Mkunwa and Hiyari.

## Kenya – Migori County

The incidence of CBSD leaf symptoms was high (above 50%) in most of the fields visited and in all varieties, except 'Migyera'. CMD was also present at high incidences, ranging from 40 - 69% in all varieties except Migyera which had a mean incidence of 15% [lower than in Uganda] (Table 1). The favoured varieties, Weite, Nyakasanya and Manchoberi represented about 55% of the plants examined during the survey. Root necrosis at a severity of three or above was rarely seen, except in a few highly susceptible varieties which were grown in only one or two fields. No root necrosis was seen in the varieties 'Nakasanya' and one referred to by farmers as 'Agriculture MH'. Another variety, 'Mwitamajera' had slight necrosis which did not reach score three. The varieties showing most necrosis were Agric 2, 'Nyakahonda' and 'Amakuria'. The variety Nyakasanya had one of the highest incidences of CBSD leaf symptoms but no root necrosis (Table 2). Two varieties stand out as exhibiting the type of tolerance to CBSD that has been seen elsewhere i.e. high susceptibility to infection by the virus but low propensity for root necrosis – Nyakasanya and Mwitamajera. The only variety to show both low leaf incidence and low root necrosis was Migyera.

Losses for individual varieties ranged from 0 - 1.6% of the crop, with a total loss estimate from all the varieties examined in Migori County of 7% (Table 2).

## Malawi

As in Uganda, the number of varieties grown was very few in the two villages surveyed in the Lake region of Malawi, with a heavy reliance on the variety '20:20' which accounted for more than 50% of the surveyed crop. CBSD leaf incidence was very high , reaching over 90% in some fields. The variety 'Sauti' had the lowest incidence but still reached 63% in one village. CBSD incidence was 90% in both villages for the variety 20:20. Mean CMD incidence across the varieties was 27% but only 1-3% of the plants of Sauti showed CMD symptoms (Table 1).

Despite the high incidence of CBSD leaf symptoms, root necrosis incidence and severity was low, even in 20:20. 'Matakolembwende' was more prone to root necrosis than either Sauti or 20:20 and Sauti was the least prone (Table 2). Total crop loss in the two villages was 7%.

## Household Crop Loss

Table 3 shows the crop loss estimates based on percentage of each necrosis grade in the roots examined and adjusted for the proportion of necrotic tissue that must be removed before processing or cooking. The losses range from 0.8% in Kachaka village in Malawi to 6.4% in Maranje village in Southern Tanzania. If we assume that a root necrosis severity score of three or above constitutes a total crop loss, then the mean loss of root yield due to CBSD across 14 villages in four countries in eastern Africa, was estimated at 7%. This figure goes down to 3.2% if it is assumed that households are able to remove the necrotic portions of the roots before cooking.

#### Discussion

In Uganda Migyera and TME 204 are both 'improved' varieties and Migyera in particular has become widely distributed. It was first released during the 1990s as a CMD-resistant variety and became widely distributed during the CMD epidemic at that time. Its popularity and adoption has further increased, as it turned out to be resistant to CBSD.

The almost total reliance on a single variety (Migyera) is due to its resistance to CBSD, most of the susceptible varieties having been rejected by farmers. However, this makes the communities highly vulnerable to food insecurity should Migyera succumb to CBSD at a later stage or to any other pest or disease. A similar situation exists in Malawi where much of the cassava germplasm has been lost due to its being prone to CBSD root necrosis. The predominant varieties remaining are only three in number with heavy reliance on the variety 20:20. Cultivation of the variety Sauti is likely to increase due to its resistance to both CBSD and CMD.

Most of the cassava crops grown in Sengerema District of the Lake Zone in Tanzania are of varieties with a low propensity for CBSD root necrosis and this is unlikely to have come about by chance. CBSD root necrosis has such a conspicuous impact on the household's ability to use them for consumption, that those with a high propensity for root necrosis have been rapidly discarded. Crop losses to CBSD root necrosis averaged 7% for the three villages surveyed but this might have a significant impact on food security, given that root yields were already very low due to the exhausted sandy soils in the area.

The low incidence of CMD in the Lake Zone may be due to the dissemination of CMDresistant varieties. The origin of the varieties is often unknown by farmers once they are given a local name and become distributed from farmer to farmer. Otherwise it is unusual to find such a low incidence of CMD on local 'land races'.

The variety Kifuu cha Nazi (data not shown due to low sample numbers) seen in the Southern Zone of Tanzania was interesting in having a relatively low incidence of leaf symptoms but a high incidence of severe root necrosis. This has implications in breeding for resistance to CBSD, as a lower susceptibility to infection may not be useful if the variety develops severe root necrosis in those plants that do become infected.

Because farmers eventually replace varieties highly prone to root necrosis with those that are less prone, the impact of CBSD is less than would otherwise be the case. Much of the crop can be lost when the predominant varieties are highly prone to root necrosis which is likely to be the case in the new epidemic areas. A second study linked to the survey reported herein, investigated the effect of CBSD root necrosis on the use of cassava for processing into flour. Roots with a necrosis severity score of three or above were unsuitable for flour production. When cassava was scarce processors were sometimes forced to use roots with mild CBSD symptoms by cutting out the necrotic portions. However, this would have a negative impact on the profitability of the processing enterprise (Abayomi et al. 2015).

Farmers can decrease the risk of losses to CBSD by adopting varieties that are less prone to root necrosis and taking cuttings for re-planting only from plants that are not showing CBSD leaf symptoms. However, in the longer term, research continues to develop high yielding varieties with true resistance [i.e. less infection and no root necrosis] to CBSD combined with resistance to CMD.

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**Table 1** Disease incidence of CBSD and CMD based on leaf symptoms and severity ofCBSD based on root necrosis

**Table 2** Crop loss estimates due to CBSD root necrosis for main varieties grown [assuming a score of 3 or above represents a total loss]

**Table 3**. Proportion of roots with CBSD necrosis score of 3, 4 and 5 and crop loss estimate based on proportion of root tissue lost when necrotic areas are cut-out<sup>1</sup>

Fig 1. Longitudinal sections of cassava roots showing severity grades 1-5 for CBSD root necrosis

Country	Village	Varieties [top 3 in village]	Mean CBSD % [leaf symptoms]	Mean CMD %	Mean CBSD root necrosis Score [1-5]	% roots with CBSD score above 3
Uganda	Nandere	Migera TME204	4 0	38 8	1.02 1.25	1.6 0.0
	Natoto	Namuyangu Migera Bibina	100 0 92	7 57 75	2.88 1.12 1.73	56.0 7.3 22.6
	Kakuja	Migera Kabaratakere	0 75	12 38	1.02 1.50	1.1 33.0
MEAN**			45%	39%		
Tanzania [Lake]	Kijuka	Mkombozi	25	0	1.43	10.5
	<b>,</b>	Mwanaminzi Malamogonzobe	61 77	5 5	1.36 1.02	4.0 0.8
	Nyampande	Busanagulwa Lufaili	72 41	0 2	1.57 1.70	15.7 20.5
	Nyamezeze	Lufunganya Ngarabuto	57 94	2 13	1.52 1.22	0.4 3.3
MEAN**		Lufunganya Mwanaminzi	93 85 67%	0 0 3%	1.29 1.26	6.8 3.4
Tanzania [Mtwara]	Mkunwa	Kigoma Red Vincent Nakuchima	36 57 52	19 2 20	1.23 1.44 1.20	6.9 12.6 5.5
	Maranje	Musa Nakuchima	32 31	14 34	1.43 1.38	13.8 16.0
	Hiyari	Lihipukalyene Kigoma Red Mtukane	49 52 41	20 3 9	1.73 1.44 1.68	18.7 13.0 17.5
MEAN**		Nachinyaya	74 47%	8 13%	1.35	2.3
Kenya	Migori County	Merry go round Migyera	52 8	41 15	1.10 1.14	1.7 5.0
		Nakasamuel Obarodak	60 78	40 55	1.53 1.69	8.0 12.0
		Ondielo Agric MH	64 60	69 52	1.31 1.07	4.0 0.0

**Table 1** Disease incidence of CBSD and CMD based on leaf symptoms and severity of CBSD based on root necrosis

**Table 2** Crop loss estimates due to CBSD root necrosis for main varieties grown [assuming a score of 3 or above represents a total loss]

Country	Village	Variety	Variety Frequency %	Necrosis score % >3	Crop loss %	Total yield loss in village %
Uganda	Nandere	Migera	83	1.6	1.3	
		TME204	7	0.0	0.0	
		Namuyangu	1	56.0	0.6	1.9
	Natoto	Migera	57	7.3	4.2	
		Bibina	43	22.6	9.7	13.9
	Kajuka	Migyera	93	1.1	1.0	
		Kabaratakere	7	33.0	2.3	3.3
		Area Mean				6.4
Tanzania [Lake]						
	Kijuka	Mkombozi	8	10.5	0.8	
		Mwanaminzi	52	4.0	2.1	
		Malamogonzobe	22	0.8	0.2	3.1
	Nyampande	Busanagulwa	24	15.7	3.8	
		Lufaili	47	20.5	9.6	
		Lufunganya	12	0.4	<0.1	13.5
	Nyamezeze	Ngarabuto	60	3.3	2.0	
		Lufunganya	22	6.8	1.5	
		Mwanaminzi	6	3.4	0.2	3.7
		Area Mean				6.8
Tanzania [Southern]						
	Mkunwa	Kigoma Red	33	6.9	2.3	
		Vincent	33	12.6	4.2	
		Nakuchima	11	5.5	0.6	7.1
	Maranje	Musa	29	13.8	4.0	
	-	Nakuchima	24	16.0	3.8	
		Liphipukalyene	16	18.7	3.0	10.8
	Hiyari	Kigoma Red	38	13.0	4.9	
		Mtukane	27	17.5	4.7	
		Nachinyaya	15	2.3	0.3	9.9
		Area Mean				9.3
Kenya						
-	Migori	Merry go round	9	1.7	0.2	
	County	Migyera	4	5.0	0.2	
		Nakasamuel	2	8.0	0.2	
		Obarodak	2	12.0	0.2	
		Ondielo	4	4.0	0.2	
			2		0.0	
		Agric MH	2	0.0	0.0 1.6	
		Agric 2		79.4		
		Nyakasanya	7	0.0	0.0	
		Chombololo	2	0.0	0.0	

**Table 2** Crop loss estimates due to CBSD root necrosis for main varieties grown [assuming a score of 3 or above represents a total loss]

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		Bibina	43	22.6	9.7	13.9
	Kajuka	Migyera	93	1.1	1.0	
		Kabaratakere	7	33.0	2.3	3.3
		Area Mean				6.4
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		Mwanaminzi	52	4.0	2.1	
		Malamogonzobe	22	0.8	0.2	3.1
	Nyampande	Busanagulwa	24	15.7	3.8	
		Lufaili	47	20.5	9.6	
		Lufunganya	12	0.4	<0.1	13.5
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		Ondielo	4	4.0	0.2	
			2		0.0	
		Agric MH	2	0.0	0.0 1.6	
		Agric 2		79.4		
		Nyakasanya	7	0.0	0.0	
		Chombololo	2	0.0	0.0	

**Table 3**. Proportion of roots with CBSD necrosis score of 3, 4 and 5 and yield loss estimate based on proportion of root cut-out<sup>1</sup>

Country	Village	Total No of roots examine	% with score 3 % ved	with score 4 %	ś with score
Tanzania [Lake Zone]	Kijuka	465	3.6	0.9	0.2
	Nyampande	538	10.2	2.6	3.6
	Nyamezeze	480	4.5	1.4	0.5
Tanzania [Southern Zone]	Mkunwa	709	7.2	1.5	1.6
	Maranje	733	10.4	3.1	4.6
	Hiyari	735	4.6	2.5	2.9
Malawi	Chambogho	292	10.4	3.9	0.0
	Kachaka	667	2.9	0.3	0.0
Kenya	Migori County	1427	5.1	1.8	1.0

<sup>1</sup>Uganda not included because of the dominance of a single variety [Migyera] that is resistant to CBSD

<sup>2</sup>Estimated from research that shows on average 25%, 35% and 58% of tissue is removed when necrotic areas are cut necrosis grades 3, 4 and 5 (Abayomi et al. 2015)

#### SCORE 1: NO NECROSIS SYMPTOMS



SCORE 2: TRACE OF NECROSIS



SCORE 3: CLEARLY DEFINED AREAS OF NECROSIS BUT NECROTIC AREAS CAN BE EASILY REMOVED



SCORE 4: MOST OF ROOT NECROTIC BUT MAY STILL BE POSSIBLE TO EMOVE NECROTIC AREAS FOR HOME CONSUMPTION



SCORE 5: MOST OR ALL OR ROOT NECROTIC AND UNSUITABLE FOR HUMAN CONSUMPTION



Fig 1. Longitudinal sections of cassava roots showing severity grades 1 – 5 for CBSD root necrosis