Pathogenicity of *Sclerotium rolfsii* on cocoyam varieties in Ghana

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ABSTRACT

The pathogenicity of five strains of *Sclerotium rolfsii* on four varieties of *Xanthosoma mafaffa*, identified as 'Amankani fitaa', 'Amankani fufuo', 'Amankani kyirepe' and 'Amankani pa', and *Colocasia esculenta* (Synonym: *Colocasia antiquorum*) in Ghana was investigated using the prescribed pathogenicity/ infection tests. All the cocoyam varieties were susceptible to the five strains at different rates. After 20 days of inoculation, all cocoyam plants were infected; leaves were dead and the cormels were heavily rotted. The rot progressed from top downwards. However, cormel rot was greater at the basal region than at the apical region. Strains SrXA1 and SrXA2 caused the greatest rot in wound-inoculated cormels, while infection of 3-month-old plants proceeded fastest in soils inoculated with strains SrEL1 and SrEL2. The pathogenicity was greatest on *X. mafaffa* var. 'Amankani fitaa' and least on *X. mafaffa* var. 'Amankani kyirepe,' especially by the rotting activity of strains SrXA1, SrEL1 and SrEL2. Considering the pathogenicity of all five strains on the cocoyam varieties tested, *C. esculenta* was most vulnerable to the tested *S. rolfsii* strains in Ghana.

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Introduction

Sclerotium rolfsii, a facultative soil parasite of economic importance, causes disease of several world economic crop plants as a result of its ability to produce oxalic acid and cell-wall degrading enzymes, and its fast growth rate (Anahosur, 2001; Punja, 1985). It causes damping-off of seedlings and soft rot at the collar region, stem or root rot of crop plants (Aycock, 1966; Bowen, Hagan & Weeds, 1992; Cilliers Herselman & Pretorius, 2000; Cilliers, Pretorius & Wyk Van, 2003; Punja, 1985). In Ghana, it has been found to cause cormel and root rot on *Xanthosoma mafaffa* and *Colocasia esculenta* (Addison & Chona, 1971). The disease takes the form of a wet root rot, yellowing and wilting of leaves and failure to form cormel, followed in severe cases by death. It affects plants of all ages. *Sclerotium rolfsii* also causes post-harvest rot of the cormel. Decaying cormlets are usually coated with a typical mycelial felt of the fungus. *Sceratium rolfsii* invades the host through wounds and natural openings, and by direct penetration of intact surface tissue of the host.

Xanthosoma mafaffa, *C. esculenta*, *Manihot esculenta*, *Dioscorea* spp., and *Ipomea batatas* provide the main source of carbohydrate for most people in rural and urban communities in Ghana. Among root crops in Ghana, *X. mafaffa* can be regarded as important because they keep well in the field and in storage; and are, therefore, available throughout the year. The uses of cocoyam varieties in Ghana are many. The tender leaves are the main if not the only source of spinach for the inhabitants of the humid forest of Ghana and most urban community dwellers. The protein content per 100 g dry weight of the cocoyam leaf is 22.17 g (Maduewesi & Onyike,

1980). The cormel supplies easily digestible starch and is known to contain substantial amounts of protein, vitamin C, thiamine, riboflavin, and niacin (Maduewesi & Onyike, 1980). The leaves and tender parts of the stem are relished by livestock. The peelings of the cormel are also fed dried or fresh to goats, sheep, cattle, and pigs. The cormel is used in different forms (Karikari, 1971). It is peeled and boiled and pounded into 'fufu' and eaten with soup, a popular Ghanaian food. The boiled cormel is often also eaten directly with stew. The unpeeled cormel is roasted and the skin removed when ready for eating. The boiled or roasted form may also be mashed and palm oil added and eaten with either fish or roasted groundnut.

Addison & Chona (1971) estimated the damage caused by the fungus in Ghana in the range of 5-30 per cent. Now, interest in cultivating cocoyam by farmers has declined considerably because of the large quantity of *X. mafaffa* and *C. esculenta* destroyed annually by the fungus in the country. The reduction in the cultivation of these crops is affecting the food security of the country because most people depend on them, as well as the export market for non-traditional crops from the country.

In most pathogenic fungi such as *S. rolfsii,* variation within species is common. Many parasites exist in various forms, strains, and varieties associated with specific hosts; thereby widening the host range and spectrum of host species. A typical example is *Fusarium oxysporum,* in which the various forms are named after their specific host plants (Booth, 1971). Strains of *S. rolfsii* have been identified to be distinct in their ability to use carbon and nitrogen compounds, as well as in their enzyme activity, which indicates variation in the production of oxalic acid, and causes damping-off of seedlings and soft rot at the collar regions of economic crops (Ludwig & Haltrich, 2002; Punja & Damiani, 1996; Punja,1985).

There have been limited studies on the susceptibility of the various cocoyam varieties cultivated in Ghana to this widespread soil facultative parasite. This paper reports the pathogenicity of *S. rolfsii* strains, with special reference to collar rot and cormel rot in *X. mafaffa* varieties: 'Amankani fitaa' (white cocoyam), 'Amankani fufuo' (light-coloured cocoyam), 'Amankani kyirepe' (hard cocoyam), and 'Amankani pa' (proper cocoyam); and *C. esculenta* (kooko) in Ghana.

Materials and methods

Isolation of S. rolfsii *strains, maintenance of stock cultures and preparation of inocula*

Mature brown-coloured sclerotia were carefully removed with a pair of sterile fine forceps from the host plant and placed in distilled water to wash off any adhering soil particles. The sclerotia were then surface-sterilized by immersing them in 1.0 per cent sodium hypochlorite for 3 min, rinsed after that in three changes of sterile distilled water and then inoculated onto sterile potato dextrose agar (PDA) in sterile petri dishes. After 4 days the growing mycelium was subcultured. A second subculturing applied to ensure a completely pure culture.

Five different strains of *S. rolfsii* were collected at different locations in Legon and Achimota from two host plants and designated according to their sources as follows: Strain SrXLL from *X. mafaffa* at Legon campus; Strain SrXA1 from *X. mafaffa* at West Legon; Strain SrXA2 from *X. mafaffa* at Achimota; Strain SrEL1 from *Elaeis guineensis* at Legon campus; Strain SrEL2 from *E. guineensis* at West Legon. Stock culture of each *S. rolfsii* strain was maintained on PDA slants in MaCartney's tubes in the refrigerator, and subcultured fortnightly. Whenever inoculum was required for experiment, the fungus was raised on PDA in sterile Petri dishes for 4 days and inoculum collected from the advancing edge of the culture of the mycelium. All chemicals used in the investigation were from Oxoid Limited, Basingstoke, Hampshire, England, United Kingdom.

Cocoyam varieties

Five varieties of cocoyam in Ghana were used in the study: four varieties of *X. mafaffa*, 'Amankani fitaa' (white cocoyam), 'Amankani fufuo' (light-coloured cocoyam), 'Amankani kyirepe' (hard cocoyam), and 'Amankani pa' (proper cocoyam); and *C. esculenta*. All varieties of cocoyam in Ghana have been described (Abbiw, 1990; Karikari, 1971). The cocoyam varieties were supplied by the Agriculture Research Station of the University of Ghana at Kade, Ghana. Both cormels and petioles were used in the study. The cocoyam varieties were raised in garden loam soil in black polythene bags (36.5 cm × 22.5 cm) with drainage holes at the bottom for 12 weeks in a greenhouse before use.

Infection of cocoyam plants growing in S. rolfsii *-inoculated soils*

Four varieties of *X. mafaffa*, 'Amankani fitaa', 'Amankani fufuo', 'Amankani kyirepe' and 'Amankani pa'; and *C. esculenta* were grown in separate pots in garden loam soil of *p*H 5.0 and 25.0 per cent water holding capacity. The plants were allowed to grow for 3 months and then inoculated. The inoculum for each pot consisted of mycelium raised in 30 ml potato dextrose broth (PDB) for 6 days at 30 °C. The contents of one flask were stirred into the soil of one pot (36.5 cm × 22.5 cm) and the mouth of the pot covered with black polythene bag for 2 days to enable the fungus become established in the soil. Owing to the drift of the culture filtrate to acidic *p*H during growth of *S. rolfsii*, the mycelium was first washed with sterile distilled water before being used as inoculum. There were four replicate plants in each test. The cumulative number of petioles infected was recorded after 4, 8, 12, 16 and 20 days. In the control treatment, no inoculum was used.

Rotting of wound-inoculated cormels of cocoyam varieties by five S. rolfsii *strains*

Mature cocoyam cormels were inoculated with the different strains of *S. rolfsii* and the rotted areas measured after 8 days. Mycelium was raised in PDB at 30 °C for 4 days and used as inoculum. The culture filtrate was poured off and the mycelium rinsed with sterile distilled water. The mycelium was then transferred into a sterile Petri dish containing 10 ml sterile distilled water and macerated with a pair of blunt forceps. The cocoyam cormels were surface-sterilized and placed in 24.0 cm × 12.0 cm × 6.0 cm plastic boxes with tight-fitting lids. A plug, 3 mm deep, was then removed with a 5-mm cork borer from opposite sides, at the apical and basal halves of the cormel. Into each of the four cavities was placed the same amount of macerated mycelium, using a microspatula. The plug of cocoyam tissue removed was replaced and the wound sealed with thin vaseline. The inoculated cormels were incubated at 30 °C for 8 days. There were five replicates of each cormel of the four varieties of *X. mafaffa*, 'Amankani fitaa', 'Amankani fufuo', 'Amankani kyirepe' and 'Amankani pa'; and cormels of *C. esculenta*. The control cormels were not inoculated. Transverse sections of the cormels were cut through the site of inoculation, and the depth and the diameter of rot immediately beneath the cormel covering were measured, and the mean calculated for each treatment.

Degradation of blocks of tissues of cormels of cocoyam varieties by the five strains of S. rolfsii

Another way of assessing the ability of facultative parasites to attack host organs is to inoculate different organs *in vitro*. This experiment also used the apical and basal regions of the cormels separately. Cormels of the cocoyam were peeled and each was cut transversely into two halves. Mini-blocks, measuring 1.0 cm × 1.0 cm × 0.5 cm, were cut from the two regions and surface-sterilized with 5 per cent sodium hypochlorite for 5 min and rinsed in three changes of sterile distilled water, and drained on sterile filter paper. The initial fresh weights of the mini-blocks were determined using electronic balance (Electronic Balance ER-108A, A & D Co. Ltd, Tokyo, Japan). Five mini-blocks were placed on 4-day old PDA culture plates of *S. rolfsii*, and

incubated at 30 °C for 8 days. Three Petri plates of every treatment were withdrawn after 2, 4, 6 and 8 days. After the incubation period, the mycelium covering the blocks were carefully removed with a pair of fine forceps and the decomposing blocks were carefully lifted with a spatula and transferred to aluminium foil cups and dried in an electrically heated oven (Gallenkamp oven 300 plus series, A. Gallenkamp and Co. Ltd, London, England) at 80 °C for 48 h. The percentage loss in dry weight was calculated.

Data analysis

The data were analysed statistically using one-way and multifactor analysis of variance (ANOVA). The least significance difference (LSD) of the means was set at confidence limits of 95 per cent ($P < 0.05$) (Kershaw, 1973).

Results

Infection of cocoyam plants growing in S. rolfsii-*inoculated soils*

All the replicates of cocoyam plants were infected and some leaves were dead 20 days after inoculation, except the control treatment. The plants were dug at the end of the experiment and bisected longitudinally and examined. The cormels of all the treatments rotted heavily, the rot progressing from the top downwards. The plants were not infected at the same rate by the different *S. rolfsii* strains (Table 1). Infection proceeded slowest in *X. mafaffa* var. 'Amankani fufuo', in which some leaves still remained uninfected by the 16th day after inoculation for all *S. rolfsii* strains. The percentage infection was in the range of 84.6 – 93.3 per cent by 16 days of inoculation. Strains SrEL1 and SrEL2 had infected all the leaves of *X. mafaffa* var. 'Amankani fitaa' by the 12th day. The percentage infection was 84.6 – 100.0 (Table 1). This variety could be considered the most susceptible. All the leaves of the remaining three varieties had been infected by the 16th day by all the *S. rolfsii* strains (85.7 – 100%). The results showed that infection progresses fastest in the first 8 days after inoculation (30.8 – 78.6%), whereas infection rate in 4 days for all the cocoyam varieties was 30.8 – 53.8 per cent. None of the control plants showed any sign of infection.

Rotting of wound-inoculated cormels of cocoyam varieties by S. rolfsii strains

Fig. 1 and Tables 3,4,5 and 6 indicate that the basal region of the cormels rotted to a greater extent than the apical region, and the diameter of the rot, measured immediately beneath the cormel covering, was greater than the depth of the rot. The diameter and depth of the rot at the apical and basal regions were greatest in all the cocoyam varieties inoculated with *S. rolfsii* strain SrXA1 and SrXA2). (Tables 3,4,5 and 6). The two strains were, therefore, the most potent. Strain SrEL1 was the least active in all the varieties, except *X. mafaffa* var. 'Amankani fitaa' (Fig. 1a) and 'Amankani pa' (Fig. 1b), followed by strains SrEL2 and SrXLL. The diameter of the rot caused by Strain SrEL1 was in some cases markedly and significantly smaller statistically than those of rots caused by the rest (Table 2). The cormels of the different cocoyam varieties rotted to varying degrees by the different *S. rolfsii* strains. The deepest rot was observed in *X. mafaffa* var. 'Amankani pa' cormels (Fig. 1b), and the broadest rot diameter in cormels of *C. esculenta* inoculated with any of the *S. rolfsii* strains (Fig. 1c). None of the control-inoculated cormels showed any signs of rotting.

It was observed that the rot did not extend uniformly from the inoculated spot. The rot was wider near the exterior of the cormel, and relatively lesser penetration inwards was observed as shown by the depth and diameter measurements (Fig. 1). Because of the nature of the rot, it was impossible to calculate the linear rate of advance of the strains through the cormels. Cormels of *C. esculenta* were the most susceptible, and the greatest extent of rot was observed when they were inoculated especially with strains SrXA2 and SrXA1 (Fig. 1c). Among the *X. mafaffa* varieties, 'Amankani fitaa' and 'Amankani kyirepe' rotted to a lesser extent than 'Amankani fufuo' and 'Amankani pa' (Fig. 1a, b).

TABLE 1
Infection of Varieties of Xanthosoma mafaffa *and* Colocasia esculenta *Growing in Soil Inoculated with Different* S. rolfsii *Strains*

Cocoyam variety	*Strain*	*No. of petioles at time of inoculation*	*% petioles rotted in indicated number of days*				
			4	*8*	*12*	*16*	*20*
X. mafaffa	SrXLL	13	46.2	76.9	84.6	100.0	*
'Amankani fitaa'	SrXA1	13	38.5	69.2	76.9	100.0	*
	SrXA2	14	42.9	78.6	85.7	100.0	*
	SrEL1	13	46.2	76.9	100.0	*	*
	SrEL2	14	50.0	78.6	100.0	*	*
	Control	12	0	0	0	0	0
X. mafaffa	SrXLL	13	46.2	61.5	76.9	92.3	100.0
'Amankani fufuo'	SrXA1	13	46.2	61.5	69.2	92.3	100.0
	SrXA2	14	42.9	64.3	71.4	92.9	92.9
	SrEL1	15	46.7	66.7	80.0	93.3	93.3
	SrEL2	13	46.2	69.2	76.9	84.6	100.0
	Control	12	0	0	0	0	0
X. mafaffa	SrXLL	14	42.9	71.4	78.6	85.7	85.7
'Amankani kyirepe'	SrXA1	13	30.8	69.2	84.6	100.0	*
	SrXA2	12	41.7	75.0	83.3	100.0	*
	SrEL1	13	46.2	76.9	92.3	100.0	*
	SrEL2	13	38.5	76.9	92.3	100.0	*
	Control	12	0	0	0	0	0
X. mafaffa	SrXLL	12	50.0	75.0	83.3	100.0	*
'Amankani pa'	SrXA1	12	41.7	66.7	75.0	91.7	100.0
	SrXA2	13	46.2	61.5	69.2	92.3	100.0
	SrEL1	14	35.7	71.4	78.6	92.9	100.0
	SrEL2	13	53.8	69.2	76.9	92.3	100.0
	Control	12	0	0	0	0	0
C. esculenta	SrXLL	11	45.5	63.6	72.7	100.0	*
	SrXA1	14	50.0	71.4	78.6	100.0	*
	SrXA2	14	42.9	64.3	78.6	100.0	*
	SrEL1	14	42.9	64.3	71.4	92.9	92.9
	SrEL2	13	46.2	69.2	84.6	92.3	92.3
	Control	12	0	0	0	0	0

*: Dead cocoyam petioles

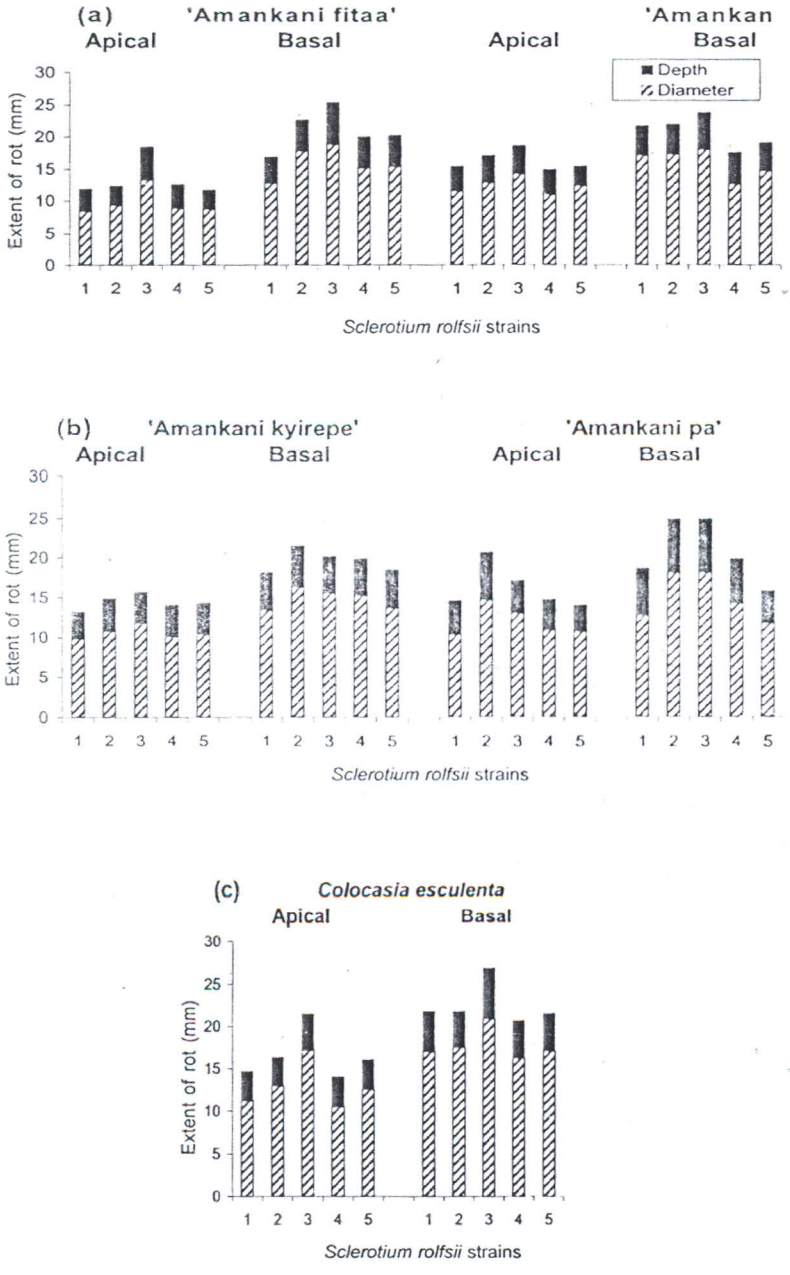

Fig. 1. Rotting at apical and basal region of wound-inoculated cormels of *Xanthosoma mafaffa* varieties (a) '*Amankani fitaa*' and '*Amankani fufuo*' (b) 'Amankani kyirepe' and 'Amankani pa' and (c) *Colocasia esculenta* by the five strains of *S. rolfsii* at 30°C for 8 days (1 = *S. rolfsii* strain SrXLL; 2 = *S. rolfsii* strain SrXA1; 3 = *S. rolfsii* strain SrXA2; 4 = *S. rolfsii* strain SrEL1; 5 = *S. rolfsii* strain SrEL2).

TABLE 2

Degradation of Tissues of the Apical and Basal Regions of the Cormels of the Different Cocoyam Varieties by the Different S. rolfsii *Strains at 30 °C over 8 Days*

S. rolfsii *strain*	*Period of Incub-ation*	*Percentag weight loss (%) of the two regions of cormels of* Xanthosoma mafaffa *variety*								*Percentage weight loss of the two regions of cormels of* Colocasia esculenta	
		'Amankani fitaa'		*'Amankani fufuo'*		*'Amankani kyirepe'*		*'Amankani pa'*			
		Apical	*Basal*	*Apical*	*Basal*	*Apical*	*Basal*	*Apical*	*Basal*	*Apical*	*Basal*
SrXLL	2	11.5±0.01	15.6±0.01	11.1±0.01	12.2±0.01	9.3±0.01	12.5±0.01	11.1±0.0	16.3±0.01	11.8±0.01	14.6±0.01
	4	28.3±0.01	34.8±0.01	14.8±0.02	17.4±0.01	12.5±0.01	19.1±0.01	19.2±0.01	25.5±0.01	17.3±0.01	25.5±0.01
	6	38.5±0.01	45.7±0.01	17.0±0.01	24.0±0.01	23.6±0.01	29.2±0.02	30.2±0.01	37.5±0.01	33.3±0.0	44.6±0.02
	8	44.0±0.0	46.0±0.01	30.0±0.02	40.0±0.02	32.0±0.01	40.0±0.0	34.6±0.02	38.8±0.02	34.6±0.01	48.8±0.02
SrXA1	2	11.3±0.01	19.6±0.01	14.8±0.0	18.8±0.00	15.7±0.01	19.1±0.0	13.5±0.02	18.0±0.01	15.4±0.02	18.8±0.01
	4	19.2±0.01	27.7±0.01	15.1±0.01	19.6±0.01	18.2±0.01	23.9±0.02	17.0±0.01	25.0±0.01	18.9±0.0	28.3±0.01
	6	35.8±0.01	44.4±0.01	26.0±0.01	30.6±0.0	21.8±0.02	30.0±0.01	28.8±0.01	39.6±0.01	28.8±0.02	35.6±0.01
	8	42.3±0.01	52.2±0.01	37.3±0.01	46.7±0.01	30.9±0.01	37.5±0.01	33.3±0.01	40.0±0.02	32.1±0.01	42.6±0.01
SrXA2	2	9.6±0.01	17.4±0.0	17.3±0.01	22.2±0.01	12.7±0.01	16.7±0.01	15.7±0.01	18.0±0.01	15.1±0.01	22.2±0.01
	4	13.2±0.01	26.7±0.01	28.8±0.01	31.1±0.01	21.4±0.02	29.2±0.01	25.0±0.01	27.7±0.03	23.1±0.01	33.3±0.01
	6	34.5±0.01	41.3±0.01	30.8±0.01	40.0±0.01	25.9±0.03	33.3±0.01	29.4±0.01	34.7±0.01	35.8±0.01	48.9±0.01
	8	43.1±0.01	54.3±0.0	38.0±0.0	47.7±0.01	34.5±0.01	44.7±0.01	34.6±0.01	40.8±0.01	43.4±0.01	56.5±0.01
SrEL1	2	15.1±0.0	20.0±0.0	13.0±0.0	19.6±0.0	7.5±0.01	10.4±0.01	11.8±0.01	16.7±0.01	13.2±0.01	18.8±0.01
	4	19.2±0.0	27.7±0.01	23.1±0.01	31.0±0.02	18.2±0.01	20.8±0.01	17.6±0.01	23.4±0.02	18.9±0.01	24.5±0.02
	6	35.8±0.01	43.5±0.01	32.7±0.01	41.3±0.0	18.5±0.0	25.0±0.01	28.0±0.00	34.0±0.01	23.1±0.01	47.9±0.02
	8	45.1±0.0	52.2±0.01	38.9±0.0	51.1±0.02	34.0±0.02	41.3±0.01	37.3±0.02	47.9±0.01	42.3±0.01	17.9±0.01
SrEL2	2	13.2±0.0	16.7±0.01	15.1±0.01	20.0±0.01	14.5±0.03	15.2±0.01	11.3±0.01	17.5±0.01	9.8±0.01	16.7±0.01
	4	26.7±0.0	22.2±0.01	26.4±0.01	28.3±0.0	17.3±0.02	21.3±0.03	15.4±0.01	22.2±0.01	15.4±0.01	20.8±0.02
	6	30.2±0.01	40.0±0.01	35.8±0.01	45.7±0.0	22.6±0.03	23.4±0.01	30.2±0.02	41.3±0.02	19.2±0.01	31.1±0.02
	8	42.6±0.02	51.1±0.01	42.6±0.02	52.2±0.01	30.2±0.01	34.0±0.01	36.5±0.01	45.7±0.01	30.8±0.01	37.5±0.01

TABLE 3

Rotting of Wound-inoculated Cormels of 'Amankani fitaa' by Five Strains of S. rolfsii *at 30 °C in 8 Days*

S. rolfsii *isolates*	*Rotting at inoculated apical region of cormel*		*Rotting at inoculated basal region of cormel*	
	Extent of rot (mm) ± S. E.		*Extent of Rot (mm) ± S. E.*	
	Diameter	*Depth*	*Diameter*	*Depth*
SrXLL	8.4±0.05a	3.4±0.03b	12.8±0.09a	4.0±0.03a
SrXA1	9.3±0.08c	3.0±0.03a	17.7±0.07c	4.9±0.03b
SrXA2	13.3±0.06d	5.0±0.04d	18.8±0.09d	6.5±0.04c
SrEL1	8.8±0.06b	3.7±0.03c	15.1±0.08b	4.8±0.03b
SrEL2	8.7±0.05b	3.0±0.03a	15.3±0.11b	4.9±0.03b
Control	0.0	0.0	0.0	0.0

By the calculated confidence limits at 95%, means in vertical rows bearing the same letters are not significantly different.

TABLE 4

Rotting of Wound-inoculated Cormels of 'Amankani fufuo' by Five Strains of S. rolfsii *at 30 °C in 8 Days*

S. rolfsii *strain*	*Rotting at inoculated apical region of cormel*		*Rotting at inoculated basal region of cormel*	
	Extent of rot (mm) ± S. E.		*Extent of rot (mm) ± S. E.*	
	Diameter	*Depth*	*Diameter*	*Depth*
SrXLL	11.5±0.05b	3.8±0.03b	17.1±0.11c	4.6±0.03bc
SrXA1	12.9±0.09d	4.1±0.03c	17.2±0.09c	4.8±0.04c
SrXA2	14.2±0.44e	4.4±0.02d	18.0±0.08d	5.8±0.04d
SrEL1	11.0±0.07a	3.9±0.03b	12.7±0.10a	4.8±0.03a
SrEL2	12.4±0.07c	3.0±0.02a	14.6±0.08b	4.4±0.03b
Control	0.0	0.0	0.0	0.0

By the calculated confidence limits at 95%, means in vertical rows bearing the same letters are not significantly different.

TABLE 5

Rotting of Wound-inoculated Cormels of 'Amankani kyirepe' by Five Strains of S. rolfsii *at 30 °C in 8 Days*

S. rolfsii *strain*	*Rotting at inoculated apical region of cormel*		*Rotting at inoculated basal region of cormel*	
	Extent of rot (mm)±S.E		*Extent of rot (mm)±S.E*	
	Diameter	*Depth*	*Diameter*	*Depth*
SrXLL	9.8±0.06a	3.4±0.03a	13.4±0.06a	4.8±0.04c
SrXA1	10.8±0.06c	4.1±0.04b	16.4±0.09c	5.0±0.03c
SrXA2	11.8±0.08d	3.9±0.02b	15.6±0.09b	4.5±0.03b
SrEL1	10.1±0.08ab	3.9±0.03b	15.3±0.07b	4.5±0.03b
SrEL2	10.4±0.07b	3.9±0.03b	13.6±0.08a	4.9±0.04a
Control	0.0	0.0	0.0	0.0

By the calculated confidence limits at 95%, means in vertical rows bearing the same letters are not significantly different.

TABLE 6

Rotting of Wound-inoculated Cormels of 'Amankani pa' by Five Strains of S. rolfsii *at 30 °C in 8 Days*

S. rolfsii *strain*	*Rotting at inoculated apical region of cormel*		*Rotting at inoculated basal region of cormel*	
	Extent of rot (mm)±S.E		*Extent of rot (mm)±S.E*	
	Diameter	*Depth*	*Diameter*	*Depth*
SrXLL	10.4±0.03a	4.3±0.04c	12.7±0.20b	6.0±0.10c
SrXA1	14.7±0.20d	6.0±0.10d	18.1±0.12d	6.8±0.07c
SrXA2	13.0±0.07c	4.3±0.04c	18.1±0.20d	6.8±0.10d
SrEL1	10.9±0.05b	4.0±0.03b	14.4±0.07c	5.4±0.04b
SrEL2	10.7±0.20ab	3.4±0.03a	11.8±0.08a	4.1±0.04a
Control	0.0	0.0	0.0	0.0

By the calculated confidence limits at 95%, means in vertical rows bearing the same letters are not significantly different.

Degradation of blocks of tissues of comels of different cocoyam varieties by five strains of S. rolfsii

Blocks of tissues of the cormels of the different cocoyam varieties were degraded at different rates (Table 2). The blocks from the basal half of the cormels were degraded to a greater extent than those from the apical half. Differences in percentage loss in dry weight by the 8th day of incubation ranged from 2.0 per cent in *X. mafaffa* var. 'Amankani fitaa' inoculated with *S. rolfsii* strain SrXLL to 14.2 per cent in *C. esculenta* inoculated with strain SrXLL (Table 2). Generally, for degradation of apical and basal regions, blocks of *X. mafaffa* var. 'Amankani fitaa' lost the greatest dry weight and *X. mafaffa* var. 'Amankani kyirepe' the least, especially by the activity of strains SrXA1, SrEL1 and SrEL2.

The varieties did not show the same order of susceptibility as was observed in the wound-inoculation experiments. The *X. mafaffa* var. 'Amankani kyirepe' again consistently showed the smallest loss in mean tissue dry weight (Table 2). However, in contrast to the trend in the wound - inoculation experiments, *C. esculenta* showed a lesser loss in mean dry weight than *X. mafaffa* var. 'Amankani fitaa', a less susceptible variety in the earlier experiment. This variety showed the greatest loss in mean dry weight (Table 2).

Discussion

The rotting of cormels indicates that the fungus entered the plant through the base of the outermost petiole and migrated downwards into the cormel, and transversely inwards to infect the inner petioles of the plant. The direction of infection of the petioles was centripetal. Cormels could be infected in two ways. The mycelium from the soil would first form infection cushions on the petioles, penetrate the epidermis of the petioles and grow in the petiole and, subsequently, invade the cormel. This is aided by the production of a number of extracellular, cell wall degrading enzymes including cellulases. In addition, an important pathogenesis event is the production of large amounts of oxalic acid (Cilliers *et al.*, 2000, 2003; Punja & Damiani, 1996). Blocks of tissues of the cormels placed on *S. rolfsii* cultures provided greater surface area to volume ratio to the hyphae, and oxygen concentration was not restricted as in the usual inoculation method. The mycelium may also enter the cormels through wounds. The length of period for infection to occur depends on the efficiency of bleaching the epidermis and the rate of growth and activity in the petioles.

The probability that factors in infected tissues could differ from observations made in pure cultures had been noted by many workers. Studies found that there were more compounds in apple fruits rotted by *Botrytis cinerea, Penicillium expansum, Pyrenochaeta furfuracea,* and *Sclerotinia fructigena* than in sound apples (Cole & Wood, 1961). The numbers of these compounds were greater in extracts of apple rotted by *P. expansum* and *P. furfuracea* than in extracts of rots produced by *B. cinerea* and *S. fructigena.* This shows that tissues rotted by different fungi, and also different strains, would contain different products from the breakdown of polysaccharides, which may have different effects on further activity of the rotting organisms. Alternatively, this indicates specificity of phytoalexins induced by different fungi when they invade same or different plant tissues.

The course of events in the rotting process of plant tissues by facultative parasites is well known (Kershaw, 1973). The parasite begins to grow into healthy plant tissue and starts to produce pectinesterase and chain-splitting enzymes. The latter cause some degradation of cell wall pectic substances, and the pectinesterase de-esterifies some high molecular-weight pectinases to form pectates. The cells of the host are killed, possibly as a result of the action of pectic enzymes on the cell walls; and phenols are brought together with host and parasite phenolases to form coloured oxidative products, some of which inactivate the chain-splitting enzymes (Anahosur, 2001; Cilliers *et al.*, 2003; Wood, 1967). The slower growth of the fungi into the inner regions of the cormels might be due to decreasing oxygen concentration with

increasing depth. Different amounts of oxidative products might have been formed in the rotting cormels of the different varieties, resulting in difference in their susceptibility.

In a study, all four strains of *S. rolfsii* tested on 12 crops infected all the plants (Epps, Patterson & Freeman, 1951). The relative degree of susceptibility of the crops to the four strains varied only slightly. The authors stated that all four strainscould kill essentially all the plants if vigorous inoculum was used. Furthermore, they reported that between two strains B and R, B strain was slightly more virulent than the R strain, but the difference was inconsistent and occasionally in the other direction. The results of the investigation reported in this paper provide another instance of the nature of strains of *S. rolfsii,* of which varieties *X. mafaffa* and *C. esculenta* are susceptible to varying degrees. The importance of the relationship between *S. rolfsii* strain and varieties *X. mafaffa* and *C. esculenta* involves not only the susceptibility of the variety, but also its role as substrate for sclerotium formation; hence the survival of the strain. Sclerotium production should naturally be influenced by compounds in the cormels, because glucose and other compounds have been found to influence sclerotium formation in *S. rolfsii* (Bowen *et al.*, 1992; Kershaw, 1973).

Conclusion

Relevant studies have been duly carried out on the pathogenicity of the five *S. rolfsii* strains on four varieties of *Xanthosoma mafaffa* ('Amankani fitaa', 'Amankani fufuo', 'Amankani kyirepe' and 'Amankani pa') and *Colocasia esculenta*. The five strains differed in their pathogenicity, and it is reasonable to suggest that, out of the five cocoyam varieties tested, *C. esculenta* was vulnerable to many *S. rolfsii* strains in Ghana. Commercial cultivation of cocoyam should focus on *X. mafaffa* varieties. To safeguard the stability of the soil ecosystem, it is proposed that good agricultural practices such as crop rotation should be used, which could be relied on to prevent onset of epidemics. The disease is easily detectable either by the death of the large leaves or by the conspicuous clusters of sclerotia on the infected plants. Infected plants should be uprooted and destroyed, the top soil in the area should be dug in, and after harvesting, the thrash should be burnt. As most parts of the plant is eaten, very little thrash is left behind, and its disposal is less formidable than it would seem. In addition to these field practices, only uninfected cormels should be stored so that the crop of the next planting season would be raised with clean materials.

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