Passion fruit collar rot disease occurrence in major growing districts of Uganda

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Abstract — Introduction. In Uganda, passion fruit (*Passiflora* spp.) production occupies some 500 ha, with an annual production of 3,500 t concentrated in the small scale sector. Its productive life span is primarily limited by diseases, amongst which collar rot caused by *Fusarium solani* is considered to be the most economically important. A study was undertaken to establish the occurrence of the disease in 16 major passion fruit growing districts. **Materials and methods**. Six fields were visited for each of the districts surveyed. Collar-rotting stem samples were collected from fields showing clear disease symptoms. In the laboratory, causal organisms were isolated and identified. **Results**. Out of the 16 districts surveyed, four had no collar rot attack, five had localised infection and seven had widespread collar rot infection. The disease was limited to the warm lowlands. Local purple (*P. edulis* f. *edulis*) was found to be attacked earlier than hybrid purple (*P. edulis* f. *edulis* x *P. edulis* f. *flavicarpa*). Isolates were consistently found to be *Fusarium solani* and to be growing best at 25–30 °C. Koch's postulates proved that it induces the disease. **Discussion**. The presence of virulent *F. solani* isolate in the soil could explain limited yield and production of the small purple passion fruit (*P. edulis* f. *edulis*), a susceptible species, in the lowland areas, as opposed to widespread production of the tolerant Kawanda hybrid passion fruit. The impact of factors like soil pH, inoculum density and grown species could influence disease ingress; thus, these factors need to be assessed in further field and laboratory studies. Emphasis should be put on evaluation of various passion fruit varieties for resistance. © Éditions scientifiques et médicales Elsevier SAS

Uganda / *Passiflora* / plant diseases / *Fusarium solani* / farm surveys / identification

Présence de la maladie de la pourriture rose du collet (prc) chez la grenadille, dans les principaux sites de production ougandais.

Résumé — Introduction. En Ouganda, 3 500 t de grenadilles (*Passiflora* spp.) sont produites chaque année par de petites exploitations, sur quelques 500 ha. Le développement de la plante est limité principalement par des maladies dont la (prc), causée par *Fusarium solani,* considérée comme économiquement la plus grave. Une étude a cherché à établir la distribution de cette maladie dans les 16 principaux districts producteurs de grenadilles. **Matériel et méthodes**. Pour chacun des districts contrôlés, 6 champs ont été visités. Des échantillons de tiges atteintes de (prc) ont été collectés. Les organismes incriminés par la maladie ont été isolés et identifiés en laboratoire. **Résultats**. Parmi les 16 districts visités, quatre n'ont montré aucune attaque de (pcr), cinq présentaient quelques infections et sept étaient largement colonisés par la maladie. Celle-ci s'est trouvée limitée aux zones de basse altitude. La grenadille violette locale (*P. edulis* f. *edulis*) a été attaquée plus tôt que l'hybride violet (*P. edulis* f. *edulis* × *P. edulis* f. *flavicarpa*). Le champignon isolé a été radicalement identifié comme du *Fusarium solani* ; son développement optimal se fait à 25–30 °C. L'application des postulats de Koch a montré qu'il induisait la maladie. **Discussion**. La présence d'isolats virulents de *F. solani* dans le sol pourrait expliquer les rendements limités de la grenadille violette (*P. edulis* f. *edulis*), espèce sensible, par rapport à la production plus importante de l'hybride Kawanda tolérant. L'impact sur la maladie de facteurs tels que le pH du sol, la densité d'inoculum et les espèces présentes devra être évalué dans de prochaines expérimentations en champ ou en laboratoire. L'effort devra porter sur la recherche de variétés résistantes. © Éditions scientifiques et médicales Elsevier SAS

Ouganda / *Passiflora* / maladie des plantes / *Fusarium solani* / enquête sur exploitations agricoles / identification

1. introduction

At present, some 500 ha are under passion fruit production in Uganda. The crop is produced by small-scale farmers with orchard sizes ranging 0.25–3 ha. Yield is 7 $t \cdot ha^{-1}$ and annual production is estimated to be 3,500 t of which about 150 t is yearly exported to Europe. The major factors limiting marketable yield being reported are diseases and poor post-harvest handling.

Passion fruit collar rot is an economically important disease to passion fruit growing in Uganda [1]. The disease causes a canker around the stem at the soil level possibly leading to plant mortality. Initial disease symptoms in the field are general weakness and reversible wilting on sunny dry days. On farm, the presence of the disease has been frequently reported by passion fruit growers, extension workers and horticultural researchers. The disease is claimed to cause over 50% crop loss.

As early as 1976, collar rot disease was reported attacking passion fruit in Uganda [2]. Elsewhere, it was reported in Australia [3], in South Africa [4], in Hawaii, in South Florida [5] and in Mauritius [6]. Due to its growing impact and economic importance, it was deemed necessary to study the magnitude of infestation and to establish the causal organism.

2. materials and methods

In 1993, sixteen major passion fruit growing districts in Uganda were surveyed. These were Lake districts of Tororo, Iganga, Mukono, Kampala, Mpigi, Masaka and Rakai; in central Uganda, Luwero, Kiboga and Mubende districts; in western highlands, Kasese, Kabarole, Hoima and Kiboga districts and in eastern highlands, Mbale district. For each district, six passion fruit growing farmers were visited with specific emphasis on those with infested orchards.

The size of orchard, species cultivated and total number of plants collar-rotten were recorded, in addition to soil type, vegetation and management practices. Samples of collar-rotten stems as well as soil were collected from visited fields. Stem sections were taken from the rotten area frontier and carried in paper bags. Soil samples were collected from the base of collar rotten plants in aluminium foil bags [7].

To isolate the causal organism from soil samples, passion fruits were used as baits [7]. Fruit was surface-sterilised with 10% sodium hypochlorite for 15 min. About 5 g of soil were put inside each surface-sterilised fruit and incubated at 28 °C for 7 d in a Gallenkamp incubator. Surface-sterilised stem sections were incubated at 28 °C for 3 d. The growing fungal culture was isolated onto PDA (Oxoid) media plates. Using aerial mycelium tips, isolation of the fungus was made by inoculating PDA plates. The growing fungus was subcultured using single spore semi-mechanical technique [7]. Water agar was prepared and spread even onto slides to form a thin layer (2 mm) and inoculated with conidia using a wet needle. After incubation at 28 °C for 15 h, germinating single spores were picked at × 100 magnification. To purify isolates, each spore was carefully picked and inoculated onto PDA plates. For each isolate, 20 germinating spores were picked.

The fungus was identified taking conidia size, length and width ratio, shape, septation and structure of top and foot cells, the nature of aerial mycelium, pigmentation, spore colour, presence of chlamydospores, sporodochia, pionnates, nature of phiallides and growth rate on PDA [8–10].

Koch's postulates were verified by drench-inoculating pots/sterilised substrate with isolate at conidia suspension concentration of 50,000 $conidia \cdot mL^{-1}$. Four pots, each with four *Passiflora edulis* f. *edulis* susceptible plants, were inoculated to test for isolate pathogenicity. Inoculation was done as soon as seeds germinated. The number of wilted plants was recorded and cause established.

3. results

Daily ambient temperatures in the region were found to vary from an average minimum 13 °C in the western highlands to an

average maximum 30 °C in the Lake Victoria basin lowlands. On average, rainfall was estimated to vary between 1125 and 1375 mm annually. Soil types varied between sandy loam and sand clay loams, with volcanic soils in the highlands and alluvials in western lowlands of Kasese district. Vegetation changes from forest to wooded savanna away from Lake Victoria basin. However, most forests have been reduced to cultivated land and pockets of bushland (*table I*).

It was noted that out of the 16 districts surveyed, four had no collar rot disease, five had localised infection, while seven had widespread infection of passion fruit orchards (*table I*). Mpigi district had highest level of infection with an average of five farmers having completely lost 3 ha of 2 year-old plants due to wilt caused by collar rot.

Unlike highland areas planted with local small purple species (*P. edulis* f. *edulis*), the Lake basin mostly has large purple Kawanda hybrid clones. According to all 72 respondent farmers interviewed, local small purple plants in the lowland districts were attacked by collar rot after 2 years from planting date.

In Mbale district, there was a remarkable variation between lowland and highland in the occurrence of passion fruit diseases. Lowlands had collar rot, brown spot and woodness virus, while highlands had no collar rot but had anthracnose, brown spot and woodness virus (*table I*).

For control of the disease, some farmers had splice-grafted seedlings with *P. edulis* f. *flavicarpa* acting as rootstock, and hybrid clones as scion. The yellow passion fruit is said to be tolerant to collar rot disease [4]. However, plants with graft union under soil surface were found to have collar-rotted.

On PDA medium, fungus with whitish grey aerial mycelium grew abundantly from diseased stem sections and fruit baits. The isolated fungus was observed to have septate and branching hyphae. Mycelium was leatherly and peeling off the PDA. Within, a medium, cream, tan, light brown or magenta pigmentation was produced. By growing the isolate under different temperatures (5, 10, 15, 20, 25, 30 and 35 °C), it was observed that its colour varied accordingly. At 10 and 15 °C, cultures had cream pigmentation, whereas they were cream to pinkish or light magenta at 20 °C; magenta at 25 and 30 °C; and again cream at 35 °C. When the same culture plate was put under varying temperatures, cream or magenta colours developed in alternating rings. Therefore, colour was confirmed not to be reliable in species identification. On the other hand, in vitro growth of the isolate studied using single spore cultures grown on PDA plates at the same temperatures as above (i.e., 5, 10, 15, 20, 25, 30 and 35 °C) indicated that the isolate grew best between 25 °C at a radial growth rate of 0.424 $mm \cdot h^{-1}$, and 30 °C at a mean rate of 0.4 $mm \cdot h^{-1}$. At 5 and 35 °C, growth was stopped (*table II*). The fungus grown in plates kept at 5 and 35 °C grew luxuriantly when transferred to 25 °C.

Both microconidia and macroconidia were found in aerial mycelium. Phiallides occurred on long septate and branched conidiophores. There were more microconidia (69%) than macroconidia (31%) in 2 week-old cultures. Three-septa macroconidia were more abundant, i.e., 15% of total number observed, than any other. There were 8% of 2-septa macroconidia (*table III*).

Macroconidia were very slightly bent, sausage-shaped or cylindrical with round apical cells slightly beaked and marked foot cells. Microconidia were observed to be cylindrical, oval, round or even oblong. Thick-walled chlamydospores were abundant, occurring in singles, doubles or even in chains. In all cases, the morphology of phiallides, conidiophores and conidia was consistent with the description of *Fusarium solani* by Joffe [9] and Nelson et al. [10]. Pathogenicity tests indicated that out of 16 *P. edulis* f. *edulis* plants inoculated by drenching, collar rot and wilt symptoms were reproduced on ten plants within 2 months, whereas another six plants were simply stunted. *Fusarium solani* was recovered from wilted plants, thus fulfilling Koch's postulates.

Table I.
Geographical factors of districts visited in Uganda, with passion fruit species grown, existing diseases and their level of occurrence.

District altitude (m)	Temperature (°C)	Annual rainfall (mm)	Natural vegetation	Soil type	Disease susceptibility				
					Collar rot	Brown spot	Anthracnose	Woodness virus	Variety
Mukono 1500	17–30	125	Forest / Savanna	Sand, clay, loam	++	+	+	+	Hp
Mpigi 1500	17–30	125	Forest / Savanna	Sand, clay, loam Sand, loam	+++	+	+	+	Hp^+ / Lp
Mbale 1200–3000	15–30	125–140	Savanna / Montane	Sand, clay, loam Sand	– +	+ +	++ –	++ +	Hp Lp^+
Tororo 1200	17–35	125	Woody savanna	Sand, loam	+	+	+	+	Hp^+
Iganga 1200	16–30	125	Forest / Savanna	Sand, loam	+	–	–	+	Hp+ / Lp
Mubende 1500	16–28	100	Forest / Woody savanna	Sand, loam Sand, clay, loam	+	+	–	+	Hp / Lp^+
Kiboga 1700	15–28	100	Woody savanna	Sand, clay, loam	+	+	–	+	Lp^+ / Hp
Kasese 1500–3000	15–27	115–250	Savanna Montane	Alluvial Ferric, humic Volcanic	–	++	–	+ +	Lp^+
Kabalore 1000–3000	13–25	140–250	Woody savanna Montane	Sand, clay, loam Volcanic, alluvial	–	+	++	+	Lp^+
Masaka 1300	17–28	115	Forest / Woody savanna	Sand, clay, loam	++	+	+	+	Hp^+ / Lp
Rakai 1500	17–28	115	Forest / Woody and grassy savanna	Sand, loam Sand, clay, loam	+	+	+	+	Hp^+ / Lp
Kampala 1500	18–30	125	Forest / Savanna	Sand, clay, loam	+	+	–	+	Hp^+
Jinja 1200	18–30	125	Forest / Savanna	Ferric Sand, loam	+	+	–	+	Hp^+
Luwero 1200	18–28	115	Forest / Woody savanna	Sand, loam Sand, clay, loam	++	+	+	+	Hp^+ / Lp
Hoima 1200	17–28	140	Forest / Woody savanna	Sand, clay, loam Ferric	–	+	–	+	Hp / Lp^+
Kamuli 1200	18–28	115	Woody savanna	Sand, loam	–	+	–	+	Hp

Hp: big purple hybrid passion fruit; Lp: *Passiflora edulis* f. *edulis*; Hp^+ or Lp^+: major species grown.

Table II.
Radial growth rate ($mm \cdot h^{-1}$) of *Fusarium solani* culture at varying temperatures on PDA medium (at 5 °C, the radial growth is mil, whatever the growth duration).

Growth duration (h)	Temperature used for the isolate growth (°C)					
	10	15	20	25	30	35
12	0	0	0	0	0	0.05
24	0	0.03	0.18	0.25.	0.25	0.05
36	0	0.08	0.20	0.33	0.38	0.05
48	0.03	0.10	0.30	0.41	0.44	0.05
60	0.04	0.13	0.33	0.43	0.44	0.05
72	0.05	0.15	0.33	0.48	0.45	0.05
84	0.07	0.18	0.38	0.50	0.48	0.05
96	0.08	0.18	0.40	0.50	0.48	0.05
108	0.08	0.19	0.40	0.52	0.49	0.05
120	0.09	0.19	0.38	0.50	0.48	0.05
132	0.09	0.20	0.39	0.53	0.48	0.05
144	0.09	0.20	0.39	0.53	0.48	0.05
Mean radial growth rate	0.03	0.13	0.30	0.43	0.43	0.42

Table III.
Conidia characteristics observed on fungi isolated from diseased passion fruit stem sections and fruit baits and grown on PDA medium.

Septa number	Length (µm)	Width (µm)	Percentage (%)
0	7.2	3.0	47
1	15.0	4.0	22
2	20.2	3.6	8
3	31.0	4.4	15
4	36.0	4.5	6
5	37.2	3.6	2

4. discussion

The fact that most of the lowland lake shore areas were once mostly covered by forests and thickets and are now used for passion fruit production, among other crops, gives a possibility for *Fusarium solani* to have survived saprophytically on decaying vegetation debris prior to attacking the passion fruit host. As such, the Lake basin had most incidences of collar rot while other areas mostly had woodness virus, anthracnose (*Glomerella cingulata*) and brown spots (*Alternaria passiflorae*).

In the identification of *Fusarium solani* isolates from rotten passion fruit stem sections, culture colour obtained on PDA is not a parameter which can be used because a change from one growth temperature to another was followed by changes in culture pigmentation. This leaves morphological characteristics as the most reliable and conventional parameters for the identification of this Ugandan isolate. Some sources have reported the collar rot causal organism to be *Nectria haematococca*, the perfect stage of *Fusarium solani*. From current results, it is evident that *F. solani*, the imperfect stage, is a virulent form. It was consistently iso-

lated from infected samples collected from surveyed passion fruit growing districts. Ploetz [5] reported *N. haematococca* as having caused sudden wilt in Southern Florida, a disease which could be different from the collar rot found in Uganda which results into gradual wilting, culminating into total plant death once the canker eats away living tissue around the stem. According to Lutchmeah and Muraphur [6], *F. solani* was isolated from cankers at stem base collar region and on upper vines of infected passion fruit plants in Mauritius, whereas, during the current study in Uganda, cankers on uppervines were found to be caused by either *Glomerella cingulata* or *Alternaria passiflorae*, whereas *F. solani* was only but repeatedly isolated from collar rot cankers at infected passion fruit plant stem bases.

The presence of a virulent *F. solani* isolate in the soil could explain limited yield and production of the small purple passion fruit (*P. edulis* f. *edulis*), a susceptible species, in the lowland areas, as opposed to widespread production of the tolerant big purple Kawanda hybrid passion fruit (farmer observation). Most infection due to *F. solani* was confirmed to be in lowland areas with temperatures ranging 20–32 °C for most parts of the year (*table I*). This conforms with laboratory findings that the characteristic optimum growth temperature for *F. solani* is between 25 and 28 °C. This gives a possible answer for the survival of pathogen inoculum in the soil and consequent successful infection of passion fruits. One can still wonder, however, as to whether other factors like soil pH, inoculum density and grown species could influence disease ingress. These factors' impact needs to be assessed in further field/laboratory studies. Emphasis should be put on evaluation of various passion fruit varieties for resistance. Promising resistant varieties should then be recommended for use in lowlands areas as rootstock for productive but sensitive varieties. Tentatively, lowlands which have had no collar rot disease incidence should be encouraged to use straight away only grafted seedlings, when available, as a preventive measure. Those with established orchards from cuttings should be encouraged to have an integrated control programme developed to combine fungicide drenches with management practices. It is also recommended that an appropriate environment-friendly disease control programme be initiated to alleviate the present situation.

references

[1] Ssekyewa C., Fina Opio A., Swinburne T.R., Van Damme P.L.J., Abubakar Z.M., Sustainable management of collar rot disease of passion fruits in Uganda, Int. J. Pest Manage. 45 (1999) 173–177.

[2] Emechebe A.M., Mukiibi J., Nectria collar and root rot of passion fruit in Uganda, Plant Dis. Rep. 60 (1996) 227–231.

[3] Gardnar D.E., Pathogenicity of *Fusarium oxysporum* sp. *passiflorae* to Banana Poka and other *Passiflora* spp. in Hawaii, Plant Dis. 73 (6) (1989) 476–478.

[4] Cole D., Fusarium wilt, Passion Fruit Newsletter 5 (1990), 2 p.

[5] Ploetz R.C., Sudden wilt of passion fruit in Southern Florida caused by *Nectria haematococca*, Plant Dis. 75 (1991) 1071–1075.

[6] Lutchmeah R.S., Muraphur F.B., Sudden wilt of yellow passion fruit *P. edulis* f. *flavicarpa* caused by *Fusarium solani*, FAO Plant Protection Bulletin 41 (2) (1993) 126–127.

[7] Anonymous, Plant pathologist's pocket book, 2nd ed., Commonwealth Mycological Institute (CMI), Commonwealth Agricultural Bureaux, Farnham Royal, Slough, UK, 1983, pp. 324–352.

[8] Wollenweber H.W., Reinking O.A., Die Fusarien, ihre Beschreibung, Schadwirkung und Bekampfung, Paul Prey, Berlin, Germany, 1935.

[9] Joffe A.Z., Taxonomic problems of the genus *Fusarium*, In: Fusarium species: their biology and toxicology, John Wiley and Sons, Interscience publication, Inc. New York, USA, pp. 386–440.

[10] Nelson P.E., Tousson T.A., Marassas W.F.O., *Fusarium* species. An illustrated manual for identification, The Pennsylvania State University Press, University Park, USA and London, UK, 1983.

Presencia de la enfermedad de la pudrición rosa del cuello (prc) en la granadilla, en los principales sitios de producción de Uganda.

Resumen — Introducción. En Uganda, en unas 500 ha, pequeñas plantaciones producen 3.500 t de granadillas (*Passiflora* spp.) cada año. El desarrollo de la planta se halla limitado principalmente a causa de enfermedades entre ellas la (prc), causada por *Fusarium solani,* considerada como la más grave económicamente. Un estudio procuró establecer la distribución de esta enfermedad en los 16 principales distritos productores de granadillas. **Material y métodos**. Para cada uno de los distritos controlados, se visitaron seis campos. Se recolectaron muestras de tallos afectados de (prc). Se aislaron y se identificaron los organismos incriminados por la enfermedad en laboratorio. **Resultados**. Entre los 16 distritos visitados, cuatro mostralon ningún ataque de (pcr), cinco presentaban unas infecciones y siete estaban ampliamente colonizados por la enfermedad. Esta se encontró limitada a las zonas de baja altitud. La granadilla violeta local (*P. edulis* f. *edulis*) fue atacada más temprano que el híbrido violeta (*P. edulis* f. *edulis* × *P. edulis* f. *flavicarpa*). El hongo aislado fue radicalmente identificado como *Fusarium solani*; su desarrollo optimo se hace a 25–30 °C. La aplicación de los postulas de Koch mostró que inducía la enfermedad. **Discusión**. La presencia de aislados virulentos de *F. solani* en el suelo podría explicar los rendimientos limitados de la granadilla violeta (*P. edulis* f. *edulis*), especie sensible, respecto a la producción más importante del híbrido Kawanda tolerante. El impacto de factores tales como el pH del suelo, la densidad de inoculo y las especies desarrolladas, pudiendo tener una influencia en la enfermedad, tendrá que evaluarse en próximas experimentaciones en campo o en laboratorio. El esfuerzo tendrá que abarcar la investigación de variedades resistentes. © Éditions scientifiques et médicales Elsevier SAS

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