



Survey and conventional management methods of bacterial wilt disease in open fields and greenhouses in Tanzania

Agatha Aloyce^{1,2} · Patrick Alois Ndakidemi^{1,2} · Ernest Rashid Mbega^{1,2}

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Abstract

A study was conducted from January to February 2018 to determine bacterial wilt disease (BWD) incidence and severity in open-field and greenhouse environments in twelve tomato growing districts in Tanzania. About 220 farmers were interviewed to assess their knowledge on BWD by using a semi structured questionnaire. Results indicated significant ($p < 0.05$) difference of BWD incidence and severity among districts. Similarly, BWD incidence and severity were significantly ($p < 0.05$) higher in greenhouses than in the open field environments implying that BWD is a major challenge in tomato under greenhouse than in open-field environments. Most of the farmers were not certain about BWD symptomology and management. Majority (>80% of 220 respondents) of farmers could not identify sources of BWD in environment and do not adhere to sanitation measures recommended for greenhouse tomato production. 90% of the interviewed farmers ventured into greenhouse tomato production by imitating from neighbors without technical guidance. To manage BWD, majority (70%) of farmers use chemicals which they reported as ineffective, 13% use botanical, 10% do crop rotation which was reported to be not practical because of land scarcity and long time that *Ralstonia solanacearum* can survive. Rest (7%) of farmers do not use any BWD management measure. There was no report of either use of disease resistant cultivars or biological control as a strategy for BWD management in the study area. There is therefore need to develop techniques for farmers to manage the BWD by exploring promising options such as use of effective botanical extracts.

Keywords *Ralstonia solanacearum* · Soil-borne · Incidence · Severity

Introduction

Bacterial wilt disease (BWD) caused by a soil-borne pathogen *Ralstonia solanacearum* (Smith), is a deadly disease of tomato and other crops worldwide (Fegan and Prior 2005; Alvarez et al. 2010). It is a devastating disease accounting up to 100% tomato yield losses depending on cultivar, soil type, cropping environment and pathogen

strain (Kelman 1953; Hayward 1991). Economically, BWD has been estimated to affect about 1.5–2 million ha in 80 countries causing an annual loss of approximately \$950 million (Elphinstone 2009). It is among the serious tomato production challenges in different areas in the world including the East Africa countries (Aloyce et al. 2017). In Uganda for instance, BWD has been associated with a loss of about 88% of tomato (Katafiire et al. 2005). In Tanzania, information about tomato BWD is scarce and hence worthwhile to be generated for the development of improved disease management strategies.

Ralstonia solanacearum causes bacterial wilt infection by entering plants through wounds or injuries caused by insects, nematodes or simply by cultivation (Inoue and Nakaho 2014). Infection proceeds to the inner cortex level of primary roots, with bacteria forming large intercellular pockets, and cortical cells next to them displaying features of degeneration (Vasse et al. 1995). Once in the vascular cylinder, it infects the intercellular spaces of vascular parenchyma adjacent to xylem vessels, which eventually are invaded and degraded. The

✉ Agatha Aloyce
aloycea@nm-aist.ac.tz

¹ Department of Sustainable Agriculture and Biodiversity Ecosystem Management, School of Life Sciences and Bioengineering, Nelson Mandela African Institution of Science and Technology (NM-AIST), P.O. Box 447, Arusha, Tanzania

² Centre for Research, Agriculture Advancement, Teaching Excellence and Sustainability (CREATES) in Food and Nutrition Security, Nelson Mandela African Institution of Science and Technology, P.O. Box 447, Arusha, Tanzania

pathogen can then break into and fill xylem vessels and the surrounding parenchyma cells are highly degraded by hydrolytic enzymes (Vasse et al. 1995; Saile et al. 1997). Within the xylem vessels, the bacterium moves throughout the stem to the upper parts of the plant while it is multiplying to over 10^{10} cells per cm of stem (Saile et al. 1997). Extensive multiplication in the xylem vessels leads to clogging of the vessels thereby inhibiting flow of water and nutrients and the whole plant wilts and dies (Agrios 2005; Shamayeeta and Sujata 2016).

After death of the plant, *Ralstonia solanacearum* is released into the environment, where it can survive in plant debris, soil and/or water until it gets into contact with a new host. In presence of susceptible hosts and suitable environmental conditions such as temperature and relative humidity, the released bacterium develops disease infection (Agrios 2005). Disease incidence and severity increases in the presence of suitable temperatures (>29 °C), relative humidity ($>85\%$) and wet soils (Castilla and Hernández 2007; Alvarez et al. 2008; Mrema et al. 2017), conditions which are common in greenhouses.

It is also worth noting that greenhouse production system is one of the knowledge intense production systems (Opdam et al. 2005; Tanny et al. 2008). Inadequate knowledge contributes to the increased incidence and severity of bacterial wilt of tomato in greenhouse in developing countries (Qian et al. 2011; Allen et al. 2016). Farmers should understand and follow specific principles of BWD management in greenhouses (Castilla and Hernández 2007). Contrarily, most farmers who have ventured into greenhouse tomato production have limited knowledge on different aspects such as specific greenhouse operation principles, the best greenhouse structures and management of insect pests and diseases (Mwaniki et al. 2017) whereby management of diseases is considered to be one of the knowledge demanding aspects.

Unskilled farm workers for instance can introduce *Ralstonia solanacearum* into greenhouse via their shoes, cultivation, weeding or pruning tools (Alvarez et al. 2010; Shamayeeta and Sujata 2016; Prior et al. 2016; Gaofei et al. 2017; Aloyce et al. 2017). Once *Ralstonia solanacearum* are in the greenhouse, they multiply and spread quickly because of suitable conditions (Kinyua et al. 2014; Mwaniki et al. 2017). Other sources of *Ralstonia solanacearum* in the greenhouse include contaminated soil, seedlings, irrigation water, plant debris and host weeds (Alvarez et al. 2008; Heuvelink et al. 2008; VonZabeltitz 2011; Singh et al. 2014; Radhi et al. 2016). Furthermore, greenhouse tomatoes are often not rotated with another crop (Yuliar and Koki 2015; Wei et al. 2017) because tomato is a priority crop for greenhouse production to the majority of farmers in Tanzania. This increases disease incidence and severity due to the buildup of pathogen inoculum in the soil. The present study aimed to determine tomato

BWD incidence and severity in main tomato growing districts and under greenhouse and open field environments in Tanzania. Knowledge of farmers about BWD management in greenhouses and management methods used against BWD in Tanzania were also evaluated during the study.

Materials and methods

Incidence and severity of BWD in Tanzania

A field survey was conducted in twelve districts covering the main agro-ecological zones of Tanzania to understand the incidence and severity of BWD. Fields were surveyed when plants were at flowering stage and it involved mainly Tanya tomato variety due to its high popularity in the study area (Mbega et al. 2012). A multistage random sampling procedure was used. The first involved selecting the wards within the district, then the villages and the fields. Four (4) wards were selected at random in a district and five (5) fields including greenhouses were identified from each village. Within the field, five (5) plots of 50 m² (5 m × 10 m) were sampled by critically observing symptoms of BWD and through bacterial streaming test (Agrios 2005; Kinyua et al. 2014). Plants with BWD symptoms were counted in each plot. Disease severity was recorded on a 1–5 scale (Hyakumachi et al. 2013; Sang et al. 2016) where 1 = No symptom, 2 = Top young leaves wilted, 3 = Two leaves wilted, 4 = Four or more leaves wilted and 5 = Plant dies. The agro-ecological parameters namely temperature and relative humidity of the study area were measured and recorded during survey to understand the relationship of BWD incidence and severity with these parameters.

Knowledge of farmers on principles of BWD management in greenhouses in Tanzania

To capture information on the number of farmers using various management methods and their knowledge on principles of tomato BWD management in greenhouses, about twenty tomato growing farmers were selected randomly per district and were interviewed by using a semi structured questionnaire with both close and open ended questions. In addition to the questions on the management methods employed by each farmer, farmers were also provided with key principles of BWD management practiced and responses scored on a 1–4 (Table 1).

In order to cross-check information collected from the farmers and ensure that responses reflect farmers' knowledge and experience, twenty (ten district extension and research officers) development agents were selected as key informants and interviewed by using open-ended semi structured questionnaire. These development agents worked very closely

Table 1 Criteria used to score farmers' knowledge on principles of tomato BWD management in greenhouse

Score	Knowledge level	Description of criteria
1	No	Farmers could not mention any principle of tomato diseases management in greenhouse
2	Low	Farmers named one principle, one tomato disease and one risk of not complying following
3	Medium	Farmers named BWD, described one or more of BWD symptoms, described at least one identification method, described at least one type of damage caused by BWD
4	High	Farmers named BWD, described one or more of BWD symptoms, described at least one identification method, described at least one type of damage caused by BWD and describe at least one way of BWD dissemination

with farmers and are familiar with principles of tomato production in greenhouses.

Data analysis

BWD incidence in a district was calculated by using the formula

$$\text{Wilt incidence (\%)} = \frac{\text{Mean of wilted plants in a district}}{\sum \text{plants assessed in a district}} \times 100$$

Average BWD severity per district was calculated using the formula

$$\text{Severity (\%)} = \frac{5A + 4B + 3C + 2D + E}{5N} \times 100$$

where A = average number of plants on scale 5 in the district; B = number of plants on scale 4; C = number of plants on scale 3; D = number of plants on scale 2; E = number of plants on scale 1; N = total plants evaluated in the district.

Data of disease incidence and severity were pooled together by calculating the average of incidence and severity per district and subjected to the analysis of variance (ANOVA). Means of disease incidence and severity were separated by using the Tukey's procedure whereas the least significant difference (LSD) was used to compare means of management methods ($p = 0.05$), all facilitated by the Costat data analysis software program.

Mean score for farmers' responses was computed as

$$\text{Mean score (MS)} = \frac{\text{Responses scored}}{\text{The highest score}}$$

$$MS = \frac{4 + 3 + 2 + 1}{4} = 2.50$$

Using the interval score of 0.05, the upper and lower limits were determined as $2.50 + 0.05$ and $2.55 - 0.05 = 2.45$ respectively. On the basis of this, $MS < 2.45$ were ranked 'low',

those $2.45 \geq MS \leq 2.54$ were ranked 'medium' while the $MS \geq 2.55$ were considered 'high'.

Results

Incidence and severity of BWD in Tanzania

Out of 4800 plants assessed in all districts, 2648 (55%) were infected by the BWD at different levels of disease incidence and severity which significantly differed among districts ($p < 0.05$) (Table 2). Incidence (44.6%) and severity (59.3%) were significantly higher in Arumeru as compared with other districts in which incidence and severity ranged from 5.8–20.8% and 10.7–46% respectively (Table 2). Mean values of temperature and relative humidity (RH) in the BWD most affected districts were 30 °C and 88% as compared with 20.5 °C and 75% °C in the BWD less affected districts, respectively (Table 2). Similarly, incidence and severity of BWD were significantly ($p < 0.05$) higher in the greenhouses than in the open field tomato production environments (Table 3).

Mean temperature and RH values in all greenhouses visited were > 29 °C and $> 85\%$ respectively except in Mbeya, Tandahimba and Chake Chake (Fig. 1). In linking the BWD incidence and severity in greenhouses with temperature and RH, the results indicated that incidence and severity increased along with temperature and RH increases (Fig. 1).

Knowledge of farmers on principles of BWD management in greenhouse in Tanzania

The mean score of farmers' level of knowledge for different principles practiced varied from 0.0–3.12 (Table 4). Respondents indicated that practices such as testing of soil and irrigation water to determine their suitability for tomato production were not considered necessary and thus not done. Most respondents recognized BWD problem and believe to be caused by the cold weather and have named it as ice disease "barafu". They can describe the

Table 2 BWD incidence and severity, temperature and relative humidity in different districts in Tanzania

District	Incidence (%)	Severity (%)	Temperature (°C)	Relative humidity (%)
Arumeru	44.60a	59.30a	31	89
Babati	38.70a	44.00a	29	87
Manyoni	22.60b	36.00b	27	85
Nyamagana	14.48c	25.90c	28	81
Mvomero	27.80b	33.80b	26	80
Kilolo	10.60b	20.80c	22	79
Temeke	19.10c	14.65c	29	83
Chake Chake	22.4b	30.10b	28	83
Mbeya urban	8.80d	9.70d	21	80
Kibondo	11.7c	14.30c	27	81
Tandahimba	6.30d	4.10d	20	70
Kongwa	23.10b	31.20b	28	80
Mean	20.85	26.99	26.33	81.50
F-test	*	*		
CV (%)	19.60	20.07		

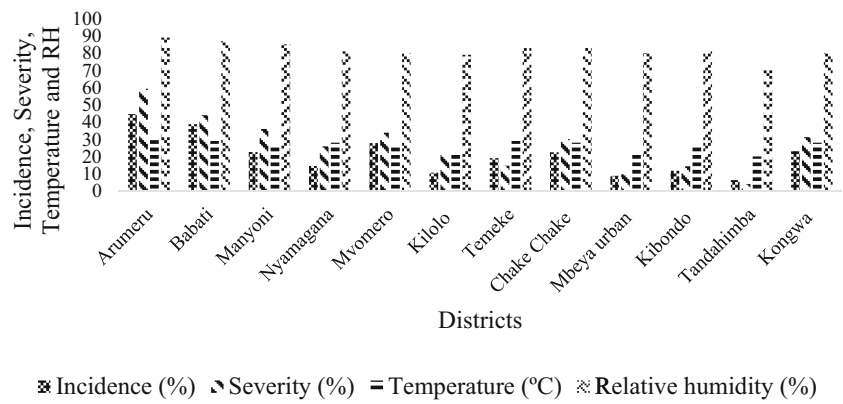
* = Means are significant at $p = 0.05$. Mean of incidence and severity with the same letter within the column are not significantly different based on Tukey's test ($p = 0.05$)

Table 3 BWD incidence and severity in open-field and greenhouse in different districts in Tanzania

District	Environment	Incidence (%)	Severity (%)	District	Environment	Incidence (%)	Severity (%)
Arumeru	Greenhouse	58.71a	54.33a	Temeke	Greenhouse	21.47a	19.49a
	Open field	23.89b	24.97b		Open field	7.63b	15.16a
	Mean	41.30	39.65			14.55	17.33
	F test	*	*		F test	**	ns
Babati	Greenhouse	24.59a	52.81a	Chake Chake	Greenhouse	18.40a	36.80a
	Open field	4.11b	21.19b		Open field	12.00b	24.30a
	Mean	14.35	37.00			15.20	30.55
	F test	**	**		F test	*	ns
Manyoni	Greenhouse	24.80a	52.00a	Mbeya	Greenhouse	9.67a	11.70a
	Open field	13.80b	34.00b		Open field	7.04a	8.43a
	Mean	19.30	43.00			8.36	10.07
	F test	*	*		F test	ns	ns
Nyamagana	Greenhouse	26.34a	42.85a	Kibondo	Greenhouse	16.52a	25.62a
	Open field	12.96b	24.49b		Open field	12.78a	19.65b
	Mean	19.65	33.67			14.65	22.64
	F test	*	**		F test	ns	*
Mvomero	Greenhouse	26.30a	34.31a	Tandahimba	Greenhouse	8.48a	15.70a
	Open field	13.33b	22.83b		Open field	2.42b	7.62b
	Mean	19.82	28.57			5.45	11.66
	F test	*	*		F test	*	***
Kilolo	Greenhouse	17.44a	38.50a	Kongwa	Greenhouse	25.81a	28.53a
	Open field	15.73a	36.42b		Open field	9.50b	13.31b
	Mean	16.59	37.46			17.66	20.92
	F test	ns	ns		F test	**	**

*** = Means are significant at $p = 0.001$, ** = Means are significant at $p = 0.01$ and * = significant at $p = 0.05$. Mean of incidence and severity with the same letter(s) within the column are not significantly (ns) different based on Tukey's test ($p = 0.05$)

Fig. 1 Relationship of BWD incidence, severity, temperature and relative humidity in greenhouses in Tanzania



disease by symptoms but they could not identify its source in greenhouses or predict its outbreak but believed it is not a new tomato disease in greenhouses. Furthermore, results of mean score of respondents' knowledge was ranked as low in all districts (Table 5).

BWD management methods

Results revealed that a number of management methods are used by farmers such as chemicals (70%), botanicals (13%) and crop rotation (10%) (Fig. 2). Imidacropid, metalaxyl,

Table 4 Mean score for principles of BWD management in greenhouse in Tanzania

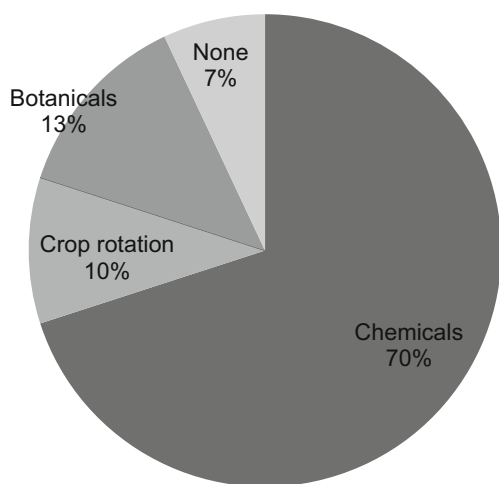
Stage	Principle	Mean score (Max = 4)
Before growing cycle	Test soil for its status – soil microbes and fertility	0.00
	Identify source of irrigation water and test its suitability for tomato irrigation	0.00
	Clear the screen-house of plant debris and weeds	2.11
	Wash and disinfect benches, potting tables, storage shelves, tools and equipment	0.44
During growing cycle	Avoid recontamination with pathogen	0.33
	Purchase certified seeds from reliable sources	0.03
	Use cell grown seedlings grown by using sterilized growth media	2.27
	Use resistant cultivars whenever possible	1.41
	Close the greenhouse door all the time	0.96
	Have a foot bath at the door with disinfectant	2.43
	Disinfect farm tools and equipment e.g. hoe, pruning knives and scissors before use or when moving from one plant to another	1.80
	Avoid unnecessary visits/visitors inside the greenhouse	0.23
	Maintain a disease prevention program for tomato plants	0.19
	Inspect plants for diseases - look for localized symptoms such as root lesions, cutting end rot, leaf spots, and shoot blights	0.46
	Monitor seedlings for damping off	0.31
	Inspect incoming seedlings	3.12
	Properly identify the diseases	2.93
	Select a well-drained soil	1.27
	Apply water for optimum plant growth	2.08
	Space plants for good air movement and sunlight	1.79
	Irrigate early enough in the day to allow foliage to remain dry overnight	1.06
	Do not reuse growing media	0.01
	Use clean gloves	0.34
	Use un-contaminated (disinfected) drip irrigation system	0.06
Use separate screen-houses for vegetable plants and ornamental plants	0.01	
Maintain required relative humidity and temperature	0.53	
End of cycle	Discard unused seedlings, fruits and plant debris	0.05

Table 5 Farmers' level of knowledge and rank on BWD management in greenhouses in Tanzania $N = 220$

District	High(4)	Medium(3)	Low(2)	No(1)	Mean score	Level	Rank
Arumeru	2(0.40)	6(0.90)	9(0.90)	3(0.15)	2.35	Low	1
Babati	0(0.00)	1(0.15)	10(1.00)	9(0.45)	1.60	Low	6
Nyamagana	1(0.20)	6(0.90)	9(0.90)	4(0.20)	2.20	Low	2
Manyoni	0(0.00)	2(0.30)	8(0.80)	9(0.45)	1.55	Low	7
Mvomero	0(0.00)	0(0.00)	3(0.30)	17(0.85)	1.15	Low	8
Kilolo	3(0.60)	6(0.90)	6(0.60)	5(0.25)	2.35	Low	1
Temeke	0(0.00)	5(0.75)	10(1.00)	5(0.25)	2.00	Low	4
ChakeChake	0(0.00)	3(0.45)	11(1.10)	6(0.30)	1.85	Low	5
Kibondo	0(0.00)	0(0.00)	7(0.18)	13(0.65)	0.83	Low	9
Tandahimba	0(0.00)	0(0.00)	3(0.3)	17(0.85)	1.15	Low	8
Kongwa	0(0.00)	3(0.45)	11(1.10)	6(0.30)	1.85	Low	5
Mbeya Urban	1(0.20)	4(0.60)	11(1.1)	4(0.20)	2.10	Low	3

profenofos and thiamethoxam were regularly reported chemicals used by farmers which are applied on tomato on weekly basis in the morning from transplanting to harvesting. Most of respondents (90%) (Fig. 2) reported that chemicals are ineffective against BWD and declared their willingness to adopt other management methods if available. Moreover, some farmers (7%) do not use any method to manage BWD (Fig. 2).

Statistically, the number of farmers using various methods to manage BWD were significant ($p < 0.05$) (Table 6). The crude extract from Neem (*Azadirachta indica*) leaves and seeds was a commonly used botanical that was applied at least 2–3 times on weekly basis. Farmers reported to be uncertain on the performance of botanical extracts on BWD management.

**Fig. 2** Number of farmers using different management methods for BWD in Tanzania

Discussion

BWD is a serious problem of tomato production affecting about 55% of tomato plants in Tanzania. The ability of farmers to talk about BWD could indicate how economically important this disease is to them. *Ralstonia solanacearum* is transmitted through soil, surface water, infected plant materials, insects, farm workers and cultural practices such as weeding and pruning or mono-cropping (Alvarez et al. 2010; Remenant et al. 2010; Yang et al. 2012). Suitable environmental conditions such as temperature and RH that are $>29^{\circ}\text{C}$ and 85% respectively accelerate infection development by favoring multiplication of *Ralstonia solanacearum* cells (Alvarez et al. 2008; Jonathan et al. 2014).

Higher incidence and severity of BWD disease in Arumeru as compared with other districts could be attributed by factors such as soil and strain type, mono-cropping, poor

Table 6 Mean comparison of number of farmers using different management methods against BWD in Tanzania

Method	Mean
Chemical	12.83a
Do not use any practice	3.67b
Botanicals	2.42b
Implement crop rotation	1.83b
Mean	5.19
F- test	***
LSD	4.73
CV (%)	14.04

*** = significant means at $p = 0.001$. Mean with the same letter within the column are not significantly different based on LSD ($p = 0.05$)

phytosanitary practices, suitable temperature and RH for *Ralstonia solanacearum* cell multiplication (Alvarez et al. 2010; Mrema et al. 2017).

Similarly, higher incidence and severity of BWD under greenhouse than open field environments could be ascribed by increased temperature and RH in greenhouse than in open field environment. Temperature range of 25–30 °C enhances development of lateral roots in tomato and thus significantly increasing the entry points of *Ralstonia solanacearum* which increases disease incidence and severity in greenhouses as compared with the open field environments (Agrios 2005). This is unfortunate and a major challenge to farmers in developing countries who in most cases have inadequate knowledge of BWD management especially in the greenhouse. Therefore, there is a need of manipulating temperature and relative humidity in the greenhouses and empower farmers with needed skills to improve management of BWD in Tanzania.

Results of the present study implied that there is inadequate understanding of farmers on principles of BWD management in greenhouses which could be due to the fact that most farmers live in rural areas where resource-poor and less educated farmers are found (Mwaniki et al. 2017). Respondents were affirmative that they ventured into greenhouse tomato production by imitating from neighbors without technical guidance. Allen et al. (2016) reported that adequate knowledge and technical guidance are key for successful management of BWD in greenhouses. Limitation in knowledge on key principles of BWD management can contribute to increase in disease incidence and severity (Qian et al. 2011; Allen et al. 2016). Soils and water for example constitute a major source of *Ralstonia solanacearum* (Ndakidemi 2007; Alvarez et al. 2008; Kinyua et al. 2014; Yuliar and Koki 2015), but farmers in the study area never consider testing soils and irrigation water suitability for the tomato production. Because of inadequate knowledge, farmers in the study area were unaware of the effect of temperature and RH on BWD development in the greenhouse, instead they thought it is a cold weather problem. By maintaining environmental conditions such as temperature, RH and soil in the greenhouse unsuitable for BWD development, the incidence and severity can be decreased (Alvarez et al. 2010; Mbega et al. 2012). This could be achieved by developing appropriate greenhouse structures to allow proper air circulation, irrigation and drainage systems.

Farmers also indicated inadequate understanding about importance of using quality seeds and seedlings, immediately uprooting infected plants and discarding debris, disinfecting contaminated tools, identifying sources of *Ralstonia solanacearum*, using foot bath and importance of disease outbreak prediction and prevention program. Weeding is important as some weeds such as *Solanum dulcamara*, *Ipomoea* sp. and *Portulaca oleracea* are hosts of *Ralstonia solanacearum*

that remain asymptomatic (Pradhanang et al. 2005) and aid in long term survival of the pathogen in the environment. Burning of plant debris could deny *Ralstonia solanacearum* of a source of food as it enters the saprophytic stage of its life cycle and hence infection is reduced. All these are important in managing BWD and farmers are thus required to be guided after empowering them with the needed knowledge.

Although majority (70%) used chemical pesticides, they reported that chemicals are not effective. One of the reasons for this could be limitation in farmers' knowledge in which farmers can use inappropriate chemical, rate or application (Ngowi et al. 2007; Aloyce 2013). Other contributing factors could be pathogen genetic transformation and resistance to chemicals. Apart from being ineffective, use of chemicals as pesticides have been claimed to be environmentally unsafe (Yuliar and Koki 2015). Some of the alternative methods for BWD management include use of botanicals, resistant/tolerant varieties and proper sanitation measures. Farmers reported to use some plant extracts to manage BWD however, they are unsure of their effectiveness. None of the farmers reported to use resistant varieties or biological control agents. Information on performance of specific methods used by farmers to manage BWD is limited in Tanzania and thus further research is recommended. Research is recommended to explore promising botanical extracts for which farmers have shown adoption interest to manage BWD in Tanzania.

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Compliance with ethical standards

Conflict of interest Authors declare that they have no conflict of interest.

Ethical approval All procedures performed in study involving human participants, farmers in this respect were in accordance with the ethical standards of the institutional and national research committee.

Informed consent Informed consent was obtained from all individual farmers included in the study.

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