INNSPUB

International Journal of Agronomy and Agricultural Research (IJAAR)
ISSN: 2223-7054 (Print) 2225-3610 (Online)
http://www.innspub.net
Vol. 9, No. 1, p. 92-99, 2016

RESEARCH PAPER OPEN ACCESS

Effectiveness of organic substances in the control of powdery mildew (*Sphaerotheca fuligenia*) of butternut (*Cucurbita moschata* PEPO)

Munyaradzi Shamuyarira[1], Lovejoy Tembo[1, 2*], Sommerset Mhungu[1]

[1]*Faculty of Agriculture, Women's University in Africa, Harare, Zimbabwe*

[2]*Faculty of Agriculture, Zimbabwe Open University, Harare, Zimbabwe*

Article published on July 23, 2016

Key words: Powdery mildew, Sodium bicarbonate, Cattle milk, Dilute acetic acid, Garlic.

Abstract

Powdery mildew (*Sphaerotheca fuligenia*) is an economically important disease of cucurbits requiring sustainable means of management. A field experiment was set up to evaluate the effectiveness of cattle milk, sodium bicarbonate, dilute acetic acid and garlic in controlling powdery mildew in butternut (*Cucurbita moschata pepo*). The trial was laid out as a randomized complete block design with 3 replicates and 6 treatments. The treatments were applied at the following concentrations; 1. Cattle milk at 100ml/liter of water, 2. Dilute acetic acid at 6ml/litre of water, 3. Bicarbonate of soda at 10g/litre of water, 4. Garlic at 20g/litre of water, 5. Tubuconazole in the form of folicur at 0.5ml per liter of water and 6. Control which was not sprayed. A significant difference ($P<0.01$) was recorded at 60days after planting with respect to disease incidence and milk had the lowest incidence of 12.9% and the highest incidence of 37.3% was recorded in the control treatment. Milk and sodium bicarbonate managed to suppress both disease incidence and severity. This study confirms the fungicidal properties of milk and sodium bicarbonate, garlic, and dilute acetic, suggesting their use as fungicides for disease control for sustainable powdery mildew management.

* **Corresponding Author:** Lovejoy Tembo ✉ ltembo13@gmail.com

Introduction

Powdery mildew is a common fungal disease among plants species and the causal organisms are obligate biotrophic parasites that belong to phylum Ascomycota (Hacquard, 2014). On cucurbits, the disease is commonly caused by two fungal species, *Golovinomyces cichoracearum* and Podos*phaera fusca (*Syn. *Podoshaera xanthii)*, and these can induce identical symptoms but can be distinguished easily under light microscopy (Thomason and Gibson, 2006). The disease causes development of whitish powder-like growths on leaf surfaces, petioles and stems (Perez-Garcia *et al.,* 2009).

Powdery mildew is widespread and causes serious damage to almost all cucurbits under both field and greenhouse conditions (Perez-Garcia *et al.,* 2009) causing heavy yield and quality losses (Shi *et al.,* 2007). Powdery mildew infections reduce yield by lowering the plant's vigour and increasing the number of sun-scotched fruits (Daughtrey *et al.,* 2006). It reduces farm net profits as it increases production costs since application of fungicides is the main practice in most cucurbit crops for managing the disease. Most of these fungicides are of a synthetic nature and have a systemic and/or contact action. Synthetic fungicides require frequent applications to effectively control fungal diseases such as powdery mildew. Frequent application of fungicides produces strains tolerant to fungicides due to selection pressure (McGrath, 1996).

For adequate protection against powdery mildew, fungicides with translaminar action are used (Baldwin and Waldenmaier, 1992). Unfortunately these pose the highest risk for development of resistance as they affect a single site on the fungi's metabolic pathway. This creates high chances for development of resistance and subsequent crop damage (O' Brien, 1994). Consequently, horticultural farmers resort to using stronger fungicide concentrations which impact negatively on the environment. The use of increased concentrations can also result in phytotoxicity. The collateral effect of fungicides in general may be in varying degrees of toxicity to humans, animals and the environment (Gullino and Wardlow, 1999; Acero *et al.,* 2011). Qualitative resistance can also occur resulting in complete loss of disease control that cannot be regained through using higher rates or more frequent fungicide applications (McGrath, 2001). Leaf infection by powdery mildew interferes with photosynthesis and respiration, leading to reduced fruit set, inadequate ripening, and poor flavor development. Consequently fruits have low sugar content, poor flavour, and do not store well (Kabir *et al.,* 2011).

There is therefore need to come up with sustainable powdery mildew control strategies (McGrath and Shishkoff, 1999). Thus, the objective of this research was to investigate the effectiveness of cattle milk, garlic, bicarbonate of soda and vinegar as fungicides for the control of powdery mildew in butternut.

Materials and methods

Experimental site

A field trial was conducted at Green Finger Commercial Nursery, Harare, Zimbabwe. The site has red alluvial clay soils and is situated at an altitude of 1506 meters above sea level, experiences mean annual temperatures ranging from 10 to 26 ^{o}C and receives an annual rainfall ranging between 800 and 1000mm. Annual humidity averages 57.6% and an average of 7.6 hours of sunshine are received per day.

Experimental design and treatments

The experiment was laid out as a Randomized Complete Block Design with three replicates and six treatments. The treatments were as follows: Cattle milk at 100ml/ L^{-1} water, dilute acetic acid at 6ml/ L^{-1} water, bicarbonate of soda at 10g/ L^{-1} water, garlic at 20g/L^{-1} water, Tubuconazole in the form of Folicur at 0.5ml/L^{-1} water and a negative control which was not sprayed

Trial management

Fertilizer, Compound C (5:13:12 N: P: K) was applied at the rate of 660kg ha^{-1}. Squash butternut cultivar Waltham was directly seeded and three seeds were

planted per station at a spacing of 0.4m apart within the row and 1.5m between the rows.
The gross plot was composed of three rows. Each row had nine plants, and the middle rows were the net plots with sampling being done on three plants in the middle. Plants were thinned to one plant/station three weeks after emergence. Top dressing was carried out at the rate of 34kg N ha^{-1} at appearance of first fruits.

Inoculation and spraying

Powdery mildew was inoculated in the plots approximately a month after planting by collecting leaves with powdery mildew spores and rubbing the leaves on the plants so that conidia remained on the leaf surfaces (Sinclair and Dingra, 1995). Inoculation was done late in the afternoon to prevent desiccation of the conidia and also to benefit from the night dew which increases humidity on the leaf surface. Two leaves from the bottom were inoculated.

Sprays were administered at weekly intervals beginning a week after inoculation to allow disease infestation. All sprays were mixed just before spraying except for the garlic spray which required to be prepared a day before spraying by crushing 0.1 kg garlic and soaking them in one litre of water. Further dilution of the garlic solution with four litres of water was done prior to spraying. The leaves were drenched with the spray using a knapsack sprayer.

Disease severity assessment was carried out according to the percentage leaf area covered by the disease and allocating scores. A rating scale of zero to five was used where; 0=no sporulation, 1=0-20% leaf cover, 2=20-40% leaf cover, 3=40-60% leaf cover, 4=60-80% leaf cover and 5=80-100%leaf cover. Disease scoring was done by assessing six young leaves, six middle aged leaves and six old leaves per plot.

Data collection

Disease incidence was also assessed by recording the number of affected leaves and representing them as a percentage of the whole plant (Chaube and Pundir, 2009). Data on disease severity were collected by allocating scores as described by Trigiano *et al.* (2006).
Foliar assessment began a week after the first spray. Yield parameters assessed include; fruit count, average fruit weight, and yield per plot, fruit length and diameter.

Statistical analysis

Data were analyzed using Genstat version 8.1. Analysis of variance (ANOVA) was carried out for the overall treatment effects and pair wise comparison between means were determined using the least significant difference (LSD) at P = 0.05. Square root transformations were carried out on proportions before they were subjected to analysis of variance to normalize the data.

Results and discussion

Disease incidence

At 60 days after planting there was a significant difference (P<0.001) among the treatments with respect to disease incidence. Folicur had the lowest disease incidence of 3.9% followed by milk with 12.9% and the untreated control had the highest incidence of 37.3% (Fig.1). However dilute acetic acid and garlic were not significantly (P>0.05) different from the untreated control and sodium bicarbonate had a high disease incidence of 24%.

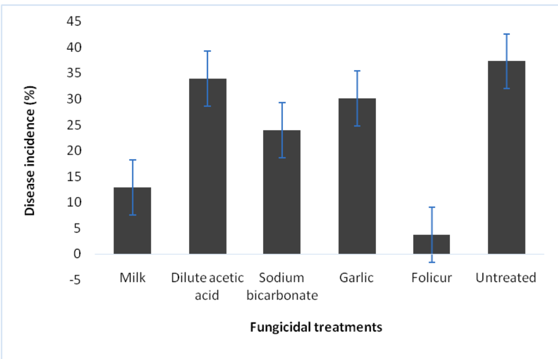

Fig. 1. Disease incidence in relation to fungicidal treatment.

Vertical bars represent standard error bars of means.

Milk contains amino acids and salts which have germicidal effects and these may have contributed to incidence reduction (Pasini *et al.*, 1997). Milk also contains super oxide anions which antagonize fungi. These are generated in the presence of ultraviolet light when milk is exposed to sunlight (Korycha-Dahl and Richardson, 1978). Lacteferin, a milk component has the ability to antagonize fungi (Crisp *et al.,* 2006). Bettiol (1999), in his researches on zucchini powdery mildew observed leaf surface pH increase as another possible mode of action which could have resulted in milk lowering disease incidence. His results concur with observations of Ziv and Zitter (1992).

Milk helps the establishment of a protective oily barrier and antagonistic organisms which could also have helped in incidence reduction (McGrath and Shishkoff, 1999). Milk could also have induced systemic resistance to the pathogen (Reuveni *et al.,* 1993) or induced the production of biocidal compounds which lower disease incidence (Tzeng and DeVay, 1989).

Incidence reduction through the use of bicarbonate of soda concurred with the results observed by Williams and Williams (1997) when powdery mildew in grapes was controlled using 1 % solution of sodium bicarbonate. Williams and Williams (1992) also observed that soda at 2% concentration and 1% Sunoil had positive results in reducing powdery mildew in euonymus. Bicarbonate of soda was effective possibly because it altered the pH on the leaf surface and prevented spore germination. In this experiment the efficacy of sodium bicarbonate could have been increased by raising the rate as no phytotoxic effects were observed. The addition of surfactants such as oils and other film forming substances could also have lowered the disease incidence (Homma *et al.,* 1981; Reuveni *et al.,* 1995).

Incidence reduction through the use of garlic sprays may have been compromised because garlic sprays are normally applied as a preventative rather than a curative measure. Thus, its action could have been limited because of pre-existing powdery mildew spores from the inoculum and wind transferred spores as powdery mildew spores are easily dispersed. Also the short reproductive cycle could have facilitated high incidence levels as treatments began a week after inoculation (New Zealand Institute for Crop and Food Research, 2001). Application of garlic solution before inoculation was seen to assist in increasing the effectiveness of the solution in inhibiting *P. infestans* in potatoes (Bekepe *et al.,* 2006**)**. This behaviour was also observed in this experiment as the application after the disease was established provided negligible control.

Dilute acetic acid may also have failed to reduce disease incidence possibly because a lower rate was used or because it may be better as a preventative rather than a curative spray. Thus its primary action may be on spore germination than as a desiccant, as a result its use may be confined to preventative treatments rather than as curative.

Disease severity

Powdery mildew severity was significantly different throughout the growing period ($P<0.001$) among the fungicidal treatments. The untreated control had the highest disease severity across the growing period. Seven days after the first spray, bicarbonate of soda treatments had lower disease severity from garlic and dilute acetic acid treated plots while milk and folicur treated plots had the lowest severity, with the latter having the lowest disease severity throughout the growing period. Observations from day 14 did not show any significant differences among dilute acetic acid, garlic and bicarbonate of soda ($P>0.05$). At day 28 bicarbonate of soda treated plots had significantly lower ($P<0.05$) disease severity compared to garlic and dilute acetic acid treated plots. Milk and Folicur maintained a consistently lower disease severity, while the untreated control had the highest severity the observation period (Fig. 2).

Generally increased disease severity over time may have been as a result of increased susceptibility as older leaves are more susceptible to powdery mildew as well as multiplication of disease spores (Cheah *et al.,* 1996). Increase in canopy growth could also have provided shade for lower leaves and created a favourable micro climate with higher moisture content for the disease (Enright and Cipollini, 2007).

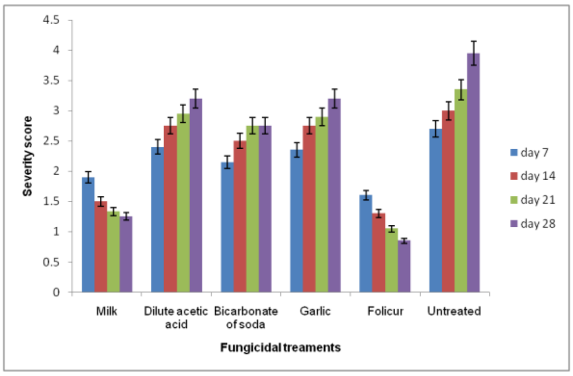

Fig. 2. Disease severity in relation to the fungicidal treatment.

Vertical bars represent standard error bars of means. Scores from 0-5 were used to represent percentage leaf cover. 0= no spores, 1= 0-20%, 2= 20-40%, 3=40-60%, 4=60-80%, 5= 80-100%.

Dilute acetic acid and garlic solution did not reduce disease severity probably the timing of the applications may have been inappropriate (Bekepe *et al.,* 2006). Lack of surfactants could also have resulted in the failure of the garlic and dilute acetic acid spray to reduce severity of the disease. Addition of a sticker such as oil could have increased their efficacy as oil contributes to the inhibition of conidia germination (Ko *et al.,* 2003). Shorter spraying intervals may also have been beneficial in the absence of a sticker as the compounds are contact in action.

No severity increase was recorded on the bicarbonate of soda treated plots on day 21 and day 28 possibly because bicarbonate of soda had managed to alter the pH on the leaf surface to prevent further spore germination and mycelial growth of the pathogen (Ko *et al.,* 2003).

The success of milk in reducing powdery mildew severity consistently throughout the growing period may possibly be attributed to the creation of an oily barrier and its fungi antagonistic properties (McGrath *et al.,* 1996). Surface pH alterations and the presence of super oxide anions could also have reduced disease severity (Bettiol 1999; Korycha-Dahl and Richardson, 1978).

The consistent reduction in severity in the folicur treated plots may mean that powdery mildew isolates in Zimbabwe are still sensitive to demethylation inhibitor fungicides and that they can be used without any problems until such a time when resistant strains occur.

Number of fruits per plant

Fungicidal treatments showed significant differences ($P<0.001$) in the number of fruits per plant. Garlic and milk had significantly higher ($P<0.05$) fruit numbers per plant than the other fungicidal treatments (Fig. 3).

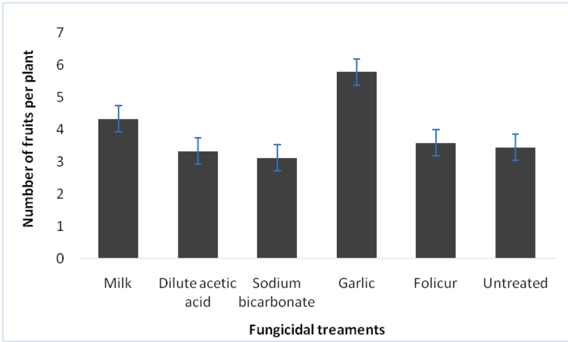

Fig. 3. Number of fruits per plant in relation to the fungicidal treatment.

Vertical bars represent standard error bars of means.

Significant increases in the number of fruits in the garlic sprayed plots can be attributed to the growth regulatory properties present in the garlic solution which facilitated an increase in the number of flowers to produce more fruits (Morsy *et al.,* 2009). Higher fruit numbers in the garlic treated plots are also explained by results obtained by (Grozav and Foarce, 2005) where essential oils of garlic significantly increased height and root length of both monocotyledonous and dicotyledonous plants. The changes in growth characteristics could have enhanced the flowering processes, as a result more fruits were observed. On the other hand, milk could have attracted pollinators such as bees resulting in a high number of fruits due to good pollination. Milk contains salts, sugars and enzymes which could also have facilitated improved growth which translated to more fruits

Yield per plot

Yield was significantly affected ($P<0.001$) by the application of different fungicides. Higher yields were recorded in plots sprayed with garlic compared to the rest of the plots (Fig. 4).

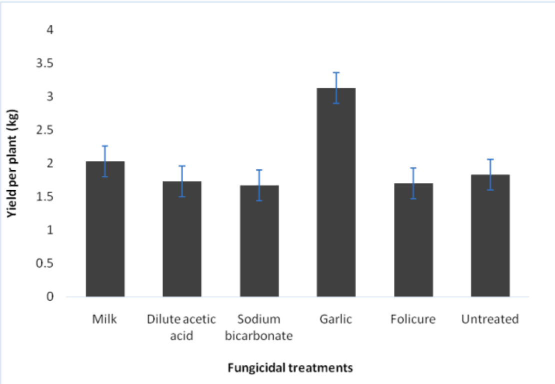

Fig. 4. Effect of fungicidal treatment on yield of butternut.

Vertical bars represent standard error bars of means.

The high yield was due to the high number of fruits rather than improved fruit weight as no significant differences were observed with respect to fruit length, diameter and weight. Garlic performed better than the other fungicides probably due to the fact that it has some properties that can alter growth of plants (Morsy *et al.,* 2009). These results concur with the findings of Grozav and Foarce, (2005) where plant growth characteristics were increased through the use of garlic extracts.

Conclusion

Treatments with folicur had the lowest powdery mildew severity and incidence while those subjected to the milk and sodium bicarbonate managed to significantly suppress the disease. All treatments had a lower diseases severity compared to the untreated control showing that the biorationals have fungicidal properties. High yields were observed in the garlic treated plots. In this experiment, powdery mildew did not affect the size of the fruit, its weight and sucrose content.

Acknowledgement

The authors are grateful to Green Finger Commercial Nursery for providing land for carrying out this research.

References

Acero FJF, Carbu M, El-Akhal MR, Garrido C, Gonzalez-Rodriguez VE, Cantoral JM. 2011. Development of proteomics-based fungicides: New strategies for environmentally friendly control of fungal diseases. International Journal of Molecular Science **12**, 795-816.

Baldwin RE, Waldenmaier CM. 1992. Evaluation of selected fungicides for the control of powdery and downy mildew in squash. Fungicide and Nematicide Tests **47**, 131.

Bekepe K, Sommartya T, Rakvidhyasastra V, Singburaudom N, Sukprasert P, Berga, L. 2006. Crude garlic extract effect on the growth of mycelia, germination of zoospores and sporangia and time of application on the infection of *Phytophthora infestans* (Mont.) de Bary of Potato under controlled conditions in Ethiopia. Kasetsart Journal of Natural Science **40**, 729-737.

Bettiol W. 1999. Effectiveness of cow's milk against zucchini squash powdery mildew (*Sphaerotheca fuliginea*) in greenhouse conditions. Crop Protection **18**, 489-491.

Chaube HS, Pundir VS. 2009. Crop diseases and their management. Second edition. PHI Learning Private Limited, New Delhi 172-175.

Cheah LH, Page BBC, Cox JK. 1996. Epidemiology of powdery mildew (*sphaerotheca fuliginea*) of squash. Proceedings of the 49th New Zealand Plant Protection Conference 147-151.

Crisp AD, Wicks TJ, Troup BG, Scot ES. 2006. Mode of action of milk and whey in the control of grapevine powdery mildew. Australasian Plant Pathology **35,** 487-493.

Daughtrey ML, Hodge KT, Shishkoff N. 2006. In: Trigiano RN, Windham MT, Windham AS. Plant pathology. Concepts and laboratory exercises. CRC Press, New York 219-220.

Enright SM, Cipollini D. 2007. Infection by Powdery Mildew *Erysiphe crucuferarum* (Erysiphaceae) strongly affects growth and fitness of *Alliaria petiolata* (Brassicaceae). American Journal of Botany **94(11)**, 1813-1820.

Grozav M, Foarce A. 2005. Preliminary study on the biological activity of *Allium sativum* L. essential oil as potential plant growth regulators. Electronic Journal of Environmental, Agricultural and Food Chemistry **4**, 1138-1142.

Gullino ML, Wardlow LR. 1999. Ornamentals. In: R. Albajes, M.L. Gullino, J.C. van Lenteren, and Y. Elad (eds.) Integrated pest and disease management in greenhouse crops. Kluwer, Netherlands 48-60.

Hacquard S. 2014. Chapter Four-The genomics of powdery mildew fungi: past achievements, present status and future prospects. Advances in Botanical Research **70**, 109-142.

Homma Y, Arimoto Y, Misato T. 1981. Effects of sodium bicarbonate on each growth stage of cucumber powdery mildew fungus (*Sphaerotheca fuliginea*) in its life cycle. Journal of Pesticide Science **6**, 201-209.

Kabir L, Sang-Woo K, Jin HJ, Yun SK, Kyoung SK, Youn SL. 2011. Inhibition effects of silver nanoparticles against powdery mildew on cucumber and pumpkin. Microbiology **39(1)**, 26-32.

Ko WH, Wang SY, Hsieh TF, Ann PJ. 2003. Effects of sunflower oil on tomato powdery mildew caused by *Oidium neolycopersici*. Journal of Phytopathology **151**, 144-148.

Korycha-Dahl M, Richardson T. 1978. Phytogeneration of superoxide anion in serum of bovine milk and in model systems containing riboflavin and amino acids. Journal of Dairy Science **61,** 400-407.

McGrath MT, Shishkoff N. 1999. Evaluation of biocompatible products for managing cucurbit powdery mildew. Crop Protection **18**, 471- 478.

McGrath MT. 1996. Successful management of powdery mildew in pumpkin with disease threshold based fungicide programmes. Plant Disease **80**, 910-916.

McGrath MT. 2001. Fungicide resistance in cucurbit powdery mildew: expertise and challenges. Plant Disease **85 (3)**, 236-245.

Morsy SM, Dragham EA, Mohamed GM. 2009. Powdery Mildew Diseases and Growth Characteristics of Cucumber. Egyptian Journal of Phytopathology **3**, 35- 46.

New Zealand Institute for Crop & Food Research. 2001. Powdery mildew in squash Fact sheet **133,** 1-2.

O'Brien RG. 1994. Fungicide resistances in population of cucurbit powdery mildew (*Sphaerotheca fuliginea).* New Zealand Journal of Crop and Horticultural Science **22**, 145-149.

Pasini C, D'Aquila F, Curir P, Gullino ML. 1997. Effectiveness of antifungal compounds against rose powdery mildew *(Sphaerotheca pannosa var. rosae)* in glasshouses. Crop Protection **16**, 251- 256.

Reuveni M, Agapo V, Reuveni R. 1995. Suppression of cucumber powdery mildew (*Sphaerotheca fuliginea*) by foliar sprays of phosphate and potassium salts. Plant pathology **44**, 31- 39.

Reuveni M, Agapov V, Reuveni R. 1993. Induction of systemic resistance to powdery mildew and growth increase in cucumbers by phosphates. Biological Agriculture and Horticulture **9**, 305-315.

Shi Z, Wang F, Zhou W, Zhang P, Jian Fan Y. 2007. Application of Osthol induces a resistance response against powdery mildew in pumpkin leaves. International Journal of Molecular Science **8**, 1001-1012.

Sinclair JB, Dingra OD. 1995. Basic Plant Pathology Methods. Second Edition. CRC Press, Florida 170-176.

Trigiano RN, Windham MT, Windham AS. 2006. Plant Pathology: Concepts and Laboratory Exercises CRC press Washington DC.

Tzeng DD, Devay JE. 1989. Biocidal activities of mixtures of methiomine and riboflavin against plant pathogenic fungi and bacteria and possible modes of action. Mycologia **81**, 402- 412.

Williams G, Williams P. 1992. More on baking soda/horticultural oil vs. fungal disease. HortIdeas. June 69.

Williams G, Williams P. 1997. Sodium bicarbonate for control of mildew on grapes. HortIdeas. June p. 70.

Ziv O, Zitter TA. 1992**.** Effects of bicarbonates and film-forming polymers on cucurbit diseases. Plant Disease **76**, 513-517.