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Research Application Summary

Botanical fungicide for sustainable production of fresh vegetables

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Abstract

Pests and diseases are a major challenge to vegetable production. Growers rely on chemical pesticides for management but despite their efficacy, synthetic pesticides have negative effects on the applicator, consumers and environment. Some of the chemical pesticides are highly toxic to humans and non-targeted organisms, and their compounds are often non-biodegradable and therefore accumulate in the environment resulting to pollution and loss of biodiversity. Therefore, there is need for safe but efficacious alternatives to chemicals in integrated pest management. Compounds of plant origin have been found safe and have been commercialized mainly for the management of insect pests. This study aims at developing a botanical fungicide from rhizomes of turmeric and ginger for the management of a range of fungal plant pathogens. Turmeric (*Curcuma longa*) and ginger (*Zingiber officinale*) rhizomes collected from farmers will be ground and extracted in alcohol. The extracts will be tested for efficacy against fungal plant pathogens in vitro. The mode of action of the active compounds on fungal spore germination, cellular structures, germ tube and hyphal growth will be determined by microscopy. The phytochemical composition of the active extracts will be analysed by high performance liquid chromatography (HPLC). The most active extract will be formulated using locally available carriers and evaluated for efficacy against fungal plant diseases under farm field conditions. It is envisaged that a broad spectrum botanical fungicide will be identified and this would contribute to effective management of fungal diseases in vegetables and reduce the usage of synthetic pesticides, thus positively impacting on human health, environment and trade.

Key words: Botanical pesticides, *Curcuma longa*, fungal pathogens, vegetables, *Zingiber officinale*

Résumé

Les pestes et les maladies sont un défi majeur pour la production de légumes. Les producteurs comptent sur les pesticides chimiques pour la gestion, mais malgré leur efficacité, les pesticides synthétiques ont des effets négatifs sur l'applicateur, les consommateurs et l'environnement. Certains pesticides chimiques sont fortement toxiques aux humains et organismes non ciblés, et leurs composés sont souvent non biodégradables et par conséquent s'accumulent dans l'environnement, entraînant une pollution et une perte de biodiversité. Par conséquent, il faut des alternatives sûres mais efficaces aux produits chimiques dans la gestion intégrée des pestes. Les composés d'origine végétale se sont révélés sûrs et ont été commercialisés principalement pour la lutte contre les insectes nuisibles. Cette étude vise à développer un fongicide botanique à partir de rhizomes de curcuma et de gingembre pour la gestion d'une gamme d'agents fongiques phytopathogènes. Les rhizomes de curcuma (*Curcuma long*a) et de gingembre (*Zingiber officinale*) collectés auprès des agriculteurs seront broyés et extraits dans l'alcool. L'efficacité des extraits sera testée contre les pathogènes fongiques des plantes in vitro. Le mode d'action des composés actifs sur la germination des spores fongiques, des structures cellulaires, du tube germinatif et la croissance des hyphes sera déterminé par microscopie. La composition phytochimique des extraits actifs sera analysée par Chromatographie liquide à haute performance (HPLC). L'extrait le plus actif sera formulé en utilisant des supports disponibles localement et évalué pour son efficacité contre les maladies fongiques des plantes dans des conditions du champ agricole. Il est envisagé d'identifier un fongicide botanique à large spectre, ce qui contribuerait à une gestion efficace des maladies fongiques dans les légumes et réduirait l'utilisation de pesticides de synthèse, ayant ainsi un impact positif sur la santé humaine, l'environnement et le commerce.

Mots clés: Pesticides botaniques, Curcuma longa, pathogènes fongiques, légumes*, Zingiber officinale*

Introduction

Botanical pesticides are derivatives of plants which possess compounds with pesticidal properties (Vidyasagar and Tabassum, 2013). These natural products have been used in the past by farmers to manage crop pests and diseases until development of synthetic pesticides. Farmers heavily rely on synthetic chemicals for crop protection due to their efficacy and availability (Mahmood *et al.*, 2016). However, synthetic pesticides have high toxicity and pollute the environment after use due to their unbiodegradable nature. They also have negative effects on non- targeted organisms such as natural enemies of crop pests. Their toxicity is of great concern as they are a health hazard to humans and animals. Presence of pesticide residues on crop produce also has negative implications on export trade especially with niche markets such as the European Union (Mishra *et al.,* 2016). Therefore, alternative pest control approaches, including natural sources, that are safe to consumers, environment and enhance compliance to the stringent market requirements are rapidly gaining attention.

The major sources of botanical pesticides include neem (*Azadirachta* indica), tobacco (*Nicotiana tabacum*), pyrethrum (*Tanacetum cinerariifolium*) and species of Ryania and Schoenocaulon. These among other plants belong to families that have been studied and shown to possess strong pesticidal properties. These plants contain active compounds such as alkaloids, tannins, terpenes, and saponins that act upon different pests in varied ways (Vidyasagar and Tabassum, 2013). However, most of the available botanical pesticides are used as insecticides only targeting the insect category of pests. Research has shown that some plants have fungicidal properties which make them effective against plant pathogens including bacteria, nematodes, viruses and fungi (Lengai and Muthomi, 2018). The aim of this study is to develop a botanical fungicide from turmeric (*Curcuma longa*) and ginger (*Zingiber officinale*) for use in different vegetable crops with the goal of coming up with a safe, cost-effective product that would enable farmers produce pesticide residue-free fresh vegetables and improve access to lucrative markets.

Turmeric and ginger were considered for this study based on findings of previous research that showed their potential efficacy against fungal plant pathogens (Muthomi *et al.,* 2017). Crude extracts from turmeric and ginger rhizomes reduced growth of selected fungal pathogens in vitro (Figure 1). Considering that turmeric and ginger belong to the same family (Zingiberaceae), their active compounds could possess synergistic properties when used in combination against plant fungal pathogens. Phytochemical analyses have shown that turmeric has compounds including vital elements and oils which make it a versatile plant. The varied phytochemical composition of turmeric explains diverse utilization of this plant as a spice, medicine, antioxidant, anti-inflammatory agent, among other uses (Yadav and Tarun, 2017). Ginger also has compounds, some of which are similar to those of turmeric, which make the plant useful as a spice and treatment of several human ailments (Prasad and Tyagi, 2015). The quantities of those compounds are nonetheless different.

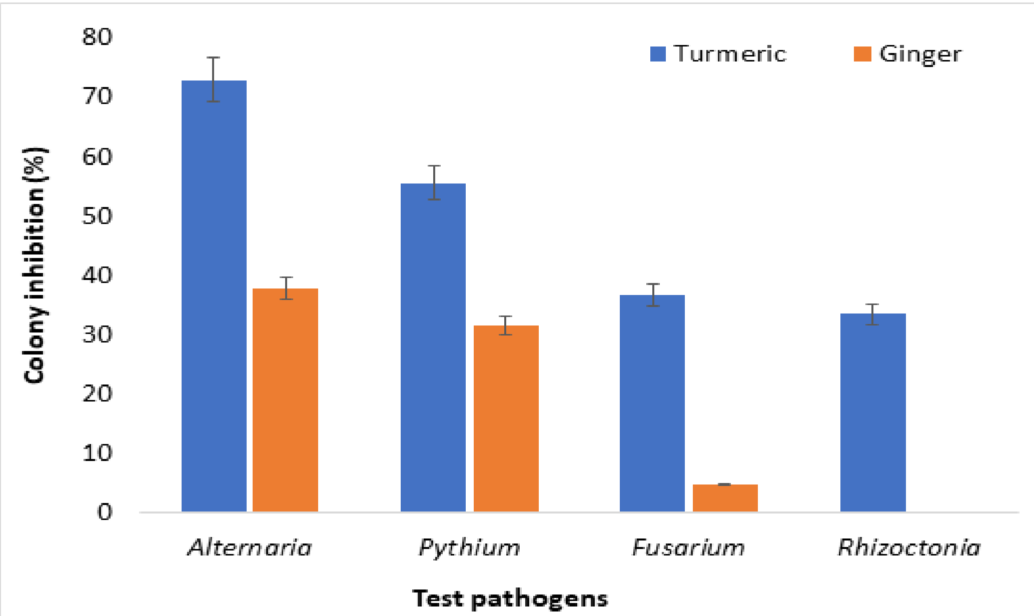

Figure 1. Percent reduction in colony diameters of fungal pathogens grown on agar media amended with extracts from turmeric and ginger rhizomes. (Muthomi *et al.,* 2017).

Research approach

Rhizomes of turmeric and ginger will be collected in northern Tanzania, dried in an oven and ground into powder. The powder (100 g) will be extracted with 95% alcohol for 30 minutes, filtered through double layer cheese cloth and Whatman No. 2 filter paper. The alcohol will be evaporated under vacuum and the resulting extracts stored at 4 ^{o}C and used for *in vitro* assay against fungal plant pathogens of vegetables according to Muthomi *et al.* (2017).

The test fungal plant pathogens will be isolated from various vegetable crops and purified on artificial culture media. The extracts will be tested for activity against the isolated plant pathogens in vitro by poisoned food technique. The culture medium will be amended with the extract and each of the test fungal pathogens will be plated separately on the amended medium. Controls will comprise of fungal pathogens cultured on medium without the extract and on medium amended with a known fungicide as a positive check. Fungal pathogen growth will be monitored over 20 days and antifungal activity will be determined by measuring fungal colony diameter and antifungal activity will be determined by calculating the percentage mycelial growth inhibition as follows:
The constituent compounds of the active extracts will be determined and identified by HPLC analysis while the mode of action will be determined by microscopy and in vitro assays.

$$\% \, Inhibition = \frac{Radial\ growth\ of\ control - Radial\ growth\ of\ treatment}{Radial\ growth\ of\ control} * 100$$

Microscopic studies will entail testing the effect of the active compounds on fungal pathogen spore germination, germ tube elongation and cellular structures. The in vitro assays will be used to determine the effect of the extracts on mycelial growth and spore production by culturing the test fungal pathogens on growth media amended with the active extracts followed by measurement of radial growth and spore concentration in suspension harvested from the test cultures. The activity of the extracts will be compared to assays containing a conventional fungicide and sterile distilled water as controls. The most effective extracts will be considered for formulation and the product will be tested for efficacy in vitro together with the raw extracts and a standard fungicide. Optimization of the materials for formulation will be done until a stable product is ready for field trials (Ahmad and Khan, 2012; Araujo *et al.,* 2014).

The crude and formulated extracts will be subjected to field trials to determine their efficacy in managing fungal diseases of tomato in small holder production in Kenya and Tanzania. The experimental treatments will comprise the crude rhizome extracts, the formulated product, a synthetic fungicide, commercial botanical pesticide, and distilled water as controls. The experiments will be laid out in randomized complete block design in three replicates over two cropping cycles. Susceptible tomato varieties will be used. Application of the treatments will be done weekly, commencing one week after transplanting until the end of harvesting. The rates of application of the botanical pesticides will be determined based on activity while those of the positive controls will be as indicated by the manufacturer. Data on distribution, incidence and severity of observed diseases and quantity of tomato fruit grades will be collected. The effectiveness of the treatments will be measured by ability to reduce the diseases levels in comparison to the controls. Harvesting will be done when tomatoes show harvest index and the fruits will be weighed and graded. The fruits will then be grouped into marketable and unmarketable categories observing and recording the differences between the treatments and the controls.

Contribution of a botanical fungicide in vegetable production. Development of a safe and efficacious botanical fungicide will contribute greatly to the incomes of small holder farmers who produce vegetables either for domestic consumption or lucrative markets. Botanical pesticides have little to zero toxicity due to their swift degradable nature. This also somehow contributes to the Africa's Agenda 2063 and Sustainable Development Goals of not only food safety but also climate change mitigation, through environmental conservation by reducing pollutants. There are stringent requirements regarding the quality and safety of the produce countries export. Presence of chemical residues on produce is not allowed and detection risks destruction of the exported consignment, at the cost of the exporter. Furthermore, some of the chemicals farmers rely on for utter effectiveness have been banned. Besides, consumers of fresh vegetables in lucrative markets have expressed their preference for organically produced food and are willing to pay an extra dime for them. This hence challenges farmers to offer better quality and safe produce in order to reap more benefits. Accordingly, a broad-spectrum fungicide will make a great contribution towards safe and quality production of fresh vegetables.

Involvement of farmers during the field experiments will augment their awareness on usage of botanical pesticides. Some farmers use botanical products based on their traditional knowledge and therefore a formulated product will present them with a chance to improve on a practise they already know. It will also create further interest and awareness to those who are unaware of such practices. Other farmers could also be interested in growing the study plants for sale. Sourcing materials for pesticide production from within the local environment should make the end product affordable to the average farmer. Moreover, production of the source plants such as turmeric and ginger would be done in arid and semi-arid areas (ASALs) to avoid competition with other uses such as spices. This could be a livelihood opportunity for inhabitants of those ASALs not to mention that it could reduce pressure on the land where food is grown.

Conclusion

Sustainable management of pests and diseases in vegetable crops is a contribution to the larger goal of sustainable agricultural development. Use of botanical pesticides is one of the many strategies for integrated pest management which is ecologically and economically acceptable. Production of a broad-spectrum botanical pesticide for use in vegetable production will be a step towards maximizing on yield of vegetables as a source of food as well as income, in a sustainable manner. Regulation of successful products should be reviewed and not handled the same way as the chemical pesticides. This will spur more innovations regarding natural products for plant protection. The knowledge in Africa regarding use of medicinal plants both for human medicine and crop protection is immense and it needs to be tapped. The continent has the capacity to revolutionize its pest management strategies based on its indigenous knowledge and engagement of the scholars it has in its academic and research institutions working in conjunction with industrial sector (Stevenson *et al.,* 2017).

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References

Ahmad, I. and Khan, M. S. A. 2012. Microscopy in mycological research with especial reference to ultrastructures and biofilm studies. pp. 646-659. In: Current Microscopy Contributions to Advances in Science and Technology. Méndez-Vilas, A. (Ed.). Formatex.

Araújo, K. M., Lima, A., Silva, J. N., Rodrigues, L. L., Amorim, A. G. N., Quelemes, P. V., Santos, R. C., Rocha, J. C., Andrades, E. O., Leite, J. R. S. A., Mancini-Filho, J. and Trindade, R. A. 2014. Identification of phenolic compounds and evaluation of antioxidant and antimicrobial properties of *Euphorbia tirucalli* L. *Antioxidants* 3: 159-175.

Lengai, G.M.W. and Muthomi, J.W. 2018. Biopesticides and their role in sustainable agricultural production. *Journal of Biosciences and Medicines* 6: 7-41.

Mahmood, I., Imadi, S. R., Shazadi, K., Gul, A. and Hakeem, K. R. 2016. Effects of pesticides on environment. pp. 253-269. In: Plant, Soil and Microbes. Springer, Cham.

Mishra, R. K., NaimuddinK. K., Sujaynand, G. K., Jagdeeswaran, R., Saabale, P. R., Akram, M. and Singh, N. P. 2016. Production and popularization of biological control agents to enhance pulse production: an eco-friendly approach. ICAR-Indian Institute of Pulses Research, Kanpur

Muthomi, J. W., Lengai, G. M. W., Wagacha, J. M. and Narla, R. D. 2017. In vitro activity of plant extracts against some important plant pathogenic fungi of tomato. *Australian Journal of Crop Science* 11 (6): 683-689.

Prasad, S. and Tyagi, A. K. 2015. Ginger and its constituents: role in prevention and treatment of gastrointestinal cancer. Gastroenterology research and practice. *Gastroenterology Research and Practice* pp.1-11.

Stevenson, P. C., Isman, M. B. and Belmain, S. R. 2017. Pesticidal plants in Africa: a global vision of new biological control products from local uses. *Industrial Crops and Products* 110: 2-9.

Vidyasagar, G. M. and Tabassum, N. 2013. Antifungal investigations on plant essential oils: a review. *International Journal of Pharmacy and Pharmaceutical Sciences* 5 (2): 19-28.

Yadav, R. P. and Tarun, G. 2017. Versatility of turmeric: A review of the golden spice of life. *Journal of Pharmacognosy and Phytochemistry* 6 (1): 41-46.