

Effects of leaf powder and ethanol extracts of selected plants on growth and incidence of onion pink root rot disease fungus (*Phoma terrestris*)

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ABSTRACT

Onion is one of the most important vegetable crops grown throughout the world and it suffers from several diseases. Pink root rot disease caused by *Phoma terrestris* is a limiting factor for onion production. The aim of this study was to investigate the effects of basil, neem, lantana and argel extracts at 0.5, 1.0 and 2.0 mg/l on fungal growth and disease incidence compared to the fungicide Premis FS (25%). All plant leaf extracts showed significant reductions in the growth of *Phoma terrestris*. Basil showed a complete inhibition zone of mycelial growth at 2.0 mg/l, followed by neem, lantana and the least inhibition was showed by argel. The fungicide Premis FS (25%), as a control, also caused a reduction of mycelial growth. All treatments of plant extracts and the fungicide Premis FS (25%) significantly reduced pink root rot disease compared to the control under greenhouse conditions. The highest reduction of the disease incidence was achieved by the fungicide Premis FS (25%), followed by the extracts of basil, neem, lantana and the least effect was shown by argel. In conclusion, it is recommended to control onion pink root rot disease using basil leaf extract at 2.0 mg/l.

INTRODUCTION

Onion (*Allium cepa* L.) is one of the most important vegetable crops grown all over the world and consumed in various forms. In the Sudan, onion is the most important vegetable crop occupying about 33% of the total area under vegetables.

Onion is subject to a number of diseases such as *Fusarium* basal rot, *Botrytis* bulb rot, black mold rot, downy mildew, rust, iris yellow spot virus, onion yellow dwarf virus, bacterial soft rot and pink root rot disease (*Phoma terrestris*) which is a limiting factor for onion production. It is found worldwide in the soil and can be 45cm deep (Babadoost, 1990). Onion diseases are mostly controlled by the use of synthetic fungicides (Bollen, 1979; Shivpuri and Gupta, 2001; Mathur and Sharma, 2006; Mathur *et al.*, 2007). The application of these chemicals cause pollution, affect wild life and develop resistance in the microbial population.

Integrated fungal disease management program emphasize the use of environmentally safe, cost effective and available alternatives such as plant extracts for more effective control of fungal diseases. Recently, workers have reported the use of plant extracts for controlling onion diseases (Bajwa *et al.*, 2003; Sharma and Sain, 2005; Singh and Singh, 2005; Singh *et al.*, 2007). Therefore, our present study investigated the effects of basil (*Ocimum basilicum* L), neem (*Azadirachta indica* L.), argel (*Solenostemma argel* L.) and lantana (*Lantana camara* L) against onion pink root rot causal agent (*Phoma terrestris*).

MATERIALS AND METHODS

Laboratory experiment

Leaves of basil (*Ocimum basilicum* L), neem (*Azadirachta indica* L), argel (*Solenostemma argel* L.) and lantana (*Lantana camara* L.) were collected from Gezira area in central Sudan. The collected leaves of tested plants were dried under shade and ground to a fine powder using mortar and pestle, kept in plastic bags and labeled for extraction. Ethanol extraction was done using soxhlet apparatus. Plant extracts were used at 0.5, 1.0 and 2.0 mg/l. The extracts were applied to potato dextrose agar media.

The pathogenic fungus (*Phoma terrestris*) was isolated from infected roots of onion. Pathogenicity test was carried out according to Koch's postulate. Mycelia disc of 5 mm diameter using a sterile cork-borer of the fungus, was placed centrally on the PDA media, which was previously mixed with the extract solutions according to the method described by Abdel-Rahim, *et al.*(1997). The fungicide Premis FS was used as a control. All treatments were incubated for 7 days at room temperature for fungal growth. The diameter of the radial growth of the fungus was measured and calculated as a percentage from the growth diameter of the control at the end of the incubation period. The experiments were arranged in a completely randomized design with two replicates. Inhibition percentages were calculated according to the formula:

$$\text{Mycelial growth inhibition (\%)} = \frac{D_c - D_t}{D_c} \times 100$$

where:

D_c = diameter of control.

D_t = diameter of test.

Greenhouse experiment

Thirty grams of leaf powder of tested plants were used for the treatment of infested soil (4kg / pot). Five seedlings of onion were planted in the pots and kept under greenhouse condition. Each

treatment was replicated three times. Treatments were arranged in a completely randomized design. After 30 days, disease incidence was calculated using the following formula:

$$\text{Disease incidence (\%)} = \frac{\text{Number of infected plants}}{\text{Total number of plants}} \times 100$$

Data were statistically analyzed using the standard analysis of variance procedure. Means were separated using Duncan's Multiple Range Test at 5% level of significance.

RESULTS AND DISCUSSION

Laboratory experiment

Table 1 shows the main effects of plant extracts on fungal growth and disease incidence in the greenhouse. Basil leaf extract was the most effective in reducing fungal growth and disease incidence, whereas argel was the least effective. Basil extract showed the best fungal growth inhibition (27.1 mm) compared to the control (65.8mm), and the fungicide Premis FS (43.1mm), followed by neem (32.8 mm) and lantana (41.9mm), whereas the argel extract was the least one(51.8mm). Inhibition of fungal growth by plant extracts obtained here agreed with the findings of many workers who reported that onion diseases were controlling by using plant extracts (Bajwa, *et al.*, 2003;Sharma and Sain, 2005; Singh and Singh, 2005; Singh *et al.*, 2007).

Greenhouse experiment

Table 1 shows the effects of leaf extracts of selected plants on the percentage of disease incidence. Basil resulted in the lowest percentage of disease incidence followed by neem, lantana and lastly argel. The fungicide Premis FS resulted in zero disease incidence. Needless to say, the untreated control resulted in 100% disease incidence.

Table 1. Main effects of plant extracts and the fungicide Premis FS (25%) on the diameter of fungal growth and disease incidence.

| Treatment | Diameter of fungal growth | Disease incidence |
|------------------|---------------------------|-------------------|
| Basil | 27.1c | 24.4c |
| Neem | 32.8c | 37.7bc |
| Lantana | 41.9b | 42.2bc |
| Argel | 51.8a | 44.4b |
| Premis FS (25%) | 65.8a | 00.0d |
| Infected control | 43.1b | 100a |
| SE ± | 1.58 | |

* Means followed by the same letter in a column are not significantly different according to Duncan's Multiple Range test at $P \geq 0.05$.

Management of plant diseases is an essential process through which incidence of plant disease, crop losses and cost of production are minimized; contributing to sustained crop production. Among the control measures, chemical control may appear more effective due to its direct lethal effects on fungal growth. However, chemical fungicides cause damage to the environment, increase of production cost and health risk. Natural plant extracts may contribute to minimize the adverse effects of plant diseases on crop production without the risk of synthetic chemicals. Many studies demonstrated the positive effects of natural extracts against bacterial diseases (Ahmed, 2005) and

nematodes (Gamal *et al.*, 2008). Basil extracts reduced both fungal growth and pink root rot disease which is clearly suggested to be used to control plant diseases.

Table 2 shows the interaction effects of plant extracts and their concentrations on the diameter of fungal growth and the percentage of mycelial growth inhibition. Neem and basil leaf extracts resulted in the smallest diameter of fungal growth and the highest mycelial growth inhibition, followed by lantana and argel. Lantana leaf extract was as effective as that recorded for Premis (25%). Generally, the high concentration of leaf extracts gave the best results for all plant types compared to the lower concentration.

Table 2. Interaction effects of plant extracts and their concentrations on the diameter of fungal . and mycelial growth inhibition percentage.

| Treatment | Concentration (mg/l) | | | | | |
|---------------|--------------------------------|-------------------------------|--------------------------------|-------------------------------|--------------------------------|-------------------------------|
| | 0.5 | | 1.5 | | 2.0 | |
| | Diameter of fungal growth (mm) | Mycelia growth inhibition (%) | Diameter of fungal growth (mm) | Mycelia growth inhibition (%) | Diameter of fungal growth (mm) | Mycelia growth inhibition (%) |
| Basil | 46.0 b | 30.0 | 35.0 ab | 46.0 | 00.0 a | 100.0 |
| Neem | 42.0 b | 34.0 | 34.2 ab | 47.0 | 21.5 a | 67.0 |
| Lantana | 53.5 bc | 18.0 | 45.3 b | 31.0 | 27.0 ab | 58.0 |
| Argel | 64.0 c | 20.0 | 50.3 bc | 24.0 | 41.0 b | 37.0 |
| Premis Fs 25% | 52.0 bc | 20.0 | 44.3 b | 33.0 | 32.5 ab | 50.0 |
| Control | 65.5 c | 00.0 | 65.8 c | 00.0 | 65.8 c | 00.0 |
| SE ± | | | | 2.73 | | |
| CV(%) | | | | 8.84 | | |

* Means followed by the same letter in a column are not significantly different according to Duncan's Multiple Range test at $P \geq 0.05$.

The ultimate goal of control measures is to keep the diseases losses below the economic level by reducing the pathogen population multiplication. Natural extracts succeeded in reducing fungal growth as well as disease incidence. Basil extracts resulted in an acceptable level of disease incidence without adverse effects on the environment which may be encountered by the use of synthetic fungicides.

Many plants products have been reported as having antimicrobial activities against plant pathogenic fungi (Sokovicet, *et al.*, 2009). Extracts of many plant species such as *Lawsonia alba* L, *Parthenium hysterophorus* L and *Moringa oleifera* L were reported to have antifungal activities against soil-borne pathogens such as *Alternaria porri*, *Fusarium oxysporium* and *Stemphylium vesicarium* (Gaikwad *et al.*, 2014).

CONCLUSION

In conclusion, it is recommended to use basil extracts at 0, 2 mg/l for the control of onion pink root rot disease. Further research work is needed to identify the active ingredients responsible for fungicidal activity.

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بمرض تأثير مسحوق الأوراق ومستخلصات الإيثانول لبعض النباتات على نمو ونسبة الإصابة (*Phoma terrestris* عفن جذور البصل الوردي)

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الخلاصة

يعتبر محصول البصل من أهم المحاصيل المزروعة حول العالم ولكنه يعاني من عدد من
من *Phoma terrestris* الأمراض. يعتبر مرض عفن جذور البصل الوردي الذي يسببه الفطر
العوامل المحددة لإنتاج البصل. هدفت الدراسة لمعرفة تأثير مستخلصات الريحان و الحرجل و
اللانثانا والنيم بتركيزات صفر و 1 و 2 مليجرام/لتر على نمو الفطر ونسبة الإصابة بالمرض في
كشاهد. أظهرت الدراسة نقصاً معنوياً في نمو الفطر (25%) Premis FS وجود المبيد الفطري
. ثبط الريحان في التركيز العالي (2 مليجرام/لتر) نمو الفطر تثبيطاً كاملاً *Phoma terrestris*
تلاه النيم و اللانثانا وأخيراً الحرجل. كذلك نتج عن معاملة النباتات المصابة في المشتل
الأعلى (25%) Premis FS بالمستخلصات النباتية نقصاً في نسبة الإصابة وكان المبيد الفطري
في التأثير على معدل الإصابة ثم تلاه الريحان ثم النيم و اللانثانا و آخرها الحرجل. خلصت
الدراسة للتوصية بمعاملة البصل المصاب بمرض تعفن جذور البصل الوردي بمستخلص أوراق
الريحان بالتركيز العالي 2 مليجرام/لتر

