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METABOLITES OF *PENICILLIUM CITRINUM* AS POTENT HERBICIDES AGAINST PARTHENIUM WEED

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A B S T R A C T

This study was undertaken to assess the herbicidal activity of metabolites of *Penicillium citrinum* against parthenium weed (*Parthenium hysterophorus* L.). Two growth media *viz.* malt extract (ME) broth and potato dextrose (PD) broth were used for preparation of fungal metabolites by growing the fungus in these media for 15 days. The metabolites were evaluated in laboratory bioassays using their original (100%) and diluted (50%) concentrations, and compared with a negative control as well as with original and diluted growth media. In general, both the growth media had insignificant effect on germination and various growth parameters except root length where original ME broth significantly increased this parameter. Both the concentrations of the metabolites prepared in different growth media significantly ($P \leq 0.05$) suppressed germination and various shoot/root growth parameters over control. Metabolites prepared in PD broth exhibited more activity than the metabolites prepared in ME broth. There was up to 31%, 77%, 92% and 77% decrease in germination, shoot and root lengths, and seedlings dry weight due to metabolites in ME broth, while these parameters were decreased by 57%, 89%, 94% and 93% due to application of metabolites in PD broth, respectively.

Keywords: Fungal metabolites, Natural herbicides, *Parthenium hysterophorus, Penicillium citrinum*.

INTRODUCTION

Partheniumis a noxious weed of Asteraceae, generally grows on wastelands and also in some crops (Javaid and Anjum, 2005; Javaid and Khan, 2020). It is present in many countries of the world including Pakistan (Riaz and Javed, 2011). It lies at the 10th position in the ranking of the most hazardous weeds (Li *et al.,* 2018), and the 7th most destructive weed in Australia, Asia and Africa (Kaur *et. al.,* 2014). It is adapted to establish in a variety of environmental conditions (Kumari, 2014). It has the ability to reduce production of pasture, disturb biodiversity in natural communities, and adversely affect the health of animals and man (Khan *et al.,* 2019). Many weed management strategies have been utilized by farmers to minimize the yield losses caused by *P. hysterophorus* (Adkins and Shabbir, 2014). Among these, chemical control is the most widely adapted method to control parthenium and other weeds (Javaid, 2007; Bajwa *et al*., 2015). However, it causes environmental pollution and human health risks, harmful for the pollinators and affects the beneficial microorganisms present in the soil (Travlos *et al.,* 2011). Thus, chemical approach is threatening to agricultural sustainability and must be replaced with environment friendly approaches (Javaid *et al*., 2020).

Researchers are now searching for environment friendly weed management tactics to exclude the option of synthetic and chemical herbicides (Ellison and Cock, 2017; Javaid *et al.,* 2020). One of these alternative approaches is the use of fungal metabolites either in crude form or as purified compounds (Javaid *et al.,* 2013; Akbar and Javaid, 2013). There are various reports in

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which fungal metabolites successfully seized the growth of various weeds including parthenium (Javaid and Ali, 2011; Javaid *et al.,* 2017), through the production of antifungal compounds namely holadysenterine (Akbar *et al.,* 2014), drazepinone (Evidente *et al.,* 2005), and ophiobolin A (Evidente *et al.,* 2006). The herbicidal activity of various fungal species including *Trichoderma*, *Dreschlera*, *Alternaria* and *Aspergillus* against parthenium has been reported in previous literature (Akbar and Javaid, 2012, 2013; Javaid *et al*., 2013, 2017; Bashir *et al.,* 2018). Application of metabolites of *Alternaria macrospora* and *A. alternata* caused significant damaging to parthenium development (Saxena and Kumar, 2010; Kaur *et al.,* 2014). Recently, Bashir *et al.* (2018) evaluated metabolites of *Aspergillus niger* against *P. hysterophorus,* which were very effective in controlling the growth of *P. hysterophorus. Penicillium citrinum* is known to produce various secondary metabolites (Jha *et al.,* 2016). However, their herbicidal activity against parthenium has not been studied so far. Thus, the present study was designed to evaluate the herbicidal activity of secondary metabolites of *Penicillium citrinum* against parthenium.

MATERIALS AND METHODS

Preparation of fungal metabolites: Culture of *P. citrinum* was procured from First Culture Bank of Pakistan. Sub-culturing of the fungus was done by using 2% malt extract agar medium. Fungal metabolites were prepared under antiseptic conditions by inoculating 2% ME and 2% PD broths with the fungal inoculum in separate flasks of 250-mL volume (Bashir *et al.,* 2018). The fungal inoculated flasks were incubated at 25 °C for 15 days at a static condition. As incubation period completed, separation of the fungal mat from broth was made by sterilized muslin cloth and the obtained liquid cultures were filtered through A4 filter paper and then through Millipore filter papers. Distilled water was added to original metabolites (100%) to make lower concentration (50%) of the fungal metabolites in ME and PD.

Laboratory bioassays: Seeds of *P. hysterophorus* were collected during December 2019 as seeds of winter season show 100% germination percentage (Javaid *et al.,* 2010). The surface sterilized seeds (using 2% sodium hypochlorite) of parthinium were placed in a Petri plate of 9-cm diameter on a filter paper bed and 2.5 mL of concentrated and diluted growth media as well as fungal metabolite were applied to each respective Petri plate. For a control, same amount of water was applied in another set of Petri plates. Each treatment was replicated four times with 25 seeds in each plate. These plates were adjusted in a growth chamber at 25 °C following completely randomized design (CRD). Data regarding germination, shoot and root lengths, and fresh and dry biomasses were recorded after ten days.

STATISTICAL ANALYSIS

Data about germination and growth of parthenium were analyzed by one-way ANOVA followed by application of Tukey's HSD test at $P \leq 0.05$ using software Statistix 8.1.

RESULTS AND DISCUSSION

Effect of *P. citrinum* metabolites on germination: The effect of metabolites of *P. citrinum,* prepared in ME and PD broths, on germination of parthenium seeds is presented in Figure 1, 2A and 3A. In general, the effect of both the growth media was insignificant on germination. On the other hand, both the concentrations (original or 100% and diluted or 50%) of the metabolites of *P. citrinum* prepared either in ME broth or PD broth, significantly reduced germination. However, there was marked difference in herbicidal activities of the metabolites prepared in the different media. Metabolites of the fungus prepared in PD broth were more suppressive towards germination than the metabolites prepared in PD broth. Original and diluted metabolites in PD broth reduced germination by 57% and 49%, respectively, over control (Figure 3A). By contrast, original and diluted metabolite in ME broth reduced germination by 31% and 21%, respectively (Figure 2A). Previous reports also showed that metabolites of different fungal species reduced germination of parthenium weed. Javaid and Adrees (2009) assessed the herbicidal activity of metabolites of 9 fungal species against parthenium. Among these, metabolites of *Alternaria alternata*, *Drechslera rostrata* and *Cladosporium* sp. significantly retarded the germination of parthenium seeds up to 90%, 50% and 73%, respectively. Similarly, metabolites of *Alternaria japonica* and *Aspergillus niger* significantly reduced germination of parthenium (Javaid *et al.,* 2017; Bashir *et al.,* 2018).

In the present study, two growth media were used for the preparation of metabolites of *P. citrinum.* There was a pronounced difference in herbicidal activities of the metabolites prepared in the two different media. Metabolites prepared in PD broth showed greater herbicidal activity than the metabolites prepared in ME

broth. These findings are in line with the results of previous similar studies. Javaid *et al.* (2017) used ME and PD broths for preparation of metabolites of *Alternaria japonica.* Bioassays with these metabolites showed a markedly higher herbicidal activity of PD metabolites than ME metabolites against parthenium. Similar differences in activity of metabolites of *A. niger* and *Trichoderma* spp., prepared in different growth media have also been reported against parthenium (Javaid *et al.,* 2013; Bashir *et al.,* 2018).

Effect of *P. citrinum* metabolites on shoot length: The effect of *P. citrinum* metabolites on shoot length of parthenium seedling is illustrated in Figure 1, 2B and 3B. In negative control, shoot length was 2.14 cm. Both the growth media showed an insignificant effect on shoot length. Shoot length in original and diluted ME was 2.09 cm and 2.10 cm, and that of in PD broth it was 2.00 cm and 2.08 cm, respectively. However, metabolites prepared in different growth media variably and significantly reduced this growth parameter as compared to control. Shoot length in original and diluted fungal metabolites of ME broth was 0.5 cm and 0.64 cm that was 77% and 70% lower over control (Figure 2B). Likewise, hoot length in original and diluted metabolites of PD broth was 0.24 cm and 0.42 cm that was 89% and 80% lower than in control (Figure 3B). Similar reduction in shoot length of parthenium has also been reported due to metabolites of *Fusarium oxysporum, F. solani* and *Drechslera australiensis* (Javaid and Adrees, 2009),

Figure 1. Effect of metabolites of *Penicillium citrinum,* prepared in malt extract broth and potato dextrose broth, on growth of parthenium.

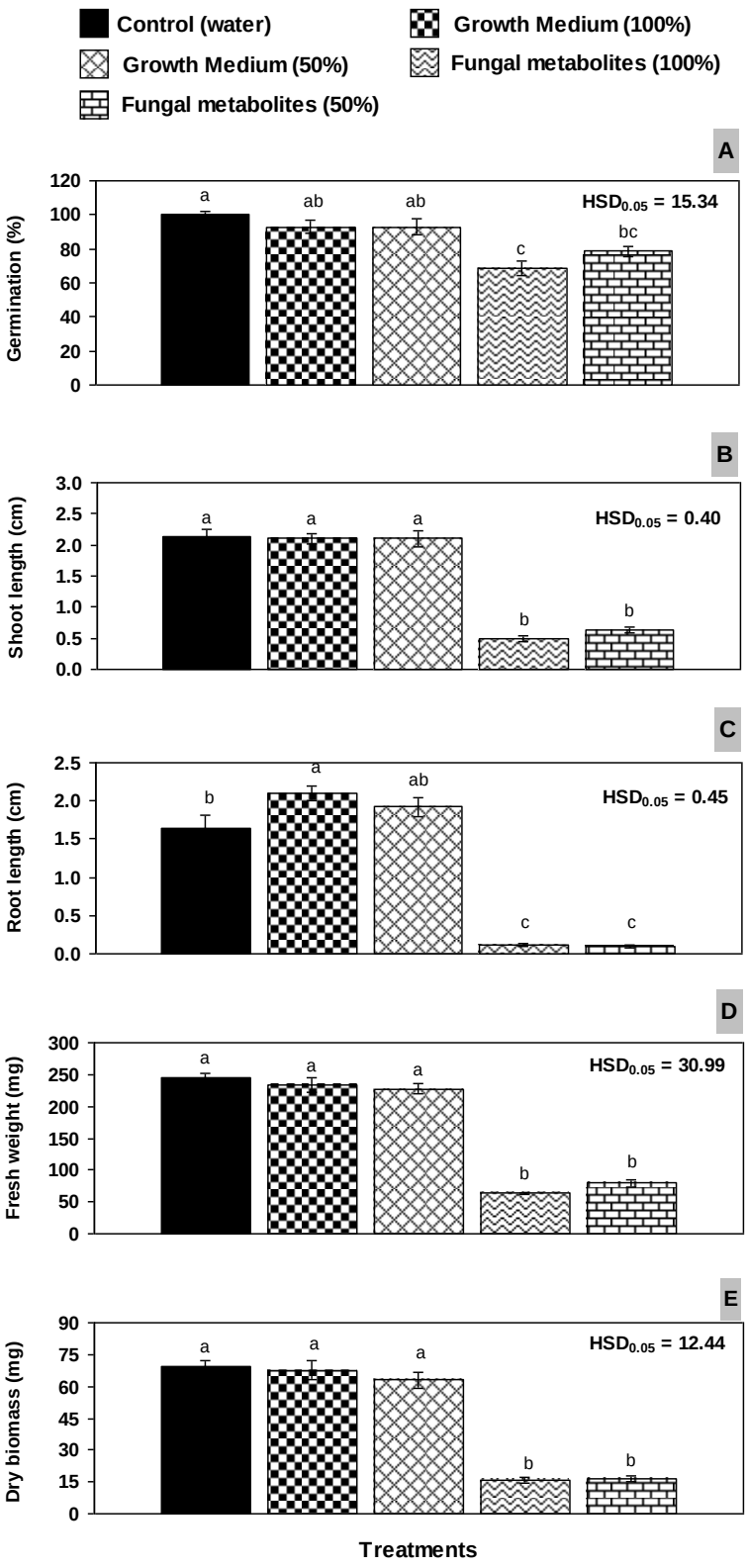

Figure 2. Effect of metabolites of *Penicillium citrinum,* prepared in malt extract broth, on germination and growth of parthenium. Vertical bars show standard errors of means. Values with different letters show significant difference (P≤0.05) as determined by Tukey's HSD test.

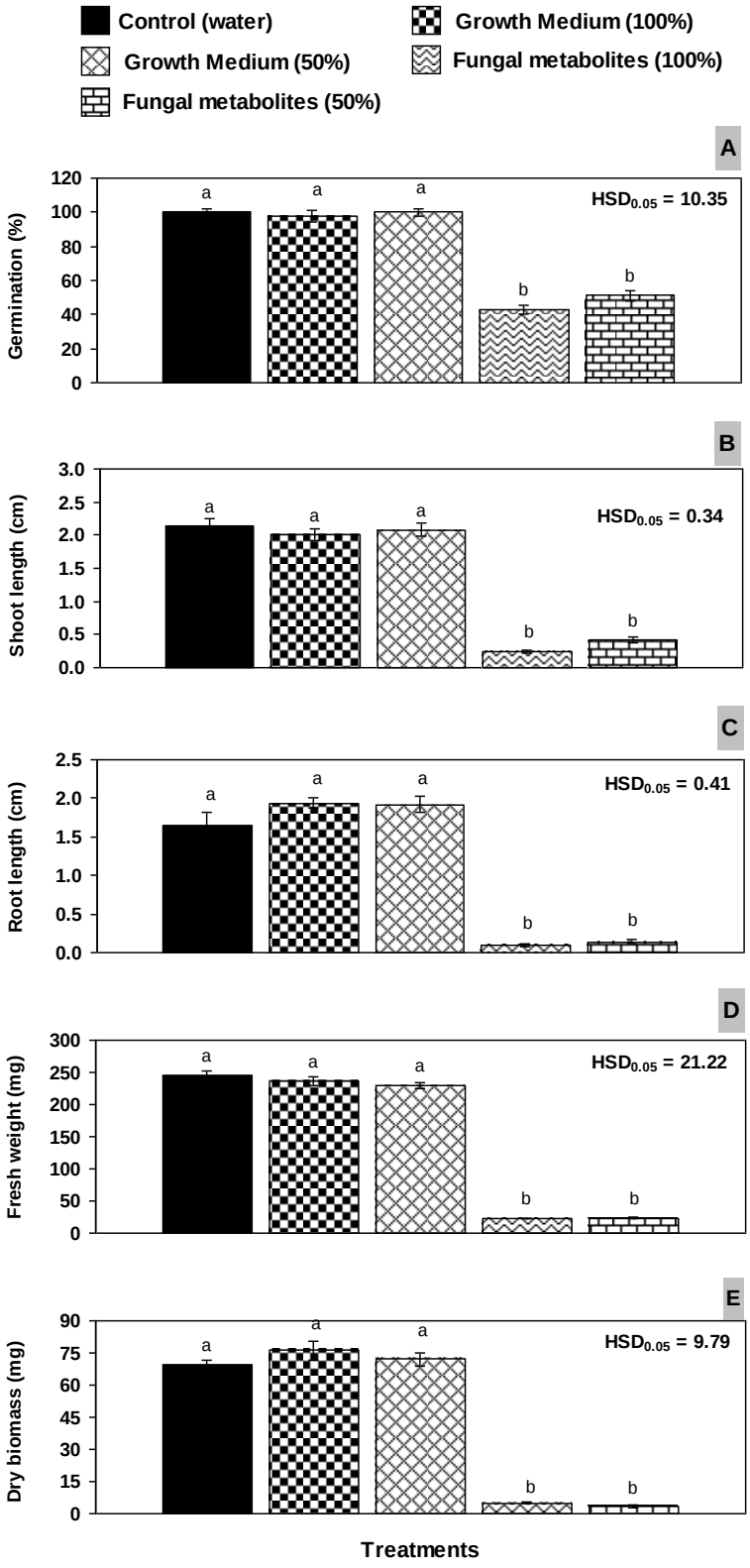

Figure 3. Effect of metabolites of *Penicillium citrinum,* prepared in potato dextrose broth, on germination and growth of parthenium. Vertical bars show standard errors of means. Values with different letters show significant difference (P≤0.05) as determined by Tukey's HSD test.

Effect of *P. citrinum* metabolites on root length: Root length in control was 1.64 cm. Both the growth media stimulated root length. In case of PD broth, the increase in root length was insignificant. By contrast, there was a significant ($P \leq 0.05$) increase in root length due to original ME broth while the effect of diluted ME was insignificant. Metabolites of *P. citrinum* produced in either of the two growth media significantly declined root length to variable extents. In case of ME broth, there was 92% and 94% decrease in root length while in case of PD broth, there was 94% and 91% suppression in root length due to original and diluted metabolites of *P. citrinum* (Figure 1, 2C and 3C). In general, root growth was more susceptible to the application of fungal metabolites than the shoot growth. It might be ascribed to the fact that growth inhibiting substance from the surrounding are first absorbed by the roots and therefore they show more abnormal behavior to application of fungal metabolites than the shoot (Javaid and Shah, 2007). Similar differential response of shoot and root growth to the application of fungal metabolites (Javaid *et al.,* 2011) or allelopathic plant extracts (Javaid and Khan, 2020) has also been reported in many previous studies.

Effect of *P. citrinum* metabolites on biomass: The effect of both the growth media on biomass of parthenium was insignificant. On the other hand these parameters were significantly suppressed by application of fungal metabolites prepared in either of the two growth media. Fungal metabolites, original and diluted, prepared in ME broth declined dry biomass of parthenium by 77% and 76%, while those prepared in PD broth reduced this parameter by 93% and 95%, respectively, over control (Fig 2D & E, 3D & E). Earlier, metabolites of *A. niger, A. japonica* and *Trichoderma* spp. prepared in different growth media showed variable effects on biomass of parthenium (Javaid *et al.,* 2013, 2017; Bashir *et al.,* 2018).

CONCLUSION

Metabolites of *P. citrinum* possess herbicidal activity against parthenium. Metabolites prepared in PD broth showed higher herbicidal activity than those prepared in ME broth. Root growth was more susceptible to fungal metabolites than the shoot growth. Further studies are required to identify the potential herbicidal constituents in metabolites of *P. citrinum* prepared in PD broth.

REFERENCES

Adkins, S. and A. Shabbir. 2014. Biology, ecology and management of the invasive parthenium weed (*Parthenium hysterophorus* L.). Pest Management Science, 70: 1023-1029.

Akbar, M. and A. Javaid, 2013. Prospects of using fungal metabolites for the management of *Rumex dentatus,* a problematic weed of wheat. International Journal of Agriculture and Biology, 15: 1277-1282.

Akbar, M. and A. Javaid. 2012. Herbicidal activity of fungal culture filtrates against *Chenopodium album* L. and *Avena fatua* L. Journal of Animal and Plant Sciences, 22: 977-982.

Akbar, M., A. Javaid, E. Ahmad, T. Javed and J. Clary. 2014. Holadysenterine, a natural herbicidal constituent from *Drechslera australiensis* for management of *Rumex dentatus* Journal of Agricultural and Food Chemistry, 62: 368-372.

Bajwa, A.A., G. Mahajan and B.S. Chauhan. 2015. Nonconventional weed management strategies for modern agriculture. Weed Science, 63: 723-747.

Bashir, U., A. Khan and A. Javaid. 2018. Herbicidal activity of *Aspergillus niger* metabolites against parthenium weed. Planta Daninha, 36: Article ID e018167123.

Ellison, C.A. and M.J.W. Cock. 2017. Classical biological control of *Mikania micrantha*: the sustainable solution. CAB International, 162-190.

Evidente, A., A. Andolfi, A. Cimmino, M. Vurro, M. Fracchiolla and R. Charudattan. 2006. Herbicidal potential of ophiobolins produced by *Drechslera gigantea*. Journal of Agricultural and Food Chemistry, 54: 1779-1783.

Evidente, A., A. Andolfi, M. Vurro, M. Fracchiolla, M.C. Zonno and A. Motta. 2005. Drazepinone, a trisubstituted tetrahydronaphthofuroazepinone with herbicidal activity produced by *Drechslera siccans*. Phytochemistry, 66: 715-721.

Javaid, A. 2007. Efficacy of some common herbicides against parthenium weed L. Pakistan Journal Weed Science Research, 13: 93-98.

Javaid, A. 2010. Herbicidal potential of allelopathic plants and fungi against *Parthenium hysterophorus* - a review. Allelopathy Journal, 25: 331-334.

Javaid, A. and Ali, S., 2011. Alternative management of a problematic weed of wheat *Avena fatua* L. by metabolites of Trichoderma. Chilean Journal of Agricultural Research, 71: 205-211.

Javaid, A. and H. Adrees. 2009. Parthenium management by cultural filtrates of phytopathogenic fungi. Natural Product Research,23: 1541-1551.

Javaid, A. and I.H. Khan. 2020. Potential use of *Coronopus didymus* in parthenium management. Pakistan Journal of Weed Science Research,26: 37-45.

Javaid, A. and M.B.M. Shah. 2007. Phytotoxic effects of aqueous leaf extracts of two Eucalyptus spp. against *Parthenium hysterophorus* L. Science International (Lahore), 19: 303-306.

Javaid, A. and T. Anjum. 2005. *Parthenium hysterophorus* L.- A noxious alien weed. Pakistan Journal of Weed Science Research,11: 171-177.

Javaid, A., A. Javaid and M. Akbar. 2011. Herbicidal potential of culture filtrates of *Drechslera* spp. against *Parthenium hysterophorus.* Chilean Journal of Agricultural Research,71: 634-637.

Javaid, A., G. Shafique, S. Ali and A. Shoaib. 2013. Effect of culture medium on herbicidal potential of metabolites of Trichoderma species against *Parthenium hysterophorus*. International Journal of Agriculture and Biology, 15: 119-124.

Javaid, A., S. Shafique and S. Shafique. 2010. Seasonal pattern of seeds dormancy in *Parthenium hysterophorus* L. Pakistan Journal of Botany, 42: 497-503.

Javaid, A., T. Mubeen, U. Bashir and A. Shoaib. 2017. Management of parthenium weed using metabolites of *Alternaria japonica*. Planta Daninha, *35*: Article ID e017161195.

Javaid, N., M.H. Shah, I.H. Khan, A. Javaid and S.M. Waleed. 2020. Herbicidal activity of *Ageratum conyzoides* against parthenium. Pakistan Journal of Weed Science Research, 26: 137-146.

Jha, S.N., P. Jaiswal, M.K. Grewal, M. Gupta and R. Bhardwaj. 2016. Detection of adulterants and contaminants in liquid foods-a review. Critical Reviews in Food Science and Nutrition,56: 1662-1684.

Kaur, M., N.K. Aggarwal, V. Kumar and R. Dhiman. 2014. Effects and management of *Parthenium hysterophorus*: a weed of global significance. International Scholarly Research Notices, 2014: Article ID 368647.

Khan, N., D. George, A. Shabbir, S.W. Adkins. 2019. Suppressive plants as weed management tool: managing *Parthenium hysterophorus* under simulated grazing in Australian grasslands. Journal of Environmental Management, 247: 224-233.

Kumari, M., 2014. *Parthenium hysterophorus* L.: A noxious and rapidly spreading weed of India. Journal of Chemical, Biological and Physical Sciences, 4: Article ID 1620.

Li, J., M. Li, X. Gao and F. Fang. 2018 Corn straw mulching affects *Parthenium hysterophorus* and rhizosphere organisms Crop Protection, 113: 90-96.

Riaz, T. and A.Javaid. 2011. Prevalence of alien weed *Parthenium hysterophorus*L. in grazing and wastelands of district Attock. Pakistan. Journal of Animal and Plant Sciences,21: 542-545.

Saxena, S. and M. Kumar. 2010. Evaluation of *Alternaria alternata* ITCC4896 for use as mycoherbicide to control *Parthenium hysterophorus*. Archives of Phytopathology and Plant Protection, 43: 1160-1164.

Travlos, I.S., C.N. Giannopolitis and G. Economou. 2011. Diclofop resistance in sterile wild oat (*Avena sterilis* L.) in wheat fields in Greece and its management by other post-emergence herbicides. Crop Protection, 30: 1449-1454.

Contribution of Authors:

Arshad Javaid	:	Supervised the work, did statistical analysis, and finalized the paper
Iqra H. Khan	:	Did experimental work
Shahbaz Ahmed	:	Contributed in manuscript writing
Malik F. H. Ferdosi	:	Collection of materials, contributed in writing
Syeda F. Naqvi	:	Contributed in drafting and reviewing the manuscript