Fungicidal Management of Stemphylium blight of Onion caused by *Stemphylium vesicarium* (Wallr.) Simmons

Bhavya Mishra* and Rajesh Pratap Singh

Department of Plant Pathology, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar-263145, Uttarakhand, India.

http://dx.doi.org/10.13005/bbra/2539

(Received: 12 August 2017; accepted: 05 September 2017)

Onion is an important commercial crop grown all over the world. It is attacked by many diseases which cause yield losses and result in lowering the quality and export potential of the produce. Stemphylium blight caused by *Stemphylium vesicarium* (Wallr.) Simmons is one such disease, which has become an economic threat since past few years, especially in Northern and Eastern India. In the present study, field evaluation as well as *in vitro* study was conducted for evaluating some fungicides for the management of Stemphylium blight as well as to observe their effects on yield and quality parameters of onion. The results of the *in vitro* study revealed that fluopyram + tebuconazole gave complete mycelial inhibition of test fungus at 50 ppm concentration. Results of the field experiments showed that all the treatments significantly reduced the disease severity. However, strobilurins were found to be more effective, giving more than 50% disease control over check. Keeping in view the overall performance of the fungicides, it was concluded that the combination products azoxystrobin 25% + flutriafol 25% SC and fluopyram 20% + tebuconazole 20% SC can be recommended for the management of Stemphylium blight of onion under field conditions.

**Keywords:** Stemphylium, triazoles, strobilurins, management, fungicides, *in vitro*.

---

Onion is the most widely cultivated species of the genus *Allium* and is one of the most important commercial crops grown all over the world. India is the second largest producer of onion in the world, but falls behind other countries in terms of productivity. Among the various reasons, diseases and pests are an important constraint in onion production. The crop is attacked by many diseases which cause yield losses and also result in deteriorating the quality and export potential of the produce. Stemphylium blight caused by *Stemphylium vesicarium* (Wallr.) Simmons is one such disease, which was not a major economic threat earlier, but has become a serious problem throughout the country since recent past, especially in Northern and Eastern India including Uttarakhand. Surveys conducted by NHRDF indicated that Stemphylium blight was more severe in the winter/summer than in the rainy season with 1.3-100 per cent incidence (Gupta et al., 1994) and sometimes may even cause 100 per cent crop losses (Singh et al., 1992). Leaf blight can lead to premature defoliation the crop, thereby making it more susceptible to secondary and post-harvest infections. Disease intensity is higher in seed crop than in bulb crop. Various management strategies like cultural practices, field sanitation, and biological control have been suggested. All these methods are effective only when employed well in advance as precautionary measure (Kata, 2000). But once the disease has appeared, these methods become less effective. In such situation, chemical control offers a good choice to grower for managing the disease. Chemical pesticides
have been used since long and have the advantage of providing quick, effective and economic management of plant diseases. Therefore, the present study was undertaken to evaluate the *in vitro* and field efficacy of some new generation fungicides against Stemphylium blight of onion. Since yield and quality parameters are the main focal point of any agronomic practice, therefore whether the applied disease control measures are effective or not, is often judged by the final harvested produce. So the effect of fungicide application on the yield of the produce was also assessed.

**MATERIALS AND METHOD**

**In vitro Study**

The present investigations on *in vitro* bioassay of different fungicides were carried out using Completely Randomized Design (CRD). Evaluation of triazole and strobilurin fungicides was carried out for their efficacy to inhibit the mycelial growth of the test fungus *Stemphylium vesicarium* by “Poisoned food technique” as described by Sharvelle (1961). The triazole fungicides viz. tebuconazole (25EC), propiconazole (25EC), difenoconazole (25EC), flutriafol (25SC); fluopyram (50SC) is a novel compound belonging to a new chemical class of succinate dehydrogenase inhibitors; strobilurin fungicides viz. azoxystrobin (25SC) and combination products viz. fluopyram + tebuconazole (40SC) and azoxystrobin + flutriafol (25SC) were tested at 50 parts per million (ppm), 100 ppm and 200 ppm concentrations. Stock solution of the fungicides, each of 10000 ppm was prepared and required volume of stock solution was added to double strength PDA medium just before pouring so as to obtain the desired concentration of the “poisoned” medium. 20 ml of the poisoned medium was poured into sterile Petri plates. After solidification, the plates were then inoculated with 5 mm mycelial discs from 10 days old culture of the test fungus cut by a sterilized cork borer and incubated at 24 ± 1°C. Three replications were maintained for each treatment. The fungus grown on un-amended PDA served as control. The radial growth of the colony was recorded after 10 days and percent mycelial growth inhibition was calculated as described by Vincent (1927).

\[ I = \frac{C - T}{C} \times 100 \]

Where,

- \( I \) = Inhibition percent
- \( C \) = Growth of the test fungus in control (mm)
- \( T \) = Growth of the test fungus in treatment (mm)

**Field Study**

Field experiment was conducted at Vegetable Research Centre of G.B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand during *rabi* crop seasons in 2013-14 using Randomized Block Design (RBD) with three replications. Three foliar sprays of fungicides at 15 days interval, beginning at 45 days after transplanting were given. The treatments used were:

Observations on disease severity were recorded one week after each spray. Disease severity in field was monitored on ten selected plants per plot in three replicated plots using 0-5 scale. Percent disease index (PDI) was calculated by the following formula given by Wheeler (1969).

\[ \text{PDI} = \frac{\text{Sum of all disease ratings of 6 leaves} \times \text{maximum rating value}}{\text{Total number of leaves}} \times 100 \]

Per cent disease control (PDC) was worked out in a similar was as per cent mycelial inhibition, i.e. by subtracting the disease severity in treatment from that of check and then dividing by disease severity in check. Harvesting of each treatment was done separately and the yield and bulb size was recorded. Total yield was recorded and marketable yield was calculated by discarding those bulbs which were having bolters, or were highly deformed or rotten and of unacceptable, non-marketable quality. Grading was done on the basis of bulb size into three grades viz. A (diameter more than 55 mm), B (diameter 45-55 mm) and C (diameter less than 45 mm).

**Statistical Analysis**

The results of the lab studies and field experiment were statistically analyzed using the statistical package developed by GBPUAT, Pantnagar (STPR 2 and 3). The analysis of replicated data on the mycelial growth of the test fungus was done as per CRD and that of the field replicated data on disease severity and yield was done as per RBD.
RESULTS AND DISCUSSION

In vitro Study

Eight fungicides were evaluated at three different concentrations i.e. 50 ppm, 100 ppm and 200 ppm to see their effect on mycelial growth of *Stemphylium vesicarium*. Data pertaining to colony diameter and percent inhibition of radial growth presented in Table 2 revealed that all the fungicides significantly inhibited the growth of the test fungus at all tested concentrations. Fluopyram, tebuconazole and their combination product fluopyram + tebuconazole were found most effective at all the concentrations, giving 100 per cent growth inhibition, followed by propiconazole. At 50 ppm concentration (Fig 1), azoxystrobin was found less effective giving 29.20 per cent mycelial inhibition. Same trend followed at 100 and 200 ppm concentration, where highest inhibition (100%) was by fluopyram, tebuconazole and fluopyram + tebuconazole. Azoxystrobin and azoxystrobin + flutriafol were found at par giving the least per cent inhibition in all concentrations. Among the triazoles, tebuconazole provided 100 percent growth inhibition at all concentrations and was found most effective followed by propiconzole, flutriafol and difenoconazole. Fluopyram was found quite effective but its combination with azoxystrobin reduced its efficacy. It was almost at par with azoxystrobin. It showed that azoxystrobin

![Fig. 1. Effect of fungicides on the radial growth of *Stemphylium vesicarium* at 50 ppm](image)
and its combination azoxystrobin + flutriafol were not effective in inhibiting the mycelial growth of *Stemphylium vesicarium*. The effect of fungicide treatment as well as that of fungicide concentration and their interaction was found significant in inhibiting the mycelial growth of the test fungus.

The result of present study is in accordance with the reports by Collina et al. (2006) on *in vitro* sensitivity of *Stemphylium vesicarium* to various fungicides which showed that best activity on mycelial growth was obtained with anilinopyrimidines, tebuconazole, flutriafol, difenoconazole and propiconazole. Mishra and Gupta (2012) conducted *in vitro* study and found that azoxystrobin (0.1 %), propiconazole (0.1 %) were effective. Sensitivity of *S. vesicarium* to strobilurin fungicides has also been confirmed by Alberoni et al. (2006). However, the present study showed that azoxystrobin was less effective as compared to triazoles. The reports of triazoles being effective were supported by Mohan et al. (2004) who recorded the highest pathogen inhibition by triazole fungicides. Out of the 3 triazoles tested, tebuconazole was the most effective for *S. botryosum* followed by difenoconazole and hexaconazole. In the present study also, tebuconazole was found to be best, giving complete inhibition of *S. vesicarium*, followed by propiconazole and flutriafol, which confirms the superiority of triazole fungicides over others in inhibiting the growth of *S. vesicarium* and indicates that there is significant difference in the efficacy within the members of triazole group also.

### Table 1. List of fungicides and their doses used for the field studies

| S. No. | Fungicide                  | Dose (g a.i. ha⁻¹) |
|--------|----------------------------|-------------------|
| 1.     | Fluopyram 50% SC           | 75                |
| 2.     | Tebuconazole 25% EC        | 75                |
| 3.     | Propiconazole 25% EC       | 125               |
| 4.     | Difenoconazole 25% EC      | 125               |
| 5.     | Azoxystrobin 25% SC        | 100               |
| 6.     | Flutriafol 25% SC          | 100               |
| 7.     | Fluopyram 20%SC + Tebuconazole 20% SC | 150 |
| 8.     | Azoxystrobin 25% SC + Flutriafol 25% SC | 25 |

*all values are mean of three replication

### Table 2. Effect of different fungicides and their concentrations on the mycelial growth of *S. vesicarium*

| Treatments                      | Colony Diameter (mm)* | Growth Inhibition (%) |
|---------------------------------|-----------------------|-----------------------|
|                                 | 50ppm     | 100ppm   | 200ppm | 50ppm     | 100ppm   | 200ppm   |
| Fluopyram                       | 0         | 0        | 0      | 100       | 100      | 100.00   |
| Propiconazole                   | 8.33      | 6.00     | 2.67   | 81.75     | 86.86    | 94.16    |
| Tebuconazole                    | 0         | 0        | 0      | 100       | 100      | 100.00   |
| Difenoconazole                  | 18.17     | 17.00    | 13.17  | 60.22     | 74.09    | 78.83    |
| Fluopyram+Tebuconazole          | 0         | 0        | 0      | 100       | 100      | 100.00   |
| Azoxystrobin                    | 32.33     | 28.67    | 25.5   | 29.20     | 37.23    | 44.16    |
| Flutriafol                      | 16.17     | 11.83    | 9.67   | 64.6      | 74.09    | 78.83    |
| Azoxystrobin+Flutriafol         | 31.33     | 29.00    | 26.17  | 31.39     | 36.5     | 42.70    |
| Check                           | 45.67     | 45.67    | 45.67  | nil       | nil      | nil      |

S. Em ± CD (p=0.05)

| Treatments | Concentrations | Treatment X Concentration | CV |
|------------|----------------|--------------------------|----|
| 0.55       | 1.56           | 0.32                     | 0.90|
| 0.95       | 2.70           | 10.79                    |    |
Table 3. Field evaluation of fungicides for management of Stemphylium blight of onion

| Treatments                        | Percent Disease Index* | Percent Disease Control |
|-----------------------------------|------------------------|-------------------------|
|                                   | 45 DAT | 60 DAT | 75 DAT | 45 DAT | 60 DAT | 75 DAT |
| Fluopyram                         | 10.36  | 32.02  | 48.90  | 46.46  | 46.95  | 38.50  |
| Propiconazole                     | 13.41  | 37.14  | 59.88  | 30.65  | 38.46  | 24.69  |
| Tebuconazole                      | 10.80  | 35.60  | 47.55  | 44.15  | 41.01  | 40.20  |
| Difenoconazole                    | 12.19  | 35.15  | 54.61  | 37.00  | 41.75  | 31.31  |
| Fluopyram+Tebuconazole            | 9.49   | 32.51  | 42.71  | 50.96  | 46.14  | 46.28  |
| Azoxystrobin                      | 8.79   | 26.26  | 45.01  | 54.55  | 56.50  | 43.39  |
| Flutriafol                        | 12.28  | 35.68  | 49.25  | 36.51  | 40.88  | 38.05  |
| Azoxystrobin+Flutriafol           | 6.51   | 28.02  | 45.98  | 66.34  | 53.58  | 42.17  |
| Check                             | 19.34  | 60.35  | 79.51  | -      | -      | -      |
| S.Em ±                           | 0.47   | 1.79   | 0.66   | -      | -      | -      |
| CD (p=0.05)                       | 1.41   | 5.37   | 1.99   | -      | -      | -      |
| CV                               | 7.13   | 8.65   | 2.18   | -      | -      | -      |

*all values are mean of three replications, DAT= Days After Transplanting

Table 4. Effect of fungicide application on the yield and bulb size of onion

| Treatments                        | Total Yield* (t ha⁻¹) | Marketable Yield* (t ha⁻¹) | A | Bulb Grades (%) |
|-----------------------------------|-----------------------|-----------------------------|---|-----------------|
|                                   |                       |                             |   |                 |
| Fluopyram                         | 20.38                 | 19.51                       | 4.79 | 16.79 | 78.42 |
| Propiconazole                     | 21.19                 | 20.59                       | 2.21 | 19.92 | 77.87 |
| Tebuconazole                      | 23.83                 | 22.26                       | 7.65 | 24.98 | 67.38 |
| Difenoconazole                    | 22.69                 | 22.01                       | 5.73 | 18.62 | 75.65 |
| Fluopyram+Tebuconazole            | 22.65                 | 22.05                       | 8.28 | 18.37 | 73.34 |
| Azoxystrobin                      | 21.06                 | 19.97                       | 2.90 | 23.01 | 74.10 |
| Flutriafol                        | 24.97                 | 23.51                       | 5.78 | 17.95 | 76.27 |
| Azoxystrobin+Flutriafol           | 22.28                 | 21.56                       | 3.74 | 18.69 | 77.57 |
| Check                             | 16.32                 | 15.98                       | 1.95 | 14.50 | 83.55 |
| S.Em ±                           | 1.21                  | 1.11                        | -   | -    | -    |
| CD (p=0.05)                       | 3.64                  | 3.32                        | -   | -    | -    |
| CV                               | 9.69                  | 9.23                        | -   | -    | -    |

*all values are mean of three replications
offered disease control, but within the triazole group there is remarkable difference in efficacy against Stemphylium blight of onion. Flutriafol was most effective followed by tebuconazole and difenoconazole. Propiconazole was found least effective. Mohan et al. (2004) who recorded that out of the 3 triazoles tested, tebuconazole was the most effective for *S. botryosum* followed by difenoconazole and hexaconazole, thus showing the high degree of variation among the same group of fungicides. This study supports our findings, that there are differences in the efficacy of triazole fungicides and tebuconazole was found superior over other triazoles like propiconazole. Results of field trials by Tesfaendrias et.al (2012) showed that azoxystrobin + difenoconazole (Quadris top), fluopyram + pyremethanil (Luna Tranquility), difenoconazole (Inspire) were the most effective in reducing stemphylium leaf blight, which is supporting the findings of the present study that combination products especially a combination of strobilurin and triazole (azoxystrobin + flutriafol) are providing a better disease control.

Strobilurin fungicides are quinone outside inhibitors and are effective in inhibiting the germination of fungal spores. Although their effect against mycelial growth of the fungus is also documented, but inhibiting spore germination is what they do best. On the other hand, triazole fungicides inhibit ergosterol biosynthesis in pathogenic fungi. Because spores already contain ergosterol, the triazole fungicides are generally not very effective in preventing spore germination. Triazole fungicides work best by inhibiting fungi’s mycelial growth (Bradley, 2011). This might be the probable reason of difference in the in *vitro* and field results discussed above. Strobilurins have the novel mode of action and thus perform better under field conditions rather than providing direct mycelial inhibition in *vitro*. Moreover, strobilurin fungicides have been researched for their potential physiological effects like delayed senescence, altered amounts of plant hormones, increased activity of antioxidative enzymes which further strengthens the plants in fields and thus quite possibly provide better disease management.

**Effect on Yield**

All the treatments were separately harvested and their yields were recorded and bulbs were graded on the basis of size. The results presented in Table 4 shows the effect of fungicidal sprays on yield and bulb size. The data revealed that all the treatments were significantly effective over control in increasing the yield but within the treatments, all the treatments were found at par in terms of marketable yields. However, the higher total as well as marketable yields were obtained in the triazole treatments (flutriafol, tebuconazole and fluopyram + tebuconazole). The lowest total and marketable yield were obtained in case of check (16.32 and 15.98 t ha⁻¹). The highest share of A grade bulbs in the marketable yield (8.28 %) was in case of fluopyram + tebuconazole although all treatments were at par. These results are in accordance with the studies of Mohan et al. (2004) working on chilli and onion, who recorded that the triazole fungicides also produced the highest yields.

**CONCLUSION**

Laboratory studies revealed that fluopyram + tebuconazole at 50 ppm completely inhibited the radial growth of *S. vesicarium*. Under field conditions, azoxystrobin + flutriafol was found most effective in reducing the disease severity and providing better disease control. Keeping in view the overall performance of all the tested fungicides, the combination products were found to be superior in managing the disease. Therefore, azoxystrobin 25% + flutriafol 25% SC @ 25 g a.i. ha⁻¹ and fluopyram 20% + tebuconazole 20% SC @150 g a.i. ha⁻¹ can be recommended for the management of Stemphylium blight of onion under field conditions.

**ACKNOWLEDGEMENTS**

Authors are thankful to the Department of Plant Pathology, G.B. Pant University of Agriculture and Technology, Pantnagar (Uttarakhand) for providing the necessary funding, laboratory and field research facilities during the present studies and furnishing the investigation with valuable information.

**REFERENCES**

1. Alberoni, G., Collina, M. and Brunelli, A. *Stemphylium vesicarium* sensitivity to dicarboximide and strobilurin fungicides. Giornate-Fitopatologiche. 2006; 2:105-110.
2. Bhatia, J.N. and Chahal, D. Studies on effectiveness of certain new fungicides in controlling Stemphylium blight of onion seed crop. Agricultural Science Direct. 2014; 34(3): 237-239.

3. Bradley, C.A. Providing Some Clarity on Fungicide Products. The Bulletin. Pest management and crop development information for Illinois. 2011; No. 2 Article 4/April 8. http://bulletin.ipm.illinois.edu/print.php?id=1454 Accessed on 11-08-2017.

4. Collina, M., Alberoni, G. and Brunelli, A. In vitro sensitivity of Stemphylium vesicarium to fungicides. Bulletin OILB/SROP. 2006; 29(1): 155-161.

5. Gupta, R. P., Srivastava, K. J., Pandey, U. B. and Midmore, D. J. Diseases and insect pests of onion in India. International Symposium on Alliums for the Tropics. Acta Horticulturae. 1994; 358: 265-269.

6. Kata, J. Physical and cultural methods for the management of soil borne pathogens. Crop Protection. 2000; 19: 725-731.

7. Mishra, R. K. And Gupta, R. P. In vitro evaluation of plant extracts, bio-agents and fungicides against Purple blotch and Stemphylium blight of onion. Journal of Medicinal Plants Research. 2012; 6(45): 5658-5661.

8. Mohan, C., Thind, T. S., Prem, Raj and Arora, J. K. Promising activity of triazoles and other fungicides against fruit rot of chilli and Stemphylium blight of onion. Plant Disease Research Ludhiana. 2004; 19(2): 200-203.

9. Sharvelle, E. G. The nature and use of modern fungicides. Burgess Publishing Co., Minnesota, USA, 1961; 308 p.

10. Singh, D., Dhiman, J. S., Sidhu, A. S., Hari Singh, D. and Singh, H. Current status of onions in India: strategies for disease resistance breeding for sustained production. Onion Newsletter Tropics. 1992; 4: 43-44.

11. Tesfaendrias, M.T.; Paibomesai, M.; Celetti, M. and McDonald, M.R. 2012. The battle against Stemphylium leaf blight of onion in Ontario, Canada. http://www.hort.cornell.edu/expo/proceedings/2014/Onions/Stemphylium%20Tesfaendrias.pdf. Retrieved on 15-06-2015.

12. Ureba, B. M. J., Prados-Ligero, A. M. and Melero-Vara, J. M. Effectiveness of tebuconazole and procymidone in the control of Stemphylium leaf spots in garlic. Crop Protection. 1998; 17 (6): 491-495.

13. Vincent, J. M. Distortion of fungal hyphae in the presence of certain inhibitors. Nature. 1927; 159: 850.

14. Wheeler, B. E. An Introduction to Plant Diseases. John Wiley and Sons Ltd, London, UK. 1969; pp 43-46.