

## Efficacy of Fluopicolide 6.25 % + Propamocarb Hydrochloride 62.5% SC against cucumber downy mildew disease caused by *Pseudoperonospora cubensis*

K. Nagendran\*, Sujoy Saha<sup>1</sup>, A.B. Rai and B. Singh

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### Abstract

The widely distributed and devastating oomycete, *Pseudoperonospora cubensis* (Berkeley & Curtis) Rostovtsev is the causal agent of cucurbit downy mildew, infecting over 40 host species. The present study was undertaken to evaluate different doses of Fluopicolide 6.25% + Propamocarb Hydrochloride 62.5% SC against downy mildew of cucumber under field condition in two consecutive seasons. The pooled data indicates that combination dose of Fluopicolide 6.25 % + Propamocarb Hydrochloride 62.5% SC @ 1500 ml/ha provided the best disease control (82.43%) amongst all the treatments along with increasing the yield by 39.8%. The combination was not phytotoxic on cucumber up to the dose of 3000 ml/ha.

**Keywords:** Fluopicolide, cucumber, downy mildew, Propamocarb hydrochloride, synergism

### Introduction

Cucumber (*Cucumis sativus*) is the fourth most widely cultivated vegetable crop in the world and is known for its several uses. Besides being an excellent source of vitamin K and Molybdenum, cucumbers are rich in pantothenic acid, minerals and vitamin C. It helps to fight inflammation, stress and obesity as well as supports digestive and heart health (Kumar et al. 2010). The most widespread and serious pathogen hampering the yield of cucumber is *Pseudoperonospora cubensis* (Berkeley & Curtis) Rostovtsev., first described by Berkeley in 1868 from Cuba. The exceptional ability of the spores of *P. cubensis* to survive uncongenial conditions coupled with the difficulty of covering the underside of the foliage

near the ground with fungicides restricted the efficacy of fungicidal control of downy mildew in the 1970's. This brought to fore the need towards breeding for resistance as a potential alternative. Cucumber yield losses from downy mildew remained minimal compared to other diseases till 2004, when a more virulent race of *P. cubensis* caused more than 40% loss for cucumber growers (Colucci et al. 2006). Thus the host resistance was no longer effective due to which the control of downy mildew demanded an intensive fungicide program as the first line of defense (Savory et al. 2011).

The chemical control of downy mildews, including that of the cucurbits, got a major break-through with the discovery of the systemic action of the acylalanine group on these diseases (Palti and Cohen 1980). Phenylamides, strobilurins, carboxylic acid amides (CAAs), benzamides, cyanoacetamide-oxime and carbamates are among the fungicide chemistries commonly used to control this disease. Since their discovery in 1977, phenylamide fungicides such as metalaxyl contributed to effective control of diseases caused by members of the order Peronosporales (Gisi and Cohen 1996). However, 2 years after introduction, the first metalaxyl-resistant isolates of *P. cubensis* were reported in Israel (Reuveni et al. 1980). Strobilurin fungicides, such as azoxystrobin and trifloxystrobin, were launched in 1996 (Sauter et al. 1999) and represented a new and important class of chemicals that provided control of fungal diseases caused by oomycete, ascomycete, and basidiomycete pathogens. However, reduced efficacy of strobilurins was also reported in *P. cubensis* in Japan in 1999 (Ishii et al. 2001). The systemic carboxylic acid amide fungicides were developed primarily to control downy mildew and were effective in controlling cucurbit downy mildew until resistance in *P. cubensis* was reported in 2004 in China (Zhu et al. 2007). In the late 1990s, the pyridinylmethylbenzamide fungicide, fluopicolide (2,6-dichloro-N-[[3-chloro-5-(trifluoromethyl)-2-pyridinyl]methyl])

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Indian Institute of Vegetable Research, Varanasi – 221305, Uttar Pradesh

<sup>1</sup>National Research Centre for Grapes, Pune – 412 307, Maharashtra

\*Corresponding author, Email: krishnagendra@gmail.com

benzamide), the first of a new generation of fungicides with a novel mode of action, was developed to control a wide spectrum of oomycete diseases (Briggs *et al.* 2006). Mixtures of single-site (systemic) and multi-site (protectant) fungicides have been recommended for disease control programs in an effort to prolong the efficacy of systemic fungicides by delaying build up of resistant strains in pathogen populations (Skylakakis 1981). Considering this, a field trial was envisaged to evaluate the efficacy of the synergistic combination of fluopicolide and propamocarb hydrochloride against downy mildew of cucumbers. Propamocarb hydrochloride is a carbamate fungicide with specific activity against oomycetous pathogens. Thus, the combination Fluopicolide 6.25% + Propamocarb Hydrochloride 62.5% SC (F+PH) was evaluated for its efficacy against downy mildew of cucumber.

### Materials and Methods

The trial was conducted at the research farm of ICAR-Indian Institute of Vegetable Research at Varanasi, Uttar Pradesh, India for two consecutive kharif seasons (2014 and 2015). Twenty eight days old seedlings of cucumber var. Swarna Sheetal were transplanted in plots of area 5m×5m. Standard package of practices were followed to raise the crop. Seven different treatments comprising of 3 combination doses of Fluopicolide and Propamocarb hydrochloride @ 1000, 1250 and 1500 ml/ha, 2 solo doses of Fluopicolide 48 % SC@ 200 ml/ha and Propamocarb Hydrochloride 72.2 % SL@ 1250 ml/ha, a standard check fungicide Cymoxanil 8% + Mancozeb 64%-72% WP @ 1500 ml/ha and an untreated control were laid down in the randomized block design (RBD) with three replications. Fungicide application began with the visibility of initial disease symptoms i.e. 35 days after planting for downy mildew (10-12 leaf stage) and repeated once after 15 days. Ten plants from each replication excluding the border rows were taken at the beginning of each spray and scored for disease using 0-4 rating scale (0- healthy or no disease, 1- 1-10%, 2- 11-25%, 3 -26-50%, 4 -51-% and above area infected) of Thind *et. al* (1991). The Percent Disease Index (PDI) was calculated based on the observation using the formula,

The harvesting was done after fruit maturity and fruit yield was calculated in quintals per hectare. All the data obtained were statistically analyzed. For phytotoxicity observations, the cucumber plants were treated with the combination dose of Fluopicolide and Propamocarb hydrochloride @ 1500 and 3000 g/ha doses. Observations on leaf injury, wilting, vein clearing, necrosis, epinasty and hyponasty were recorded on ten randomly selected cucumber plants from each plot at

0, 1, 3,5, 7, 10 and 15 days after application (DAA). The level of phytotoxicity was estimated by visual assessment on below mentioned scale of 0-10 ( ).

### Results and Discussion

The analyzed data presented in Table 2 clearly indicates that the two doses of Fluopicolide 6.25 %+Propamocarb Hydrochloride 62.5% SC (F+PH) @ 1250 ml/ha and 1500 ml/ha provided a significant disease control of downy mildew of cucumber with significant increase in the yield over its solo doses as well as the untreated control. The dose of F+PH@ 1250g/ha manifested a disease control of 75.48% for downy mildew with an yield increase of 33.3%,while, the higher dose of F+PH @ 1500 g/ha gave a disease control of 82.43% with a subsequent increase in yield by 39.8% as compared to untreated control. The percent increase in yield obtained from both these doses of F+PH significantly surpassed the solo doses of Fluopicolide 48% SC@ 200 ml/ha and Propamocarb Hydrochloride 72.2% SL @ 1250 ml/ha clearly hinting towards synergy. No phytotoxicity symptoms were developed on leaves up to 15 days of spray in the two mentioned doses of the combination of F+PH. This indicated that the two test combinations are not phytotoxic to cucumber up to the mentioned doses.

The results obtained help to elucidate the point that combination dose of F+PH surpassed the solo doses of the same fungicides in controlling downy mildew of cucumber, thereby indicating that this combination is more effective in controlling the disease. The major homologous component of this combination is the Benzamide fungicide, Fluopicolide, which functions by delocalizing spectrin-like proteins of the pathogen (FRAC code list 2016, <http://FRAC.info>). Fluopicolide is a systemic fungicide known to affect all growth stages of oomycetous pathogen, including release and motility of zoospores, cyst germination, mycelial growth and sporangial production (Jackson *et al.* 2010). Fluopicolide-treated hypha or zoospores of *P. infestans* showed a fast perturbation of a spectrin-like protein (Toquin *et al.* 2006) but the specific target protein of fluopicolide remains unclear.

*P. cubensis* being a widely devastating foliar disease of cucumber, many fungicides were targeted for its control.

**Table 1:** Phytotoxicity rating scale

Score	Phytotoxicity (%)	Score	Phytotoxicity (%)
0	No phytotoxicity	6	51 – 60
1	0 – 10	7	61 – 70
2	11 – 20	8	71 – 80
3	21 – 30	9	81 – 90
4	31 – 40	10	91 – 100
5	41– 50		

**Table 2:** Effect of different doses of Fluopicolide 6.25 %+ Propamocarb Hydrochloride 62.5% SC on downy mildew of cucumber

T. No.	Treatments	Dose (ml/ha)	Per cent Disease Index*				Fruit yield of Cucumber (Q/Ha)			
			2014-15	2015-16	Mean	PDC <sup>a</sup>	2014-15	2015-16	Mean	PIY <sup>b</sup>
1.	Untreated control	-	68.05 (55.58)	71.6 (57.9)	69.82	0.00	63.80	33.47	48.64	-
2.	Fluopicolide 6.25 %+Propamocarb Hydrochloride 62.5% SC	1000	17.73 (24.9)	29.6 (32.97)	23.68	66.08	75.83	43.97	59.90	23.1
3.	Fluopicolide 6.25 %+Propamocarb Hydrochloride 62.5% SC	1250	12.13 (20.38)	22.1 (28.01)	17.12	75.48	78.29	51.43	64.86	33.3
4.	Fluopicolide 6.25 %+Propamocarb Hydrochloride 62.5% SC	1500	11.60 (19.91)	12.9 (20.91)	12.27	82.43	78.89	57.13	68.01	39.8
5.	Fluopicolide 48 % SC	200	28.97 (32.55)	34.8 (36.14)	31.88	54.34	68.00	43.03	55.52	14.1
6.	Propamocarb Hydrochloride 72.2 % SL	1250	36.77 (37.32)	35.1 (36.34)	35.95	48.51	69.37	42.17	55.77	14.6
7.	Cymoxanil 8% + Mancozeb 64%-72%WP	1500	23.10 (28.69)	28.2 (32.04)	25.65	63.26	73.46	49.27	61.37	26.1
	CD(0.05)	-	2.01	7.01	-	-	2.40	4.75	-	-

\* The percent disease index shown in the table is the reading taken 10 days after second spray; Values in the parentheses are arcsine transformed  
a: Per cent disease control ; b: per cent increase in yield

**Table 3:** Observations on phytotoxicity of Fluopicolide and Propamocarb hydrochloride 62.5% SC on cucumber

S. No.	Treatments	Dosage (g/ ha)	Yellowing & Necrosis (Days after spray)					Vein Clearing (Days after spray)					Wilting & Leaf injury (Days after spray)					Epinasty and Hyponasty (Days after spray)					Stunting (Days after spray)				
			1	3	5	7	14	1	3	5	7	14	1	3	5	7	14	1	3	5	7	14	1	3	5	7	14
			1	Untreated control	NIL	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
2	Fluopicolide 6.25 %+Propamocarb Hydrochloride 62.5% SC	1500	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
3	Fluopicolide 6.25 %+Propamocarb Hydrochloride 62.5% SC	3000	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Note: Observations were recorded at 1, 3, 5, 7, 10 & 15 days after the spray of fungicides

The results obtained from fungicide efficacy trials indicate that fungicides with active ingredients zoxamide, cymoxanil, famoxadone and cyazofamid are the most efficacious ones in managing the downy mildew on cucumber (Colucci 2008). Interestingly, the multi-site fungicides including dithiocarbamates (e.g. mancozeb), phthalimides (folpet), chloronitriles (chlorothalonil) and copper formulations still account for about 50% of the downy mildew fungicide market. Among the single-site fungicides, four chemical classes the Quinone outside inhibitors (QoIs; 'strobilurins', mainly azoxystrobin, famoxadone, fenamidone), the phenylamides (PAs, mainly mefenoxam), the carboxylic acidamides (CAAs; mainly dimethomorph, iprovalicarb, benthialicarb, mandipropamid) and the cyanoacetamid-oximes (cymoxanil) are the market leaders (Gisiand and Sierotzki 2008) against downy mildew. Among the chemical products evaluated in a study against downy mildew of cucumber, the pyridinylmethyl-benzamide (fluopicolide), carbamate (e.g., propamocarb) and quinone inside inhibitor (QiI) (e.g., cyazofamid) groups of fungicides were the most effective, whereas the CAA (Dimethomorph and mandipropamid) group was the least effective (Ojiambo et al. 2010).

Fluopicolide's are also highly effective against other

oomycetes such as *Phytophthora infestans*, for which consistent control has been reported (Cooke and Little 2006). Treatments involving these fungicides result in lower disease severity and higher yield than the non-treated control. In addition, a locally systemic + protectant fungicide program was compared to a protectant-only program with respect to delay of fungicide application. Results indicated that the locally systemic + protectant program was more effective than the protectant only program when applied before disease detection (Colucci 2008). Fluopicolide in combination with propamocarb hydrochloride is superior to metalaxyl and mancozeb in controlling potato late blight (Cooke and Little 2006). The present study is in sync with the aforementioned finding i.e. Fluopicolide in combination with propamocarb hydrochloride is superior to Cymoxanil 8% + Mancozeb 64%-72%WP in controlling oomycetous pathogen, *P.cubensis*.

## Conclusion

Fluopicolide 6.25 %+Propamocarb Hydrochloride 62.5% SC @ 1250-1500 g/ha as a foliar spray manifested significantly higher per cent control of downy of cucumber over the solo doses, increased the yield and devoid of any phytotoxic effects on cucumber. The

combinations may be recommended for the management of downy mildew diseases and subsequently included in the Good Agricultural Practices (GAP) of cucumber.

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### सारांश

व्यापक रूप से वितरित और विनाशकारी उमाइसीजीट स्फूडोपेरेनोस्पोरा क्यूबेनसीस (बरकेले एवं करटीस) रोस्टोभट्सेप कद्दू वर्गीय सब्जियों में मृदुरोमिल आसिता रोग का कारक है जो 40 से अधिक प्रजातियों को संक्रमित करता है। वर्तमान अध्ययन में फ्लूपीकोसाइड (6.25 प्रतिशत) और प्रोपामोकार्ब हाइड्रोक्लोराइड (62.5 प्रतिशत एस सी) के विभिन्न मात्राओं का मूल्यांकन ककड़ी की मृदुरोमिल आसिता के प्रति क्षेत्र दशा में लगातार दो अवधि में किया गया। एकत्रित आँकड़ा से स्पष्ट होता है कि सभी उपचारों में फ्लूपीकोसाइड और प्रोपामोकार्ब की संयोजक में (1500 मिलीलीटर/हेक्टेयर) रोग के नियंत्रण एवं उपज वृद्धि करने में सबसे अच्छा है।

### References

- Briggs G, Mansfield D, Moloney B, Gary S and Wegmann T (2006) The discovery and chemistry of fluopicolide: a new standard for oomycetes disease control. *PflanzenschutzNachr. Bayer (Ger. Ed.)* 59:141-152
- Colucci SJ, Wehner TC and Holmes GJ (2006) The downy mildew epidemic of 2004 and 2005 in the eastern United States. *Proc. Cucurbitaceae* 403-411.
- Colucci S (2008) Host range, fungicide resistance and management of *Pseudoperonospora cubensis*, causal agent of cucurbit downy mildew. MS Thesis, North Carolina State University, Raleigh USA
- Cooke LR and Little G (2006) Evaluation of fluopicolide-containing formulations for the control of potato late blight in Northern Ireland. *PflanzenschutzNachr. Bayer (Ger. Ed.)* 59: 303-316.
- Gisi U and Cohen Y (1996) Resistance to phenylamide fungicides: a case study with *Phytophthora infestans* involving mating type and race structure. *Annu Rev Phytopathol* 34: 549-572.
- Gisi U and Sierotzki H (2008) Fungicide modes of action and resistance in downy mildews. *European Journal of Plant Pathology* 122: 157-167. Doi: 10.1007/s10658-008-9290-5
- Ishii H, Fraaije BA, Sugiyama T, Noguchi K, Nishimura K, Takeda T, Amano T and Hollomon D W (2001) Occurrence and molecular characterization of strobilurin resistance in cucumber powdery mildew and downy mildew. *Phytopathology* 91: 1166-1171
- Jackson KL, Yin JF, Csinos AS and Ji PS (2010) Fungicidal activity of fluopicolide for suppression of *Phytophthora capsici* on squash. *Crop Prot.* 29: 1421-1427.
- Kumar D, Kumar S, Singh J, Narender, R, Vashistha B and Singh N (2010) Free Radical Scavenging and Analgesic Activities of *Cucumis sativus* L. Fruit Extract. *Journal of Young Pharmacists* 2(4): 365-368. doi:10.4103/0975-1483.71627.
- Ojiambo PS, Paul PA and Holmes GJ (2010) A quantitative review of fungicide efficacy for managing downy mildew in cucurbits. *Phytopathology* 100: 1066-1076.
- Palti J and Cohen Y (1980) Downy mildew of Cucurbits (*Pseudoperonospora cubensis*): The fungus and its hosts, distribution, epidemiology and control. *Phytoparasitica* 8(2): 109-147.
- Savory EA, Granke LL, Quesada-Ocampo LM, Varbanova M, Hausbeck MK and Day B (2011) The cucurbit downy mildew pathogen *Pseudoperonospora cubensis*. *Molecular Plant Pathology*, 12: 217-226. doi:10.1111/j.1364-3703.2010.00670.x
- Reuveni M, Eyal M and Cohen Y (1980) Development of resistance to metalaxyl in *Pseudoperonospora cubensis*. *Plant Dis.* 64: 1108-1109
- Sauter H, Steglich W and Anke T (1999) Strobilurins: evolution of a new class of active substances. *Angew. Chem. Int. Ed. Engl.* 38: 1328-1349.
- Skylakakis G (1981) Effects of alternating and mixing pesticides on the buildup of fungal resistance. *Phytopathology* 71: 1119-1121.
- Thind TS, Singh PP, Sokhi SS and Grewal RK (1991) Application timing and choice of fungicides for control of downy mildew of musk melon. *Plant Disease Research* 6: 49-53.
- Toquin V, Barja F, Sirvan C, Gamet S, Latorse MP, Zundel JL, Schmitt F and Beffa R (2006) A new mode of action for fluopicolide: modification of the cellular localization of spectrin-like protein. *Pflanzenschutz Nachr. Bayer* 59: 171-184.
- Zhu SS, Liu XL, Wang Y, Wu XH, Liu PF, Li JQ, Yuan SK and Si NG (2007) Resistance of *Pseudoperonospora cubensis* to flumorph on cucumber in plastic houses. *Plant Pathol.* 56: 967-975.