

**CULTURAL VARIABILITY AMONG NINE ISOLATES OF
UROCYSTIS AGROPYRI ON WHEAT**

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Abstract

Four temperatures and five different media were tested for cultural variability of nine geographical isolates of *Urocystis agropyri* (Preuss.) Schroet causing flag smut of wheat. *Urocystis agropyri* is extremely slow growing pathogen and each isolate responded differently at different temperatures. All the isolates/collections grew well at 20°C after 45, 60, and 70 days after incubation. Ambala isolate was fast growing. PDA medium was found to be most suitable for the growth of all the nine isolates/collections. The average mycelial growth was also maximum on Potato Dextrose Agar Medium followed by Corn Meal Dextrose Agar, Oat Meal Dextrose Agar, Grain Meal Dextrose Agar and minimum on Wheat Meal Dextrose Agar.

Key Words: Variability, media, isolates, wheat.

Introduction

Flag smut of wheat caused by *Urocystis agropyri* (Preuss) Schroet is a potential danger to wheat production as it may cause yield losses upto 40-60% in the field (Bhatnagar *et al.*, 1978). The incidence of smut disease has been reported from the states of Punjab, Haryana, Himachal Pradesh, Madhya Pradesh, Uttar Pradesh, Delhi, Bihar, and Rajasthan of India (Goel *et al.*, 1977). Because of continuous cultivation of susceptible varieties, the inoculum load of the pathogen gradually build up in the soil and owing to its persistence for many years in soil pose a serious threat to wheat cultivation, particularly in north-western plains zone of India. Use of resistant varieties is one of the important alternatives to overcome the disease problem. A variety which exhibits resistance in one area may show susceptibility in another area due to variation in pathogen (Paulkar and Raut, 2004). For breeding resistant varieties, knowledge of variability in pathogen is essential. The information on variation in cultural characters viz., effect of temperature and media on growth characteristics is meager in India. So, in the present study, emphasis have been given on cultural variability among isolates of *Urocystis agropyri* collected from different areas and states of India.

Materials and Method

Flag smut caused by *Urocystis agropyri* was collected from 9 different locations of 5 states i.e., Ambala, Bhiwani, Hisar (Haryana); Durgapura, Sriganaganagar,

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Sikar (Rajasthan); IARI (Delhi), Ludhiana (Punjab), and Palampur (H.P.). The seed of highly susceptible variety C 306 smeared with dry teliospores powder (previous crop season) of flag smut pathogen/collections individually 20g/kg seed was sown separately in 2m × 5m plots at three dates i.e., 1st week of December, 3rd week of December and 1st week of January to get the diseased samples at different intervals for isolation. The pathogen was isolated in pure culture. For isolation, 0.5 cm portions of unbroken freshly smutted tissues were taken and surface sterilized with 0.1% mercuric chloride solution for 1-2 minutes. After 2-3 thorough washings with distilled and sterilized water, the bits were transferred aseptically on PDA slants and incubated at 20 ±1°C for growth of pathogen for further studies.

The mycelial bits from slants of different isolates were transferred to the centre of Petri dish containing PDA medium and incubated at 15, 20, 25, and 30°C for fungal growth. The linear growth of the mycelium was measured in milli meter after 45, 60, and 75 days of incubation at different temperatures.

To study the effect of media on growth of *U. agropyri*, five different media i.e., Potato Dextrose Agar (PDA), Oat Meal Dextrose Agar (OMDA), Wheat Meal Dextrose Agar (WMDA), Corn Meal Dextrose Agar (CMDA), and Grain Meal Dextrose Agar (GMDA) were used for the growth of different isolates by incubating at 20±1°C for 60 days.

Results and Discussion

Urocystis agropyri is extremely slow growing pathogen and each isolate responded differently at different temperatures. It is apparent from the data of Table 1 that by and large growth of all the isolates/collections was maximum at 20°C after 45, 60, and 70 days of incubation (DAI) followed by 25°C, 15°C, and minimum at 30°C. Among the isolates, Ambala isolate was fast growing with maximum mycelial growth after 45, 60, and 75 DAI at all the temperature levels. The maximum linear growth of 21.1 mm was recorded in Ambala isolate, while minimum growth of 13.9 mm of Bhiwani and Palampur isolates after 75 days of incubation. In Sriganaganagar, Hisar, and Sikar isolates, the linear growth ranged between 19.0 and 19.3 mm. However, the linear growth of isolates of Delhi, Ludhiana, and Durgapura ranged 16.1-16.4 mm.

PDA medium was found to be most suitable for the growth of all the nine isolates, whereas on other media i.e., OMDA medium Ambala, Bhiwani, Hisar, Durgapura, Sikar, and Ludhiana isolates; WMDA medium Ambala, Bhiwani, Durgapura, and Sikar isolates; CMDA medium: Ambala, Bhiwani, Hisar, Durgapura, Sikar, and Palampur isolates and on GMDA medium Ambala,

Bhiwani, Hisar, Durgapura, and Palampur isolates grew well. However, the growth of Sriganaganagar and Delhi isolates was observed only on PDA, whereas Ambala, Bhiwani, and Durgapura isolates grew well on all the 5 test media (Table 2).

Table 1. Effect of temperature on linear growth of different isolates of *Urocystis agropyri* on PDA.

Isolate/ collections	Linear growth (mm)											
	15°C			20°C			25°C			30°C		
	45DAI	60DAI	75DAI	45DAI	60DAI	75DAI	45DAI	60DAI	75DAI	45DAI	60DAI	75DAI
Ambala	17.2	17.7	18.3	19.1	20.4	21.1	17.4	17.8	17.9	15.3	15.6	15.8
Bhiwani	9.9	11.2	11.9	12.2	13.3	13.9	10.1	11.9	12.2	8.4	9.1	9.3
Hisar	13.1	13.7	14.6	17.2	18.6	19.1	13.3	14.2	14.8	11.6	11.8	12.1
Durgapura	10.8	12.1	12.9	13.1	15.3	16.4	11.9	12.4	12.8	9.7	10.1	11.3
Sriganaga nagar	13.9	14.5	15.2	16.9	18.2	19.3	15.6	16.1	16.4	13.5	13.8	14.2
Sikar	13.3	13.7	14.5	17.1	18.5	19.0	13.9	14.4	14.8	11.5	11.9	12.3
Delhi	12.9k	13.2	13.5	15.2	15.6	16.1	13.1	14.2	15.1	8.7	9.1	9.3
Ludhiana	11.8	12.1	13.0	15.3	15.9	16.2	12.2	12.8	13.2	9.4	9.8	10.9
Palampur	10.3	10.7	11.2	13.3	13.7	13.9	12.1	12.6	12.2	11.3	11.7	12.2
Mean	12.6	13.2	13.9	15.4	16.6	17.2	13.2	14.0	14.3	11.0	11.4	11.9

DAI = Days after incubation

Each figure is the average of 3 replicates

Table 2. Effect of different media on growth of *U. agropyri* isolates.

Isolate/ collections	Medium				
	PDA	OMDA	WMDA	CMDA	GMDA
Ambala	+	+	+	+	+
Bhiwani	+	+	+	+	+
Hisar	+	+	-	+	+
Durgapura	+	+	+	+	+
Sriganaganagar	+	-	-	-	-
Sikar	+	+	+	+	+
Delhi	+	-	-	-	-
Ludhiana	+	+	-	-	-
Palampur	+	-	-	+	+

+ = Growth observed

- = No growth

PDA containing glucose probably supported the best growth of the pathogen. Fungi responds differently to nutritional factors and to a great extent depends on function of the substrate (Rani and Kumar, 2007).

It is also evident from Table 3 that average mycelial/colony growth of flag smut isolates was affected by media used. PDA medium was found to be the best

Table 3. Comparative cultural growth of *U agropyri* isolates on different media after 60 days of incubation.

Isolates/collections	Mycelial colony diameter (mm)					Mean
	PDA	OMDA	WMDA	CMDA	GMDA	
Ambala	20.6(4.55)	19.0 (4.36)	17.6 (4.20)	15.8 (3.98)	18.3 (4.28)	18.26 (4.28)
Bhiwani	13.4 (3.67)	13.0 (3.61)	11.6 (3.42)	12.7(3.57)	13.1 (3.63)	12.76 (3.58)
Hisar	19.0 (4.37)	18.6 (4.32)	0.0 (0.31)	18.2 (4.27)	17.9 (4.24)	14.74 (3.50)
Durgapura	13.1 (3.63)	12.6 (3.56)	11.7(3.43)	11.8 (3.45)	12.9 (3.60)	12.42 (3.53)
Sriganganagar	16.2 (4.03)	0.0 (0.31)	0.0 (0.31)	0.0 (0.31)	0.0 (0.31)	3.24 (1.06)
Sikar	14.1 (3.76)	0.0 (0.31)	14.0 (3.75)	13.8 (3.72)	0.0 (0.31)	8.38 (2.37)
Delhi	12.9 (3.60)	0.0 (0.31)	0.0 (0.31)	0.0 (0.31)	0.0 (0.31)	2.58 (0.97)
Ludhiana	15.7 (3.98)	15.4 (3.93)	0.0 (0.31)	0.0 (0.31)	0.0 (0.31)	6.22 (1.77)
Palampur	12.8 (3.60)	0.0 (0.31)	0.0 (0.31)	11.2 (3.36)	11.7 (3.43)	7.14 (2.20)
Mean	15.3 (3.91)	8.73 (2.34)	6.10 (1.82)	9.27 (2.59)	8.21 (2.27)	--
C D (p = 0.01)	Isolates = 0.070 Media = 0.052		Isolates X Media = 0.156			

Each figure is an average of 3 replicates

Figures in parentheses are $\sqrt{n+1}$

with average mycelial growth followed by CMDA, OMDA, GMDA, and minimum on WMDA. The Ambala isolate had maximum mycelial growth on PDA, while minimum on CMDA after 60 days of incubation at 20°C. This may be due to that each isolate has a different ability to utilize the specific substrates for their growth and multiplication (Suriachandraselvan and Seetharaman, 2003; Paulkar and Raut, 2004). However, the average mycelial growth of different isolates on other media ranged between 12.6 and 19.0 mm on OMDA, 11.6 and 17.6 mm on WMDA, 11.2 and 18.2 mm on CMDA and 11.7 and 18.3 mm on GMDA after 60 days of incubation at 20 °C. The results of present investigations are in accordance with earlier reports of Moller and Hockenhull (2001); Tanina *et al.* (2004).

References

- Bhatnagar, G. C., R. B. L. Gupta and V. L. Mishra. 1978. Effect of flag smut caused by *Urocystis agropyri* on yield components of wheat cultivars in Rajasthan, India. *Plant Dis. Reprtr.* **62**: 348-350.
- Goel, L. B., D. V. Singh, K. D. Srivastava, L. M. Joshi and S. Nagarajan. 1977. Smuts and bunts of wheat in India. Division of Mycology and Plant Pathology, IARI., New Delhi, India, p.38.
- Moller, K. and J. Hockenhull. 2001. Biometry of oospores and intraspecific variation of four *Pyihium* species. *Mycol. Res.* **105** : 1206-12 15.

- Paulkar, P. K. and B. T. Raut. 2004. Variability among the isolates of *Fusarium oxysporum* f. sp. *ciceri*. *J. Mycol. Pl. Pathol.* **34**(1) : 20-23.
- Rani, J. S. and M. R. Kumar. 2007. Morphological and cultural variability among isolates of *Pythium aphanidermatum* — incitant of damping off in tomato. *J. Mycol. Pl. Pathol.* **37**(1): 45-48.
- Suriachandraselvan, M. and K. Seetharaman. 2003. Effect of culture media on growth and sclerotial production of different isolates of *Macrophomina phaseolina* infecting sunflower. *J. Mycol. Pl. Pathol.* **33**(2) : 226-229.
- Tanina, K., M. Tojo, H. Date, H. Nasu and S. Kasuyama. 2004. *Pythium* rot of chigensal (*Brassicu campestris* L. Chinensis group) caused by *Pythium ultimum* avr. *ultimum* and *Pythium aphanidermatum*. *J. Gen. Pl. Pathol.* **70**: 188-191.