Effect of different meteorological parameters on germination of inoculum of teliospores of *Tilletia indica* in rice-wheat rotation system in Punjab, India

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ABSTRACT

Dynamics of inoculum in relation to meteorological parameters affecting occurrence of Karnal bunt epidemic in wheat in Punjab was studied during the years 2001-02 and 2002-03. There existed an intricate relationship between germination of teliospores of *Tilletia indica*, the causal organism of karnal bunt of wheat and meteorological parameters. The ambient and soil temperature showed negative relationship with the germination of teliospores. Increase in temperature beyond 25 °C resulted decrease in germination of teliospores. Relative humidity had significant positive relationship with germination of teliospores and maximum teliospore germination was recorded when relative humidity was more than 75 per cent. Different models were developed for prediction of disease possibility based on viability of inoculums depending on meteorological conditions. Based on humid thermal ratio, the best fit models for forecasting viability and germination of teliospore of *T. indica* throughout the year and during crop season were developed which explained 65 to 89 per cent and 60 to 52 percent of the variations, respectively.

Key words: Tilletia indica, model, meteorological parameters, karnal bunt, wheat

Karnal bunt incited by *Tilletia indica* is widely distributed and economically important disease of wheat. Karnal bunt was first reported from Karnal (Haryana), India by Mitra (1931). It has since been reported in Pakistan, Afghanistan, Iraq, Mexico, Nepal (Warham *et al*, 1986). Karnal bunt is mainly distributed at latitudes 25°N-30°N in India, Pakistan and Mexico; at 27°N-30°N in Iran; and extends to 34°N in India, Pakistan, and a few isolated areas of the U.S.

Karnal bunt infected grains in an ear are randomly, completely or partially converted into sori enclosed by pericarp. The infected portion of grains is grey initially and turn black later on. The sori containing black powdery mass of teliospores emit fishy odour due to presence of trimethylamine. The teliospores mixed with soil at the time of harvesting and threshing and those along with the infected seed constitute the primary inoculum (Chahal and Pannu, 1997). The disease causes losses in yield, seed vigour and deteriorates quality of the produce and appearance of grains. Quantitative losses ranged between 0.2 - 0.6 per cent of total produce during epiphytotic years. The importance of karnal bunt lies in fact that causal agent being seed, soil and air borne is difficult to manage once introduced in an area. Therefore, wheat importing countries have imposed strict quarantine measures and insist on a zero tolerance limit on shipment of wheat from karnal bunt prone regions. Farmers have to bear repeated setbacks as their seed get rejected for want of meeting certifications standards (0.25%).

Environmental factors play a major role in occurrence of the disease by influencing over wintering and germination

of teliospores; production, multiplication and dissemination of sporidia, infection and establishment of the pathogen and development of the disease (Gill *et al.*, 1993). Moderate temperature (19-23 °C), high humidity (>70%), cloudiness or rainfall during anthesis favoured disease development in susceptible host variety (Aujla *et al.*, 1977, Nagarajan, 1991). Moderate temperatures, high relative humidity or free moisture, cloudiness, and rainfall during anthesis favoured disease development (Agarwal *et al.*, 1976).

For infection to occur the fungus requires specific optimum temperature as well as optimum conditions of moisture and / or humidity. In general, karnal bunt disease infection, disease progression, and survival are favoured by cool temperatures during flowering, moist conditions (wheather through rainfall or irrigation), low soil moisture during fallow periods and short, mild or absent winters. According to Zhang et al., (1984), the optimal temperature for teliospore germination was between 15 and 22 °C. Similarly, Singh (1994) cited an optimum range of 15-25 °C. Teliospore germination was arrested by dessication but resumed when soil moisture was restored (Smilanick et al., 1985). Teliospore germination apparently required a minimum of 82% relative humidity and preferably free water (Singh, 1994). Infection was favoured by periods of high humidity, rain showers, and cool temperatures during the two to three week period of anthesis (Singh, 1994).

However in addition to meteorological factors, the availability of viable inoculum is of foremost importance during vulnerable stage of the crop. Therefore forecasting

Table 1: Percent germination of teliospores of *Tilletia indica* incubated at room temeperature or 20±2 °C and previously exposed to meteorological parameters under field conditions under the wheat-rice rotation system from May 2001 to April 2003

Exposure condition		Percent germination of teliospores incubated at		Tilletia indica incubated at 20±2 °C			
room temeperature							
Months-Fortnight		2001-2002	2002-2003	2001-2002	2002-2003		
May	I	-	-	-	8.2		
	II	-	-	-	8.0		
June	I	-	-	-	8.4		
	II	-	-	-	8.4		
July	I	-	-	-	8.7		
	II	-	-	-	9.0		
August	I	-	-	-	8.8		
	II	-	-	-	8.0		
September	I	-	-	-	8.1		
	II	-	-	-	8.0		
October	I	-	-	-	8.0		
	II	-	-	-	9.0		
November	I	2.4	3.2	12.5	12.0		
	II	5.1	5.3	13.5	13.6		
December	I	5.5	5.6	14.0	14.2		
	II	11.0	11.2	19.0	19.2		
January	I	10.5	10.6	18.5	17.8		
-	II	9.2	9.0	17.6	17.6		
February	I	9.1	9.1	17.3	17.1		
	II	11.0	11.2	19.0	19.2		
March	I	9.5	9.6	17.9	17.3		
	II	1.1	0.0	10.4	10.3		
April	I	-	-	9.0	8.8		
	II		-	8.1	8.0		

^{&#}x27;-' indicates no germination of teliospores observed

models based on meteorological parameters predicting viabilities and germination of teliospores is of fundamental importance for successful management of disease. Keeping in this view, the present studies were undertaken to study the effect of meteorological parameters on build up of inoculum of T. *indica*.

MATERIALS AND METHODS

Collection of infected wheat grains

Post-harvest surveys were undertaken in different grain markets of Punjab. Infected grains were collected, dried and stored in air-tight glass vials at room temperature.

Exposing teliospores to meteorological parameters under field conditions

The teliospores of karnal bunt of wheat were trapped in three layered nylon cloth and placed in upper layer of soil filled in the earthen pots. These earthen pots were buried in the field. Seventy-two earthen pots containing teliospores were buried at 5 cm depth in the rice filed in a randomized block design with three replications in May 2001 and May 2002. The pots were exposed to field conditions for the four crop seasons comprising two wheat and two rice crops from 2001 to 2003. The observations were made as follows.

Per cent germination of teliospore

At fortnightly intervals, starting from May 2001 up to April 2003, three earthen pots were removed from field and brought to the laboratory. Nylon cloth bags containing teliospores were taken out of pot and the teliospores were floated over the surface of sterilized distilled water in Petri plates (50 x 17 mm). These Petri plates were incubated at both room temperature and 20±2 °C. The germination of teliospores was recorded under light microscope on surface of water after 7 days. For each replication, the numbers of total and germinated spores were recorded from 3 different microscopic fields and percentage of germinated teliospores was

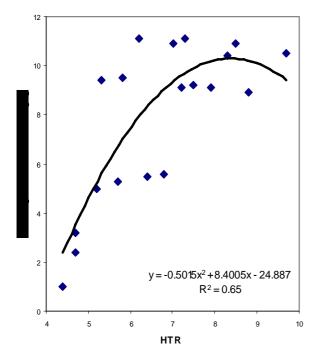


Fig.1: Relationship between HTR and germination of Teliospores of *Tillletia indica* through out the year

Fig.2: Relationship between HTR and germination of Teliospores of *Tillletia indica* during the wheat crop season

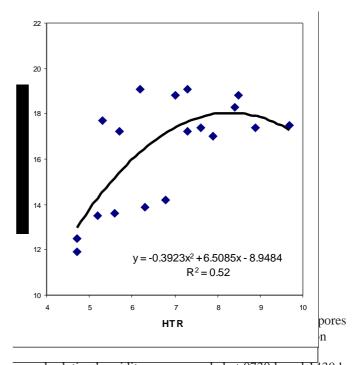


Fig.3: Relationship between HTR and viability of teliospores of *Tillletia indica* throughout the year

calculated.

Meteorological observations

Daily observations of meteorological parameters, viz morning (minimum) and evening (maximum) air temperature

and relative humidity were recorded at 0730 h and 1430 h from May 2001 to April 2003. Soil moisture was recorded on weekly basis and after any rainfall event. Morning relative humidity and mean atmospheric temperature revealed high correlation with teliospore germination. A single transformed

Table 2: Relationships between Germination of Teliospores previously exposed to ambient meteorological parameters under field conditions and incubated at room temperature

variable was formed by taking ratio of morning relative humidity and corresponding mean atmospheric temperature

Humid Thermal Remainder of the state of the stat

Coefficient of correlation

and was referred to as humid thermal ratio.

The coefficient of correlation among different variables, viz. minimum and maximum air temperature, surface soil temperature and relative humidity were calculated following the method suggested by Snedecor and Cochran (1967).

Regression analysis

Simple regression analysis was carried out as suggested by Panse and Sukhatme (1978). Multi-regression equation, exponential functions and second degree polynomial were

Air Temperature	Maximum	Y = 26.40 - 0.
	Minimum	Y = 14.79 - 0
	Mean	Y = 22.13 - 0
Soil temperature	Morning	Y = 15.34 - 0
•	Evening	Y = 22.44 - 0
	Mean	Y = 20.26 - 0.
Relative Humidity	Morning	Y = 47.01 + 0
•	Evening	Y = 15.28 + 0
	Mean	Y = 13.61 + 0
Humid Thermal Ratio		Y = -3.76 + 1
Miniple indufregression ting the t	eliospore germinati	0 Y = $17.94 - 0$.
repariconshigs between germination		Y = 3.97 - 0.6
of teliospores and other		Y = 3.60 - 0.5
meteorological parameters AND DIS	CUSSION	Y = 2.89 - 0.7
		$0.137 X_7$

Teliospores previously exposed to ambient meteorological parameters for

Meteorological variables

Multiple linear regression

meteorological parameters

of teliospores and other

relationships between germination

	10, 11,
Tellospores previously exposed to ambient meteorological param	neters fi
and 15 November 2002 to 15 March 2003.	
Air Temperature and at 20±2 °C (Table Maximum Was correlated with 12 = 22 Minimum Was correlated with 13 = 13	2.29 - 0.
meteorological parameters. Among the three types $q = 10$	3.14 - 0.
Mean Y = 19	9.52 - 0.
Soliantitative response functions, viz. symple linear, log-linear = 13	3.65 - 0.
and quadratic, the simple linear system explains the maximum $\mathbf{r}_{=1}$	
variation indicated by highest R^2 values and was, therefore $\chi = 1$	7.85 - 0.
Retarted Homidary eloping predictive functions. $Y = 32$	2.65 + 0
Evening $Y = 2$.	26 + 0.1
Correlation coefficient has prevailing $= 5$.	29 + 0.1
Humid Thermal Ratio $Y = -$	752 + 1.

Y = 16.80 - 0

Y = 8.05 - 0.6

Y = 6.28 - 0.4Y = 6.75 - 0.7

 $0.834 X_7$

Table 3: Relationships between Germination of Teliospores previously exposed to ambient meteorological parameters under field conditions and incubated at 20 °C temperature

Meteorological variables		Equation	r^2
Teliospores previously exposed t	to ambient meteorolo	ogical parameters from May 2001 to April 20	03.
Air Temperature	Maximum	$Y = 27.88 - 0.559 X_1$	0.87
	Minimum	$Y = 19.62 - 0.486 X_2$	0.76
	Mean	$Y = 23.94 - 0.543 X_3$	0.85
Soil temperature	Morning	$Y = 20.49 - 0.457 X_4$	0.78
	Evening	$Y = 25.80 - 0.426 X_5$	0.83
	Mean	$Y = 23.75 - 0.463 X_6$	0.85
Relative Humidity	Morning	$Y = 8.65 + 0.0.244 X_7$	0.42
	Evening	$Y = 9.98 + 0.162 X_8$	0.41
	Mean	$Y = 2.33 + 0.155 X_9$	0.43
Humid Thermal Ratio		$Y = 3.94 + 1.75 X_{10}$	0.84
Multiple linear regression		$Y = 23.91 - 0.262 X_3 - 0.241 X_6$	0.85
relationships between germination		$Y = 23.51 - 0.239 X_3 + 0.388 X_7$	0.84
of teliospores and other		$Y = 27.94 - 0.502 X_6 + 0.374 X_7$	0.85
meteorological parameters		$Y = 26.71 - 0.288 X_3 - 0.330 X_6$	0.85
		+ 0.354 X ₇	
Teliospores previously exposed and 15 November 2002 to 15 Ma		ogical parameters from 5 November 2001 to	15 March 20
Air Temperature	Maximum	$Y = 28.19 - 0.559 X_1$	0.48
7 III Tomperature	Minimum	$Y = 20.67 - 0.543 X_2$	0.29
	Mean	$Y = 25.70 - 0.644 X_3$	0.46
Soil temperature	Morning	$Y = 20.96 - 0.445 X_4$	0.30
2 on temperature	Evening	$Y = 25.80 - 0.409 X_5$	0.40
	Mean	$Y = 24.41 - 0.484 X_6$	0.41
Relative Humidity	Morning	$Y = 15.20 + 0.335 X_7$	0.22
	Evening	$Y = 11.70 + 0.101 X_8$	0.24
	Mean	$Y = 5.59 + 0.155 X_9$	0.22
Humid Thermal Ratio		$Y = 9.11 + 1.07 X_{10}$	0.38
Multiple linear regression		$Y = 25.74 - 0.692 X_3 - 0.394 X_6$	0.42
relationships between germination		$Y = 21.28 - 0.604X_3 + 0.804X_7$	0.44
of teliospores and other		$Y = 19.91 - 0.450 X_6 + 0.417 X_7$	0.37
meteorological parameters		$Y = 20.33 - 0.699 X_3 - 0.659 X_6$	0.39
2 1		$+ 0.403 X_7$	

meteorological parameters and germination of teliospores of T. indica incubated at room temperature are shown in Table 2 and for incubation at controlled temperature of 20 ± 2 °C are shown in Table 3.

A significant negative correlation (Table 2 and 3) was found between air temperature and germination of teliospores. It indicated that low day time temperature coupled with other favourable conditions, i.e. high relative humidity and cloudiness for more than 3.5 hours per day was conducive for germination of teliospores; production and multiplication of secondary sporidia. It was found that with increase in temperature, the germination of teliospores decreased and there was no teliospore germination at temperatures above 30 ± 2 °C. The low temperature during

reproductive and grain filling stages leads to delay in the maturity of the grains which may result in prolonged predisposed condition of the plants, multiplication of the sporidia and spread of disease within infected spikes (Joshi *et al.*, 1981)

Soil temperature

A significant negative correlation was also found between soil temperature and germination of teliospores (Table 2 and 3). The teliospores in soil germinate, producing primary sporidia. Soil temperature influenced the production of sporidia (Aujla *et al.*, 1990) and the low soil temperature (20±2 °C) was favorable for germination of teliospores. Thus with increase in soil temperature, there was decrease in germination of teliospores subsequently decrease in the

production of sporidia.

Relative humidity

There was a positive and significant correlation (Table 2 and 3) between the teliospore germination and relative humidity. It indicated that high relative humidity played a vital role in disease epiphytotic situation and seemed to be a major determining factor in the disease outbreak. Teliospore germination was maximum when average relative humidity was above 75 per cent. These studies were in accordance with Mavi *et al.*, (1992) and Jhorar *et al.*, (1993). Joshi *et al.*, (1981) also observed that rains during grain filling period favoured the disease by increasing the moisture in air and thus creating congenial conditions for the disease.

Humid thermal ratio (HTR)

There was a positive significant correlation (Table 2 and 3) between teliospore germination and humid thermal ratio. Forecasting models based on humid thermal ratio were developed by considering the period of whole year or only the wheat growing period of 15 November to 15 March. A set of 25 relationships were tested using the 'CURVEFIT' programme. The best fit relationships which explained maximum variation are presented in Fig. 1 &2. The same procedure was adopted to establish relationships between the viability of teliospores and humid thermal ratio Fig. 3 & 4. The curvilinear relationships were best fit as compared to linear models and explained maximum variations.

CONCLUSION

In addition to meteorological factors the availability of viable inoculums is of foremost importance during the vulnerable stage of the crop. Therefore the amount of disease that will develop in a growing season can be predicted only if we know the viability of teliospores available in soil during that particular year. Greater the survival of teliospores and proportionately more severe can be karnal bunt outbreak in the following season in combination with favourable meteorological parameters. Availability of favourable conditions can induce germination resulting into exhausting of inoculums beforehand and non appearance of disease despite of the availability of favourable conditions and presence of susceptible hosts. Therefore meteorological conditions prevailing during the crop period as well as before and after should be taken into consideration.

REFERENCES

Agarwal V. K., Singh A. and Verma H. S. (1976). Outbreak of Karnal bunt of wheat. *F.A.O. Plant Prot. Bull.*, **24** (3): 99-100.

- Aujla S. S., Sharma I. and Gill K. S. (1990). Effect of soil moisture and temperature on teliospore germination of Neovossia indica. *Indian Phytopath*, **43**: 223-25.
- Aujla S. S., Sharma Y. R., Chand K. and Sawney S. S. (1997). Influence of weather fartors on the incidence and epidemiology of Karnal bunt disease of wheat in the Punjab. *Indian J Ecol* **4**: 71-74.
- Chahal S. S. and Pannu P. P. S. (1997). Present status of Karnal bunt of wheat. Achievements and prospects in mycology and plant pathology. Chahal S S, Parashar I B, Randhawa H S and Arya S (eds.), International Book Distributors, Dehradun, p. 99-106.
- Gill K. S., Sharma I. and Aujla S. S. (1993). Karnal bunt and Wheat Production. Punjab Agricultural University, Ludhiana, India, p. 153.
- Jhorar O. P., Mavi H. S., Sharma I. and Aujla S. S. (1992). A biometeorological model for forecasting Karnal bunt of wheat. *Pl Dis. Res.*, **7**: 204-09.
- Jhorar O. P., Sharma I., Mavi H. S., Aujla S. S. and Nanda G. S. (1993). Forecasting models for effective application of fungitoxicants in the management of Karnal bunt. *Indian J Mycol Pl Pathol.*, **23**: 78-89.
- Joshi L. M., Singh D. V. and Srivastava K. D. (1981). Meteorological conditions in relation to incidence of Karnal bunt of wheat in India. Proc 3rd Intn Symp Plant Pathology, IARI, new Delhi, December 14-18, pp. 11.
- Mavi H. S., Jhorar O. P., Sharma I., Singh G., Mahi G. S., Mathauda S. S. and Aujla S. S. (1992). Forecasting Karnal bunt disease of wheat A meteorological method. *Cereal Res Commun* **20**: 67-74.
- Mitra, M. (1931). A new bunt of wheat in India. *Ann. Appl. Biol.*, **18**:178-179.
- Nagarajan S. (1991). Epidemiology on Karnal bunt of wheat incited by Neovossia indica and an attempt to develop a disease prediction system. Technical report Wheat Programme. CIMMYT, Mexico, march, p. 1-62.
- Panse V. G. and Sukhatme P. V. (1978). Statistical methods for agricultural workers. ICAR, New Delhi, p. 381.
- Singh, A. (1994). Epidemioogy and Management of Karnal bunt disease of wheat, 165 pp. G.B. Pant University, Research Bulletin No. 127.
- Smilanick, J. L., Hoffman, J. A. and Royer, M. (1985). Effect of temperature, pH, light and dessication on teliospore

- germination of Tilletia indica. *Phytopathology*, **75**: 1428-1431.
- Snedecor G. W. and Cochran W. G. (1967). Statistical Methods. Oxford and IBH, New Delhi, p.593.
- Warham, E. J., Mujeeb-Kazi, A., and Rosas, V. (1986). Karnal bunt (Tilletia indica) resistance screening of Aegilops
- species and teir practicl utilization for Triticum aestivum improvement. *Can. J. Plant Pathol.*, **8**: 65-70.
- Zhang, Z., Lange, L. and Mathur, S. (1984). Teliospore survival and plant quarantine significance of Tilletia indica (causal agent of Karnal bunt) particularly in relation to China. OEPP/EPPO Bull. **14**:119-128.

Received: March 2010; Accepted: April 2010