A model for *Tilletia indica* (Karnal bunt)—*Triticum aestivum* (Wheat) system under changing environmental conditions

Ritu Bala1 · Jaspal Kaur1 · Parminder Singh Tak2 · Sarabjot Kaur Sandhu3 · Pushpinder Paul Singh Pannu2

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Abstract
Karnal bunt (KB) of wheat incited by *Tilletia indica* Mitra is now gaining importance from the last few years due to its increasing incidence. Regular surveys are conducted to collect wheat grains samples from different grain markets of Punjab, India. Since weather plays a significant role in the initiation as well as the development of Karnal bunt. Thus, the variation in Karnal bunt incidence worked out and is being interpreted in relation to the prevailing environmental conditions during the most susceptible stage for the two decades (1991–92 to 2014–15) for the Punjab, India. The incidence of Karnal bunt was correlated with the weather parameters during the February and March of the corresponding year. The correlation analysis revealed the positive role of rainfall, rainy days, evening relative humidity, and Humid thermal index of March and the negative role of sunshine hours of February in the development and incidence of Karnal bunt. By using these parameters, a multiple regression model was developed and validated for forecasting the disease. The regression analysis showed a coefficient of determination of 0.77 and a D.W value of 1.88. The detailed analysis of historical data for more than two decades divulged the amount of total rainfall as well as the number of rainy days of March as the most critical factor for the Karnal bunt development.

Keywords Karnal bunt · Correlations · Multiple regression analysis · Rainfall · Rainy day

Introduction
Karnal bunt (KB) caused by *Tilletia indica* Mitra of wheat, a serious non-tariff barrier in world wheat trade is gaining importance due to its increasing incidence in the North-Western Plain Zone (NWPZ) of India over the last few years. The disease was first identified from the NWPZ i.e., Karnal, Haryana during 1930 (Mitra 1931). Punjab, an important wheat-growing area of NWPZ and very near to the Karnal reported the presence of this disease since the 1940s. The disease has appeared in varying intensities and severities over the years in Punjab with the highest ever incidence during the year 2014–15 (Kaur et al. 2018). The reason behind this rise in Karnal bunt incidence may be attributed to highly conducive environmental conditions during the most susceptible stage of the host along with the abundant spore load of the *Tilletia indica* in the soil and lack of resistant varieties. A well-known fact regarding this disease is that all the stages of the life cycle of the pathogen, *T. indica* i.e., germination of teliospores, production and multiplication of sporidia, their dissemination, lodging on the host surface at the vulnerable stage, infection, and establishment depend upon suitable environmental conditions (Singh and Gogoi 2011). Further, the disease has multiple modes of transmission and thus cannot be managed by a single method of disease control. Furthermore, in the disease triangle of the Karnal bunt, we always have the presence of two out of three factors that can lead to disease development i.e., 1) susceptible host (as all the cultivated varieties are susceptible), 2) virulent pathogen (abundant inoculum is present in the soil) and whenever environment become congenial, the disease will develop.

Thus, the occurrence and development of the disease always vary due to environmental factors. The role of...
suitable weather conditions during flowering, which is the most susceptible stage for infection, has been earlier documented by several workers. High humidity (70%), low temperature (19–23 °C), continuous rainy/foggy and cloudy weather for > 13 days from ear emergence to anthesis have been reported to favor the disease in different years at many places resulting in disease epiphytotic (Munjal 1971; Aujla et al. 1977; Singh and Prasad 1978).

Several disease predictions models have also been developed establishing a significant correlation between KB incidence and weather parameters (Jhorar et al. 1992; Singh et al. 1996; Smily 1997; Workneh et al. 2008; Wei-chuan and Gui-ming 2010; Singh and Karwasra 2016). But due to shifts in climatic conditions as well as the agricultural practices due to intensive agriculture over the last two decades, there is a dire need to develop a forecasting model based on various weather parameters under present environmental conditions. Therefore, the effect of weather parameters regarding the Karnal bunt incidence in Punjab, India over the last two decades was analyzed and interpreted in the present study.

**Materials and methods**

**Surveys for Karnal bunt incidence**

Wheat samples were collected by regular surveys conducted from different grain markets situated in all the districts of Punjab annually in April/May. The samples were collected from the grain markets falling in almost all the districts of the state. Every year 80–120 grain markets were surveyed and 15–30 samples were taken from each market. Samples of 500 g to 1 kg of grains from different heaps of wheat were done randomly and collected in paper bags. For calculating the disease intensity/severity of infection in the seed lots, from each sample, 2000 grains were drawn for counting the number of bunted/healthy seeds and percent infection was worked out.

**Weather parameters**

The historical data of weather parameters used in the analysis were obtained from the Department of Agrometeorology, Punjab Agricultural University, Ludhiana. The various weather parameters included maximum temperature (TMAX), minimum temperature (TMIN), morning relative humidity (RHM), evening relative humidity (RHE), total rainfall (TRF), number of rainy days (RD), and sunshine hours (SSH) for the two months i.e., February and March. The Humid Thermal Index (HTI) was also taken under consideration and was calculated as described by Jhorar et al. (1992) to estimate disease incidence.

\[
HTI = \frac{ERH}{TMX}, \text{ where } ERH \text{ is the evening relative humidity at } 15.00 \text{ h and } TMX \text{ is the mean maximum temperature.}
\]

The data of the Karnal bunt incidence and weather parameters were compiled and analyzed for more than two decades (1991–92 to 2014–15) and validated for the next four years (2015–16 to 2020–21(except 2019–20 due to covid-19 restrictions). Multiple correlations between weather parameters and incidence of the disease were worked out and further analysis was carried out by backward stepwise regression method.

**Results**

**Surveys**

The extensive surveys conducted all over Punjab by the scientists of the Wheat section, Department of Plant Breeding and Genetics, PAU, Ludhiana since 1980. These surveys showed that there was variation in Karnal bunt incidence for all the years under consideration in the Ludhiana district of Punjab. Out of 23 years, the Karnal bunt incidence was high in 13 years and the seed was not able to meet the seed certification standards (KB infection was < 0.05%). Further, data on average infection from 1991–92 to 2014–15 showed three high KB years i.e., 1996–97, 2006–07, and 2014–15 with 0.64, 0.95, and 2.20 percent infection respectively. The highest Karnal bunt incidence ever recorded in Punjab was 2.20% during the year 2014–15 (Fig. 1).

**Correlations**

Percent incidence of Karnal bunt disease showed a positive relationship with weather parameters namely Humid thermal index, minimum temperature, evening relative humidity, rainfall, and the number of rainy days while maximum temperature, morning relative humidity, and sunshine hours showed a negative relationship with the disease in February and March months (Figs. 2 and 3). Further the correlation relationship between the minimum and maximum temperature, morning relative humidity for both the months while rainfall, rainy days, and sunshine hours of February was very poor or weak. A significant positive correlation of 0.64 was found between the rainfall of March along with rainy days and Karnal bunt incidence as depicted in Figs. 2 and 3. There is a moderate positive correlation between the evening relative humidity of February \((r = 0.40)\) as well as of March \((r = 0.46)\). This showed that high relative humidity during the daytime played a vital role in disease occurrence. There is a positive correlation between the humid thermal index of March \((r = 0.44)\) and negative correlation \((-0.46)\) of sunshine hours during February with the disease (Figs. 2 and 3).
Development and validation of regression model

Further for the prediction of Karnal bunt incidence, multiple stepwise regression analysis was carried out. In total, ten regression models were developed using the stepwise regression method on highly correlated variables to predict KB incidence, and a model with maximum adjusted $r^2$ of 0.77 and D.W value of 1.88 was selected and further validated (Supplementary material). The regression analysis showed that these elements result in a coefficient of determination $r^2 = 0.77$ and D.W value of 1.88. The critical factors in this model were again rainfall of the March having a positive correlation coefficient of 0.64. Apart from this, the rainy days of March also showed a positive correlation (0.46) with the disease incidence. The evening relative humidity, Minimum temperature, Humid thermal index, and rainy days of February have a weak but positive correlation respectively while the maximum temperature of February has a negative and weak correlation with the disease (Fig. 0.3). The selected model was statistically significant, $F = 7.085$ at 0.01 percent level of significance (Table 1). Simultaneous multiple regression to investigate the best predictions of Karnal bunt incidence in Ludhiana, Punjab is.

\[
\text{Disease Incidence (DI)} = 16.646 - 6.414 (\text{HTIF}) + 0.311 \times \text{TminF} - 0.9116 \times \text{TmaxF} + 0.318 \times \text{FRhe} - 0.196 \times \text{RDF} + 0.022 \times \text{RFM} + 0.1 \times \text{RDM}. 
\]

The equation generated in this case was used to predict the KB value of all the years under consideration. Except for a very few years, the model has been able to predict the disease incidence quite satisfactorily (Fig. 4a, b). The selected model based on the data of the 23 years was also validated for the next five years (2015–16 to 2020–21) and showed a good agreement between the observed and predicted disease incidence during the period under investigation which was within the ±50% of the reported one (Table 2).

Discussion

The detailed surveys conducted over more than two decades showed the presence of pathogen *Tilletia indica* in almost all the years in all the districts (Sharma et al. 1998, 2004;
indicated the presence of established inoculum in the soils of Punjab since the 1980s. An evaluation of the soil samples from different places of Punjab reported an average teliospore density of $5 \times 10^3$ to $16 \times 10^3$ I per $250 \text{cm}^3$ of soil. This points out that in the endemic area the availability of viable teliospores to initiate the disease is not a limiting factor (Nagarajan et al. 1991). Furthermore, the importance and distribution of Karnal bunt in India appear to be related to periodic widespread cultivation of susceptible or tolerant cultivars, although the impact of weather during these periods is also to be considered.

Before 1968, indigenous tall wheat grown in the main wheat belt was susceptible to Karnal bunt and the disease was often widespread, although usually not severe. The semidwarf wheat Kalyansona, PV18, and Sonalika (introduced in the late 1960s) were comparatively more resistant than indigenous wheat and so the incidence of KB had been reduced to some extent. In 1975, high-yielding but Karnal bunt susceptible cultivars were released, and as their popularity grew, disease severity increased throughout northern India. After 1982, cultivars with KB tolerance were introduced and efforts were made to diversify the cultivars grown by farmers. These changes coincided with a reduction in disease incidence up to 1989. However, the situation changed again with the widespread use of Karnal bunt susceptible cultivars. Wheat variety HD 2329 was the most predominant in the state from 1990 to 1996 and 80 percent of the collected samples during the years represented this variety (Sharma et al. 1998). All the wheat varieties under cultivation from 1996 till date except PBW502 (2004) and DBW17 (2008) are susceptible to this disease and thus the role of susceptible varieties has been indicated to play an important role for high disease in most of the years under study (unpublished personal data). Thus, continuous growing of KB susceptible varieties of wheat over a larger area over a long period may have enhanced the inoculum load of the $T. indica$ in the soil of Punjab, and the same is true for the Ludhiana district. Thus among the commercial/popular set of cultivars management strategies may be more relevant. So, it is imperative to breed KB-resistant varieties to reduce

### Table 1

| Model     | Sum of squares | Df | Mean square | F    | Sig |
|-----------|----------------|----|-------------|------|-----|
| Regression| 5.146          | 7  | .735        | 7.085| .001|
| Residual  | 1.556          | 15 | .104        |      |     |
| Total     | 6.703          | 22 |             |      |     |

Bala et al. 2013; Kaur et al 2018). Hence, indicated the presence of established inoculum in the soils of Punjab since the 1980s. An evaluation of the soil samples from different places of Punjab reported an average teliospore density of $5 \times 10^3$ to $16 \times 10^3$ I per $250 \text{cm}^3$ of soil. This points out that in the endemic area the availability of viable teliospores to initiate the disease is not a limiting factor (Nagarajan et al. 1991). Furthermore, the importance and distribution of Karnal bunt in India appear to be related to periodic widespread cultivation of susceptible or tolerant cultivars, although the impact of weather during these periods is also to be considered.

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this inoculum load. This will lead to depletion of soil inoculum thus disrupting the KB disease triangle by managing the host resistance.

Among all the weather parameters analyzed in the selected model, the amount of rainfall of the march is positively correlated with the disease with a correlation...
coefficient of 0.64. All the three high Karnal bunt years, 1996–97, 2006–07, and 2014–15 during the period under study also authenticated the role of high rainfall during March in disease development (Fig. 5). Also, the highest Karnal bunt incidence of 2.20% in the year 2015 was correlated with the highest rainfall (96 mm) in March. Thus, the variation in the pattern of Karnal bunt incidence over the years also suggested the role of rainfall and the number of rainy days. Other factors like HTI, Maximum temperature, rainy days, evening relative humidity, and sunshine hours of February though played a small but significant role in the disease development which may be responsible for some of the unexplained variation in the model. Further, the high rainfall of March corresponds to the ear emergence to anthesis stage and grain filling stages of the host (wheat). These stages are considered to be the most susceptible stages for the disease infection by the pathogen and also decide the further development of the disease. This can be further explained by the fact that rains during the grain filling period favor the disease by increasing the moisture in the air and thus creating congenial conditions for disease (Joshi et al. 1981). The germination of the spores of *T. indica* within a week during the last week of February and further multiplication also signify the role of rainfall during the march for the KB development (Aujla et al. 1990). The role of rainfall has been studied in an analytic comparative study of Karnal bunt epiphytotic years and KB free years indicated that a more number of foggy days, continuously reduced sunshine hours, and rainfall during the heading stage were the factors responsible for epidemic years, high showering in the last week of February and frequent rainfall during heading will result in high disease incidence (Aujla et al. 1991). Rainy days at the boot leaf stage that is important for the reoccurrence of severe KB. The role of rainfall has been also corroborated by Singh and Karwasra (2016). A clear-cut role of rainfall of the March of the previous year (A year previous to the year of KB data recording) has been depicted in Fig. 5. This helps in the buildup of KB inoculum in the previous year and disease expression in the next year (Bala and Kaur 2020).

More rainy days at the time of ear head emergence favors the disease development thus supporting the positive role of rainy days of March (Nagarajan 1991). The positive role of evening relative humidity as well as the number of rainy days in disease epiphytotic as indicated in our study has been earlier confirmed by various workers (Jhorar et al. 1992; Mavi et al. 1992). The positive role of evening relative humidity can be explained by the fact that it is more important for the Karnal bunt development than the relative humidity at night which remains high always and hence its variation did not show any effect on the disease.

Evening relative humidity during the 9th to 11th standard meteorological weeks (corresponding to the last week of Feb and the first fortnight of March) was one of the major determining factors in the disease outbreak in the Ludhiana district (Mavi et al. 1992). More cloudiness during this period helps in the growth and spread of the pathogen and leads to a severe outbreak of the disease (Munjal 1975; Aujla et al. 1987). Since evening relative humidity and maximum temperature constituted important components of HTI, the various models explaining the role of HTI in disease also supported the role of both these factors in disease development (Jhorar et al. 1992; Mavi et al. 1992; Singh and Karwasra 2016). The maximum temperature of February as well as of March is negatively correlated with the disease with a poor correlation coefficient. This negative effect of maximum temperature can be because the low daytime temperature during this period coupled with all the other favorable conditions such as rainfall, sunshine hours, rainy days, and

![Fig. 5 Rainfall and rainy days of March during the years under study and the years of validation](image-url)
evening relative humidity may help in the formation and multiplication of secondary sporidia.

The selected model explains about 77 percent of the variance in disease prediction and could be accepted as the model for predicting the disease severity could account for 67–71% of the variation observed in the field plot from which the input data was collected. Considering the range of environments and the general nature of their prediction equation, an error of up to 30% may be reasonable (Burleigh et al. 1972). Earlier, two models developed under Punjab conditions (Jhorar et al. 1992; Mavi et al. 1992) when validated for the period of the study showed a good agreement with our model (Fig. 6). The change in the equation may be due to the high cropping intensity and changed practices over the period. These practices may have delayed the sowing of the wheat crop and thus delayed anthesis which whenever coincides with the high rainfall in March lead to the high Karnal bunt incidence.

The unexplained variation in disease incidence over the years despite the good rainfall suggested that there are factors responsible for preponing teliospore germination and sporidia multiplication after sowing of the crop to December might help in averting the possibilities of disease outbreak in the endemic areas even if there are conducive conditions prevail at flowering. Therefore, for the precision of disease prediction models, meteorological conditions from sowing to wheat growth need to be taken into account (Sharma and Nanda 2003). The disease was maximum in those varieties which came into flower during the period from the fourth week of February to the first week of March. Apart from this, several other aspects of pathogen biology i.e., inoculum threshold level, pathogen fitness at every stage, and probability of encounters between compatible sporidia may also contribute to disease escape or variation in Karnal bunt incidence over the period. Despite such variation, the rainfall and rainy days of March may serve as an indicator for the Karnal bunt incidence over a period that may be used for forecasting this disease.

**Supplementary Information.** The online version contains supplementary material available at https://doi.org/10.1007/s42360-022-00520-w.

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**Declarations**

**Conflict of interest** The authors declare that they have no conflict of interest involved in this research.

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