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CHARACTERIZATION OF ENVIRONMENTAL FACTORS CONDUCIVE FOR BROWN RUST DEVELOPMENT IN WHEAT (*Triticum aestivum* L.)

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ABSTRACT

Wheat (Triticum aestivum L.) is one of the world's most important cereal crop in terms of cultivated area and quantity of grain produced. It is the most communal staple food in Pakistan, followed by rice and maize. Wheat is susceptible to a wide range of biotic and abiotic stressors, which can lead to severe yield reductions. Rusts are economically the most significant biotic factor of wheat crop. Leaf rust, commonly known as brown rust, is a severe wheat disease in Pakistan that results in production losses ranging from 10% to 40%. In the present study, the experiment was conducted in MNS-University of agriculture Multan, in which screening of wheat breeding lines against the natural inoculum of brown rust and the relationship of leaf rust with different environmental parameters was done in two years of data. The disease severity (DS), Coefficient of infection (CI) and average coefficient of infection (ACI) was recorded by using Modified Cobb's scale. During 2020-2021, among 100 breeding lines, 16 breeding lines revealed an immune response, 14 exhibited moderately resistant response, 5 showed a susceptible response and the remaining were moderately resistant-moderately susceptible response. In season 2021-22, 19 breeding lines were immune, 19 showed a moderately resistant response, 4 breeding lines were susceptible and the remaining were moderately resistant-moderately susceptible. Correlation clearly demonstrated the response of disease development to environmental factors. Maximum temperature, minimum temperature, wind speed and sunshine hours showed a positive relationship with leaf rust severity while a negative relationship was evident in relative humidity and disease severity during both year study. This study provided evidence that there is a strong relationship between environmental factors and leaf rust severity. The environmental conditions were not ideal during the early stage of the host plant that help them to withstand the building inoculum during March-April. Therefore, the breeding lines exhibiting immune and moderately resistant response could be utilized in future breeding programs to develop leaf rust resistant varieties/ cultivars to combat this menace.

Keywords: Correlation, Coefficient of infection, Disease severity, Inoculum,

INTRODUCTION

Wheat (*Triticum aestivum* L.) is widely grown crop which also serves as a primary food and is consumed by 75% of the world's population (Grote *et al.*, 2021). Over 90% of the wheat grown worldwide is bread wheat, which is mainly utilized as flour in the making of flour-based products and a variety of baked goods

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(Gessese, 2019). It is valued for its flavor as well as its nutritional value as a source of calories, protein, and specific vitamins and minerals (Goel *et al.*, 2021). The top three countries for producing wheat individually are China, India, and Russia, which together generate about 41% of the world's total wheat (FAOSTAT, 2020). In 2020, the total global production of wheat was 760 million tons (FAOSTAT, 2020). Pakistan is among the top 10 global producers of wheat in terms of the overall production, area under wheat gowning and yield per hectare (Curtis, 2002). Wheat is the most common staple food in Pakistan, after then paddy rice and maize (Rehman *et al.*, 2015). Wheat is susceptible to biotic and

abiotic stresses, resulting in considerable reduction in yield (Yadav et al., 2021). Wheat crop is attacked by many diseases including forty fungal, thirty-two viral, and eighty-one bacterial diseases (Bakala et al., 2021). In wheat crop, fungal diseases have the significant role in damage of crop production. Fungal diseases cause the severe losses among the 31 pathogens and pests reported in wheat (Savary et al., 2019). The three rust diseases affecting wheat are leaf (brown), stem (black) and stripe (yellow) rust (Kankwatsa et al., 2017). The most prevalent rust throughout the world is wheat leaf rust (Pt), generally known as brown rust (Boltan et al., 2008). It primarily affects leaf blades, but it can also damage leaf sheaths and glumes under the most favorable conditions (Afzal et al., 2008). By lessen kernel weight and degrading grain quality, the disease impact grains and reduces yield (Figueroa et al., 2018). The Puccinia triticina is an obligatory parasite that can only produce infectious urediniospores on living leaf tissue. To complete its life cycle, leaf rust needs both primary and secondary hosts. Bread wheat and triticale are the major hosts whereas Thalictrum speciosissimum and Isopyrum fumaroides are secondary or alternate hosts. Once produced, urediniosporescan always extensively spread by air which infects the host plant hundreds of kilometers away as rust epidemics have spread across countries as a result of this spread of uredinospores (Bolton et al., 2008). Leaf rust can cause reduction in yield ranging from 10% to 40%, leading to economic losses. Leaf rust destroyed thousands of hectares of wheat crops across North America when in 1938, it hit the wheat cultivar "thatcher" which highlighting the importance of the disease. Which results in leaf rust was considered as the deadly disease in some areas of the United States, the European countries, and China where wheat is grown (Dubin and Brennan, 2009). New wheat verities affected from the leaf rust disease on a global scale. For the extension of leaf rust resistant varieties, CIMMYT estimates an expenditure ratio of 1:27 in developing leaf rust improved resistant cultivars (Huerta-Espino et al., 2011). The disease reduces the quantity and weight of wheat grains, leading to a loss of grain production (Huerta-Espino et al., 2011). During the crop season of 2000-04, wheat production losses caused by leaf rust were predicted to exceed \$350 million in the United States; from 2000-03 \$32 million and from 2008-09 \$40 million yield losses occur in Mexico and in South America, \$172 million yield losses occur. In China, leaf

rust causes annual production losses of approximately 3 million tons, whereas Pakistan's agricultural land is 80 percent infected with leaf rust, which represents a serious threat to wheat production. Pakistan suffered enormous losses of about 86 million dollars in 1970s as a result of a leaf rust during the endemic period of just a few years (Figueroa *et al.*, 2018).

Environmental factors are crucial in the spread of the rust pathogen and the outbreak of epidemics. Using genetic resistance to control wheat rust is the most efficient and cost-effective strategy (Figlan et al., 2020). Due to the health risks associated with pesticides in common foods. chemical control recommended nor advisable. The main goal of pathologists and wheat breeders is to screen out every variety and breeding line. After the green revolution, several new types were introduced, but new rust races swiftly destroyed the resistance (Mateen et al., 2015). Therefore, proper procedures and strategies must be devised to offset these significant losses in order to increase farmers' incomes and wheat yield (Mateen et al., 2015). Rust control options include seeding resistant types, fungicide use, and forecasting based on environmental parameters conducive to disease growth. Avoiding rust outbreaks in the state is a multifaceted challenge, given the limited number of cultivars available and the fact that many of their breeding lines/varieties are protected by the same immunological source at the genetic level (Mateen et al., 2015). As different ecological zones are taken into consideration, it is necessary to identify those cultivars with resistant sources so that they can be recommended as being the most suitable for cultivation in the more infected sections of the country. The Screening of wheat genotypes against isolates/races of leaf rust under natural conditions is considered the best and the cheapest way to identify these cultivars of wheat which show resistance against leaf rust. This would be helpful for future studies on the identification of resistant sources in wheat against leaf rust (Mateen et al., 2015). For monitoring of leaf rust resistant breeding lines in relation to environmental factors the study was based on the objectives to identify the resistant lines, and check the relationship of leaf rust with different environmental factors with rust response values.

MATERIALS AND METHODS

Screening of wheat genotypes against brown rust: The experiment was conducted in MNSUAM. In the research area, I had sown 100 wheat breeding lines for this purpose in augmented block design (Federer, 1956). For the screening of the wheat breeding lines against the brown rust, the 100 breeding lines were sown in the experimental area in which three lines of highly susceptible wheat rust spreader i.e. Morocco. was sown. Using a hand drill, each genotype covered 1m² field area with an 18-inch row to row spacing. In order to maintain the crop's health and vitality, agronomic techniques were applied (Ahmad et al., 2017).

Scoring of brown rust at the adult stage: Brown rust reaction, symbol field reaction, and wheat brown rust response value was recorded using a modified Cobb's scale devised by (Peterson et al., 1948). After one week interval, the severity of the disease was noted. Rust data was collected until the wheat reaches physiological maturity.

Table 1. Modified rust disease rating scale encompassing Reaction, Description, Infection type and Severity of the disease.

Sr.No.	Reactions	Description (Field response)	Infection	Response
			type	value
1.	Immune	No observable Infection	0	0
2.	Resistant	There is no uredia and no observable chlorosis or necrosis	R	0.2
3.	Moderately Resistant	Small uredia bounded by chlorotic or necrotic areas	MR	0.4
4.	Moderately resistant - moderately susceptible	Small to medium size uredia bounded via chlorotic or necrotic areas	MRMS	0.6
5.	Moderately Susceptible	Medium-sized uredia with no necrotic edges and may be some chlorosis.	MS	0.8
6.	Susceptible	No necrosis and almost no chlorosis in a large uredia	S	1.0

I= Immune, MR=moderately resistant, MRMS=moderately resistant- moderately susceptible, S=Susceptible Coefficient of infection (CI): The coefficient of infection (CI) is calculated by multiplying the response value by the infection intensity which is in percent. The coefficient of infection (ACI) is calculated by multiplying disease severity (DS) and constant values of infection type (IT). For infection types the constant values will be used founded on; R = 0.2, MR = 0.4, MRMS= 0.6, MS = 0.8 and S = 1.0 (Stubbs et al.,

AUDPC =
$$\frac{\text{N1}(\text{X1} + \text{X2})}{2} + \frac{\text{N2}(\text{X2} + \text{X3})}{2} + \frac{\text{N3}(\text{X3} + \text{X4})}{2}$$

een X1 and X2, and X1, X2, 80-90 % (Ali et al., 2017).

Where N1 is interval day between X1 and X2, and X1, X2, X3, and X4 are the rust intensities recorded on the first, second, third, and fourth recording dates, respectively.

Meteorological data collection: Conservational data consisting of temperature (maximum and minimum), rainfall, sunshine hours and relative humidity was recorded by Automatic Weather Station, MNS-University of Agriculture Multan, Pakistan. Rust severity (%) and plant response to disease was documented at seven-day intervals after the onset of disease symptoms. By using Modified Cobb's scale, the disease severity and field response was recorded (Table 1). Data was collected until the crop reaches physical maturity, and the establishment of the final rating was occurred when the disease severity reaches

1986). Average coefficient of infection: The sum of each CI value was divided by corresponding data recording years to obtain the ACI.

Calculation of AUDPC: AUDPC (Area under Disease Progress Curve) was calculated by the formula given by Milus and Line (1986). For each breeding line, area under disease was determined by using the formula:

STATISTICAL ANALYSIS

Correlation and basic linear regression analysis was used to analyze the link between climatic variables and rust severity data. Correlation analysis with statistical software was used to examine the relationship among disease severity and environmental factor. The independent variable was environmental data, whereas the dependent variable disease severity. Environmental factors that have a meaningful effect on brown rust growth was examined by graphing the data (Ali et al., 2017) and significant ranges conducive to disease severity was evaluated.

RESULTS

Screening of wheat breeding lines against brown

rust during 2020-21 and 2021-22: Adult plant response of 100 wheat breeding lines against brown rust are shown in Table 2. During 2020-21, among 100 breeding lines, 16 breeding lines showed an immune response, 14 breeding lines exhibited moderately resistant response, five breeding lines showed a susceptible response and remaining 65

were moderately resistant -Moderately susceptible shown in Table 2. During season 2021-22, out of 100 breeding lines, 19 breeding lines showed immune an response while another 19 breeding lines were moderately resistant, Whereas 4 breeding lines were susceptible and remaining were moderately resistant -moderately susceptible (Table 3).

Table 2. Reactions of breeding lines against brown rust (2020-2021)

Breeding lines	22-03-21	29-03-21	05-04-21	Disease response	ACI	AUDPC
BL-1	5	10	10	MRMS	7.5	122.5
BL-2	5	5	5	MRMS	4.5	70
BL-3	5	5	10	MRMS	6	87.5
BL-4	0	0	0	I	0	0
BL-5	5	5	10	MRMS	6	87.5
BL-6	20	20	20	MRMS	18	280
BL-7	0	5	5	MRMS	4.5	52.5
BL-8	0	0	0	I	0	0
BL-9	10	10	20	MRMS	12	175
BL-10	5	5	5	MRMS	4.5	70
BL-11	5	5	10	MRMS	6	87.5
BL-12	10	10	10	MRMS	9	140
BL-13	5	10	10	MRMS	7.5	122.5
BL-14	5	5	5	MRMS	4.5	70
BL-15	10	10	10	MRMS	9	140
BL-16	10	10	10	MRMS	9	140
BL-17	5	10	10	MRMS	7.5	122.5
BL-18	5	5	10	MRMS	6	87.5
BL-19	10	10	30	MRMS	15	210
BL-20	80	80	90	S	125	1155
BL-21	5	5	5	MRMS	4.5	70
BL-22	0	0	5	MR	1	17.5
BL-23	0	5	10	MR	2	105
BL-24	10	10	20	MRMS	12	175
BL-25	10	10	20	MRMS	12	175
BL-26	10	20	20	MRMS	15	245
BL-27	10	20	30	MRMS	18	280
BL-28	10	20	20	MRMS	15	245
BL-29	10	10	10	MRMS	9	140
BL-30	0	0	5	MR	1	17.5
BL-31	0	5	5	MRMS	3	52.5
BL-32	0	5	5	MRMS	3	52.5
BL-33	0	0	0	Ι	0	0
BL-34	5	5	5	MRMS	4.5	70
BL-35	0	0	0	I	0	0
BL-36	0	0	0	I	0	0
BL-37	20	20	20	MRMS	18	280
BL-38	10	10	10	MRMS	9	140
BL-39	10	10	10	MRMS	9	140
BL-40	30	30	30	MRMS	27	420
BL-41	10	20	30	MRMS	18	280
BL-42	5	5	10	MRMS	6	87.5
BL-43	5	10	10	MRMS	7.5	122.5

BL-44	5	10	10	MRMS	7.5	122.5
BL-45	10	20	20	MRMS	15	245
BL-46	5	10	10	MRMS	7.5	122.5
BL-47	0	10	20	MRMS	6	140
Bl-48	10	10	10	MRMS	9	140
BL-49	5	10	20	MRMS	10.5	157.5
BL-50	80	100	100	S	140	1330
BL-51	5	5	5	MRMS	4.5	70
BL-52	5	5	5	MRMS	3	70
BL-53	20	30	30	MRMS	18	385
BL-54	5	20	20	MRMS	13.5	227.5
BL-55	0	0	0	I	0	0
BL-56	5	5	5	MRMS	4.5	70
BL-57	0	<u>5</u>	5	MRMS	3	52.5
	<u></u>	<u>5</u>			6	
BL-58			10	MRMS		87.5
BL-59	5	5	5	MRMS	4.5	70
BL-60	20	30	40	MRMS	27	420
BL-61	0	0	0	1	0	0
BL-62	20	20	30	MRMS	21	315
BL-63	5	5	5	MR	3	70
BL-64	0	0	0	I	0	0
BL-65	0	5	5	MRMS	3	52.5
BL-66	0	5	10	MR	3	70
BL-67	5	5	5	MRMS	4.5	70
BL-68	0	0	0	I	0	0
BL-69	0	5	5	MRMS	3	52.5
BL-70	0	0	0	I	0	0
BL-71	0	0	0	I	0	0
BL-72	0	0	0	I	0	0
BL-73	0	0	5	MRMS	1.5	17.5
BL-74	5	5	5	MRMS	4.5	70
BL-75	0	0	0	I	0	0
BL-76	0	5	5	MR	2	52.5
BL-77	5	20	20	MRMS	13.5	227.5
BL-78	0	0	0	I	0	0
BL-79	5	20	20	MR	9	227.5
BL-80	0	0	0	I	0	0
BL-81	0	5	5	MR	2	52.5
BL-82	20	30	40	MRMS	27	420
BL-83	0	5	5	MR	2	52.5
BL-84	0	5	5	MR	2	52.5
BL-85	5	5	5	MR	3	70
BL-86	0	0	0	I	0	0
BL-87	5	5	5	MR	3	70
BL-88	20	30	30	MRMS	24	385
BL-89	5	5	5	MR	3	70
BL-89	20	30	30	MRMS	24	385
BL-90 BL-91	0	20	30	MRMS	15	245
BL-91 BL-92	10	20	20	MRMS MRMS	15	245
DL-72		ZU	۷0	CIVITIVI	13	445
-				MDMC	1 🖺	245
BL-93	10	20	20	MRMS	15	245
				MRMS MRMS MRMS	15 4.5 13	245 70 332.5

BL-96	0	10	10	MRMS	6	105
BL-97	100	100	100	S	150	1400
BL-98	80	90	100	S	130	1260
BL-99	40	50	60	S	57	700
BL-100	0	5	5	MR	2	52.5

I= Immune, MR=moderately resistant, MRMS=moderately resistant- moderately susceptible, S=Susceptible, AUDPC= Area under the disease progress curve, ACI= Coefficient of Infection Table 3. Reactions of breeding lines against brown rust (2021-2022)

		22 02 2022		D: D	A CI	AUDDC
Breeding line BL-1	16-03-22	23-03-2022	30-03-2022	Disease Response MRMS	ACI	AUDPC
BL-1 BL-2	10	10	10		6 0	140 0
BL-3	0	0	<u>0</u> 5	I MRMS	1.5	17.5
	0	0				
BL-4	0	5	5	MRMS	3	52.5
BL-5	5	10	10	MRMS	7.5	122.5
BL-6	10	10	10	MRMS	9	140
BL-7	0	0	0	I	0	0
BL-8	5	5	5	MR	3	70
BL-9	0	10	20	MRMS	9	140
BL-10	5	5	5	MR	3	70
BL-11	5	5	10	MRMS	6	87.5
BL-12	5	5	5	MRMS	4.5	70
BL-13	0	10	10	MRMS	6	105
BL-14	5	5	5	MR	4.5	70
BL-15	5	5	10	MRMS	6	87.5
BL-16	5	5	5	MRMS	4.5	70
BL-17	5	5	5	MRMS	4.5	70
BL-18	5	5	5	MRMS	4.5	70
BL-19	10	20	30	MRMS	18	280
BL-20	100	100	100	S	150	1400
BL-21	5	5	5	MR	3	70
BL-22	5	5	5	MR	3	70
BL-23	5	5	10	MR	4	87.5
BL-24	5	10	10	MRMS	7.5	122.5
BL-25	5	10	10	MRMS	7.5	122.5
BL-26	5	20	20	MRMS	13.5	227.5
BL-27	10	20	20	MRMS	15	245
BL-28	10	10	20	MRMS	12	175
BL-29	10	10	10	MRMS	9	140
BL-30	0	5	5	MR	2	52.5
BL-31	5	5	5	MRMS	9	70
BL-32	5	5	5	MRMS	9	70
BL-33	5	5	5	MR	3	70
BL-34	0	0	0	I	0	0
BL-35	0	0	0	I	0	0
BL-36	5	30	30	MRMS	21	332.5
BL-37	5	20	20	MRMS	13.5	227.5
BL-38	5	5	10	MRMS	6	87.5
BL-39	10	10	10	MRMS	9	140
BL-40	20	30	30	MRMS	24	385
BL-40	5	5	5	MRMS	4.5	70
BL-42	5	5	5	MRMS	4.5	70
BL-42	5	5	<u>5</u>	MRMS	4.5	70
BL-43	<u>5</u>	<u>5</u>	10	MRMS	6	87.5
DD-44	J	J	10	UIIVIU	U	07.5

BL-45	10	10	20	MRMS	12	175
BL-46	5	10	10	MRMS	7.5	122.5
BL-47	0	10	10	MRMS	6	105
BL-48	10	10	10	MRMS	9	140
BL-49	0	10	20	MRMS	9	140
BL-50	80	100	100	S	140	1330
BL-51	5	5	5	MRMS	4.5	70
BL-52	5	5	5	MRMS	4.5	70
BL-53	20	30	30	MRMS	24	385
BL-54	5	20	20	MRMS	13.5	227.5
BL-55	0	5	5	MRMS	3	52.5
BL-56	5	5	5	MRMS	4.5	70
BL-57	0	0	0	I	0	0
BL-58	5	5	10	MRMS	6	87.5
BL-59	0	0	0	I	0	0
BL-60	10	30	40	MRMS	24	385
BL-61	0	0	0	I	0	0
BL-62	10	20	20	MRMS	15	245
BL-63	5	5	5	MR	3	70
BL-64	0	0	0	I	0	0
BL-65	0	0	0	I	0	0
BL-66	0	5	10	MR	3	70
BL-67	5	5	5	MRMS	4.5	70
BL-68	0	0	0	I	0	0
BL-69	0	0	0	I	0	0
BL-70	0	0	0	I	0	0
BL-71	0	0	0	I	0	0
BL-72	0	0	0	I	0	0
BL-73	0	0	0	I	0	0
BL-74	5	5	5	MRMS	4.5	70
BL-75	0	0	0	I	0	0
BL-76	0	5	5	MR	2	52.5
BL-77	5	20	20	MRMS	13.5	227.5
BL-78	0	0	0	I	0	0
BL-79	5	20	20	MR	9	227.5
BL-80	0	5	5	MR	2	52.5
BL-81	5	5	5	MR	3	70
BL-82	20	40	40	MRMS	30	490
BL-83	0	5	5	MR	2	52.5
BL-84	0	0	0	I	0	0
BL-85	5	5	5	MR	3	70
BL-86	0	0	0	I	0	0
BL-87	5	5	5	MR	3	70
BL-88	10	30	30	MRMS	19.5	350
BL-89	5	5	5	MR	3	70
BL-90	20	30	30	MRMS	24	385
BL-91	0	20	30	MRMS	15	245
BL-92	5	20	20	MRMS	13.5	227.5
BL-93	5	20	20	MSMR	13.5	227.5
BL-94	10	5	5	MRMS	5.5	87.5
BL-95	5	30	30	MRMS	19	332.5
BL-96	5	10	10	MRMS	7	122.5

BL-97	100	100	100	S	150	1400
BL-98	80	80	80	S	120	1120
BL-99	50	60	60	MRMS	75	805
BL-100	5	5	5	MR	3	70

I= Immune, MR=moderately resistant, MRMS=moderately resistant- moderately susceptible, S=Susceptible, AUDPC= Area under the disease progress curve, ACI= Coefficient of Infection

Data Recorded based on Area under Disease Progress Curve (AUDPC): Ten breeding lines BL-35, BL-61, BL-64, BL-68, BL-70, BL-71, BL-72, BL-75, BL-78 and BL-86 were immune to brown rust and these cultivars showed the

AUDPC values as 0 and response value is also 0. The response value for susceptible lines BL 20, BL-50, BL-97, and BL-98 were 1.0 and their AUDPC values 1400, 1330, 1120, and 1155 respectively given in Table 4.

Table 4. Area Under Disease Progress Curve and Breeding lines reaction against brown rust:

Sr.No	Ranges of	Breeding line (2020-21)	Breeding line (2021-	Level of Resistance
	AUDPC		22)	or Susceptibility
1.	0-100	BL-04, BL-08, BL-33, BL-35	BL-02, BL-07, BL-34	I
		BL-36, BL-55, BL-61, BL-64	BL-35, BL-57, BL-59	
		BL-68, BL-70, BL-71, BL-72 BL-75, BL-78, BL-80,	BL-61, BL-64, BL-65	
		BL-86	BL-68, BL-69, BL-70	
			BL-71, BL-72, BL-73	
			BL-75, BL-78, BL-84	
			BL-86	
2.	101-300	BL-79	BL-79, BL, 23	MR
3.	301-600	BL-62, BL-95, BL-53	BL-36, BL-95, BL-88	MRMS
		BL-88, BL-90, BL-60	BL-40, BL-53, BL-60	
		BL-82, BL-40	BL-90, BL-82, BL-99	
4.	>600	BL-20, BL-50, BL-97,	BL-20, BL-50, BL-97	S
		BL-98, BL-99	BL-98	

I= Immune, MR=moderately resistant, MRMS=moderately resistant- moderately susceptible, S=Susceptible, AUDPC= Area under the disease progress curve

Relationship of environmental parameters (Maximum and minimum temperatures, relative humidity, wind speed, and sunshine) with brown rust severity during **2020-21:** The relationship of maximum temperature, minimum temperature, wind speed and sunshine hours showed a positive relationship with leaf rust severity while a negative relationship was evident in relative humidity and disease severity during season 2020-21. In case of maximum temperature, the breeding lines BL-20, BL-40, BL-50, and BL-53 showed considerable positive reaction with rise in temperature (Fig 1). The linear regression values exhibited that an increase in one degree of maximum temperature in case of BL-77, BL-91 and BL-95, the progression of disease also increased to 2.65, 4.21 and 4.41 and R2 value was 0.98 %, 0.79 % and 0.98 %, respectively (Table 5). In case of minimum temperature, the breeding lines BL-1, BL-3, BL-40, BL-82 and BL-95 showed considerable positive reaction with increase in temperature (Fig 1) and, linear regression values exhibited that an increase in one degree of minimum temperature in case of BL-91 and BL-95, the progression of disease also increased to 16.3, 15.5 and their R² value was

0.96 % and 0.97 % respectively (Table 5). On the majority of the breeding lines, a negative relationship among relative humidity and brown rust severity (%) was determined. In case of relative humidity, linear regression values in breeding line BL-66, BL-82 and BL-91, the progression of disease shows negative relationship with humidity as 2.7, 6.1 and 8.5 with R² value 0.96 %, 0.89 % and 100 % respectively (Table 5). In case of wind speed, some breeding lines like BL-09, BL-40, BL-91 and BL-95 showed that the brown rust clearly responds to wind speed (Fig 1). The linear regression values exhibited that an increase in one unit of wind speed in case of BL-01, BL-91 and BL-95, the progression of disease also increased to 5.1, 21.5 and 25.72 respectively with R² value 0.82 % (Table 5). Breeding lines BL-50 and BL-95 showed the maximum significant response with sunshine (Figure 1). Linear regression values exhibited that an increase in one hour of sun shine in case of BL-66, BL-77 and BL-95the progression of disease also increased to 16.1, 28.2 and 47.1 respectively with R² value 0.92 %, 0.94 % and 0.94 % (Table 5).

Relationship of environmental parameters (Maximum and minimum temperatures, relative humidity, wind speed, and sunshine) with brown rust severity during 2021-22: The relationship of maximum temperature, minimum temperature, wind speed and sunshine hours showed a positive relationship with leaf rust severity while a negative relationship was evident in relative humidity and disease severity during season 2021-22. In case of maximum temperature, the breeding line BL-5, BL-36 and BL-50 showed considerable reaction with increase temperature (Fig 1). The linear regression values exhibited that an increase in one degree of maximum temperature (°C) in case of BL-5, BL-26 and BL-27 the progression of disease also increased to 3.6, 10.9 and 7.3 respectively with R² value 99.2. There was a positive correlation between the maximum temperature and disease progression and in BL-21, BL-28, BL-36 and BL-50 having the significant results with the p-value of

0.05 (Table 6). In case of minimum temperature, the breeding lines BL-13, BL-26, BL, 45, BL-49, BL-50 and BL-60 responded significantly to temperature increases (Fig 1). The linear regression values exhibited that an increase in one degree of minimum temperature in case of BL-4, BL-13 and BL-26, the progression of disease also increased to 0.9, 1.3 and 2.7 respectively with R² value 97.0% (Table 6). In case of relative humidity, the breeding lines BL-26, BL, 36, BL-50 and BL-60 demonstrated a positive relationship with an increase in humidity. BL-3 and BL-28 showed the negative relationship with disease severity (Fig 1). The linear regression values exhibited the negative relationship as that an increase in one unit of relative humidity in case of BL-36 and BL-50, the progression of disease increases with R2 value 0.35 % (Table 6). In case of wind speed, some lines showed considerable positive response with brown rust disease severity like BL-26, BL-50, BL-91 and BL-95 (Fig. 1). The linear regression values exhibited that an increase in one unit of wind speed in case of BL-91 and BL-95, the progression of disease also increased to 25.5 and 22.0 respectively with R² value 0.99 % and 0.82 % (Table 6). There was a positive correlation between the wind speed and disease progression and showed the significant relationship in BL-9 and BL-66 having the significant results with the p-value of 0.05 (Table 6). In case of sun shine hours, breeding lines BL-60 and BL-88 showed the maximum significant response (Fig 1). The linear regression values exhibited that an increase in one unit of sunshine in case of BL-4 and BL-13, the progression of disease also increased to 9.4 and 18.5 with R2 value 0.94 % (Table 6). There was a positive correlation between the sunshine (h) and disease progression that showed the significant relationship in BL-3 and BL-28 having the highly significant results with the p-value of 0.00 (Table 6).

Table 5. Relationship of environmental parameters (Maximum and minimum temperatures, relative humidity, wind speed, and sunshine) with brown rust severity during 2020-21

Breeding line	Maximum	Minimum temperature(°C)	Relative humidity (%)	Wind speed (km/h)	Sunshine hours(h)
(BL)	temperature(°C)				
BL-1	BL-1 = - 20.64 + 0.8818 x	BL-1 = - 51.25 + 3.105 x		BL-01 = - 22.84 + 5.143 x	BL-1 = - 64.81 + 9.423x R ² =
	$R^2 = 0.98\%$	$R^2 = 0.97 \%$		$R^2 = 0.82\%$	0.94%
	0.99 (0.08)	0.98(0.10)		0.90(0.27)	0.97(0.15)
BL-3		BL-3 = - 31.25 + 1.976 x R ² =	BL-3 = 84.49 - 1.215 x	BL-03 = 3.60 + 0.505 x	
		0.39 %	$R^2 = 0.56\%$	$R^2 = 0.8\%$	
		0.63(0.56)	-0.75(0.45)	0.08(0.94)	
BL-7					BL-7 = - 69.81 + 9.423 x R ² =
					0.94%
					0.97(0.15)

BL-9		BL-9 = - 62.50 + 3.952 x		BL-09 = 7.21 + 1.01 x	
		$R^2 = 0.39 \%$		$R^2 = 0.8\%$	
		0.63(0.56)		0.08(0.94)	
BL-19	BL-19 = - 27.9 + 1.357 x	BL-19 = - 135.0 + 7.903 x	BL-19 = 394.6 - 5.494 x R ² =	BL-19 = 4.4 + 2.02 x	
	$R^2 = 0.14 \%$	$R^2 = 0.39 \%$	0.89%	$R^2 = 0.8\%$	
	0.38 (0.75)	0.63(0.56)	-0.75(0.45)	0.08(0.94)	
BL-20	BL-20 = 61.05 + 0.678 x				
	$R^2 = 0.14 \%$				
	0.38 (0.75)				
BL-23	BL-23 = - 35.12 + 1.221 x	BL-23 = - 92.50 + 5.081 x		BL-23 = - 29.24 + 5.648 x R ² =	BL-23 = - 120.4 + 16.15 x R ² =
	R ² = 0.62 %	R ² = 0.87 %		0.33%	0.92 %
	0.79 (0.41)	0.93(0.23)		0.57(0.61)	0.69(0.51)
BL-24			BL-24 = 112.8 - 1.532 x R ² =		
			0.53 %		
			-0.75(0.45)		
BL-26				BL-26 = - 45.69 + 10.29 x R ² =	BL-26 = - 129.6 + 18.85x
				0.82%	$R^2 = 0.94\%$
				0.90(0.27)	0.84 (0.36)
BL-27	BL-27 = -60.23 + 2.442 x			BL-27 = - 48.48 + 11.30 x R ² =	BL-27 = - 230.8 + 32.31x
	R ² = 0.62 %			0.33%	$R^2 = 0.92\%$
	0.79 (0.41)			0.57(0.61)	0.95 (0.18)
BL-28			BL-28 = 179.0 - 2.429 x R ² =	BL-28 = - 45.69 + 10.29 x R ² =	
			0.56 %	0.82%	
			-0.94(0.20)	0.90(0.27)	
BL-36			BL-36 = 544.1 - 7.662 x R ² =		
			0.89 %		
			-0.94(0.20)		
BL-40	BL-40 = -78.1 + 3.798 x	BL-40 = -300.0 + 18.06 x		BL-40 = - 34.1 + 13.32 x R ² =	
	$R^2 = 0.35 \%$	$R^2 = 0.63\%$		0.10%	
	0.59(0.59)	0.79(0.41)		0.32(0.79)	
BL-41					BL-41 = $-230.8 + 32.31x R^2 =$
					0. 92%
					0.88(0.30)
BL-45	BL-45 = - 41.28 + 1.764 x				
	$R^2 = 0.98 \%$				
	0.99(0.08)				
BL-49	$BL-49 = -39.59 + \overline{1.560} x$	BL-49 = -123.7 + 7.056 x			$BL-49 = -166.0 + 22.88x R^2 =$

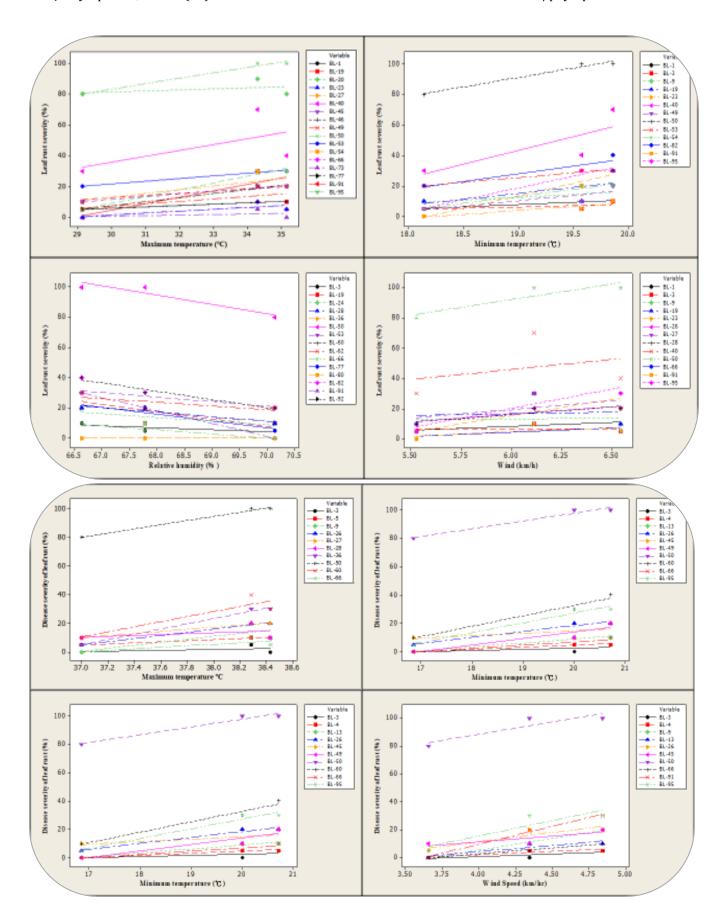
	R ² = 0.43 % 0.66(0.53)	R ² = 0.97 % 0.98(0.10)			0. 79% 0.97(0.15)
BL-50	BL-50 = - 22.56 + 3.527 x	• •	BL-50 = 511 3 - 6 129 x R ² =	BL-50 = - 31.37 + 20.57 x R ² =	• •
DL 30	$R^2 = 0.98\%$	$R^2 = 0.97 \%$	0.89 %	0.82%	0. 94%
	0.99(0.08)	0.98(0.10)	-0.94(0.20)	0.90(0.27)	0.97(0.15)
BL-53	BL-53 = - 31.28 + 1.764 x	` '	BL-53 = 235.6 - 3.065 x R ² =	0.70(0.27)	0.77 (0.13)
DL-33	$R^2 = 0.98 \%$	$R^2 = 0.97 \%$	0.89 %		
	0.99(0.08)	0.98(0.10)	-0.94(0.12)		
BL-54	0.55(0.00)	BL-54 = - 163.7 + 9.315 x	0.51(0.12)		BL-54 = - 204.4 + 28.27x R ² =
<i>DL 0</i> 1		$R^2 = 0.97 \%$			0. 94%
		0.98(0.10)			0.95(0.18)
BL-62		0.50(0.10)	BL-62 = 225.6 - 3.065 x R ² =		0.50(0.10)
			0.75 %		
			-0.75(0.45)		
BL-66	BL-66 = - 35.12 + 1.221 x		BL-66 = 192.3 - 2.747 x R ² =	BL-66 = - 29.24 + 5.648 x R ² =	BL-66 = - 120.4 + 16.15x R ² =
	R ² = 0.62 %		0.89 %	0.33%	0.92%
	0.79 (0.41)		-0.98(0.12)	0.57(0.61)	0.97(0.15)
BL-73	BL-73 = - 9.48 + 0.3391 x				
	$R^2 = 0.14 \%$				
	0.38(0.75)				
BL-77	BL-77 = - 71.92 + 2.645 x		BL-77 = 328.5 - 4.597 x R ² =		BL-77 = - 204.4 + 28.27x R ² =
	$R^2 = 0.98 \%$		0.94 %		0. 94%
	0.99(0.08)		-0.94(0.20)		0.97(0.15)
BL-80			BL-80 = 107.8 - 1.532 x R ² =		
			0.94 %		
			-0.94(0.20)		
BL-82		BL-82 = - 165.0 + 10.16 x	BL-82 = 451.3 - 6.129 x R ² =		
		$R^2 = 0.87 \%$	0.89 %		
		0.93(0.23)	-0.98(0.12)		
BL-91	BL-91 = - 121.5 + 4.205 x	BL-91 = - 297.5 + 16.37 x	BL-91 = 600.3 - 8.558 x R ² =	BL-91 = - 114.2 + 21.58 x R ² =	
	$R^2 = 0.79 \%$	$R^2 = 0.96 \%$	100 %	0.51%	
	0.89 (0.29)	0.98(0.10)	1.00** (0.20)	0.71(0.49)	
BL-92			BL-92 = 328.5 - 4.597 x R ² =		
			0.92 %		
			-0.94(0.20)		
BL-95	BL-95 = - 123.2 + 4.409 x	BL-95 = - 276.2 + 15.52 x		BL-95 = - 134.2 + 25.72 x R ² =	BL-95 = - 344.0 + 47.12 xR ² =

	$R^2 = 0.98\%$	$R^2 = 0.97 \%$		0.82%	0.94%
Cianifiaant	0.99(0.08)	0.98(0.11)		0.90(0.27)	0.97(0.15)
	* Highly Significant **	num temperature (°C) Relative	humidity unit (0/)		
	d unit: (km/h) Sun shine hou		numulty unit: (%)		
			nimum temperatures, relative h	umidity, wind speed, and suns	shine) with brown rust
	verity during 2021-22.		•	•	,
Breeding	Maximum	Minimum temperature(°C)	Relative humidity (%)	Wind speed (km/h	Sunshine hours(h)
line (BL)	temperature(°C)				
BL-3	BL-3 = - 56.6 + 1.538 x	BL-3 = - 15.69 + 0.905 x	BL-3 = 73.4 - 1.190 x	BL-3 = - 15.30 + 3.964 x R ²	BL-3 = - 50.58 + 6.731 x R ²
	$R^2 = 0.17\%$	$R^2 = 0.41 \%$	$R^2 = 0.16\%$	= 0.66%	= 0.48%
	0.42(0.72)	0.64(0.11)	-0.59(0.76)	0.81(0.38)	-1.00**(0.00)
BL-4		BL-4 = - 23.25 + 1.385 x	BL-4 = - 102.4 + 1.753 x	BL-4 = - 15.51 + 4.401 x R ²	BL-4 = - 69.81 + 9.423 x R ²
		$R^2 = 0.97 \%$	$R^2 = 0.35\%$	= 0.82%	= 0.94 %
		0.98(0.11)	-0.59(0.59)	0.90(0.27)	-0.49(0.67)
BL-5	BL-5 = - 130.2 + 3.654 x				
	$R^2 = 0.99 \%$				
	0.99**(0.05)				
BL-9	BL-9 = - 383.6 + 10.38 x		BL-9 = - 58.0 + 1.13 x	BL-9 = - 61.62 + 16.73 x	BL-9 = - 240.8 + 32.31 x R ²
	$R^2 = 0.66 \%$		$R^2 = 1.2 \%$	$R^2 = 0.99\%$	= 0.92 %
	0.81(0.38)		-0.11(0.92)	0.99*(0.05)	0.86(0.33)
BL-13		BL-13 = - 46.50 + 2.771 x	BL-13 = - 204.8 + 3.506 x	BL-13 = - 31.01 + 8.802 x	BL-13 = - 139.6 + 18.85 x
		$R^2 = 0.97 \%$	$R^2 = 35.1 \%$	$R^2 = 0.82\%$	$R^2 = 0.94 \%$
		0.98(0.11)	-0.59(0.59)	0.90(0.27)	-0.49(0.67)
BL-26	BL-26 = - 400.5 + 10.96x	BL-26 = - 64.75 + 4.156 x	BL-26 = - 302.2 + 5.259 x	BL-26 = - 41.52 + 13.20 x	BL-26 = - 204.4 + 28.27 x
	R ² = 0.99 %	R ² = 0.97 %	$R^2 = 35.1 \%$	$R^2 = 0.82\%$	$R^2 = 0.94 \%$
	0.99**(0.05)	0.98(0.10)	-0.59(0.59)	0.90(0.27)	-0.49(0.67)
BL-27	BL-27 = - 260.3 + 7.308x				
	$R^2 = 0.99\%$				
	0.42(0.72)				

BL-28	BL-28 = - 103.3 + 3.077x		BL-28 = 156.8 - 2.379 x R ²		BL-28 = - 91.2 + 13.46 x R ²
	$R^2 = 0.17 \%$		= 16.1 %		= 0.48%
	0.99**(0.05)		-0.39(0.73)		-1.00**(0.00)
BL-36	BL-36 = - 670.8 + 18.27x		BL-36 = - 507.0 + 8.77 x R ²		
	$R^2 = 0.99 \%$		= 35.1 %		
	0.99**(0.05)		-0.59(0.59)		
BL-45		BL-45 = - 21.39 + 1.809 x		BL-45 = - 20.61 + 7.929 x	
		$R^2 = 0.41 \%$		$R^2 = 0.66 \%$	
		0.64(0.55)		0.81 (0.38)	
BL-49		BL-49 = - 77.89 + 4.580 x			
		$R^2 = 0.88 \%$			
		0.93(0.22)			
BL-50	BL-50 = - 460.7 + 14.62x	BL-50 = - 13.00 + 5.541 x	BL-50 = -329.6 + 7.013 x	BL-50 = 17.97 + 17.60 x	
	$R^2 = 0.99 \%$	R ² = 0.97 %	$R^2 = 35.1 \%$	$R^2 = 0.82\%$	
	0.99**(0.05)	0.98(0.11)	-0.59(0.59)	0.90(0.27)	
BL-60	BL-60 = - 644.0 + 17.69x	BL-60 = - 114.4 + 7.351 x	BL-60 = - 252.8 + 4.63 x R ²		BL-60 = - 370.4 + 51.15 x
	$R^2 = 0.83 \% 0.91(0.26)$	R ² = 0.97 %	= 8.7 %		$R^2 = 0.99\%$
		0.98(0.10)	-0.29(0.80)		-0.74(0.42)
BL-66	BL-66 = - 191.8 + 5.192x	BL-66 = - 38.95 + 2.290 x	$BL-66 = -29.0 + 0.564 \times R^2$	BL-66 = - 30.81 + 8.365 x	
	$R^2 = 0.66 \%$	R ² = 0.88 %	= 1.2 %	$R^2 = 0.99\%$	
	0.81(0.38)	0.93(0.22)	-0.11(0.92)	0.99*(0.05)	
BL-88					BL-88 = - 269.2 + 37.69 x
					$R^2 = 0.94\%$
					-0.49 (0.67)
BL-91				BL-91 = - 92.64 + 25.53 x	BL-95 = - 344.0 + 47.12 x
				$R^2 = 0.99\%$	$R^2 = 0.94\%$
				0.99(0.06)	-0.49 (0.67)
BL-95		BL-95 = - 111.3 + 6.926 x		BL-95 = - 72.54 + 22.01 x	BL-95 = - 344.0 + 47.12 x
		$R^2 = 0.97\%$		$R^2 = 0.82\%$	$R^2 = 0.94\%$
		0.98(0.11)		0.90(0.27)	-0.49(0.67)
Significar	at * Highly Significant **				

Significant * Highly Significant **

Maximum temperature unit: (°C) Minimum temperature (°C) Relative humidity unit: (%) Wind speed unit: (km/h) Sun shine hour unit: (h)



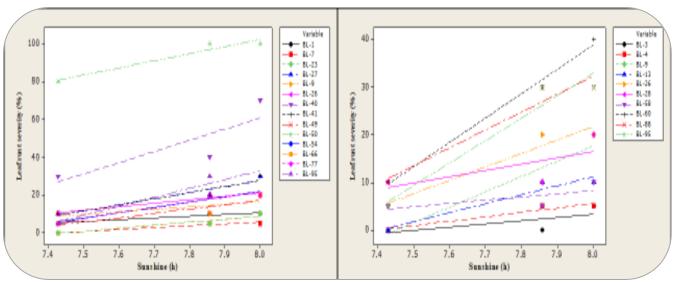


Figure 1. Relationship of environmental parameters (Maximum and minimum temperatures, relative humidity, wind speed, and sunshine) with brown rust severity during 2020-21 and 2021-22.

DISCUSSION

Present study was also focused on the environmental factors conducive for brown rust of wheat, correlation with brown rust responses and screening of wheat breeding lines against brown rust. The experiment was carried out at MNSUAM. For this objective, it was sowed 100 wheat breeding lines in the research area using an augmented block design (Federer, 1956). One hundred breeding lines of wheat were planted in the experimental region for the purpose of screening in which some lines of extremely susceptible wheat rust spreader morocco was sown.

The present study revealed that during 2020-2021, out of 100 breeding lines, 16 breeding lines show immune response as rated 0 infection type, 14 breeding lines exhibited moderately resistant response, 5 breeding lines showed susceptible response and remaining were moderately resistant-moderately susceptible. During season 2021-22, out of 100 breeding lines against brown rust, 19 breeding lines showed immune response, 19 were moderately resistant, 4 were susceptible while remaining 58 lines were moderately resistantmoderately susceptible. Previously, Mateen et al. (2015) screened 150 varieties and evaluated for resistance to wheat leaf rust and natural environmental conditions for the development of leaf rust disease of wheat. Out of 150 lines, 29, 57, and 64 lines were immune, resistant, and susceptible respectively. Similarly, Khan et al. (2002) screened 145 wheat lines for resistance to leaf rust and discovered that 39, 64, 29 and 13 lines were immune, moderately resistant, resistant, and moderately susceptible, respectively. Moreover, in another study Khan et al. (2002) screened 197 lines for leaf rust and found 89, 43, 32, 10, 16, and 7 varieties/lines that were immune, resistant, moderately resistant, moderately susceptible, and highly susceptible respectively. The present study is in agreement with the earlier findings where screened varieties/ lines were immune, resistant-moderately moderately susceptible susceptible while no resistant line was evident in our study that is inconsistent with the aforementioned studies. In present study, brown rust reaction, symbol field reaction, and wheat brown rust response value was recorded using a modified Cobb's scale devised by Peterson et al. (1948). The coefficient of infection and average coefficient of infection was calculated. AUDPC (Area under Disease Progress Curve) was calculated and Morroco showed 100 S (Susceptible: High values above 600 of AUDPC revealed high incidence of brown rust on wheat crop, while lower AUDPC values showed resistance to leaf rust in wheat breeding lines). Formerly, evidenced by the study in 2010-12, 150 varieties were screened against the severity of the leaf rust disease from this study it was discovered that Morocco's spreader had a 100S leaf rust disease severity (%) level as susceptible breeding line having the AUDPC value > 600, and the lower AUDPC value showed the resistance to leaf rust (Mateen et al., 2015). The results of the current study was supported by the previous research, as screened breeding lines showed the AUDPC value of Morocco variety greater than 600 which was evident in our study. The present study showed the correlation of environmental factors with brown rust responses. In 2020-21, the breeding lines BL-20, BL-40, BL-50, and BL-53 showed considerable positive reaction with rise in temperature and in 2021-22, the breeding line BL-5, BL-36 and BL-50 showed considerable reaction with increase in temperature. In the previous study, during 2003-2004, maximum leaf rust severity (%) was recorded at maximum air temperature (32-36 °C), and leaf rust was significantly influenced by maximum air temperature, shown by the linear relationship among maximum temperature and leaf rust severity. Whereas, during season 2004-2005, maximum leaf rust severity (%) was observed at maximum air temperature (34-35 °C), and showed the positive relationship between temperature and leaf rust severity. Moreover, Khan, (1998) discovered that at temperatures between 22 to 28 °C, environmental factors were favorable for the development of leaf rust infection as disease severity increase with increase in temperature. Similarly, Mateen et al. (2015), identified a relationship between the highest temperature and the severity of leaf rust. As temperatures raised from 28 to 32°C, the cultivars V-15, V45, V-77, V-102, and V-118 significantly responded by increase in temperature. Furthermore, the severity of leaf rust was positively correlated with maximum temperatures between 30-35°C (Sidhu and Singh, 2009). The present study was supported by the previous finding, as in a linear relationship brown rust severity increase with increase in maximum temperature which was proved in our study. In 2020-21 and 2021-22, brown rust severity (%) and minimum temperature (°C) were found to be positively correlated. With the increase in the minimum temperature, the disease severity also increased gradually. During 2020-21, the breeding lines BL-01, BL-40 and BL-82 showed considerable positive reaction with increase in minimum temperature and during 2021-22, the breeding lines BL-50 and BL-60 responded significantly to minimum temperature. Early, evidenced by, Khan, (1998), observed that the severity of leaf rust increases as the minimum temperature rises (8 to 16°C). Similarly. Mateen et al. (2015), identified that there was a positive relationship among minimum temperature and leaf rust severities. As temperature raised from 14-18°C, the varieties V-15, V-45, V-77, V-102, and V-118 showed considerable reaction. As temperature rises leaf rust severity also rises. Moreover, (Sidhu and Singh, 2009) exhibited that leaf rust severity and minimum temperatures (30-35 °C) were positively correlated. The present study is in agreement with the former findings, as in a linear relationship leaf rust severity increase with increase in minimum temperature. Current study showed that in 2020-21, breeding lines BL-3 and BL-50 showed the maximum negative relationship with relative humidity. In season 2021-22, the breeding lines BL-50 and BL-60 demonstrated a positive relationship with an increase in humidity, and BL-28 showed the negative relationship as disease severity not increases with increase in temperature. Previously, in 2003-2004 and 2004-2005, an inverse relationship between relative humidity and leaf rust severity occurred during both rating years. Brown rust decreased with increase in morning relative humidity (Sajid, 2010). Moreover, Mateen et al., (2015), found that there was positive relationship among relative humidity and leaf rust severities. With a rise in relative humidity, the varieties V-15, V-45, V-77, V-102, and V-118 significantly responded, and leaf rust values also increased. Furthermore, relative humidity had a negative correlation with the development of leaf rust (Sidhu and Singh, 2009). The present study is in agreement with the earlier findings where breeding lines in a linear relationship showed the negative relationship with the brown rust severity. In 2021-22, some breeding lines showed the positive relationship that was against by the findings of the studies mentioned earlier. Present study revealed that in 2020-21, many breeding lines showed significant response to the brown rust disease severity in BL-09 and BL-40. With the increase of wind speed, leaf rust severity also increased and during second year from 2021-22, breeding lines against wind speed showed significant response to the leaf rust disease severity in BL-50 and BL-95. Similarly, it is evidenced by Khan and Saleh, (1997) who created a multiple regression model and discovered a relation between the variables. Wheat leaf rust also indicates an increasing trend with increase in wind speed. Moreover, Mateen et al., (2015), found that the cultivars V-15, V-45, V-77, V102, and V-118 significantly reacted to rise in wind speed as leaf rust values also raised. Wind speed and leaf rust severity had a positive correlation. The present study supports the previous finding, as in a linear relationship leaf rust severity increase with increase in wind speed which was evident in our study.

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