



Effect of Temperature on Monogenic Lines of Wheat Leaf Rust Caused by *Puccinia triticina*

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WHEAT leaf rust, caused by the fungus *Puccinia triticina* Eriks., is a destructive disease found throughout common wheat production areas worldwide. Fifty wheat leaf rust monogenic lines were tested with five of *Puccinia triticina* pathotypes, i.e. BJPPQ, LQFDS, PHFPG, PTPDN, TRFDJ at four stable temperatures (30°C, 25°C, 20°C and 15°C). The wheat monogenic lines viz. *Lr* 16, *Lr* 17 and *Lr* 23 were more resistant at 25°C, while these genes were found susceptible at 15°C, 20°C and 30°C to all tested races. Eight monogenic lines, i.e. *Lr*11, *Lr*12, *Lr*13, *Lr*14a, *Lr*18, *Lr*47, *Lr*50 and *Lr*68 displayed temperature sensitivity which were completely resistant at 15°C and 20°C. *Lr*11, *Lr*12, *Lr*13, *Lr*14a, *Lr*18, *Lr*47, *Lr*50 and *Lr*68 were completely susceptible at 25°C and 30°C to all races of *Puccinia triticina*. *Lr* 34 showed temperature sensitivity to three of the tested races (LQFDS, PHFPG and PTPDN) which was resistant at 15°C and 20°C, but was susceptible at 25°C and 30°C. Genes like *Lr*1, *Lr*2a, *Lr*2b, *Lr*2c, *Lr*3ka, *Lr*3, *Lr*9, *Lr*10, *Lr*14b, *Lr*15, *Lr*10+27+31, *Lr*19, *Lr*24, *Lr*28, *Lr*33, *Lr*36, *Lr*39, *Lr*42, *Lr*51 and *Lr*67 were slightly resistant at all temperatures to some races and were susceptible to other races. The other tested monogenic lines were susceptible at all temperatures to all tested races. Further, this study will be helpful to develop resistant cultivars against leaf rust of wheat.

Keywords: Wheat, Leaf rust, Resistance genes, Temperature, *Puccinia triticina*.

Introduction

Globally, wheat is cultivated on an area of about 219 million hectares with a production of 763.2 million tons. In Egypt, 1.38 million hectares were planted with wheat in the 2019/20 which produced 8.9 million tons (FAOSTAT, 2020).

Leaf rust of wheat caused by *Puccinia triticina* Eriks. is the most widespread types of rust that affect wheat (*Triticum aestivum* L.) in Egypt and worldwide. Losses in yield of grains that resulting from leaf rust is about 15%, but may reach 30-60% under favorable conditions (Strzembicka et al., 2013; Prasad et al., 2020). In Egypt, wheat grain yield losses due to this disease reached about

32% in the highly susceptible varieties under experimental fields (El-Orabey et al., 2017; Shahin & El-Orabey, 2016). Breeding for wheat rusts resistance is continues and global importance but it also a challenge because of the interactions between resistance genes is complex. In addition, high diversity and continuous appearance of new races were occurred (Lowe et al., 2011; El-Orabey, 2018; El-Orabey et al., 2019a). In Egypt, a total of 149 leaf rust pathotypes were detected and identified during 2011/12 to 2013/14 growing seasons (El-Orabey et al., 2015). The high level of racial diversity in regional wheat leaf rust populations has made an effective and long lasting resistance in wheat genotypes very difficult to achieve.

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Genetic resistance of wheat rust diseases is characterized by one of the three main classes, i.e. race-specific or seedling resistance, which also recognized as all-stage resistance (Ellis et al., 2014); race-specific adult plant resistance (APR) and race non-specific APR, also recognized as partial or slow-rusting resistance (Tiwari et al., 2009; El-Orabey & Elkot, 2020). Race-specific resistance genes in wheat recognized to be associated with different levels of hypersensitive reaction (McIntosh et al., 2017). Moreover, race-specific resistances; seedling and adult were non-durable under high pressure of the pathogen due to mutation (Singh et al., 2011). While, race non-specific resistance genes do not give extreme levels of resistance, but can achieve when deployed by minor genes in combination (Singh et al., 2000).

At present, 88 resistance genes (R genes) for leaf rust in wheat have been known (McIntosh et al., 2017). Most of them are major resistance genes (R genes) that govern monogenic resistance according to gene-for-gene theory (Flor, 1956). The leaf rust resistance expression in wheat is in several instances sensitive to environment, specifically temperature which can change the rust reaction of resistance genes (Gousseau et al., 1985). Temperature-sensitivity of resistance gene expression has been applied in developing arguments about the function of certain genes in pathogen-host interactions (Ellingboe, 1981). Some genotypes become more susceptible by increasing temperature, some others display the reverse reaction, and other may be stable (Dyck & Johnson, 1983). This instability has caused race identification process is very difficult and complex and led to the suggestion that some genotypes, i.e. Carina, Hussar and Brevit be removed from the original differential set (Dyck & Johnson, 1983). Mains & Jackson (1926) observed the differences in the resistance of the cultivar Hussar to some physiologic races of *P. triticina* after inoculation in autumn or winter compared to inoculation in late spring season.

Environmental conditions especially in the period of post-inoculation have been exhibited to affect the interactions between wheat and pathogen. In these cases, it is not possible to recommend using leaf rust resistance gene (s) without detect the temperature affects the expression of gene in the pathogen or the host, or it affects the interaction of the two.

Thus, the aims of the present study were to determine the effect of high and low post-inoculation temperatures on the response of 50 leaf rust monogenic lines and to study the stability of leaf rust resistance genes under different temperatures.

Materials and Methods

Plant materials

Seeds of 50 leaf rust monogenic lines (Table 1) were planted in the corners of plastic pots (6 X 6cm), each pot planted with seeds of four genotypes, in a clockwise order (five to seven plants per corner). The seedlings were grown in a greenhouse at temperature ranging from a minimum of 15°C to a maximum 30°C. This experiment was carried out at Wheat Diseases Research Department, Plant Pathology Research Institute, ARC, Egypt during 2019/2020 growing season.

Inoculation and disease assessment of the tested genotypes:

Seven-days old, seedlings of the wheat monogenic lines were inoculated with the urediniospores of the five leaf rust races i.e. BJPPQ, LQFDS, PHFPG, PTPDN, TRFDJ which were more dominant during 2019/20 growing season by shaking. Inoculated seedlings were incubated in an incubator for 24hrs at 100% relative humidity and the temperature was 18-20°C. The inoculated seedlings were then transferred to growth cabinets which were maintained at constant temperatures regimes of 30°C, 25°C, 20°C and 15°C, as appropriate. All experiments with the five races were repeated three times at each temperature and all replicated disease scores were similar in all cases.

After 10-14 days, infection types (IT's) were scored for all tested genotypes using 0-4 scale (Roelfs et al., 1992; Kolmer et al., 2005). Infection types; 0= No uredinia or other macroscopic sign of infection, 0; = No uredinia but small hypersensitive necrotic or chlorotic flecks present, 1= Small uredinia surrounded by necrosis, 2 = Small to medium uredinia surrounded by necrosis or chlorosis (green islands may be surrounded by necrotic or chlorotic border), 3= Medium uredinia with or without chlorosis, 4= Large uredinia without chlorosis, IT, + = Uredinia somewhat larger than normal for the IT. Entries which showed low infection types (L) i.e. scores= 0, 0; , 1, 2 and 2+ were considered host resistant and avirulent

isolates, while those showed high infection types (H), i.e. scores= 3, 3+, 4 and 4+ were recorded as the susceptible genotype and virulent isolates.

Typical infected leaves were photographed in order to ensure consistency of infection types over time.

TABLE 1. List of leaf rust monogenic lines used in this study.

No.	Leaf rust gene	R.L. No.	Pedigree
1	<i>Lr1</i>	R.L. 6003	Thatcher ⁶ x Centenario
2	<i>Lr2a</i>	R.L. 6016	Thatcher ⁶ x Webster
3	<i>Lr2b</i>	R.L. 6019	Thatcher ⁶ x Carina
4	<i>Lr2c</i>	R.L. 6047	Thatcher ⁶ x Loros
5	<i>Lr3</i>	R.L. 6002	Thatcher ⁶ x Democrat
6	<i>Lr3ka</i>	R.L. 6007	Thatcher ⁶ x Klein Aniversario
7	<i>Lr3bg</i>	R.L. 6042	Thatcher ⁶ x Bage
8	<i>Lr9</i>	R.L. 6010	Thatcher ⁶ x Tranfer
9	<i>Lr10</i>	R.L. 6004	Thatcher ⁶ x Exchange
10	<i>Lr11</i>	R.L. 6053	Thatcher ⁶ x Hussar
11	<i>Lr12</i>	R.L. 6011	Thatcher ⁶ x Exchange
12	<i>Lr13</i>	R.L. 4031	Thatcher ⁶ x Frontana
13	<i>Lr14a</i>	R.L. 6013	Thatcher ⁶ x Selkirk
14	<i>Lr14b</i>	R.L. 6006	Thatcher ⁶ x Maria Escobar
15	<i>Lr15</i>	R.L. 6052	Thatcher ⁶ x Kenya 1483
16	<i>Lr16</i>	R.L. 6005	Thatcher ⁶ x Exchange
17	<i>Lr17</i>	R.L. 6008	Thatcher ⁶ x Klein Lucero
18	<i>Lr18</i>	R.L.6009	Thatcher ⁶ x Africa 43
19	<i>Lr19</i>	R.L. 6040	Thatcher ⁶ x <i>A. elongatum</i>
20	<i>Lr20</i>	R.L. 6092	Thatcher ⁶ x Timmo
21	<i>Lr21</i>	R.L. 6043	Thatcher ⁶ x R.L. 5406
22	<i>Lr22a</i>	R.L. 6044	Thatcher ⁶ x <i>T. tauschii</i>
23	<i>Lr22b</i>	Thatcher	Thatcher
24	<i>Lr23</i>	R.L. 6012	Thatcher ⁶ x Gabo
25	<i>Lr24</i>	R.L. 6064	Thatcher ⁶ x Agent
26	<i>Lr25</i>	R.L. 6084	Thatcher ⁶ x Transec
27	<i>Lr26</i>	R.L. 6078	Thatcher ⁶ x St-1.25
28	<i>Lr10+27+31</i>	Gatcher	<i>T. aestivum</i> Gatcher +Lr31
29	<i>Lr28</i>	R.L. 6079	Thatcher ⁶ x C77.1
30	<i>Lr29</i>	R.L. 6080	Thatcher ⁶ x CS7D-Ag#11
31	<i>Lr30</i>	R.L. 6049	Thatcher ⁶ x Terenzio
32	<i>Lr32</i>	R.L. 6086	Thatcher ⁶ x Marquis-K
33	<i>Lr33</i>	R.L. 6057	Thatcher ⁶ x P.I. 58548
34	<i>Lr34</i>	R.L. 6058	Thatcher ⁶ x P.I. 58548
35	<i>Lr35</i>	R.L. 6082	Thatcher ⁶ x <i>T. speltoides</i>
36	<i>Lr36</i>	R.L. 6083	Thatcher ⁶ x Nepawa
37	<i>Lr37</i>	R.L. 6081	Thatcher ⁶ x VPM1
38	<i>LrB</i>	R.L. 6051	Thatcher ⁶ x Bervit
39	<i>Lr38</i>	R.L. 6097	Thatcher ⁶ x T7
40	<i>Lr39</i>	TA5006	<i>Aegilops tauschii</i>
41	<i>Lr40</i>	TA5017	<i>Aegilops tauschii</i>
42	<i>Lr42</i>	WGRC11	<i>Aegilops tauschii</i>
43	<i>Lr44</i>	R.L. 6147	Thatcher ⁶ x <i>T. spelta</i>
44	<i>Lr45</i>	R.L. 6144	Secale cereal
45	<i>Lr46</i>	R.L. 6148	Pavon 76
46	<i>Lr47</i>	R.L. 4021	E 84018
47	<i>Lr50</i>	WGRC36	<i>Triticum timopheevii rmeniacum</i>
48	<i>Lr51</i>	Neepawa bc	<i>Triticum speltoides</i>
49	<i>Lr67</i>	RL6077	PI 250413
50	<i>Lr68</i>	Aurla 1	Parula

Effect of different temperatures on the response of wheat monogenic lines:

To examine the effectiveness of the tested wheat monogenic lines against the five leaf rust races, these monogenic lines were incubated at Seed Pathology Research Department, Plant Pathology Research Institute, ARC, Giza, Egypt in VELP SCIENTIFICA, FOC 225E refrigerated incubator (which programmable temperature is 0 to +50°C) under the four temperatures, i.e. 30°C, 25°C, 20°C and 15°C. The incubator has an alternative light cycle; 16hrs alternating with 8hrs dark. Alight intensity was provided with fluorescent tube for periods.

Results

Evaluation of the tested monogenic lines against five P. triticina races at different temperatures:

Effect of different temperatures, i.e. 30°C, 25°C, 20°C and 15°C on infection types of 50 leaf rust monogenic lines, each inoculated with the five races i.e. BJPPQ, LQFDS, PHFPG, PTPDN, TRFDJ of *Puccinia triticina* are shown in Tables 2-6.

Response of monogenic lines against race LQFDS:

Many changes between resistance and susceptibility were found for different wheat leaf rust monogenic lines inoculated with different five races of *P. triticina* at different temperatures (Table 2).

Leaf rust monogenic lines; *Lr16*, *Lr17* and *Lr23* were resistant at 25°C (IT 0; to 1), while these genes were susceptible (IT 3 to 4) at 15°C, 20°C and 30°C.

The seven genes; *Lr2a*, *Lr2b*, *Lr2c*, *Lr3*, *Lr3ka*, *Lr10* and *Lr24* were slightly resistant (it 0 to 2+) at all temperatures.

Eight leaf rust monogenic lines i.e. *Lr11*, *Lr12*, *Lr13*, *Lr14a*, *Lr18*, *Lr34*, *Lr50* and *Lr68* were completely resistant (IT 0 to 2) at 15°C and 20°C leaf rust race. While, these genes were completely susceptible at 25°C and 30°C (IT 3 to 4) to the same race.

Out of 50 leaf rust monogenic lines, twenty four displayed susceptible infection types at different temperatures. These monogenic lines are *Lr1*, *Lr3bg*, *Lr9*, *Lr14b*, *Lr15*, *Lr21*, *Lr22a*,

Lr22b, *Lr26*, *Lr10+27+31*, *Lr28*, *Lr29*, *Lr30*, *Lr32*, *Lr35*, *Lr36*, *Lr37*, *LrB*, *Lr38*, *Lr44*, *Lr45*, *Lr46*, *Lr47* and *Lr67* which showed IT 3 to 4. The leaf rust monogenic lines; *Lr19*, *Lr20*, *Lr25*, *Lr33*, *Lr39*, *Lr42* and *Lr51* were resistant (it 0 to 2) at all temperatures (Table 2).

Response of monogenic lines against race PHFPG:

The seedling response of the tested leaf rust monogenic lines against leaf rust race; PHFPG. Wheat leaf rust lines; *Lr16*, *Lr17* and *Lr23* were susceptible (IT 3+ to 4) at the three temperatures, i.e. at 15°C, 20°C and 30°C to race PHFPG. While, these monogenic lines were resistant (it 0 to 1) at 25°C (Table 3).

The four monogenic lines; *Lr2a*, *Lr2c*, *Lr3* and *Lr3ka* were slightly resistant (it 0 to 2+) at all temperatures.

Leaf rust monogenic lines; *Lr16*, *Lr17* and *Lr23* were resistant at 25°C (IT 0; to 1), while these genes were susceptible (IT 3 to 4) at 15°C, 20°C and 30°C.

The elven genes; *Lr1*, *Lr9*, *Lr14b*, *Lr19*, *Lr24*, *Lr28*, *Lr33*, *Lr36*, *Lr39*, *Lr42* and *Lr51* were highly resistant (it 0 to 2+) at all temperatures.

Nine leaf rust monogenic lines i.e. *Lr11*, *Lr12*, *Lr13*, *Lr14a*, *Lr18*, *Lr34*, *Lr47*, *Lr50* and *Lr68* were highly resistant (it 0 to 2) at 15°C and 20°C to leaf rust race; PHFPG. While, these genes were completely susceptible at 25°C and 30°C (IT 3 to 4) to the same race (Table 3).

Out of 50 leaf rust monogenic lines, twenty three were displayed susceptible infection types at all temperatures. These monogenic lines are *Lr2b*, *Lr3bg*, *Lr10*, *Lr15*, *Lr20*, *Lr21*, *Lr22a*, *Lr22b*, *Lr25*, *Lr26*, *Lr10+27+31*, *Lr29*, *Lr30*, *Lr32*, *Lr35*, *Lr37*, *LrB*, *Lr38*, *Lr40*, *Lr44*, *Lr45*, *Lr46* and *Lr67* which showed IT 3 to 4 (Table 3).

Response of monogenic lines against race PTPDN:

The tested leaf rust monogenic lines against leaf rust race; PTPDN. Wheat leaf rust lines; *Lr16*, *Lr17* and *Lr23* were susceptible (it 3+ to 4) at the three temperatures, i.e. at 15°C, 20°C and 30°C to race PHFPG. While, these monogenic lines were resistant (IT 0 to 1) at 25°C (Table 4).

TABLE 2. Infection types of 50 leaf rust monogenic lines tested with race LQFDS of *Puccinia triticina* under different temperatures (°C) regimes at seedling stage.

No.	Leaf rust gene	Post-inoculation temperature (°C)			
		15°C	20°C	25°C	30°C
1	<i>Lr1</i>	3	3	3	3+
2	<i>Lr2a</i>	0	1	2	2+
3	<i>Lr2b</i>	0;	1+	2	2
4	<i>Lr2c</i>	0	0	0;	1
5	<i>Lr3</i>	0;	1	2	2+
6	<i>Lr3ka</i>	0;	1	2	2+
7	<i>Lr3bg</i>	3	3	4	4
8	<i>Lr9</i>	4	4	4	4
9	<i>Lr10</i>	0	0;	1	2
10	<i>Lr11</i>	0	0	3	3+
11	<i>Lr12</i>	0;	0;	4	4
12	<i>Lr13</i>	0;	0;	4	4
13	<i>Lr14a</i>	0;	0;	3	3+
14	<i>Lr14b</i>	4	4	4	4
15	<i>Lr15</i>	3+	4	4	4
16	<i>Lr16</i>	3+	3	1	3
17	<i>Lr17</i>	4	3	0;	3+
18	<i>Lr18</i>	0;	1	3	3+
19	<i>Lr19</i>	0	0;	1	1
20	<i>Lr20</i>	0;	1	1	2
21	<i>Lr21</i>	4	4	4	4
22	<i>Lr22a</i>	3+	4	4	4
23	<i>Lr22b</i>	4	4	4	4
24	<i>Lr23</i>	4	3	0;	3
25	<i>Lr24</i>	0	0;	1	2
26	<i>Lr25</i>	0;	0;	0;	1
27	<i>Lr26</i>	4	4	4	4
28	<i>Lr10+27+31</i>	3+	3+	4	4
29	<i>Lr28</i>	4	4	4	4
30	<i>Lr29</i>	3+	3+	3+	4
31	<i>Lr30</i>	3	3+	3+	3+
32	<i>Lr32</i>	4	4	4	4
33	<i>Lr33</i>	0;	0;	0;	1
34	<i>Lr34</i>	0;	0;	3+	4
35	<i>Lr35</i>	4	4	4	4
36	<i>Lr36</i>	3+	4	4	4
37	<i>Lr37</i>	3+	3+	3+	4
38	<i>LrB</i>	4	4	4	4
39	<i>Lr38</i>	3+	3+	4	4
40	<i>Lr39</i>	0;	0;	1	1
41	<i>Lr40</i>	3+	3+	3+	4
42	<i>Lr42</i>	0;	1	1	1
43	<i>Lr44</i>	4	4	4	4
44	<i>Lr45</i>	3+	3+	4	4
45	<i>Lr46</i>	3+	3+	3+	3+
46	<i>Lr47</i>	3	3	4	4
47	<i>Lr50</i>	0;	0;	3	3+
48	<i>Lr51</i>	0;	0;	0;	1
49	<i>Lr67</i>	3	3	4	4
50	<i>Lr68</i>	0;	0;	3	3+

TABLE 3. Infection types of 50 leaf rust monogenic lines tested with race PHFPG of *Puccinia triticina* under different temperatures (°C) regimes at seedling stage.

No.	Leaf rust gene	Post-inoculation temperature (°C)			
		15 °C	20 °C	25 °C	30 °C
1	<i>Lr1</i>	0	0	0;	0;
2	<i>Lr2a</i>	1	1+	1+	2+
3	<i>Lr2b</i>	3	3+	3+	4
4	<i>Lr2c</i>	1	2	2+	2+
5	<i>Lr3</i>	0	1	2	2+
6	<i>Lr3ka</i>	0;	1	2	2+
7	<i>Lr3bg</i>	4	4	4	4
8	<i>Lr9</i>	0	0	0;	0;
9	<i>Lr10</i>	3	3	3	4
10	<i>Lr11</i>	0	0	4	4
11	<i>Lr12</i>	0;	0;	4	4
12	<i>Lr13</i>	0	0	3+	4
13	<i>Lr14a</i>	1	1	4	4
14	<i>Lr14b</i>	2	1	2	2
15	<i>Lr15</i>	4	4	4	4
16	<i>Lr16</i>	3+	4	0;	4
17	<i>Lr17</i>	3+	3+	1	3+
18	<i>Lr18</i>	0	0	4	4
19	<i>Lr19</i>	0;	0;	0;	1
20	<i>Lr20</i>	4	4	3+	4
21	<i>Lr21</i>	3+	3+	3+	4
22	<i>Lr22a</i>	4	4	4	4
23	<i>Lr22b</i>	4	4	4	4
24	<i>Lr23</i>	4	4	0	4
25	<i>Lr24</i>	1	1	2	1
26	<i>Lr25</i>	4	4	3+	4
27	<i>Lr26</i>	3+	3+	4	4
28	<i>Lr10+27+31</i>	4	4	4	4
29	<i>Lr28</i>	2	1	2	2+
30	<i>Lr29</i>	4	4	4	4
31	<i>Lr30</i>	4	3+	4	4
32	<i>Lr32</i>	3	3	3	3
33	<i>Lr33</i>	0	0	0;	0;
34	<i>Lr34</i>	0	0;	3+	3+
35	<i>Lr35</i>	4	4	4	4
36	<i>Lr36</i>	0;	1	1	1
37	<i>Lr37</i>	3+	3+	4	4
38	<i>LrB</i>	4	4	4	4
39	<i>Lr38</i>	3+	3+	4	4
40	<i>Lr39</i>	0	0	0	0;
41	<i>Lr40</i>	4	4	4	4
42	<i>Lr42</i>	0	0;	0;	0;
43	<i>Lr44</i>	3+	3+	4	4
44	<i>Lr45</i>	4	4	4	4
45	<i>Lr46</i>	3+	4	4	4
46	<i>Lr47</i>	3	3	4	4
47	<i>Lr50</i>	1	1	3+	3+
48	<i>Lr51</i>	1	1	2	2
49	<i>Lr67</i>	4	4	4	4
50	<i>Lr68</i>	0;	1	3	4

TABLE 4. Infection types of 50 leaf rust monogenic lines tested with race PTPDN of *Puccinia triticina* under different temperatures (°C) regimes at seedling stage.

No.	Leaf rust gene	Post-inoculation temperature (°C)			
		15°C	20°C	25°C	30°C
1	<i>Lr1</i>	4	4	4	4
2	<i>Lr2a</i>	0	0	1	2
3	<i>Lr2b</i>	0	0;	1	2+
4	<i>Lr2c</i>	3	3	3+	4
5	<i>Lr3</i>	4	3+	4	4
6	<i>Lr3ka</i>	0	0	1	2
7	<i>Lr3bg</i>	3	3	3	3
8	<i>Lr9</i>	4	4	3+	4
9	<i>Lr10</i>	0	0	1	1
10	<i>Lr11</i>	0	0;	3	4
11	<i>Lr12</i>	0;	0	4	4
12	<i>Lr13</i>	0	0	3+	3+
13	<i>Lr14a</i>	1	1	4	4
14	<i>Lr14b</i>	4	4	3+	4
15	<i>Lr15</i>	2	1	2	2+
16	<i>Lr16</i>	4	4	1	4
17	<i>Lr17</i>	3+	4	0	3+
18	<i>Lr18</i>	0;	1	3+	3+
19	<i>Lr19</i>	3	3	3+	4
20	<i>Lr20</i>	3+	3+	3+	4
21	<i>Lr21</i>	4	4	4	4
22	<i>Lr22a</i>	3+	4	4	4
23	<i>Lr22b</i>	4	4	4	4
24	<i>Lr23</i>	3+	4	1	4
25	<i>Lr24</i>	3+	3+	4	4
26	<i>Lr25</i>	3+	3+	3+	4
27	<i>Lr26</i>	4	4	4	4
28	<i>Lr10+27+31</i>	1	0;	1	1
29	<i>Lr28</i>	4	4	4	4
30	<i>Lr29</i>	3+	4	4	4
31	<i>Lr30</i>	4	3+	4	4
32	<i>Lr32</i>	3	4	4	4
33	<i>Lr33</i>	1	1	1+	2+
34	<i>Lr34</i>	1	1	4	4
35	<i>Lr35</i>	4	4	4	4
36	<i>Lr36</i>	3	4	4	4
37	<i>Lr37</i>	3	3+	4	4
38	<i>LrB</i>	4	4	4	4
39	<i>Lr38</i>	3+	3+	3+	3+
40	<i>Lr39</i>	1	0;	1	1
41	<i>Lr40</i>	3+	4	4	4
42	<i>Lr42</i>	0;	1+	1+	2
43	<i>Lr44</i>	4	4	4	4
44	<i>Lr45</i>	3+	3+	4	4
45	<i>Lr46</i>	4	4	4	4
46	<i>Lr47</i>	0	1	1	1+
47	<i>Lr50</i>	0	0;	3+	4
48	<i>Lr51</i>	3+	3+	3+	4
49	<i>Lr67</i>	4	3+	4	4
50	<i>Lr68</i>	0;	0;	4	4

The five monogenic lines; *Lr2a*, *Lr2b*, *Lr3ka*, *Lr33* and *Lr42* were slightly resistant (IT 0 to 2+) at all temperatures.

Leaf rust monogenic lines; *Lr16*, *Lr17* and *Lr23* were resistant at 25 °C (IT 0 to 1), while these genes were susceptible (IT 3+ to 4) at 15°C, 20°C and 30°C.

The four genes; *Lr10*, *Lr15*, *Lr10+27+31* and *Lr39* were highly resistant (IT 0 to 2+) at all temperatures.

The nine leaf rust monogenic lines i.e. *Lr11*, *Lr12*, *Lr13*, *Lr14a*, *Lr18*, *Lr34*, *Lr47*, *Lr50* and *Lr68* were highly resistant (IT 0 to 1) at 15 °C and 20 °C to leaf rust race; PTPDN. While, these genes were completely susceptible at 25°C and 30°C (IT 3 to 4) to the same race (Table 4).

Out of 50 leaf rust monogenic lines, twenty-nine displayed susceptible infection types ranging from 3 to 4 at all temperatures. These monogenic lines are *Lr1*, *Lr2c*, *Lr3*, *Lr3bg*, *Lr9*, *Lr14b*, *Lr19*, *Lr20*, *Lr21*, *Lr22a*, *Lr22b*, *Lr24*, *Lr25*, *Lr26*, *Lr28*, *Lr29*, *Lr30*, *Lr32*, *Lr35*, *Lr36*, *Lr37*, *LrB*, *Lr38*, *Lr40*, *Lr44*, *Lr45*, *Lr46*, *Lr51* and *Lr67* which showed IT 3 to 4 (Table 4).

Response of monogenic lines against race BJPPQ:

The tested leaf rust monogenic lines against leaf rust race; BJPPQ. Wheat leaf rust lines; *Lr16*, *Lr17* and *Lr23* were susceptible (IT 3+ to 4) at the three temperatures i.e. at 15°C, 20°C and 30°C to race BJPPQ. While, these monogenic lines were resistant (IT 0 to 1) at 25°C (Table 5).

The three monogenic lines; *Lr2a*, *Lr2c* and *Lr67* were slightly resistant (IT 0 to 2+) at all temperatures.

Leaf rust monogenic lines; *Lr16*, *Lr17* and *Lr23* were resistant at 25°C (IT 0 to 2), while these genes were susceptible (IT 3+ to 4) at 15°C, 20°C and 30°C.

The ten genes; *Lr1*, *Lr3*, *Lr3bg*, *Lr9*, *Lr20*, *Lr25*, *Lr26*, *Lr36*, *Lr39* and *Lr42* were highly resistant (IT 0 to 2+) at all temperatures.

The eight leaf rust monogenic lines i.e. *Lr11*, *Lr12*, *Lr13*, *Lr14a*, *Lr18*, *Lr47*, *Lr50* and *Lr68*

were highly resistant (IT 0 to 1) at 15 °C and 20 °C to leaf rust race; BJPPQ. While, these genes were completely susceptible at 25°C and 30°C (IT 3 to 4) to the same race (Table 5).

Out of 50 leaf rust monogenic lines, twenty six displayed susceptible infection types ranging from 3 to 4 at all temperatures. These monogenic lines are *Lr2b*, *Lr3ka*, *Lr10*, *Lr14b*, *Lr15*, *Lr19*, *Lr21*, *Lr22a*, *Lr22b*, *Lr24*, *Lr10+27+31*, *Lr28*, *Lr29*, *Lr30*, *Lr32*, *Lr33*, *Lr34*, *Lr35*, *Lr37*, *LrB*, *Lr38*, *Lr40*, *Lr44*, *Lr45*, *Lr46* and *Lr51* which showed IT 3 to 4 (Table 5).

Response of monogenic lines against race TRFDJ:

The tested leaf rust monogenic lines against leaf rust race; TRFDJ. Wheat leaf rust lines; *Lr16*, *Lr17* and *Lr23* were susceptible (IT 3+ to 4) at the three temperatures i.e. at 15°C, 20°C and 30°C to race TRFDJ. While, these monogenic lines were resistant (IT 0 to 1) at 25°C (Table 6).

The two monogenic lines; *Lr2b* and *Lr3ka* were slightly resistant (IT 0 to 2+) at all temperatures.

Leaf rust monogenic lines; *Lr16*, *Lr17* and *Lr23* were resistant at 25 °C (IT 0; to 2), while these genes were susceptible (IT 3+ to 4) at 15°C, 20°C and 30°C.

The nine genes; *Lr3bg*, *Lr10*, *Lr14b*, *Lr19*, *Lr24*, *Lr25*, *Lr39*, *Lr42* and *Lr67* were highly resistant (IT 0 to 2+) at all temperatures.

The eight leaf rust monogenic lines i.e. *Lr11*, *Lr12*, *Lr13*, *Lr14a*, *Lr18*, *Lr47*, *Lr50* and *Lr68* were highly resistant (IT 0 to 1) at 15°C and 20°C to leaf rust race; TRFDJ. While, these genes were completely susceptible at 25°C and 30°C (IT 3 to 4) to the same race (Table 6 and Fig. 1).

Out of 50 leaf rust monogenic lines, twenty eight displayed susceptible infection types ranging from 3 to 4 at all temperatures. These monogenic lines are *Lr1*, *Lr2a*, *Lr2c*, *Lr3*, *Lr9*, *Lr15*, *Lr20*, *Lr21*, *Lr22a*, *Lr22b*, *Lr26*, *Lr10+27+31*, *Lr28*, *Lr29*, *Lr30*, *Lr32*, *Lr33*, *Lr34*, *Lr35*, *Lr36*, *Lr37*, *LrB*, *Lr38*, *Lr40*, *Lr44*, *Lr45*, *Lr46* and *Lr51* which showed IT 3 to 4 (Table 6).

TABLE 5. Infection types of 50 leaf rust monogenic lines tested with race BJPPQ of *Puccinia triticina* under different temperatures (°C) regimes at seedling stage.

No.	Leaf rust gene	Post-inoculation temperature (°C)			
		15°C	20°C	25°C	30°C
1	<i>Lr1</i>	0;	1	1	1
2	<i>Lr2a</i>	0;	0;	1	2
3	<i>Lr2b</i>	3	3	3+	4
4	<i>Lr2c</i>	0	1	2	2+
5	<i>Lr3</i>	0;	0;	1	1
6	<i>Lr3ka</i>	3	3	4	4
7	<i>Lr3bg</i>	0;	1	1	1
8	<i>Lr9</i>	2	2	1	2+
9	<i>Lr10</i>	4	4	4	4
10	<i>Lr11</i>	0	0;	4	4
11	<i>Lr12</i>	0	0	3+	4
12	<i>Lr13</i>	1	1	4	4
13	<i>Lr14a</i>	0	0	3+	4
14	<i>Lr14b</i>	3+	3+	4	4
15	<i>Lr15</i>	3	4	3+	4
16	<i>Lr16</i>	3+	3+	0	4
17	<i>Lr17</i>	4	4	2	3+
18	<i>Lr18</i>	1	2	4	4
19	<i>Lr19</i>	4	4	4	4
20	<i>Lr20</i>	0;	1	1	1
21	<i>Lr21</i>	4	3+	3+	4
22	<i>Lr22a</i>	3+	3+	4	4
23	<i>Lr22b</i>	4	4	4	4
24	<i>Lr23</i>	3+	3+	0;	4
25	<i>Lr24</i>	4	4	4	4
26	<i>Lr25</i>	1	2	1	1
27	<i>Lr26</i>	0;	0	0	0;
28	<i>Lr10+27+31</i>	4	4	4	4
29	<i>Lr28</i>	3+	3+	3+	4
30	<i>Lr29</i>	3+	3+	3	3
31	<i>Lr30</i>	3	4	4	4
32	<i>Lr32</i>	3	4	4	4
33	<i>Lr33</i>	3	3	4	4
34	<i>Lr34</i>	4	4	4	4
35	<i>Lr35</i>	3	4	4	4
36	<i>Lr36</i>	0	2	2	2
37	<i>Lr37</i>	4	4	4	4
38	<i>LrB</i>	4	4	4	4
39	<i>Lr38</i>	3+	3+	4	4
40	<i>Lr39</i>	0	0;	1	1
41	<i>Lr40</i>	3+	3+	3+	4
42	<i>Lr42</i>	0	0	0	1
43	<i>Lr44</i>	3+	3+	3+	4
44	<i>Lr45</i>	4	3+	4	4
45	<i>Lr46</i>	3+	4	4	4
46	<i>Lr47</i>	0;	0;	3+	3+
47	<i>Lr50</i>	0	1	4	4
48	<i>Lr51</i>	4	4	4	4
49	<i>Lr67</i>	0;	1	1	2+
50	<i>Lr68</i>	0	0;	3+	4

TABLE 6. Infection types of 50 leaf rust monogenic lines tested with race TRFDJ of *Puccinia triticina* under different temperatures (°C) regimes at seedling stage.

No.	Leaf rust gene	Post-inoculation temperature (°C)			
		15°C	20°C	25°C	30°C
1	<i>Lr1</i>	3	3	3	4
2	<i>Lr2a</i>	3	3	4	4
3	<i>Lr2b</i>	0	0	1	2+
4	<i>Lr2c</i>	3	3	4	4
5	<i>Lr3</i>	4	4	4	4
6	<i>Lr3ka</i>	1	1	2	2+
7	<i>Lr3bg</i>	0	1	1	1
8	<i>Lr9</i>	3	3	3	4
9	<i>Lr10</i>	0;	0;	1	1
10	<i>Lr11</i>	0	0;	3	4
11	<i>Lr12</i>	0;	0;	4	4
12	<i>Lr13</i>	0	0	3+	4
13	<i>Lr14a</i>	0;	0;	4	3+
14	<i>Lr14b</i>	0;	1	1	1+
15	<i>Lr15</i>	3+	3+	4	4
16	<i>Lr16</i>	4	4	2	4
17	<i>Lr17</i>	3+	4	0;	3+
18	<i>Lr18</i>	0	0;	3+	4
19	<i>Lr19</i>	1	1	2	1
20	<i>Lr20</i>	3+	4	4	4
21	<i>Lr21</i>	4	4	4	4
22	<i>Lr22a</i>	3+	4	4	4
23	<i>Lr22b</i>	4	4	4	4
24	<i>Lr23</i>	3+	4	1+	4
25	<i>Lr24</i>	0;	0;	1	1
26	<i>Lr25</i>	0;	1	1	1
27	<i>Lr26</i>	4	4	4	4
28	<i>Lr10+27+31</i>	3+	4	4	4
29	<i>Lr28</i>	4	4	4	4
30	<i>Lr29</i>	3+	3+	3+	4
31	<i>Lr30</i>	4	4	4	4
32	<i>Lr32</i>	3+	4	4	4
33	<i>Lr33</i>	4	4	4	4
34	<i>Lr34</i>	3	3	4	4
35	<i>Lr35</i>	3+	4	4	4
36	<i>Lr36</i>	3	4	4	4
37	<i>Lr37</i>	3	4	4	4
38	<i>LrB</i>	4	4	4	4
39	<i>Lr38</i>	4	3+	4	4
40	<i>Lr39</i>	0;	0;	1	1
41	<i>Lr40</i>	4	4	4	4
42	<i>Lr42</i>	0;	0;	1	1
43	<i>Lr44</i>	3+	4	4	4
44	<i>Lr45</i>	4	4	4	4
45	<i>Lr46</i>	3+	3+	3+	4
46	<i>Lr47</i>	1	1	4	4
47	<i>Lr50</i>	0;	0;	3+	3+
48	<i>Lr51</i>	3+	4	4	4
49	<i>Lr67</i>	0;	0;	1	1
50	<i>Lr68</i>	0	0	3+	4



Fig. 1. Seedling reaction of the nine monogenic lines to leaf rust (left to right) i.e. *Lr11*, *Lr12*, *Lr13*, *Lr14a*, *Lr18*, *Lr34*, *Lr47*, *Lr50* and *Lr68* to race TRFDJ at different temperatures.

Discussion

Annual variation in *Puccinia triticina* races partly depends on temperature sensitive genes (Statler & Christianson, 1993). Our study provided information on sensitivity of R genes and specificity of *Puccinia triticina* races. Thus determining temperature required for mutation or host-pathogen genetic studies. Pretorius et al. (1988) indicated that, the information of temperature was very important for breeding programs planned to improving wheat genotypes with heights levels of resistance like to the parents. Our study emphasized that, the phenotypic expression resulting from the interaction between host and pathogen is adapted to a specific environment (Brwoder & Eversmeyer, 1986).

Our results were shown same results as reported by Dyck & Johnson (1983) who reported that some leaf rust resistance genes exhibited temperature sensitivity when evaluated with avirulent leaf rust races. We similarly found that some leaf rust monogenic lines show temperature sensitive (*Lr 34* and *Lr 50*) and were stable with one race of *P. triticina* but sensitive with other races. The response of resistance genes; *Lr 2a*, *Lr 2b*, *Lr 3*, *Lr 3ka*, *Lr 34* and *Lr 50* to temperature depends on the leaf rust races which were used in evaluation as reported by Dyck & Johnson (1983). These monogenic lines were stable with some races and less stable with other races according the used temperature regimes. Moreover, El-Orabey et al. (2019b) found that the *Lr 22 a*, *Lr 14b* and *Lr 28* were stable in efficacy at adult plant stage during the three growing seasons; 2017/2018, 2018/2019 and 2019/2020. Using different races of *P. triticina* in this study also explain why our data display the two monogenic lines; *Lr 16* and *Lr 17* were temperature sensitive while Brwoder (1980) grouped them as insensitive. Brwoder (1980) grouped resistance genes i.e. *Lr 11*, *Lr 12*, *Lr 13*, *Lr 14a* and *Lr 18* as highly sensitive to temperature (ineffective at high temperature). On the other hand, the wheat monogenic lines, i.e. *Lr 2a*, *Lr 2b*, *Lr 2c*, *Lr 3ka* and *Lr 24* as moderate sensitive. While, *Lr 1*, *Lr 3a*, *Lr 9*, *Lr 15*, *Lr 19* and *Lr 25* as insensitive or stable.

In the present study, the leaf rust resistance genes; *Lr 12*, *Lr 11*, *Lr 13*, *Lr 14a*, *Lr 16*, *Lr 17*, *Lr 18*, *Lr 23* and *Lr 68* were highly sensitive to temperature against all of the five *P. triticina* races used in current study. These results are in

agreement with Brwoder (1980) and Ramirez et al. (2018). Monogenic lines; *Lr 16*, *Lr 17* and *Lr 23* were effective only at 25°C but ineffective at 15°C, 20°C and 30°C. These data showed same results as reported by Dyck & Johnson (1983) who reported that genes *Lr 16* and *Lr 17* lose its resistance completely at 10°C. *Lr 26*, displays low temperature seedling resistances to leaf rust and such resistances cannot be explained by currently designated resistance genes (Datta et al., 2009).

On the other hand, other genes are well expressed at low temperatures. Moreover, our data agrees with that of Dyck & Johnson (1983) who also found that *Lr 18* became susceptible at higher temperature (25°C). In the current study, *Lr 18* was susceptible at high temperature (25°C and 30°C) for all tested races. The changes in infection type when certain leaf rust resistance genes were inoculated with specific races, especially under different temperatures, showed a complex host-parasite genetic interactions for temperature sensitivity for specific *Lr* genes and *P. triticina* races.

The results of this study confirmed the importance of temperature in genetic studies of resistance to leaf rust. Anderson (1963) showed that, partial breakdown of resistance to leaf rust with increasing temperature in the wheat cultivar South Africa 43, which has *Lr 18*. It is very difficult to locate the chromosome carrying *Lr 18* using genetic analysis method by monosomic analysis without controlling the temperature (Dyck & Samborski, 1968). The present data showed that, the chromosome identification possible if the temperature of 15°C to 20°C which suitable for the expression of most of leaf rust resistance genes.

Race identification for leaf rust survey can be also influenced by temperature sensitivity of used leaf rust differential lines (Williams & Johnston, 1965). The ineffectiveness of genes *Lr 17* and *Lr 18* at different temperature provides an observable example that could confuse race identification. Basile (1957) rejected Carina (as a source of *Lr 2b*) as being unstable, but kept Democrat (as a source of *Lr 3*). It must be understand that race identification is probably done under temperature ranging from 15 to 20°C.

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Conflict of interest: The authors declare that they have no conflicts of interest.

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تأثير درجة الحرارة على السلالات أحادية الجين لصدأ أوراق القمح الناجم عن الفطر بكسينيا تريبتسينا

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صدأ أوراق القمح، المتسبب عن الفطر بكسينيا تريبتسينا إريكس، هو مرض مدمر موجود وشائع في جميع مناطق إنتاج القمح على مستوى العالم. تم تقييم خمسون سلالة نباتية أحادية الجين لصدأ أوراق القمح باستخدام خمسة سلالات فطرية وهي BJPPQ, LQFDS, PHFPG, PTPDN, TRFDJ على درجات حرارة ثابتة (30 °م، 25 °م، 20 °م و 15 °م). سلالات القمح أحادية الجين *Lr 16*، *Lr 17* و *Lr 23* كانت مقاومة على درجة الحرارة 25 °م، بينما هذه السلالات النباتية أحادية الجين كانت قابلة للإصابة على درجات الحرارة 15 °م، 20 °م و 30 °م ضد جميع السلالات المختبرة. ثمانية سلالات القمح أحادية الجين وهي *Lr 11*، *Lr 12*، *Lr 13*، *Lr 14a*، *Lr 18*، *Lr 47*، *Lr 50* و *Lr 68* أظهرت حساسية للحرارة حيث كانت مقاومة كلياً عند درجات الحرارة 15 °م و 20 °م، بينما هذه السلالات النباتية أحادية الجين كانت قابلة للإصابة عند درجات الحرارة 25 °م و 30 °م لجميع سلالات الفطر بكسينيا تريبتسينا. أظهر *Lr 34* حساسية لدرجات الحرارة لثلاثة من السلالات المختبرة (LQFDS و PHFPG و PTPDN) والتي كانت مقاومة عند 15 °م و 20 °م، ولكن كان قابلة للإصابة للإصابة عند 25 °م و 30 °م. الجينات *Lr 1*، *Lr 2a*، *Lr 2b*، *Lr 2c*، *Lr 3ka*، *Lr 3*، *Lr 9*، *Lr 10+27+31*، *Lr 15*، *Lr 14b*، *Lr 10*، *Lr 19*، *Lr 24*، *Lr 28*، *Lr 33*، *Lr 36*، *Lr 39*، *Lr 42*، *Lr 51* و *Lr 67* كانت مقاومة قليلاً تحت جميع درجات الحرارة لبعض السلالات وكانت قابلة للإصابة بسلالات أخرى. السلالات النباتية أحادية الجين الأخرى المختبرة كانت قابلة للإصابة في جميع درجات الحرارة لجميع السلالات المختبرة. علاوة على ذلك، ستكون هذه الدراسة مفيدة لتطوير الأصناف المقاومة لصدأ أوراق القمح.