

## Supplementary material:

**Table 1:** Significant markers in the investigation

| No | Molecular Marker Name     | Linked Gene                         | Base Pairs | Annealing Temp | Locus  |
|----|---------------------------|-------------------------------------|------------|----------------|--------|
| 1  | XgWM-234                  | Lr <sub>52</sub>                    | 229,250bp  | 55 °C          | 5BS    |
| 2  | XgWM-501                  | Yr <sub>5</sub>                     | 176bp      | 60 °C          | 2B     |
| 3  | XgWM-259                  | Lr <sub>46</sub> /Yr <sub>29</sub>  | 105bp      | 55 °C          | 1BL    |
| 4  | XgWM-11                   | Yr <sub>15</sub> , Yr <sub>26</sub> | 213bp      | 50 °C          | 1B,1BS |
| 5  | XgWM-130                  | Lr <sub>34</sub> /Yr <sub>18</sub>  | 121,126bp  | 60 °C          | 7A     |
| 6  | XgWM-37                   | Lr <sub>19</sub>                    | 189bp      | 60 °C          | 7D     |
| 7  | XgWM-44                   | Lr <sub>21</sub> , Lr <sub>22</sub> | 184,176bp  | 60 °C          | 7B     |
| 8  | XgWM-437                  | Lr <sub>19</sub> /Sr <sub>25</sub>  | 109,111bp  | 50 °C          | 7D     |
| 9  | XgWM-210                  | Lr <sub>21</sub> , Lr <sub>22</sub> | 303,189bp  | 60 °C          | 2B,2D  |
| 10 | XgWM-295                  | Lr <sub>34</sub>                    | 254,258bp  | 60 °C          | 7D     |
| 11 | XgWM-533                  | Sr <sub>2</sub>                     | 316bp      | 60 °C          | 2-3-B  |
| 12 | SCS-265                   | Lr <sub>19</sub>                    | 130bp      | 60 °C          | 7D     |
| 13 | SCS-253                   | Lr <sub>19</sub>                    | 736bp      | 60 °C          | 7D     |
| 14 | Xbrac-352                 | Lr <sub>34</sub> /Yr <sub>18</sub>  | 150,211bp  | 60 °C          | 7DS    |
| 15 | STS-638                   | Sr <sub>15</sub> , Lr <sub>20</sub> | 232bp      | 55 °C          | 7A     |
| 16 | STM-773                   | Sr <sub>32</sub> ,Sr <sub>36</sub>  | 192,162bp  | 64 °C          | 2B     |
| 17 | CFA 2019 Sr <sub>22</sub> | Sr <sub>22</sub>                    | 234,500bp  | 55 °C          | 7AL    |
| 18 | Xgdm-87                   | Lr <sub>50</sub>                    | 110bp      | 50 °C          | 2B     |
| 19 | S30-13L                   | Lr <sub>51</sub>                    | 790bp      | 52 °C          | 1S     |
| 20 | Lr <sub>1</sub>           | Lr <sub>1</sub>                     | 110bp      | 60 °C          | 5DL    |
| 21 | Lr <sub>47</sub>          | Lr <sub>47</sub>                    | 450,380bp  | 55 °C          | 7AS    |
| 22 | KSUD-14                   | Lr <sub>21</sub> ,Lr <sub>40</sub>  | 885bp      | 54 °C          | 1DS    |

**Table 2:** Leaf rust resistance markers validation for wheat genotypes. Positive sign shows the presence of rust resistance genes in genotypes while, Negative sign shows the absence of rust resistance genes in genotypes

| Genotypes | gWM-234<br>Lr52 | gWM-44<br>Lr21,Lr22 | gWM-37<br>Lr19 | gWM-295<br>Lr34 | gWM-210<br>Lr21,Lr22 | Lr1 | Lr51 | Lr47 | gdm-87<br>Lr50 | KSUD -14<br>Lr21,Lr40 | SCS-253 | SCS-265 |
|-----------|-----------------|---------------------|----------------|-----------------|----------------------|-----|------|------|----------------|-----------------------|---------|---------|
| V-4189    | +               | +                   | +              | -               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| Lasani    | +               | +                   | +              | +               | +                    | -   | +    | +    | -              | +                     | +       | +       |
| V-4188    | +               | +                   | +              | -               | +                    | +   | +    | +    | +              | -                     | +       | +       |
| V-4022    | +               | +                   | +              | +               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| AARI-10   | +               | +                   | +              | +               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| AS-02     | +               | +                   | +              | +               | +                    | -   | +    | +    | -              | +                     | +       | +       |
| Inqlab-91 | +               | -                   | +              | +               | -                    | +   | +    | -    | +              | -                     | +       | +       |
| Sehar     | +               | -                   | -              | -               | -                    | +   | +    | +    | +              | +                     | -       | -       |
| Uqab-02   | +               | +                   | +              | +               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| Pasban-90 | +               | +                   | +              | -               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| TW-76001  | +               | +                   | +              | +               | +                    | -   | +    | +    | -              | -                     | +       | +       |
| TW-76002  | +               | +                   | +              | +               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| TW-76003  | +               | +                   | +              | +               | +                    | -   | +    | +    | +              | +                     | +       | +       |
| TW-76004  | +               | -                   | +              | -               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| TW-76005  | +               | -                   | +              | -               | -                    | +   | +    | +    | -              | +                     | +       | +       |
| Parwaz-94 | +               | -                   | +              | -               | -                    | +   | +    | +    | +              | -                     | +       | +       |
| TW-76007  | +               | +                   | +              | -               | +                    | +   | -    | +    | +              | +                     | +       | +       |
| TW-76008  | +               | +                   | +              | -               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| TW-76009  | +               | +                   | +              | -               | +                    | +   | +    | +    | +              | +                     | +       | +       |
| TW-76010  | +               | +                   | +              | +               | +                    | +   | +    | +    | +              | +                     | +       | +       |

**Table 3:** Strip rust resistance markers validation for wheat genotypes. Positive sign shows the presence of rust resistance genes in genotypes while, Negative sign shows the absence of rust resistance genes in genotypes.

| Genotypes | XgWM-11<br>Yr <sub>15</sub> , Yr <sub>26</sub> | XgWM-501<br>Yr <sub>5</sub> | XgWM-259<br>Lr <sub>46</sub> /Yr <sub>29</sub> | XgWM-130<br>Lr <sub>34</sub> /Yr <sub>18</sub> | Xbrac-352<br>Lr <sub>34</sub> /Yr <sub>18</sub> |
|-----------|--|-----------------------------|--|--|---|
| V-4189    | -  | +                           | -  | +  | +   |
| Lasani    | -  | +                           | +  | +  | +   |
| V-4188    | -  | +                           | -  | +  | +   |
| V-4022    | +  | -                           | +  | +  | +   |
| AARI-10   | -  | +                           | +  | +  | +   |
| AS-02     | +  | +                           | +  | -  | -   |
| Inqlab-91 | -  | -                           | +  | +  | +   |
| Sehar     | +  | +                           | -  | +  | +   |
| Uqab-02   | +  | -                           | +  | +  | +   |
| Pasban-90 | +  | +                           | -  | +  | +   |
| TW-76001  | +  | -                           | +  | +  | +   |

|           |   |   |   |   |   |
|-----------|---|---|---|---|---|
| TW-76002  | + | - | + | + | + |
| TW-76003  | + | + | + | + | + |
| TW-76004  | + | + | + | + | + |
| TW-76005  | - | - | - | + | + |
| Parwaz-94 | + | + | - | + | + |
| TW-76007  | + | + | - | - | - |
| TW-76008  | - | + | - | + | + |
| TW-76009  | + | + | - | + | + |
| TW-76010  | + | + | + | + | + |

**Table 4:** Stem rust resistance markers validation for wheat genotypes. Positive sign shows the presence of rust resistance genes in genotypes while, Negative sign shows the absence of rust resistance genes in genotypes.

| Genotypes | XgWM-533 Sr <sub>2</sub> | XgWM-437 Lr <sub>19</sub> /Sr <sub>25</sub> | STS-638 Sr <sub>15</sub> , Lr <sub>20</sub> | CFA 2019 Sr <sub>22</sub> | STM-773 Sr <sub>32</sub> ,Sr <sub>36</sub> |
|-----------|--------------------------|---|---|---------------------------|--|
| V-4189    | -                        | +   | +   | +                         | +  |
| Lasani    | +                        | +   | +   | -                         | +  |
| V-4188    | +                        | -   | +   | -                         | +  |
| V-4022    | +                        | -   | +   | -                         | +  |
| AARI-10   | +                        | +   | +   | +                         | +  |
| AS-02     | +                        | +   | +   | -                         | +  |
| Inqlab-91 | +                        | +   | +   | +                         | +  |
| Sehar     | +                        | +   | +   | +                         | +  |
| Uqab-02   | +                        | +   | +   | +                         | +  |
| Pasban-90 | +                        | -   | +   | +                         | +  |
| TW-76001  | -                        | +   | +   | +                         | +  |
| TW-76002  | +                        | +   | +   | +                         | +  |
| TW-76003  | -                        | -   | +   | +                         | +  |
| TW-76004  | -                        | +   | -   | -                         | +  |
| TW-76005  | -                        | -   | +   | +                         | +  |
| Parwaz-94 | +                        | -   | +   | +                         | +  |
| TW-76007  | +                        | +   | +   | +                         | +  |
| TW-76008  | +                        | +   | +   | +                         | +  |
| TW-76009  | -                        | +   | +   | +                         | +  |
| TW-760010 | +                        | +   | +   | +                         | +  |