Deciphering the genetics basis of lodging resistance in wild rice *O. longistaminata*

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Rice

Deciphering the genetics basis of lodging resistance in wild rice *O. longistaminata* --Manuscript Draft--

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Abstract:	<p>The abuse of fertilizer results in tall rice plants that are susceptible to lodging and reduced plant yield. Hence, it is important to identify and utilize the QTLs/genes for lodging resistance breeding. <i>O. longistaminata</i> exhibited a strong stem and high biomass productively, which could be the candidate gene pool for cultivars lodging resistance improvement. However, most of previous studies have focused on cultivated rice and few reports on wild rice. Here, a set of 152 BC₂F₂₀ lines derived from a cross between 93-11 and <i>O. longistaminata</i> were evaluated for lodging resistance. QTL mapping analysis combined with SNP marker derived from high-throughput sequencing identified 12 QTLs for stem diameter (SD), 11 QTLs for stem length (SL) and 3 QTLs for breaking strength (BS). Of which, 14 QTLs were firstly identified from <i>O. longistaminata</i>. A major QTL, qLR1 which was delimited to a region about 80 Kb on chromosome 1, increased stem diameter, stem length and breaking strength. And another major QTL, qLR8 that was delimited in an interval about 100 Kb on chromosome 8, significantly enhanced the breaking strength. These results provide evidence that the <i>O. longistaminata</i> can be exploited to develop lodging resistant rice lines.</p>	
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Deciphering the genetics basis of lodging resistance in wild rice *O.*

longistaminata

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Abstract

The abuse of fertilizer results in tall rice plants that are susceptible to lodging and reduced plant yield. Hence, it is important to identify and utilize the QTLs/genes for lodging resistance breeding. *O. longistaminata* exhibited a strong stem and high biomass productively, which could be the candidate gene pool for cultivars lodging resistance improvement. However, most of previous studies have focused on cultivated rice and few reports on wild rice. Here, a set of 152 BC₂F₂₀ lines derived from a cross between 93-11 and *O. longistaminata* were evaluated for lodging resistance. QTL mapping analysis combined with SNP marker derived from high-throughput sequencing identified 12 QTLs for stem diameter (SD), 11 QTLs for stem length (SL) and 3 QTLs for breaking strength (BS). Of which, 14 QTLs were firstly identified from *O. longistaminata*. A major QTL, *qLR1* which was delimited to a region about 80 Kb on chromosome 1, increased stem diameter, stem length and breaking strength. And another major QTL, *qLR8* that was delimited in an interval about 100 Kb on chromosome 8, significantly enhanced the breaking strength. These results provide evidence that the *O. longistaminata* can be exploited to develop lodging resistant rice lines.

Key words: lodging resistance, QTLs, *O. longistaminata*, wild rice, stem diameter

Introduction

Lodging in cereal crops is a major problem that results in decreased grain yield and deteriorated grain quality (Berry et al, 2004). Development of lodging-resistant varieties to cope this challenge has been widely attended to increase yield in rice, maize and other crops (Yadav et al, 2017, Zhu et al, 2013, Peng et al, 2014). Plant breeders have reduced lodging risk by introducing semi-dwarf gene *sd1*, known as “Green Revolution Gene” (Peng et al, 1999). However, recent studies showed that dwarf rice plant limit photosynthesis and biomass leading yield penalty (Murai et al, 2002). Meanwhile, it may have a negative pleiotropic effects on culm morphology (Okuno et al, 2014). In other words, the gibberellin synthesis gene (*sd1*) widely applied in Green Revolution rice also reduces the culm strength by decreasing culm diameter, which make it difficult to further improve lodging resistance by one semi-dwarf genes alone. Thus, it is important to search for alien genes favorable for breeding lodging-resistant rice.

Lodging resistance is a complex quantitative trait, which is affected by many factors, such as culm morphology, culm diameter and length, cellulose content and environment conditions. Previous studies have shown that the culm diameter and size is highly correlated with the lodging resistance of rice (Sowadan et al, 2018, Kashiwagi et al, 2006, Kashiwagi and Ishimaru, 2004, Fan et al, 2018). Ookawa et al. (Ookawa et al, 2010) identified four QTLs for bending moment at breaking and section modulus of the fourth internodes derived from two indica rice with strong culms. Kashiwagi et al. (Kashiwagi et al, 2008) detected five QTLs for pushing

1 67 resistance from a backcross between Nipponbare and Kasalath. Shailesh Yadav et al.
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3 68 (Yadav et al, 2017) mapped 12 QTLs for lodging resistance from a backcross between
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6 69 Swarna and Moroberekan. Moreover, three functional genes including *prl5*, *SCM2*,
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9 70 and *SCM3* have been identified to regulate stem diameter. Interestingly, *SCM2*
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12 71 increased not only culm strength, but also spikelet number. However, relative to
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15 72 cultivated rice, few reports about lodging resistance were detected from wild rice, let
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18 73 alone the underlying molecular mechanism. Hence, exploitation of new QTLs or
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21 74 genes contributed to lodging resistance from the wild rice help to enrich the genes for
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24 75 rice improvement of lodging resistance.

25 76 The perennial wild species *O. longistaminata*, which showed large stem diameter,
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28 77 thick stem wall, and high biomass production, is believed help to improve rice
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31 78 lodging resistance (Khush, 1997). In this study, we conducted QTLs analysis of
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34 79 potential lodging resistance of *O. longistaminata* by evaluation of stem diameter, stem
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37 80 length, and breaking strength using an advanced backcross inbred line (BIL)
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40 81 population derived from a cross between *O. longistaminata* and 93-11. Totally, 26
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43 82 QTLs for lodging resistance were detected, of which, 12 QTLs for stem diameter
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46 83 (SD), 11 QTLs for stem length (SL), and 3 QTLs for breaking strength (BS) were
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49 84 derived from *O. longistaminata*. These novel QTLs will lay the foundation for
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52 85 breeding strong lodging resistant rice, and broaden our understanding of the genetic
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55 86 basis of rice for lodging resistance.

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58 88 **Materials and methods**

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89 **Plant materials and field experiments**

90 The recurrent indica rice 93-11, wild rice *O. longistaminata* and 152 *O.*
91 *longistaminata* BILs were used in this research (Jin et al, 2018). Field experiments
92 were conducted in Linshui, Hainan province during the rice-grown season from late
93 October to late-March and in Wuhan, Hubei province during mid-May to
94 late-September in 2017 and 2018. Each BIL line were planted 5 rows, each row
95 planted 10 plants in a density of 20 cm × 16.5 cm. Three replications were carried out
96 by randomized complete block design for each experiment.

98 **Measurement lodging resistance related traits**

99 After heading two weeks, five plants of each BILs and 93-11 were selected to
100 investigate the lodging resistance traits. The stem diameter (SD) of the 5th internode
101 from the top was measured using an electronic vernier caliper in the field. Stem length
102 of the internode (SL) from the top was measured from the ground to the basal of
103 panicle. The breaking strength (BS), a parameter for the physical strength of the stem,
104 was measured at the last internode of the plants using a plant lodging tester.

106 **Statistical analysis**

107 All the data were analyzed using SPSS 20.0 statistical software. The means values of
108 each trait were used for further QTL analysis. The standard deviation of the means
109 was calculated using Microsoft Excel software. Correlation of the lodging resistance
110 traits were evaluated using Pearson's correlation.

111

112 QTL analysis and sequence analysis

113 A total of 2432 bin marker was used to construct the genetic linkage map covering the
114 whole genome as described by Jin et al (Jin et al, 2018). Inclusive Composite Interval
115 Mapping combined with additive mapping (ICIM-ADD) method was used to detect
116 more precise lodging resistance QTLs. The significant logarithm of odds (LOD) value
117 threshold for each trait was determined following the 5% permutation test with 1000
118 replicates. The putative genes on the QTL region were identified based on the RIGW
119 (<http://rice.hzau.edu.cn/rice/>).

120

121 Results

122 Phenotypic evaluation of lodging resistance-related traits

123 Three stem traits associated with lodging resistance of 93-11 and *O. longistaminata*
124 were investigated in Hainan and Wuhan during two consecutive years. And significant
125 difference for these three traits were found between the two parents in both sites
126 (Supplementary Figure 1 and Table 1). The wild rice performed better than 93-11 for
127 all of the three lodging-related traits, indicating *O. longistaminata* can efficiently
128 improve the lodging resistance of rice.

129 For the BIL population, the variation of stem diameter, stem length, and breaking
130 strength in Hainan ranged among 5.79~12.23 mm, 41.08~157.90 cm, and 5.81~29.68
131 N, with an average of 8.66 mm, 83.59 cm, and 14.95N, respectively in 2017 (Figure
132 1). Correspondingly, the CV of SD, SL, and BS was 13.51%, 35.34%, and 31.04%,

1 133 respectively (Table 1). In 2018 at Hainan, the variation of SD, SL, and BS of *O.*
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3 134 *longistaminata* BIL population ranged among 5.52~13.00 mm, 49.11~181.67 cm, and
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6 135 6.13~30.28 N, with an average of 8.68 mm, 103.03 cm, and 15.12 N, respectively.
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9 136 Correspondingly, the CV of SD, SL, and BS was 14.29%, 29.72%, and 30.75 %,
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11
12 137 respectively.

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14 138 While in Wuhan, the SD ranged from 5.27 to 12.78 mm and 5.17 to 13.25 cm,
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17 139 with the CV of 14.73% and 15.40% in 2017 and 2018, respectively. The SL ranged
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20 140 from 64.1 cm to 178.67 cm, and 78.60 cm to 191.53 cm, with the CV of 28.87% and
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23 141 33.95% in 2017 and 2018, respectively. The BS ranged from 5.41 to 30.86 N and 5.42
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26 142 to 31.33 N, with the CV of 30.97% and 31.21% in 2017 and 2018, respectively (Table
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28 143 1). These results indicate that the BS , SL and SD of *O. longistaminata* BILs all
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31 144 showed great variation among different lines, BS and SL have more genetic diversity
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34 145 than SD regardless of environments.

35 36 146 37 38 39 147 **Correlations among lodging resistance related traits**

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42 148 To illustrate the relationships among the lodging resistance-related traits so as to
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45 149 better understanding the genetic basis [16, 17], the correlation coefficients between the
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48 150 traits of the BILs were calculated using Pearson's method (Supplementary Table 2 and
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51 151 Supplementary Table 3). Results showed that the breaking strength was positively
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54 152 correlated with stem diameter and stem length in Hainan ($p < 0.001$). However, the
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57 153 breaking strength was negatively correlated with stem length at Wuhan in 2018
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60 154 (Supplementary Table 2). The negative correlation may due to the photoperiod

sensitivity of some BILs inherited from wild rice *O. longistaminata*. These results indicate that wild rice *O. longistaminata* can improve the lodging resistance without the stem length reduction. In other words, it is possible to breed high yield and high biomass cultivars with high lodging resistance, which may provide appropriate gene resources to breed super rice with taller stalk (Yuan, 2017).

QTL mapping of lodging resistance associated traits in BILs

A total of 26 QTLs for stem diameter, stem length, and breaking strength were detected in *O. longistaminata* BILs (Figure 2), among them, 14 QTLs were detected from *O. longistaminata* (Table 2 and Table 3), 12 QTLs were detected from 9311 (Supplementary Table 2 and Supplementary Table 3). Of which, eight QTLs including two for stem diameter, three for stem length and three for breaking strength were identified from *O. longistaminata* in Hainan during the two years. Two QTLs *qSD1.1* and *qSD9.1* for stem diameter was detected. The QTL *qSD9.1* was detected in two consecutive years harbor a LOD of 3.17 and 4.99, with the PVE of 6.87% and 10.85% in 2017 and 2018, respectively. *qSD1.1* was identified in 2017 explained phenotypic variation of 13.68%, with a LOD of 5.78. The three QTLs (*qSL1.1*, *qSL2.1* and *qSL2.2*) contributing to stem length were detected on chromosomes 1 and 2 in *O. longistaminata* (Figure 2). The QTL *qSL1.1* was detected in both 2017 and 2018 that explained 59.40% and 67.03% of the phenotypic variation, respectively. QTLs *qSL2.1* and *qSL2.2* explained phenotypic variation of 5.53% and 10.22%, respectively. The three QTL (*qBS1.1*, *qBS4.1*, and *qBS8.1*) contributing to stem breaking strength were

mapped on chromosomes 1, 4, and 7. Among them, QTL *qBS1.1* was repeatedly detected across two years with the PVE of 5.07% and 1.86%, respectively. While *qBS4.1* and *qBS8.1* were detected only in 2017, they explained the phenotypic variance of 7.10% and 8.70%, respectively.

A total of 10 QTLs for lodging-related traits were identified derived from *O. longistaminata* in Wuhan. Three QTLs (*qSD1.1*, *qSD8.1*, and *qSD12.1*) for stem diameter were detected on chromosomes 1, 8, and 12. The QTL *qSD8.1* was constantly detected in 2017 and 2018 harbored a LOD of 3.43 and 4.55, with the PVE of 18.33% and 8.03%, respectively. The QTL *qSD1.1* and *qSD12.1* were identified in 2018 explained phenotypic variance of 25.37% and 34.80%, respectively. Moreover, QTL *qSD12.1* shared the largest LOD and PVE for stem diameter. Five QTLs (*qSL1.1*, *qSL1.2*, *qSL2.3*, *qSL8.1*, and *qSL11.2*) that contributing to stem length were mapped on chromosomes 1, 2, 3, 8, and 11. The QTL *qSL1.1* was detected in both years and explained phenotypic variation of 7.21% and 79.89%, respectively. The QTLs *qSL1.2* and *qSL2.3* were identified in 2017 and harbored PVE of 9.12% and 8.09 %, respectively. QTLs *qSL8.1* and *qSL11.2* were mapped in 2018 with a LOD value of 3.32 and 2.50, and explained the variation of phenotypic as 4.42% and 5.01%, respectively. Two QTLs controlling breaking strength (*qBS1.1* and *qBS8.1*) were identified in 2018 explained of 11.22% and 10.22% phenotypic variations, respectively.

Co-localization of the QTLs for lodging resistance

199 To investigate the genetic effects of the QTLs responsible for lodging resistance, all
200 the QTLs identified from *O. longistaminata* in two sites were further analyzed.
201 Interestingly, the loci of *qSD1.1*, *qSL1.1* and *qBS1.1* were co-located on chromosomes
202 1 (Figure 2). Meanwhile, another QTL *qBS8.1* was detected both in Wuhan and
203 Hainan. Of which, the QTL hotspot *qSD1.1/qSL1.1/qBS1.1* explained 25.37% of the
204 lodging resistance variance for stem diameter, 79.89% for stem length, and 16.76%
205 for breaking strength, which indicates this loci may play an important role for lodging
206 resistance. What's more, this pleiotropic QTL was firstly detected in wild rice and we
207 named it lodging resistance 1 (*qLR1*). Meanwhile, another QTL hotspot *qBS8.1*
208 explained lodging resistance variance for breaking strength of 8.70% and 10.22% at
209 Hainan and Wuhan, respectively (Table 2 and Table3), was rename as lodging
210 resistance 8 (*qLR8*).

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212 **Confirmation of the *qLR1* and *qLR8* for lodging resistance**

213 To further confirm the function of the newly identified genetic locus of *qLR1* and
214 *qLR8* in *O. longistaminata*, a high-resolution mapping with several recombinant
215 including BIL 1708, 1719, 1829, 1738,1755, 1795, and 1769 was performed and the
216 *qLR1* was narrowed to an interval between BIN1-161 and BIN1-162 (Figure 3A). In
217 term of the whole BILs, the average breaking strength of the lines with *qLR1* was
218 19.60 N, which was significantly higher than those without *qLR1* (14.32N, P<0.001).
219 What's more, the average stem diameter and stem length of the lines carried *qLR1*
220 was 9.72mm and 123.12cm, was significantly larger than the lines without *qLR1*,

1 221 respectively (7.46mm, 64.46cm, $P<0.01$, and $P<0.001$, Figure 3B). These results
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3 222 indicate that the *qLR1* from *O. longistaminata* can significantly increase lodging
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6 223 resistance.
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9 224 Similarly, we delimited the *qLR8* to both tightly-linked marker BIN8-32 and
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11 225 BIN8-33 (Figure 4A). Then, we compared the genetic effects of *qLR8* on lodging
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14 226 resistance in BIL lines. Results showed that the average breaking strength of the BIL
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17 227 lines with *qLR8* group was 19.64 N in Hainan, significantly higher than that of the
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20 228 other population without *qLR8* (14.34 N), similarly results were also observed in
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23 229 Wuhan (Figure 4B). These results indicated that *qLR8* derived from *O. longistaminata*
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26 230 can significantly increase breaking strength.
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28 231 To better understand if the two QTLs had additive effects, BIL 1704, 1720, 1728,
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31 232 1730, 1742, 1779, and 1797-2, which carried *qLR1* and *qLR8* were selected. The
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34 233 breaking strength of the seven lines range from 15.92 N to 29.68 N, with an average
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37 234 of 22.55 N, was significantly higher than the average value of the *qLR1*-carrying lines
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40 235 (19.60 N), but no significant difference with *qLR8*-carrying line due to only 16 lines
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43 236 harbored *qLR8* from *O. longistaminata* (Supplementary Figure 2). These results
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46 237 indicate that two QTLs can significantly increase the lodging resistance of rice than
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49 238 either one QTL. In summary, *qLR1* and *qLR8* from *O. longistaminata* can
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52 239 significantly increase the lodging resistance of rice.
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55 241 Discussion

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58 242 Lodging resistance is an important trait that is necessary for achieving high grain
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1 243 yield in rice production (Khush, 1997). In the past 60 years, short plant stature was the
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3 244 major target for improvement of lodging resistance, but most of the dwarf genes are
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6 245 rarely applied to rice breeding due to defects caused to other agronomic phenotypes,
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9 246 such as low fertility, and short grains (Wu et al, 2018b). The “Green evolution” gene
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11 247 *sd1* is still the only dwarf source predominantly to produce semi-dwarf varieties in
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14 248 rice. Recently, some studies have raised to improve rice yield by increasing plant
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17 249 biomass (Jiang et al, 2016), and increasing plant height is an effective and feasible
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20 250 way to increase biomass in morphological viewpoint (Ma and Yuan, 2015, Ying et al,
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23 251 1998, Donald, 1968). In this study, some *O. longistaminata* BIL lines with high plant
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26 252 height also exhibited larger stem diameter, more breaking strength and strong
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29 253 resistance to lodging (Figure 3), indicating that wild rice *O. longistaminata* is a novel
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31 254 genetic resource for breeding strong lodging-resistant rice.

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34 255 Although there is over 21 QTLs for lodging resistance-associated traits had been
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36 256 reported and three of them having been cloned in cultivars (Sowadan et al, 2018,
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39 257 Ookawa et al, 2016, Mulsanti et al, 2018), less efforts were contributed to the
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42 258 detection on QTLs for lodging resistance in wild rice (Xie et al, 2017, Wu et al,
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45 259 2018a), and we know almost zero about the lodging resistance in wild rice *O.*
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48 260 *longistaminata*. In this study, we identified 12 new QTLs for lodging resistance
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51 261 derived from wild rice *O. longistaminata* for the first time, and filled the gap in this
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54 262 area. What’s more, *qLR1*, a pleiotropic QTL responsible for stem length, stem
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57 263 diameter, and breaking strength, was narrowed to a certain small region about 0.65
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59 264 cM and covered about 80 kb physical distance on chromosome 1 (Figure 3A and
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Table 3). Corresponding to the MH63RS1 reference genome (http://rice.hzau.edu.cn/rice/), the region contains only 10 predictable genes: two hypothetical protein and eight functional genes (Table 4). Meanwhile, *qLR8*, another QTL hotspot contributing to the stem breaking strength, was located in an interval about 1.34 cM and covered about 120 Kb according to MH63RS1 genome. This region contained 21 predicted genes: eight hypothetical protein and 13 functional genes (Table 4). These QTLs identified in wild rice not only get a breakthrough in the genetic basis of wild rice lodging resistance, but also establish a robust foundation for further cloning the gene for lodging resistance.

In rice breeding practice, lodging resistance is one of the most essential characters required for breeding super-high-yielding rice, and an ideal plant type is the foundation for super-high yield. Grain yield is the product of harvest index (HI) and biomass, previous studies had shown that increasing plant height can increase biomass. Here, two newly identified QTLs, *qLR1* and *qLR8* both improved breaking strength. What's more, the former can also improve stem diameter and stem length in a particular way. Take all into consideration, these two QTLs don't overlap with any previous reports. If they are pyramided together, it will greatly improve the lodging resistance in rice, indicating that these two new QTLs will serve as a candidate resource contributing to rice breeding super rice programs.

287 **Abbreviations**

288 SD: stem diameter; SL: stem length; BS: breaking strength; QTL: quantitative traits
289 loci; HI: harvest index; LR: lodging resistance; PVE: phenotypic variation explained;
290 BIL: backcross inbred line; HN: Hainan; WH: Wuhan.

291

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294

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298 (2016ZX08001004-001-002), and National Natural Science Foundation (31870322)
299 of China.

300

301 **Availability of data and meterial**

302 The datasets supporting the conclusions of this article are provided within the article
303 and its additional files.

304

305 **Author's Contributions**

306 SL and WL conceived and planned the work, WL, DD, ZY, YC and WY performed
307 phenotypic screening, SL, NL and JJ developed the population. WL analysis the
308 genotypic data. WL, ZZ and SL drafted the manuscript.

1	309	
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3	310	Ethics approval and consent to participate
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6	311	No applicable.
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11	313	Consent for publication
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36	322	or financial relationships that could be construed as a potential conflict of interest.
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 408 **Figure 1. Phenotyping of lodging-resistant traits of 152 *O. longistaminata* BILs.**

WH and HN present Wuhan and Hainan, respectively.

Figure 2. QTLs for stem diameter, stem length and breaking strength in *O. longistaminata* BILs. The most left is the scales for the measure of genetic length, left is the genetic length and bin marker distributed on each chromosome. The upward direction indicates the alleles of QTLs derived from *O. longsitaminata* showing positive effects. The downward direction indicates the 93-11 allele of the QTLs showing positive effects. The red shapes show that the QTLs were detected in Wuhan, and the blue shapes represents the QTLs were identified in Hainan. The brown shapes mean QTLs mapped both two environments.

Figure 3. Confirmation of the QTL of *qLR1*. (A) Verification of the *qLR1* using 7 BIL lines to delimit it to an interval between molecular marker between Bin1-161 and Bin1-162. Black rectangle shows the homozygous derived from *O. longistaminata*, white rectangle indicates the homozygous from 93-11. (B). *qLR1* effect value analysis of stem diameter, stem length and breaking strength in *O. longistaminata* BILs. White shapes mean the alleles from parent 93-11, black shapes indicate the homozygous genotype shared by *O. longistaminata*.

Figure 4. Validation of the QTL of *qLR8*. (A) Verification of the *qLR8* using 7 BILs to delimit it to an interval between two marker Bin8-32 and Bin8-33. (B) *qLR8* effect value analysis of breaking strength in both Wuhan and Hainan environments.

Tables

Table 1. Performance of lodging associated trait in BILs among two years

Traits	environment	year	BILs				
			Min	Max	Average	SD	CV (%)
Stem Diameter(mm)	WH	2017	5.27	12.78	7.94	1.17	14.74
		2018	5.17	13.25	7.92	1.22	15.40
	HN	2017	5.79	12.23	8.66	1.17	13.51
		2018	5.52	13.00	8.68	1.24	14.29
Stem Length (cm)	WH	2017	64.1	178.67	109.24	31.54	28.87
		2018	78.6	191.53	119.70	40.64	33.95
	HN	2017	41.08	157.90	83.59	29.54	35.34
		2018	49.11	181.67	103.02	30.62	29.72
Breaking Strength (N)	HN	2017	5.81	29.68	14.95	4.64	31.04
		2018	6.13	30.28	15.12	4.65	30.75
	WH	2017	5.41	30.86	15.50	4.80	30.97
		2018	5.42	31.33	15.51	4.84	31.21

. WH and HN present Wuhan and Hainan, respectively.

463 **Table 2. QTLs of lodging resistance derived from *O. longistaminata* in Hainan**

Investigated traits	QTLs	Chr	Pos (cM)	L/Bin	R/Bin	L/bp	R/bp	LOD		PVE		Add	
								2017	2018	2017	2018	2017	2018
Stem diameter	<i>qSD1.1</i>	1	226	1-171	1-172	35964905_36016970	35557085_35961004	5.78		13.68		0.49	
	<i>qSD9.1</i>	9	8	9-6	9-8	20987075_21077941	20967923_20984846	3.17	4.99	6.87	10.85	0.63	0.80
Stem length	<i>qSL1.1</i>	1	216	1-159	1-160	36587534_36864307	36569506_36587533	31.22	39.27	59.40	67.03	25.55	25.87
	<i>qSL2.1</i>	2	145	2-201	2-202	8863323_8895438	8841866_8863322	4.58		5.53		9.28	
	<i>qSL2.2</i>	2	119	2-247	2-148	18779567_18968062	18713595_18770186		3.77		10.22		2.57
Breaking strength	<i>qBS1.1</i>	1	216	1-161	1-162	36539238_36569505	36515725_36536296	4.87	5.07	13.70	16.76	1.86	2.06
	<i>qBS4.1</i>	4	120	4-97	4-98	19770490_20005045	19663208_19770489	2.66		7.10		2.14	
	<i>qBS8.1</i>	8	32	8-32	8-33	8075498_8118334	8118335_8196568	3.30		8.70		2.21	

466 **Table 3. QTLs of lodging resistance derived from *O. longistaminata* in Wuhan**

Investigated traits	QTLs	Chr	Pos(cM)	L/Bin	R/Bin	L/bp	R/bp	LOD		PVE		Add	
								2017	2018	2017	2018	2017	2018
Stem diameter	<i>qSD8.1</i>	8	74	8-59	8-60	9870144_9978376	20727323_21034035	3.43	4.55	18.83	8.03	1.12	1.13
	<i>qSD1.1</i>	1	216	1-161	1-162	36539238_36569505	36515725_36536296		13.29		25.37		0.65
	<i>qSD12.1</i>	12	156	12-168	12-169	22817923_22863684	22863685_22983852		51.39		34.80		1.99
Stem length	<i>qSL1.1</i>	1	216	1-160	1-161	36587534_36864307	36569506_36587533	2.77	53.00	7.21	79.89	9.11	31.64
	<i>qSL1.2</i>	1	286	1-310	1-311	23526094_23553497	23494864_23526093	3.43		9.12		10.51	
	<i>qSL2.3</i>	2	189	2-252	2-253	10475930_10505727	10520137_10562305	2.70		8.09		22.59	
	<i>qSL8.1</i>	8	78	8-59	8-60	9870144_9978376	20727323_21034035		3.32		4.42		18.61
	<i>qSL11.2</i>	11	290	11-128	11-129	1_370955	1355747_137440		2.50		5.01		23.36
Breaking strength	<i>qBS1.1</i>	1	216	1-161	1-162	36539238_36569505	36515725_36536296		4.02		11.22		1.75
	<i>qBS8.1</i>	8	32	8-31	8-32	8075498_8118334	8118335_8196568		3.78		10.22		2.57

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Table 4. Putative genes in two QTL loci of *qLR1* and *qLR8*

QTLs	Gene ID	Start site (bp)	End site (bp)	Function
<i>qLR1</i>	MH01t0727000-1	36523094	36523616	Hypothetical protein
	MH01t0727100-1	36528435	36534828	UDP-glucuronate:xylan alpha-glucuronosyltransferase 1
	MH01t0727200-1	36538440	36543325	Probable pectinesterase/pectinesterase inhibitor 51
	MH01t0727300-1	36543583	36548625	hypothetical protein OsI_04663
	MH01t0727400-1	36549701	36550356	hypothetical protein
	MH01t0727500-1	36550414	36551069	Cation/calcium exchanger 1
	MH01t0727600-1	36555859	36557636	Acyl-[acyl-carrier-protein] desaturase 1; chloroplastic
	MH01t0727700-1	36557901	36559958	Pentatricopeptide repeat-containing protein
	MH01t0727800-1	36560108	36568738	CHD3-type chromatin-remodeling factor PICKLE
	MH01t0727900-1	36569726	36571938	Putative clathrin assembly protein

<i>qLR8</i>	MH01t0165000-01	80811091	8086676	Bifunctional epoxide hydrolase 2
	MH01t0165100-01	8087305	8088199	hypothetical protein
	MH01t0165200-01	8088985	8089633	unknown protein
	MH01t0165400-01	8104092	8104277	hypothetical protein
	MH01t0165300-01	8103887	8106234	hypothetical protein
	MH01t0165500-01	8109136	8112092	putative gypsy-type retrotransposon RIRE2
	MH01t0165600-01	8115542	8118452	putative retrotransposon protein
	MH01t0165800-01	8122688	8124339	hypothetical protein
	MH01t0166000-01	8125214	8125939	hypothetical protein
	MH01t0166200-01	8129037	8129919	hypothetical protein
<i>qLR8</i>	MH01t0166300-01	8131762	8132962	hypothetical protein
	MH01t0166400-01	8134838	8137516	Disease resistance protein RGA2
	MH01t0166500-01	8140661	8144868	Dynein 8 kDa light chain; flagellar outer arm

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	MH01t0166600-01	8146496	8146816	Indole-3-acetic acid-induced protein ARG2
	MH01t0166700-01	8148809	8149501	antigen-like protein
	MH01t0166800-01	8150263	8152690	Putative AC transposase
	MH01t0166900-01	8160667	8169675	Zinc finger CCCH domain-containing protein 4
	MH01t0167000-01	8170873	8174334	Probable U6 snRNA-associated Sm-like protein LSm4
	MH01t0167100-01	8177528	8177923	Protein RALF-like 33
	MH01t0167200-01	8184849	8185401	18.6 kDa class III heat shock protein
	MH01t0167300-01	8185788	8187121	Flowering-promoting factor 1-like protein 1

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Dear Editor:

On behalf of the authors, I am submitting the manuscript entitled “Deciphering the genetics basis of lodging resistance in wild rice *O. longistaminata*” be considered for publication in “*rice*”. This article type is Original Research. The manuscript has neither been submitted for publication nor published in whole or in part elsewhere. We declare that all authors listed on the title page have read the manuscript and agreed to this submission. We assume responsibility for the validity and legitimacy of the data and its interpretation.

Lodging has caused great losses in both grain yield and quality in rice. Furthermore, it has also led to difficulties in harvest operations. The “green revolution” gene *sd1* contribute to lodging with reducing the plant height. However, short plant height has been used to shown negative effects on stem morphology in rice. Hence, it is important to search for alien genes favorable for breeding lodging-resistant rice.

O. longistaminata, which showed large stem diameter, thick stem wall, and high biomass production, is believed help to improve rice lodging resistance. Up to now, although several QTLs for cultivated rice lodging resistance had been identified. Few QTLs had been detected for lodging resistance from wild rice. In this study, we use ICIM mapping method combine with a high density bin map to identify the QTLs for lodging resistance from *O. longistaminata*. We mapped 4 QTLs for stem diameter, 7 QTLs for stem length, and 3 for breaking strength. Interestingly, the major QTL *qLR1* contributed to stem diameter, stem length, and breaking length was delimited to a region about 80 Kb. Meanwhile, another major QTL *qLR8* contributed to breaking strength were fine-mapped to a region about 100 Kb. These two regions are fairly small for rice breeding, so we consider that this work may be important for reader to breed high lodging resistance rice.

If you have any questions, please do not hesitate to contact me.

Yours sincerely,

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2019-8-20

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