

A peer-reviewed version of this preprint was published in PeerJ on 19 November 2019.

[View the peer-reviewed version](https://doi.org/10.7717/peerj.7792) (peerj.com/articles/7792), which is the preferred citable publication unless you specifically need to cite this preprint.

Wu C, Tang S, Li G, Wang S, Fahad S, Ding Y. 2019. Roles of phytohormone changes in the grain yield of rice plants exposed to heat: a review. PeerJ 7:e7792 <https://doi.org/10.7717/peerj.7792>

Response of phytohormone homeostasis to heat stress and the roles of phytohormones in rice grain yield: a review

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Rice is highly susceptible to heat stress at the reproductive stage. In this review, we first summarize recent progress in heat effects on rice grain yield during different reproductive stages. Different responses of yield traits of rice to heat stress during different reproductive stages are identified. The number of spikelets per panicle is reduced by heat stress during the early reproductive stage but is not affected by heat stress during the mid-late reproductive stage. Spikelet sterility induced by heat stress can be attributed primarily to physiological abnormalities in the reproductive organs during flowering but attributed to structural and morphological abnormalities in reproductive organs during panicle initiation. The lower grain weight caused by heat stress during the early reproductive stage was due to a reduction in non-structural carbohydrates, undeveloped vascular bundles, and a reduction in grain length and width, while a shortened grain filling duration, reduced grain filling rate, and decreased grain width affect grain weight when heat stress occurs during grain filling. Phytohormones play vital roles in regulating plant adaptations against heat stress. We discuss the processes involving phytohormone homeostasis (biosynthesis, catabolism, deactivation, and transport) in response to heat stress. It is currently thought that biosynthesis and transport may be the key processes that determine phytohormone levels and final grain yield in rice under heat stress conditions. Finally, we prospect that screening and breeding rice varieties with comprehensive tolerance to heat stress throughout the entire reproductive phase could be feasible to cope with unpredictable heat events in the future. Studies in phytohormone homeostatic response are needed to further reveal the key processes that determine phytohormone levels under heat condition.

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11 **Abstract**

12 Rice is highly susceptible to heat stress at the reproductive stage. In this review, we first summarize recent
13 progress in heat effects on rice grain yield during different reproductive stages. Different responses of yield
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22 during grain filling. Phytohormones play vital roles in regulating plant adaptations against heat stress. We
23 discuss the processes involving phytohormone homeostasis (biosynthesis, catabolism, deactivation, and
24 transport) in response to heat stress. It is currently thought that biosynthesis and transport may be the key
25 processes that determine phytohormone levels and final grain yield in rice under heat stress conditions. Finally,
26 we prospect that screening and breeding rice varieties with comprehensive tolerance to heat stress throughout
27 the entire reproductive phase could be feasible to cope with unpredictable heat events in the future. Studies in
28 phytohormone homeostatic response are needed to further reveal the key processes that determine
29 phytohormone levels under heat condition.

30 **Key words:** Rice grain yield; Heat stress; Spikelet fertility; Grain weight; Spikelets per panicle;
31 Phytohormone homeostasis

32 Introduction

33 Human activities have triggered climate change, which is characterized by global warming. The global
34 change in climate represents one of the most serious challenges that humans have had to face (Marsicek et al.,
35 2018). Crop production and food security are essential components of agriculture, and each is directly and
36 seriously affected by climate change. Rice is a staple food for approximately half of the world's population.
37 Frequent heat waves have had serious impacts on rice production (Zhang et al., 2014). An analysis of historical
38 data showed that rice grain yield decreased by 14% for every 1°C increase in the daily average temperature
39 (Aggarwal, 2009).

40 Rice plants are vulnerable to heat stress during the reproductive phase (Boote et al., 2005). Heat stress
41 will occur more frequently and unpredictably as global warming continues to worsen, which may cause rice
42 plants to suffer heat stress at any time during their reproductive stages (Marsicek et al., 2018). Previously,
43 most of the studies evaluated the responses of heat stress and the underlying mechanisms during the mid-late
44 reproductive phase (Wu & Cui, 2014). A risk assessment of the spatiotemporal variation of high temperature
45 events during the past 20 years indicated that heat events occurred in the middle and lower reaches of the
46 Yangtze River, one of the major paddy rice production areas in China, as early as mid-early July (Lin et al.,
47 2016), during which panicle formation of midseason rice occurs, resulting in disrupted panicle development
48 and finally yield loss (Wu et al., 2016). It was previously reported that rice plants respond differently to heat
49 stress during different developmental stages (Shi et al., 2015). However, few articles reached conclusions
50 concerning the different effects of heat on rice grain yield at different reproductive stages (Fahad et al., 2018).
51 Therefore, the present review offers a full conclusion regarding the effects of heat on rice grain yield during
52 the reproductive phase (panicle initiation, flowering, and grain filling), and the distinct variations in rice grain
53 yield based on the timing of the heat stress during the three reproductive stages are also identified.

54 Phytohormones play an important role in coordinating the response to heat stress in rice plants. Most of
55 the previous studies have explored the phytohormonal mechanisms of the effects of heat stress on rice grain
56 yield by analysing phytohormone levels in rice (Fahad et al., 2015). Phytohormone levels in target organs are
57 associated with processes involved in phytohormone homeostasis, e.g., biosynthesis, catabolism, deactivation,
58 and transport (phytohormones acting as mobile signals should be considered) (Sakakibara, 2010), but our work
59 on the responses of phytohormone homeostasis to heat stress is far from adequate. This review provides
60 insights into the physiological mechanisms of the rice heat stress response by considering the roles of
61 homeostasis of phytohormones (cytokinin, CTK; Indole-3-acetic acid, IAA; gibberellins, GAs; abscisic acid,
62 ABA) in grain yield.

63 Survey Methodology

64 Research papers published both in English and in Chinese were searched from the Web of Science and the
65 China National Knowledge Infrastructure (CNKI, the largest Chinese Academic Journals database) from 1950-
66 2018, respectively. There are six search terms: “rice AND high temperature/heat/warming” and “high
67 temperature/heat/warming AND phytohormone”. An initial search resulted in 26618 articles, which were
68 reduced to 8813 by limiting research to plant science, agronomy, cell biology, physiology, biology,
69 developmental biology, and environmental science. We focus mainly on response of physiological and
70 molecular aspects involving phytohormone homeostasis in rice varieties to heat stress. Then, we examined the
71 article titles, abstracts, key words to judge their relevance, and 2115 articles were considered relevant. There
72 were 15 studies in other crops (barley, wheat, tomato, etc) and Arabidopsis which were highly relevant to the
73 topic of this review, were also included.

74 **Effects of heat stress on grain yield and yield components**

75 **Heat stress definition and the maximum temperature for rice**

76 Heat stress is defined as an increase in temperature beyond a critical threshold and for a certain period of
77 time that results in irreversible damage to plant growth and development (Wu, 2016). Heat injury depends on
78 the intensity and duration of exposure to high temperatures and can be subdivided into two categories: (i) short
79 periods of exposure to extreme high temperatures and (ii) long periods of exposure to sub-high temperatures
80 (Berry & Bjorkman, 1980). Rice plants are highly susceptible to heat stress during their reproductive phase.
81 The early reproductive phase in rice is referred to as the period from panicle initiation to booting (stages R0-
82 R3 in Figure1), during which the maximum temperature for rice growth is 33.1°C. The mid-late reproductive
83 phase represents the period from heading to physiological maturity and includes flowering (stage R4 in
84 Figure1) and grain filling (stages R5-R9 in Figure1), the maximum temperature for rice growth is 37°C and
85 31.3 °C during flowering and grain filling, respectively (Counce et al., 2000; Itoh et al., 2005; Sánchez et al.,
86 2014). Heat stress reduces rice grain yield during the reproductive phase, but the impacts on grain yield and its
87 components differ when heat stress occurs at different reproductive stages (Shi et al., 2015).

88 **Effects of heat stress on the number of spikelets per panicle**

89 The reduction in the number of spikelets per panicle in heat-stressed plants is due to the attenuated
90 differentiation of secondary branches and their attached florets as well as the promotion of their degradation
91 during the early reproductive phase, but heat stress does not affect the differentiation of primary branches and
92 their attached florets (Wu et al., 2016). The differentiation of secondary branches and their attached florets is
93 more sensitive to environmental factors than that of primary branches and their attached florets (Ding et al.,
94 2014). Heat stress does not affect the number of spikelets per panicle during the mid-late reproductive phase.

95 **Effects of heat stress on spikelet fertility**

96 During the early reproductive stage, heat stress induces panicle enclosure (Wu et al., 2016). In an
97 enclosed panicle, the spikelets in the lower region are surrounded by an enclosing sheath (the lower surrounded
98 spikelets), while spikelets in the upper region of the panicle can successfully expand out of the sheath (the
99 upper expanded spikelets), as illustrated in [Figure 1](#). For the lower surrounded spikelets, spikelet sterility in
100 heat-stressed plants was associated with failure of pollination due to the physical hindrance of the enclosing
101 sheath (Wu et al., 2016). For the upper expanded spikelets, structural abnormalities in the anthers, disrupted
102 function of the septum and tapetum (Wang et al., 2016a), inhibition of microsporogenesis, reductions in the
103 starch accumulation and cytoplasm in pollen (Sakata et al., 2000), and morphological and physiological
104 abnormalities of the stigma (Takeoka et al., 1991) collectively reduced the possibility of successful pollination
105 in heat-stressed rice plants (Matsui & Omasa, 2002).

106 During the flowering stage, spikelet sterility in heat-stressed plants is associated with reduced
107 functionality of female and male organs. Impaired female and male organs account for 34% and 66%,
108 respectively, of spikelet sterility under heat stress conditions (Fábián et al., 2019). Behaviours of female and
109 male organs affect pollination and fertilization, which are impacted by heat stress: (i) the septum ruptures, and
110 stomium splits occur in the anthers. Heat stress delays or even blocks septum ruptures due to inhibition of
111 pollen swelling (Wilson et al., 2011), which is partially attributed to a disturbance in water metabolism under
112 heat stress conditions (Matsui & Omasa, 2002). (ii) Pollen sheds onto the stigmas. Heat stress disturbs anther
113 dehydration, resulting in sticky pollen grains that are retained inside the locules of dehiscent anthers, as
114 supported by our previous observation ([Figure 2B](#)). (iii) Pollen germination, pollen tube penetration, and
115 sperm delivery to the sac are affected. Heat stress affects the balance of ions (such as K^+ and Ca^{2+}) (Yan et al.,
116 2002), carbohydrate metabolism (Firon et al., 2006), and regulators (phytohormones) in pollen (Sakata et al.,
117 2010), as well as stigma vigour (Zhang et al., 2014), which collectively reduces pollen germination and pollen
118 tube polarized growth.

119 Notably, anther dehiscence is the initial step of pollination and is highly susceptible to heat stress and thus
120 was suggested as a selective marker for screening heat tolerance (Matsui & Omasa, 2002). However, we
121 observed that heat stress prevents the majority of pollen grains from escaping the dehiscent anthers ([Figure 2B](#)),
122 thus reducing the number of pollen grains available for successful pollination. This result may explain why the
123 percentage of dehiscent thecae (indicated by a basal or apical slit or aperture) was not always strongly
124 correlated with spikelet fertility under heat stress conditions (Kobayashi et al., 2011). Thus, simply screening
125 for slits or apertures in the anthers cannot ensure successful pollination in heat-stressed plants. We propose that
126 dehiscent anthers, characterized by slits or apertures, as well as low levels of residual pollen grains are
127 guarantees for effective pollination.

128 In summary, spikelet sterility induced by heat stress can be attributed primarily to physiological
129 abnormalities in the reproductive organs during flowering, and the associated mechanisms of the effects of

130 heat stress during the early reproductive phase are different, which is mainly attributed to structural and
131 morphological abnormalities in reproductive organs (Figure 3).

132 **Effects of heat stress on grain weight**

133 Rice grain weight is determined by multiplying grain volume by grain plumpness. Heat stress had adverse
134 effects on grain volume and grain plumpness during the reproductive phase (Wang et al., 2016b). The small
135 grain size in heat-stressed rice plants was characterized by both reduced grain width and reduced grain length
136 when suffering from high temperature stress during the panicle initiation stage (Takeoka et al., 1991; Wu et al.,
137 2016), but it was attributed to reduced grain thickness when suffering from high temperature stress during the
138 grain filling stage (Mohammed et al., 2013). Grain plumpness was associated with the duration of grain filling
139 and the grain filling rate. The reduction in grain plumpness induced by heat stress during the early reproductive
140 phase was associated with a reduced content of non-structural carbohydrates in stems (Li, 2012) and hindered
141 vascular bundle development (Zhang et al., 2009) but was due to the shortened duration of grain filling,
142 although the grain filling rate was increased by moderately high temperatures during the grain filling stage
143 (Cao et al., 2016; Dou et al., 2017). Notably, the early termination of grain filling induced by moderately high
144 temperatures was the result of lower sink activity due to the early senescence of the panicle, not a lack of
145 assimilation (Kim et al., 2011).

146 In conclusion, the lower grain weight caused by heat stress during the early reproductive phase is due to
147 reduced non-structural carbohydrates, undeveloped vascular bundles, and reduced grain length and width, but
148 it is attributed to a shortened grain filling duration, reduced grain filling rate, and decreased grain width when
149 exposure to heat stress occurs during grain filling (Figure 3).

150 **Responses of phytohormones to heat and their role in yield components**

151 **Phytohormone changes affect the spikelets per panicle under heat stress**

152 Heat stress reduced the content of CTK. Reductions in the CTK contents of heat-stressed rice plants were
153 due to (i) reduced long-distance transport of CTKs from the roots to the shoots; (ii) inhibited CTK biosynthesis
154 through decreased biosynthesis of related enzymes (isopentenylolation of adenosine phosphate by
155 isopentenyltransferases, IPT; cytochrome P450 mono-oxygenase, CYP735A; LONELY GUY, LOG); and (iii)
156 increased catabolism of CTK due to the higher activity of CTK metabolism-related enzymes (cytokinin
157 oxidase/dehydrogenase, CKX) and CTK glycosylation (Skalák et al., 2016; Tripathi et al., 2012; Vyroubalová
158 et al., 2009; Wu et al., 2017; Wu et al., 2016). Additional studies are necessary to understand how the
159 membrane transport of CTK is involved in the response to heat stress (Figure 4A).

160 The number of spikelets per panicle is mainly affected by heat stress during panicle initiation (Figure 3).

161 Changes in the content of CTK and CTK homeostasis in young panicles regulated the number of spikelets per
162 panicle in rice (Ha et al., 2012). Under heat stress conditions, heat-susceptible varieties showed significant
163 reductions in panicle CTKs that were accompanied by reduced spikelets per panicle in response to heat stress,
164 while a heat-tolerant variety had stable levels of panicle CTKs and a higher relative number of spikelets per
165 panicle in response to heat stress. Changes in panicle CTKs correlated with a decrease in spikelets per panicle
166 (Wu et al., 2016). We determined that enhanced transport of root-derived CTKs and stable CKX activity in
167 panicles are the two key processes that play a vital role in maintaining panicle size in rice varieties (Wu et al.,
168 2017). In Arabidopsis, reduced degradation, stable local biosynthesis, and/or enhanced transportation of CTK
169 from the roots likely contributed to the stability of CTK in target organs and facilitated adaptations to abiotic
170 stress (Ha et al., 2012). Genetic manipulation of CTK transport and degradation will be useful to stabilize
171 panicle size in rice under heat stress.

172 **Phytohormone changes affect spikelet fertility under heat stress**

173 Spikelet fertility is affected by heat stress during panicle initiation and flowering (Figure 3). In rice,
174 reduced levels of IAA in young panicles were closely related to decreased spikelet fertility, anther dehiscence
175 and pollen vigour (Wu et al., 2016). In barley and Arabidopsis, lower levels of IAA in anthers were attributed
176 to reduced expression of the IAA biosynthesis gene *YUCCA*, leading to anther indehiscence and lower pollen
177 vigour that could be rescued by the application of exogenous IAA (Sakata et al., 2010). However, in cotton
178 plants, high levels of background IAA weakened the defence response of the anthers to heat stress (Min et al.,
179 2014). It is speculated that excessively low or high levels of IAA due to disturbances in IAA homeostasis
180 during the response to heat stress are deleterious to anther dehiscence. Recently, it was found that heat stress
181 during panicle initiation reduced spikelet fertility by inducing panicle enclosure, which can be mainly
182 attributed to deficiencies in GA₁ and IAA (Wu et al., 2016). IAA regulates panicle expansion by regulating
183 GA₁ biosynthesis (Yin et al., 2007); we speculate that panicle enclosure in heat-stressed plants may be
184 attributed to reduced GA₁ biosynthesis due to the changes in IAA induced by heat stress.

185 Heat stress suppressed the biosynthesis of IAA by downregulating genes involved in IAA biosynthesis,
186 including tryptophan aminotransferase-encoding gene (*TAA1/TARs*), YUC flavin monooxygenases (*YUC*),
187 and UDP-glucosyltransferase 74b1 (*UGT74B1*) (Guo et al., 2016; Sharma et al., 2018), and reduced IAA by
188 upregulating the IAA-amino acid synthetase gene (*GH3*) in rice (Du et al., 2012), which positively regulates
189 the formation of IAA-amino acid conjugates (Mittag et al., 2015). However, there is limited information on the
190 response of genes involved in the degradation of IAA-amino acid conjugates (*ILRI*, *IAR3*) and the
191 transportation of IAA (*AUX1*, *LAX3*, *PIN1*) during heat stress in rice. In peas, monitoring an isotope labelled
192 ¹⁴C-IAA in the root tissues revealed that heat stress reduced IAA transport (Gladish et al., 2000), but the
193 sensitivity of the diffusion component (the IAA-influx/efflux gene) to temperature and IAA concentration has
194 not been determined. In summary, heat stress suppressed the biosynthesis of IAA and promoted the formation

195 of IAA-amino acid conjugates, yet information on the effects of heat on the catabolism and transport of IAA in
196 rice plants is limited (Figure 4C).

197 Heat stress reduced the content of GAs in young panicles of rice varieties (Wu et al., 2016). The GA
198 biosynthetic pathway is catalysed by several enzymes. In Arabidopsis, the GA 20-oxidase genes (*GA20ox1*,
199 *GA20ox2*, *GA20ox3*) and GA3-oxidase genes (*GA3ox1*, *GA3ox2*) that regulate the late steps of the GA
200 biosynthetic pathway were suppressed by heat stress (Toh et al., 2008). However, the genes that regulate the
201 early steps of the GA biosynthetic pathway, including ent-copalyl diphosphate synthase (*OsCPS1*), ent-
202 kaurene synthase (*OsKSI*), ent-Kaurene oxidase (*OsKO2*), and ent-kaurenoic acid oxidase (*OsKAO*), have
203 seldom been investigated under heat stress conditions in plants. The responses of the catabolism-related genes
204 (*GA 2-oxidases*, *OsGA2oxs*) and transport-related genes (*OsNPF* and *OsSWEET*) to heat stress are also limited.
205 According to existing results, heat stress suppressed the biosynthesis and promoted the deactivation of GA, but
206 few studies to date have documented the effects of heat on catabolism and the transport of GA in rice plants
207 (Figure 4D).

208 The major cause of spikelet sterility in rice was heat stress-mediated decline in the endogenous IAA level
209 in anthers, which was associated with the expression levels of IAA biosynthesis (*YUC*) and transport genes
210 (*AUX*, *PIN*) in rice under high temperature (Sharma et al., 2018). In barley and Arabidopsis, the *YUC* gene
211 affected spikelet fertility by regulating male sterility under heat stress (Sakata et al., 2010). GA is also a key
212 phytohormone for spikelet fertility, but the molecular genetic and biochemical mechanisms of GA in male
213 fertility remain largely unknown (Kwon & Paek, 2016). In thermo-sensitive genic male sterile lines, heat stress
214 suppressed the expression of the *EUI* gene, which induced the deactivation of GA (Xiao et al., 2005). Further
215 studies on the role of GA homeostasis in spikelet fertility under heat stress are needed.

216 **Phytohormone changes affect grain weight under heat stress**

217 Grain weight is impaired by heat stress during panicle initiation and grain filling (Figure 3). Formation of
218 grain weight is regulated by phytohormones under heat stress. (i) From a physiological perspective, grain
219 weight is determined by multiplying the endosperm number by the endosperm weight, which is regulated by
220 phytohormones such as CTK, ABA, and IAA (Javid et al., 2011; Nayar et al., 2013). Changes in endosperm
221 CTK, IAA, and ABA in response to heat stress lead to changes in grain weight in rice varieties (Cao et al.,
222 2016). In wheat, changes in grain weight are accompanied by changes in grain CTK content under heat stress,
223 and exogenous CTK is able to increase the final grain weight under heat conditions (Banowitz et al., 1999). (ii)
224 From an agronomic perspective, grain weight is determined by multiplying the grain filling rate by the
225 duration of grain filling, and these metrics are regulated by phytohormones during heat stress. ① Grain filling
226 rate. Changes in the grain filling rate are closely associated with the response of phytohormones such as ABA
227 and GA (Liu et al., 2007). ② Duration of grain filling. Early panicle senescence in heat stress-stressed plants is
228 the main cause of shorter periods of grain filling (Kim et al., 2011). The stay-green trait mitigated the negative

229 impacts of heat stress on grains due to an extended period of grain filling in rice (Kobata et al., 2015).
230 Enhanced CTK synthesis or increased CTK transportation from the roots to the shoots via sap in the xylem
231 delayed plant senescence (Singh et al., 1992) and may enhance heat tolerance.

232 ABA, which acts as an important mobile signal, is synthesized in the roots and transported to target
233 organs via the xylem and phloem (Kuromori et al., 2010). Heat stress had no overt effects on the ABA
234 concentration in the roots but increased the amount of continuous xylem sap flow for at least two weeks in rice
235 plants (Wu et al., 2017). Our previous data indicated that changes in ABA transportation in rice were closely
236 correlated with the effects of heat on the panicle ABA content ($r=0.66$, $P<0.05$, $n=12$). In aerial organs, heat
237 exposure increased the ABA content by upregulating ABA biosynthesis-related genes, including the
238 zeaxanthin epoxidase genes (*ABA1*, *ABA2*, *ABA4*, *AAO3*) and the 9-cis-epoxycarotenoid dioxygenase genes
239 (*NCED2*, *NCED5*, *NCED9*), but downregulated the ABA 8'-hydroxylase gene *CYP707A* (Toh et al., 2008),
240 the glucosyltransferase gene *UGT75B1*, and the glucosidase gene *AtBGI* (Dobrá et al., 2015). **Figure 4B**
241 illustrates the processes involved in ABA homeostasis that respond to heat stress. However, little is known
242 about the key processes that determine the ABA content in target organs under heat stress. ABA transport and
243 the processes involved in ABA metabolism should be studied in parallel to understand the response of local
244 ABA contents to heat stress.

245 Perspectives

246 Heat events will occur more seriously and frequently as global warming continues, and rice plants may be
247 affected by heat events at any growth stage. Although many heat-tolerant rice varieties have been identified,
248 most of these varieties endure heat injury only at specific reproductive stages. For example, the heat-tolerant
249 cv. Nagina22 exhibits relatively high grain yield under heat stress at flowering but is highly susceptible to heat
250 during the panicle initiation stage (Wu et al., 2016). Critical traits, such as high anther dehiscence (Zhang et al.,
251 2014), low stigma exertion (Wu et al., 2018), early morning flowering (Jagadish et al., 2007) are useful for
252 heat tolerance at flowering, but useless to heat stress during panicle initiation or grain filling. Further studies
253 should focus on the effect of heat stress on rice plants and the underlying mechanisms during all the
254 reproductive stages. To cope with unpredictable heat events in the future, we should screen and breed rice
255 varieties with comprehensive tolerance to heat stress throughout the entire reproductive phase.

256 The physiological and molecular mechanisms of heat effects on rice grain yield have drawn much
257 attention. The existing studies explore the phytohormonal mechanisms of the effects of heat stress on rice grain
258 yield by analysing phytohormone levels in rice. Most of previous studies focus on responses of biosynthesis
259 and catabolism of phytohormones to heat stress in rice, but studies on responses of phytohormone to heat stress
260 are relatively fewer. Full understanding of the phytohormone homeostatic response will help to reveal the key
261 processes that determine phytohormone levels and final grain yield in rice under heat stress conditions. Thus,

262 future work will provide a theoretical basis for the establishment of technology for breeding heat-tolerant
263 varieties and mitigating heat injury in rice.

264 Conclusions

265 Rice is susceptible to heat stress during the reproductive phase, e.g., panicle initiation, flowering, and
266 grain filling stage. The current study identified different responses of yield traits of rice to heat stress during
267 different reproductive stages: (i) the number of spikelets per panicle is reduced by heat stress during panicle
268 initiation stage but is not affected by heat stress during flowering and grain filling stage; (ii) spikelet sterility
269 induced by heat stress can be attributed primarily to physiological abnormalities in the reproductive organs
270 during flowering stage but attributed to structural and morphological abnormalities in reproductive organs
271 during panicle initiation stage; and (iii) the lower grain weight caused by heat stress during panicle initiation
272 stage was due to a reduction in non-structural carbohydrates, undeveloped vascular bundles, and a reduction in
273 grain length and width, while a shortened grain filling duration, reduced grain filling rate, and decreased grain
274 width affect grain weight when heat stress occurs during grain filling stage.

275 Responds of processes involving phytohormone homeostasis (biosynthesis, catabolism, deactivation, and
276 transport) play vital roles in regulating plant adaptations to heat stress. Currently, biosynthesis and transport
277 are supposed to be the key processes that determine phytohormone levels and final grain yield in rice under heat
278 stress. Prospectively, screening and breeding of rice varieties with comprehensive tolerance to heat stress
279 during panicle initiation, flowering, and grain filling could be feasible to cope with unpredictable heat events
280 in the future. In aspect of physiological mechanisms, studies in phytohormone homeostatic response are
281 needed to further reveal the key processes that determine phytohormone levels under heat condition.

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415 **Acknowledgments**

416 This work was supported by the National Key R&D Program of China (Grants No 2017YFD0300100), the
417 Natural Science Foundation of Jiangsu Province (Grants No BK20180537), the China Postdoctoral Science
418 Foundation funded project (Grants No 2017M621757), and Jiangsu Province Postdoctoral Science Foundation
419 funded project (Grants No 82171525).

420 **Additional Information**

421 Competing financial interests

422 The authors declare no competing financial interests.

423

424 Figure legends

425 **Figure 1. Illustrations of the panicle enclosure induced by heat stress, (Photo Credit: Chao Wu).**

426

427 **Figure 2. The effects of heat stress on anther dehiscence and pollen release, (Photo Credit: Chao Wu).** A:
428 the dehisced anthers with no adhered pollen under heat stress; B: the dehisced anthers with adhered pollens
429 inside the anthers under heat stress. Red arrow indicates residual pollen grains in anthers, and blue arrow
430 indicates aperture of the thecae.

431

432 **Figure 3. Illustration of the effects of heat on yield components during the reproductive phase in rice.** R0:
433 panicle development has initiated; R1: panicle branches have formed; R2: flag leaf collar formation; R3:
434 panicle exertion from boot; R4: one or more florets on the main stem panicle has reached anthesis; R5: at least
435 one caryopsis on the main stem panicle is elongation to the end of the hull; R6: at least one caryopsis on the
436 main stem panicle has elongated to the end of the hull; R7: at least one grain on the main stem panicle has a
437 yellow hull; R8: at least one grain on the main stem panicle has a brown hull; and R9: all grains that reached
438 R6 have brown hulls. Illustration of reproductive stages with morphological makers were adapted from

439 (Counce et al., 2000)

440

441 **Figure 4. Response of processes involving homeostasis cytokinin, indoleacetic acid, abscisic acid, and**
442 **gibberellin.** +, - , O , and ? indicate an increase, a decrease, and steadiness, and undefined response,
443 respectively, in a certain trait or process under heat conditions.

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445

446

Figure 1

Figure 1. Illustrations of the panicle enclosure induced by heat stress, (Photo Credit: Chao Wu).

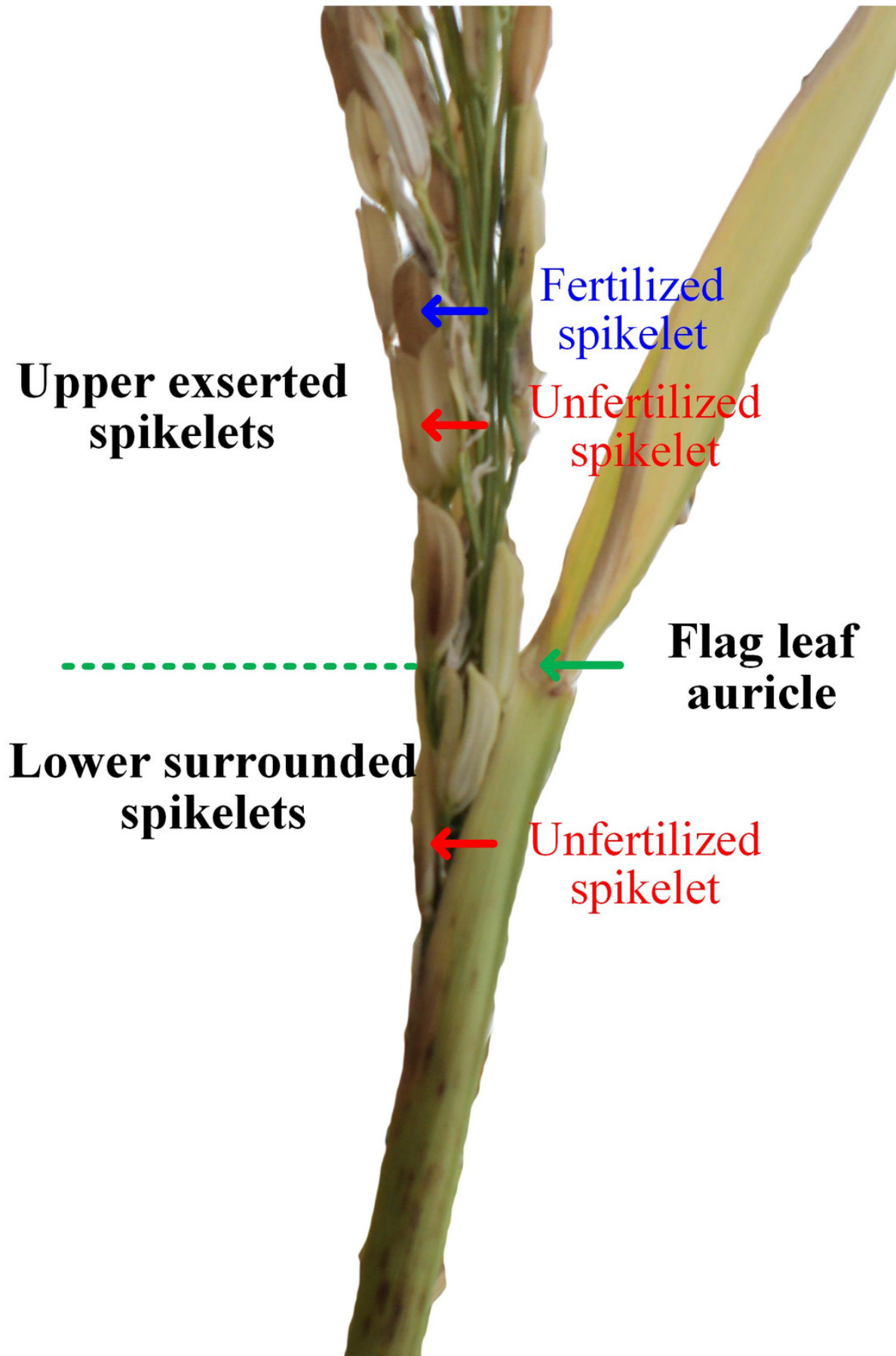


Figure 2

Figure 2. The effects of heat stress on anther dehiscence and pollen release, (Photo Credit: Chao Wu).

A: the dehisced anthers with no adhered pollen under heat stress; B: the dehisced anthers with adhered pollens inside the anthers under heat stress. Red arrow indicates residual pollen grains in anthers, and blue arrow indicates aperture of the thecae.

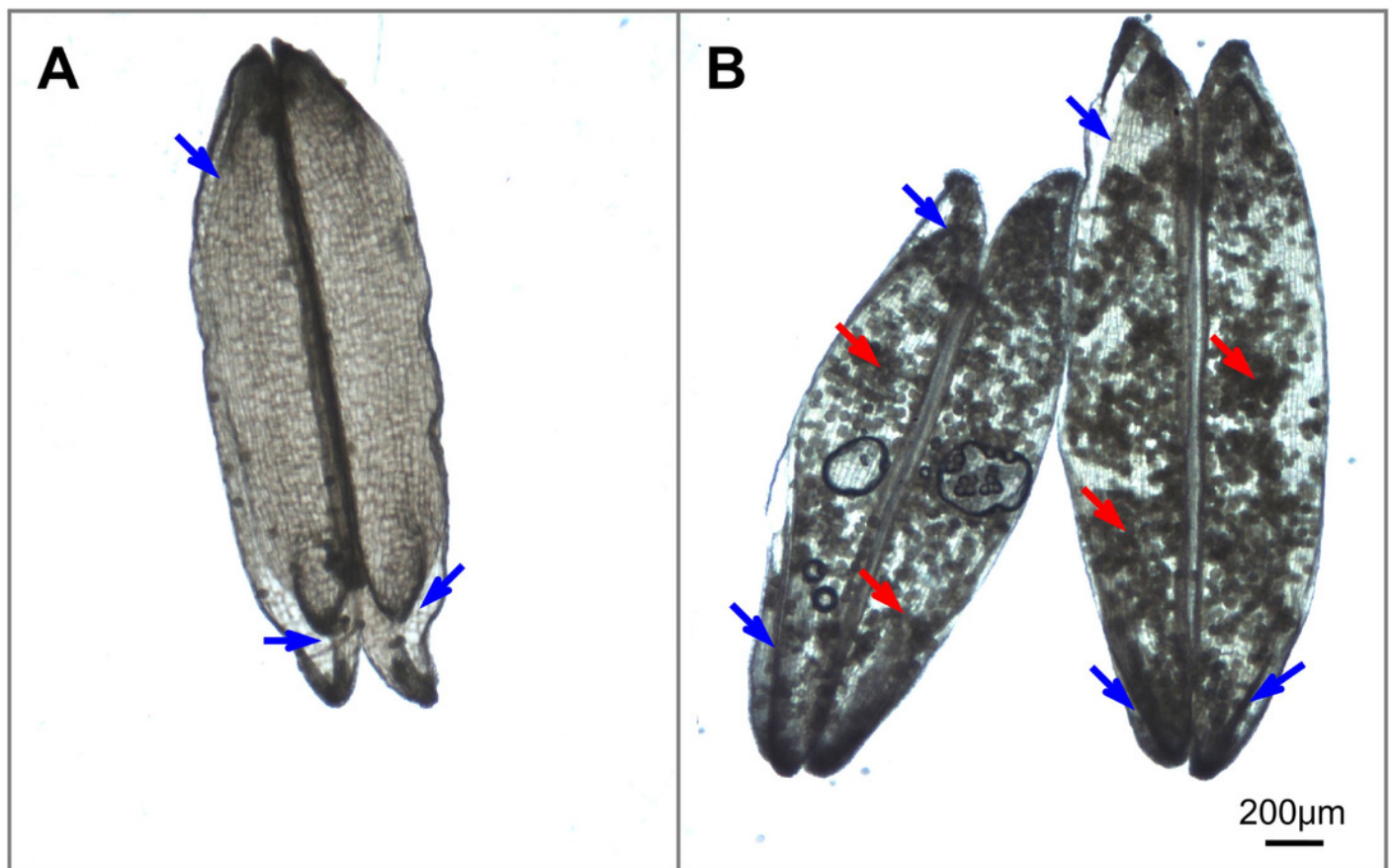


Figure 3

Illustration of the effects of heat on yield components during the reproductive phase in rice.

R0: panicle development has initiated; R1: panicle branches have formed; R2: flag leaf collar formation; R3: panicle exertion from boot; R4: one or more florets on the main stem panicle has reached anthesis; R5: at least one caryopsis on the main stem panicle is elongation to the end of the hull; R6: at least one caryopsis on the main stem panicle has elongated to the end of the hull; R7: at least one grain on the main stem panicle has a yellow hull; R8: at least one grain on the main stem panicle has a brown hull; and R9: all grains that reached R6 have brown hulls. Illustration of reproductive stages with morphological markers were adapted from (Counce et al., 2000)

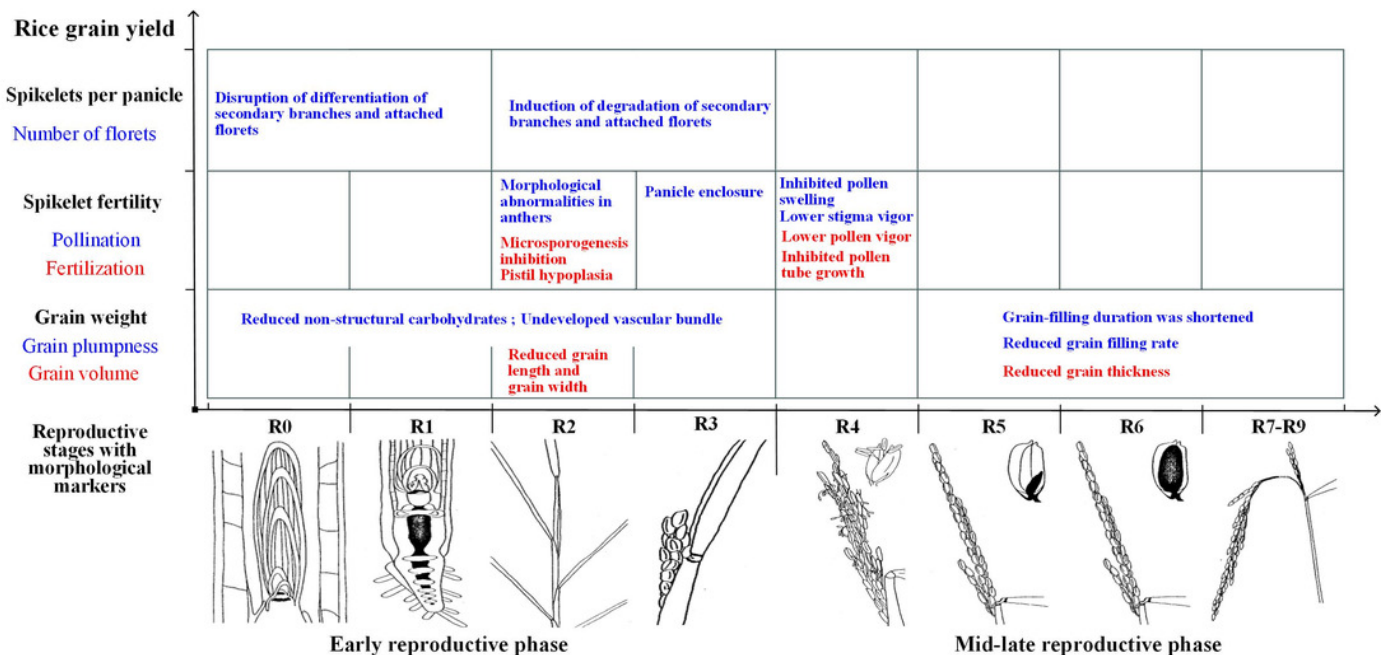


Figure 4

Response of processes involving homeostasis cytokinin, indoleacetic acid, abscisic acid, and gibberellin.

+, -, 0, and ? indicate an increase, a decrease, and steadiness, and undefined response, respectively, in a certain trait or process under heat conditions.

