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### [Short Report]

# Does Regional Temperature Difference before the Panicle Initiation Affect the Tolerance for Low Temperature-Induced Sterility in Rice?

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Key words : Cool weather damage, Oryza sativa L., Rice, Spikelet sterility, Tolerance, Water temperature, Yield.

Low temperature-induced sterility has been a major determinant for rice production in cool climates (Hayase et al., 1969; Satake, 1976; Shimono et al., 2007a; 2007b; 2007c). In the northern parts of Japan, one of the coolest rice growing areas in the world, yield loss caused by cool spell has been frequently observed at five-year intervals (Kanno, 2004). In 1993 of a severe cool year, for example, rice yield was seriously decreased to a half that of normal year in Tohoku region, mainly due to sterility.

Sensitive organ for sterility induced by low temperature is known to be the developing anther and pollen (Hayase et al., 1969; Satake, 1976). The sensitive stage is the reproductive growth period from the panicle initiation to flowering; the sensitivity is very high at the booting stage (young microspore stage) and becomes lower at the stage away from the booting stage (Hayase et al., 1969; Satake, 1976). A low temperature before the panicle initiation alone does not induce sterility. Based on this understanding, previous studies have analyzed variations in sterility observed in cool summer years only from the temperatures after the panicle initiation until flowering, but they could not fully explain the variations in sterility (Uchijima, 1976; Shimono et al., 2005; Shimono et al., 2007a; 2007b; Kanda et al., 2007). Recently, Shimono et al. (2007c) found that a low temperature especially low irrigation water temperature before the panicle initiation weakened the tolerance for sterility from a 2-yr pot study. Their finding suggests that temperature before the panicle initiation can affect variations in sterility in cool summer years through influencing the tolerance. To test this hypothesis, we grew rice under two different field conditions before the panicle initiation and evaluated their tolerance for the sterility.

#### **Materials and Methods**

Rice seedlings of cultivar Sasanishiki were transplanted to 1/5000 wagner pots which contained identical paddy soil and identical amount of fertilizer  $(N=0.6 \text{ g}, P_2O_5=1.2 \text{ g}, K_2O=0.8 \text{ g per pot})$  with one plant per pot. The pots were buried into paddies (with ca. 20 cm depth, to the equivalent level of field soil surface) of two contrast weather locations either of Morioka, Iwate (39°71'N, 141°14'E) (18 May in 2007) or Hachinohe, Aomori (40°58'N, 141°45'E) (21 May in 2007), respectively. Planting density was 22.2 plants  $m^{-2}$  (15 cm  $\times$  30 cm). Management practices including water management were in accordance with the local practices. Thus the rice plants were exposed to the field environment at respective locations. Mean air and water temperatures from transplanting to panicle initiation in paddies were ca. 2°C lower at Hachinohe (air temperature, 17.1±3.1°C and water temperature, 21.0±2.1°C mean±standard deviation) than at Morioka (air temperature,19.0±3.2°C and water temperature, 23.2±3.2°C). Solar radiation was almost identical in the two locations (20.9±8.2 MJ  $m^{-2} d^{-1}$  at Morioka, 19.7±8.8 MJ  $m^{-2} d^{-1}$  at Hachinohe). Reflecting temperature differences, the date of panicle initiation was 10 days later in Hachinohe (12 July) than in Morioka (2 July). The panicle initiation stage was estimated from leaf number on the main stem as 13.0 because previous experiment showed that the leaf number on the main stem at the panicle initiation of cultivar Sasanishiki was constant over years and temperature conditions (Shimono et al., 2007c). At the panicle initiation stage, leaf number on the main stem, leaf number index (a proportion of leaf number to final number) and plant height at the estimated panicle initiation did not significantly differ between locations (Table 1), but tiller number was significantly smaller at Hachinohe than at Morioka by 40%. Each

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Characters	Morioka	Hachinohe			
Days to heading (days)	$88.3\pm0.8$	95.7±0.9 ***			
Duration of heading (days)	$6.8\pm0.5$	$5.0 \pm 0.5$ *			
Panicle initiation					
Leaf number on the main stem	$13.0 \pm 0.2$	$13.3 \pm 0.1$ ns			
Leaf number index	$82.0 \pm 0.7$	$82.0 \pm 0.4$ ns			
Plant height (cm)	$50.0\pm0.9$	$52.0 \pm 1.0$ ns			
Tiller number	$62.2 \pm 3.3$	37.2±3.6 ***			
Heading					
Final leaf number on the main stem	$15.8 \pm 0.2$	$16.2 \pm 0.2$ ns			
Maturity					
Panicle number per plant	$40.0\pm0.9$	25.0±1.3 ***			
Spikelet number per panicle	$76.9 \pm 1.7$	86.0±2.2 **			
Spikelet number per plant	$3072\pm41$	2143±95 ***			

Table 1. Growth characteristics of rice plants grown under different locations in 2007.

Average ± SE (n=6). Duration of heading was defined as the period from 20% to 80% of whole reproductive tiller emerged. \*\*\* P<0.001, \*\* P<0.01, \* P<0.05, ns, not significant.

location had six pots.

After the day of panicle initiation, the pots were exposed to cool water treatment for inducing sterility uniformly (19.5°C, 35 cm in depth) using a water bath (1 m×1 m and 60 cm deep) in a greenhouse at Morioka. Cool water treatment system was described in detail in previous study (Shimono et al., 2007c). Briefly, water temperature was controlled at 19.5°C by supplying cool water at ca. 18°C through an electromagnetically controlled valve. A datalogger was used to record data from the thermometer and operate the valve at 2-s intervals to maintain a constant temperature and to record the average water temperature every 30 min. Water was circulated by a pump to minimize spatial variation in water temperature. A carefully calibrated Pt100 thermometer (R902-31, Chino Co., Tokyo, Japan) was used to measure water temperature. The treatments were maintained until the early grain filling stage (11 or 22 days after heading stage, at 5 Sept) to expose all panicles to the identical water temperature. Air temperature in a greenhouse was 25.9°C averaged in August when is the time panicles emerged above water level, indicating that cool water only was a source for inducing sterility. Day to heading date was significantly delayed in Hachinohe than in Morioka by 7 days (Table 1). Heading date was defined as the date of 50% of whole reproductive tiller emerged, which was determined at 2-day intervals.

At maturity, panicle number, spikelet number and spikelet sterility were measured. Sterile spikelets were carefully identified by backlighting the heads with fluorescent lightbulbs; spikelets that showed no shadowy area (i.e., no developing embryo or grain) were considered to be sterile. The percentage of sterile



Fig. 1. Low temperature-induced sterility based on whole plant (sterile spikelet number/total spikelet number,  $STR_{plant}$ ) in rice grown at different locations before the panicle initiation and exposed to cool water treatment (19.5°C, 35 cm in depth) after the panicle initiation in 2007. Bars indicate standard errors (n=6).

spikelets as the spikelet sterility was calculated based on the whole plant pooled (total sterile spikelets / total spikelets per plant) and based on the individual panicle (individual sterile spikelets/individual spikelets per panicle). Statistic analysis was conducted as individual plant as a replicate by t-test (n=6). Regressions were compared on residual variations between fitting a single line and individual lines (Mead et al., 2003).



Fig. 2. Relationship between low temperature-induced sterility based on each panicle (STR<sub>panicle</sub>) and spikelet number per panicle in rice in 2007. n=238 (Morioka) and 150 (Hachinohe). Solid line and broken line show results of regression analysis for Hachinohe and Morioka, respectively. \*\*\* P<0.001.

#### **Results and Discussion**

At maturity, panicle number per plant was significantly lower at Hachinohe than at Morioka by 37% while individual panicle size (=the spikelet number per panicle) was slightly larger at Hachinohe than at Morioka by 11% (Table 1). Spikelet number per plant was significantly smaller at Hachinohe than at Morioka by 30%.

Spikelet sterility based on the whole plant was significantly higher in Hachinohe of 86% than in Morioka of 82% (Fig. 1). The difference was significant at the 10% level, but not at the 5% level (P=0.0504). To confirm this result, we calculated the sterility per individual panicle (Fig. 2). The sterility was negatively correlated with spikelet number per panicle, and the sterility at Hachinohe was higher than at Morioka compared at the same spikelet number as well as the sterility of plant basis. The relations for two locations were significantly independent ( $F_{2, 385}$ =20.9, P <0.001), confirming that the sterility was significantly different between locations.

Cool water treatment at each location in the present study started at the panicle initiation stage of the main stem, which is known to initiate young panicle faster than most of other tillers (Hoshikawa, 1975). Although we did not examine variations in panicle initiation stage between tillers, Table 1 shows that heading duration, which was defined as the period from 20% to 80% of whole reproductive tiller emerged, differed



Fig. 3. Relationship between low temperature-induced sterility based on the whole plant (STR<sub>plant</sub>) and water temperature during vegetative growth (from transplanting to panicle initiation). Data in 2004-2005 was obtained from the results of Shimono et al. (2007c) where the sterility of rice cultivar 'Sasanishiki' was induced by cool water during reproductive growth (19.5°C, 30 cm in depth, for about two months until the all panicles had emerged above water level) and fertilizer was supplied at N=0.6 g, P<sub>2</sub>O<sub>5</sub>=12 g, K<sub>2</sub>O=0.8 g per pot. Regression line was drawn for the data of 2004-2005 (y=-5.1732 x+182.14, R<sup>2</sup>=0.830, P<0.001). Bars indicate standard errors (n=6 (2007) or 4 (2004-2005)).

only 1.8 days between location even though tiller number at this stage differed by 40%. Additionally, plant sensitivity to low temperature for sterility is known to be substantially lower around the panicle initiation stage than at the booting stage (Hayase et al., 1969; Satake, 1976). The timing of start of cool treatment in the present study would not affect sterility differences between locations through difference in variation of panicle initiation stage in tillers.

Previously water temperature rather than air temperature before the panicle initiation was found to directly affect the tolerance (Shimono et al., 2007c), so spikelet sterility based on the whole plant was plotted against water temperature averaged from transplanting to panicle initiation stage in Fig. 3 in comparison with the previous results. The present results were similar to those obtained in the previous study in which a lower water temperature increased sterility though the sterility in the present study (over 80%) was higher than in the previous study (less than 75%) at identical water temperature. This would be due to the difference in depth of cool water for inducing sterility; the water was 5 cm deeper in the present study than in the previous study which could expose the developing panicles for a longer duration to cool water.

The variations in percentage of sterility are the greatest at around 50%. The higher sterility per se in the present study would be attributed to the lower responsiveness to temperature before the panicle initiation (1.6% per °C) than in the previous study of (5.2% per °C) (Fig. 3).

In conclusion, the present results support the hypothesis that the temperature before the panicle initiation affects the tolerance for sterility induced by low temperature during the reproductive growth; lower temperature before the panicle initiation at Hachinohe could weaken the tolerance compared with that at Morioka. This finding also suggests the potential to increase the tolerance by management of water temperature before the panicle initiation. Analysis of the regional difference in sterility in cool summer years is awaited.

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