Research on Cool Injury of Paddy Rice Plants in Japan

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Introduction

Japan's northern regions, particularly the Island of Hokkaido are chronically exposed to the threat of cool injury. The regions are characterized by cool weather and short period of rice cultivation. Unseasonable low temperature and little sunshine often predominate during July and August which are the most important period for paddy rice growth. Figure 1 shows average of paddy rice yield per 10 a in Hokkaido for the past 70 years. Sixteen out of 17 low yield years were due to low temperature in summer. Particularly, there were practically no harvests in 1902 and 1913. Such catastrophic incidents of cool injury to paddy rice plants resulted in a major social problem.

With the progress in agricultural technique, such a great injury is not induced today, but still the difference in yield between average year and cool-injured year reaches as high as 150 kg per 10 a. Accordingly, the development of a new technique controlling cool injury is the most important problem for rice cultivation in Hokkaido today as well as in the old days. Moreover, cool injury is often found in paddy rice fields of high mountainous cool districts and in early season cultivation of rice plants in southwestern warm regions. Cool injury is still an important nation-wide problem.

As stated above, cool injury of paddy rice plants is caused by low summer temperature, but the influence of low temperature on paddy rice growth differs according to growth period. Previous researches have revealed that the cool injury of rice plants is classified into 2 types: delay-type and sterile-type.

In delay-type low yield is caused by grain immaturation induced by low temperature in two different stages. One is low temperature during certain vegetative growth period from young seedling stage to panicle differentiation stage. This delays heading, and the ripening stage encounters low temperature period of fall. The other is low temperature during the ripening period, although the heading is not delayed.

In sterile-type low yield is caused by the outbreak of sterility by low temperature for a short period from young panicle differentiation stage to anthesis, particularly by low temperature at the booting stage.

In Japan, the research on cool injury has

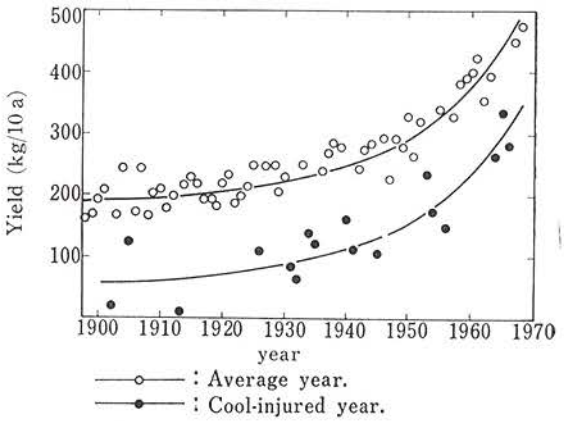

Fig. 1. Change in averages of rice yields per 10 a in Hokkaido due to technical improvement of the past 70 years.

been initiated in 1930s. Consequently, considerable reduction in injury has been achieved against delay-type cool injury by breeding of early ripening varieties and by the improvement of seedling raising method. For instance, the days in growth for early ripening varieties is more than 10 days shorter than traditional varieties. With the improvement of land bed nursery (raising young plants on dry well-drainable bed covered with vinyl film), seedlings can be transplanted more than 2 weeks earlier than in case of water bed nursery (raising young plants on submerged bed). These contribute in large measure to the prevention of growth delay. On the other hand, technical improvement against sterile-type is still insufficient and when unseasonable low temperature happens during booting stage, considerable damage can not be avoided even today. This paper covers only the problem of sterile-type and introduces the information on physico-ecological studies achieved so far on the outbreak of sterility by low temperature.

Analysis of sterility outbreak

1) Relation between growth period and sterility

It has been well-known for ages that in cool-injured year the rice yield decreases due to a great outbreak of sterility. However, up to the middle of 1930s it has not been proved about any growth stage when low temperature induces the outbreak of sterility. It has been generally assumed that the sterility resulted from the low temperature at anthesis. Therefore, the earlier researches were mostly directed to low temperature at anthesis. In 1938 it has been verified for the first time that the sterility was induced by low temperature prior to anthesis, that is, at the meiotic stage of pollen mother cells (10 to 11 days before heading)[6]. Thereafter this fact has been confirmed by many researchers[3), 7)]. These researches revealed that low temperature induces much smaller sterility at spikelet differentiation stage (around 24 days before heading) and at anthesis than at the meiotic stage of pollen mother cells.

Fig. 2 is an example of experimental results to show the relation between cooling time and sterile injury[3]. The critical period in which sterility is greatly induced by low temperature can be determined as a very short period centering around the meiotic stage of pollen mother cells (11 days prior to the heading). When this critical period is avoided, the sterility does not practically outbreak even under extreme cooling treatment such as 12°C for 6 days.

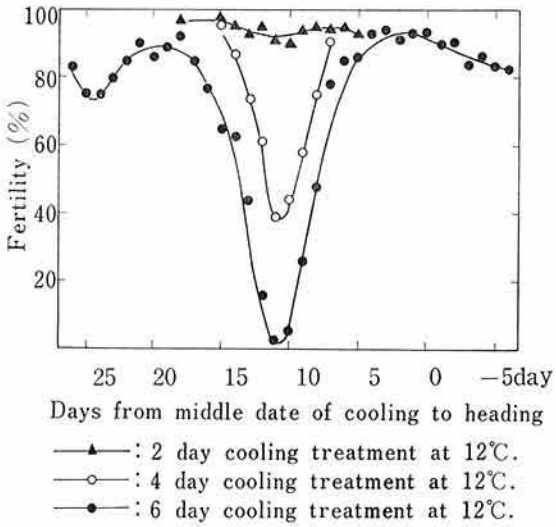

Fig. 2. Relation between growth stage and sterility outbreak.

2) Relation between the degree of low temperature and sterility

The critical temperature of inducing sterility at the meiotic stage differs with varieties and according to low temperature hours and to growth conditions, etc.

Difference in sterility among varieties: Varietal differences have been considerably recognized in sterility by low temperature at the meiotic stage. Table 1 shows a distinct difference in sterility between a very strong variety "Hayayuki" and a weak variety "Norin 20" in cool hardiness even under 12°C as well as under 15°C treatment.

The critical temperature of inducing sterili-

ty differs according to the growth condition and is comfirmed to be 15~17°C in "Hayayuki" and 17~19°C in "Norin 20" by water temperature treatment[10]. Young ears at the meiotic stage were submerged in different temperature waters.

According to yield decrease ratio and daily minimum temperatures during 5 days of the meiotic stage in the cool-injured year of 1957, the critical temperature of inducing sterility is estimated to be 16°C in a strong variety and 18°C in a weak variety[1].

From these examples it may be assumed that the critical temperature of inducing sterility is around 15~17°C for the strong varieties and 17~19°C for the weak ones.

Table 1. Varietal differences in sterility outbreak due to two low temperature treatments at the meiotic stage

Temperature \ Treated days \ Variety		Sterility (%)	
		Hayayuki	Norin 20
12°C	1	2	1
	2	3	24
	3	13	61
	4	47	96
15°C	1	2	3
	2	2	6
	3	2	41
	4	5	51
Control		2	2

Remark: Hayayuki...Very strong variety in cool hardiness.
Norin 20...Weak variety in cool hardiness.

Difference in sterility between constant and diurnal change cooling treatment: All the cooling treatments stated above have been carried out under fixed temperatures through day and night, but in the fields temperature changes diurnally. Optimum temperatures of day and night exist for each and night exist for each growth period of rice plants[8]. In order to analyze influence of temperature against sterility outbreak, it is desirable to make studies under temperature conditions similar to natural daily variation. According to the recent researches in growth cabinets, even if the average temperatures are the same, a distinct difference in sterility outbreak can be produced between the constant cooling and diurnal change cooling treatments[2]. Table 2 shows

Table 2. Sterility outbreak between constant and diurnal change cooling treatments (Var. Towada)

Treated temperature			Sterility (%)		
Average temperature	Deviation	range	4 days	7 days	10 days
	°C				
17.5°C	0	17.5	4	9	38
	5	15.0-20.0	5	11	34
	10	12.5-22.5	4	3	4
15.0°C	0	15.0	4	31	64
	5	12.5-17.5	7	28	53
	10	10.0-20.0	9	27	63

the comparison of sterility between constant and diurnal change cooling treatments when the average cooling temeprature at the meiotic stage was 17.5°C and 15.0°C. The 5°C and 10°C temperature change was made according to the sine curve programs and considerably close to natural conditions.

When the average temperature was 17.5°C, sterility was remarkably smaller in 10°C diurnal change treatment, but it was almost similar in 5°C diurnal change as compared to constant treatment. When the average cooling temperature was 15°C, there were no significant differences in sterility among 5°C and 10°C diurnal change and constant cooling treatments. From this result it may be reseanably assumed that sterility outbreak is small if the day temperature is high enough even when the night temperature is considerably low. In most earlier researches the interest was usually focused on the lowest temperature

during the night. Therefore, it is necessary to make further studies by directing the attention to low day temperature more than to low night temperature.

Difference in sterility according to growth conditions: As stated above, sterility differs with varieties and according to degree of low temperature. Moreover, it differs according to growth conditions even when the same variety is cooled under the same temperature. Table 3 shows the sterility in 17°C cooling treatment for 6.5 days during 3 year period[7]. Sterility in the same cooling treatment differs by more than twice from year to year. The cause of this cool injury difference may be assumed to result from different physiological conditions of rice plants due to different year climatic conditions. It has also been shown that abundant nitrogen supply makes rice plants more susceptible to cool injury even in the same year. Based on such a difference in growth conditions, nothing has yet been revealed as for physiological characteristics of cool injury resistance. If further research progress is made in this field, it may be expected to find some clue of preventing cool injury.

Table 3. Yearly by difference in sterility outbreak by cooling treatments at the meiotic stage*

Variety	Sterility		
	1938	1939	1940
	%	%	%
Rikuu 132	41.6	34.4	74.2
Oou 2	54.9	30.2	72.1
Shin "I"	67.9	36.8	84.0

* Cooling treatment in each year was made at 17°C for 6.5 days

Mechanism of sterility outbreak

1) Male abnormalities at anthesis

When pollinated with sound pollens, spikelets at anthesis after cooling treatment at the meiotic stage showed to have high fertility[3]. This indicates that pollens are injured by low temperature at the meiotic stage and that the fertilizing ability of pistils is sound. In rice plants cooled at the meiotic stage, male abnormalities at anthesis were observed[5]. Abnormalities were found at each different stage from anther development to fertilization. The main cause of sterility among them was considered to be partial or no dehiscence of anthers, which was resulted from pollen unripeness to a considerable degree. To clarify the mechanism of sterility outbreak it is necessary to investigate the course under which pollens delay to ripen after cooling treatment at the meiotic stage.

2) Cyto-histological abnormalities of anthers

Investigations on cool-injured field rice plants in 1941 showed many abnormalities in reproductive organs[13]. Major abnormalities among them were abnormal differentiation and degeneration of pollen and embryosac mother cells, the unpairing of chromosomes in meiosis, the inhibition of cellular wall formation and abnormal development of tapetal cells. Among them the proliferation of tapetal cells strikingly attracted attention and has been investigated. It has been assumed that sterility is mainly caused by the outbreak of abnormal proliferation in tapetal cells[14]. The reasons were that the abnormal proliferation increased along with the decline in temperature and that there was an reverse correlation between it and cool injury resistance of varieties. However, when compared with sterility, outbreak-frequency of proliferation of tapetal cells is remarkably low. From this fact it may be unreasonable to conclude that proliferation of tapetal cells is the main cause for sterility outbreak.

The tapetum is a layer tissue surrounding the outside of pollen mother cells or microspores and is surrounded by two layers of transitory tissues. Tapetum and transitory tissues play an important rôle in the process of pollen development by supplying necessary nutrition. They function through pollen mother cell division and microspore stage till their respective degeneration[9]. At the first to second contraction stages two interesting facts

were observed in cool-injured anthers. One is that abnormal tapetal cells proliferated greatly[14]. The other is that peroxidase activity and reducing substances much increased[9]. Although the relation between the abnormality of tapetum and pollen unripeness has to be verified by future researches, it can be assumed from the rôle of tapetum that a close relation exists between the two. According to the electron microscopy, the tapetum is characterized by well-developed endoplasmic reticulum (ER). In the proliferated tapetal cells these ERs were broken, while other cellular organelles such as mitochondria, plastids and Golgi bodies did not differ from those in control normal anthers[10].

3) Physiological abnormality of anthers

As for physiological researches on cool injury there are many works on an individual level such as on inhibition of nutrition uptake and translocation due to low temperature. But the anther which plays an important rôle in the outbreak of sterility is only a micro-organ which can not be analyzed by such macro-researches as an individual level. It is just recently that physiological studies of anthers were initiated to know the mechanism of sterility and there are only a little information available.

Cooling treatment at the meiotic stage decreased dry weight, protein content, free amino acids and respiratory activity of anthers from the second contraction stage (4~6 days after the meiotic stage) to heading stage where they declined to the degree practically comparable to low fertility[11]. Among the free amino acids proline declined most greatly[4]. This accords with the case of genetic-cytoplasmic male sterile plants where tapetal cells proliferate abnormaly at the microspore stage. Proline is recently thought to play a unique rôle in pollen metabolism. Therefore, pollen sterility in cool injury may be caused by similar mechanism to male sterile plants.

Technique of protection against sterility outbreak

As one practical technique to prevent sterility outbreak under low temperature at the booting stage, deep water irrigation method has been recommended[12), 15]. As irrigation water temperature is generally higher than atmospheric temperature, plant temperature is higher in portions submerged in water than above the water. Accordingly, when atmospheric temperature drops at the meiotic stage of pollen mother cells it is possible to protect the plants against sterility outbreak by submerging young ears in deep irrigation water.

The ears at the meiotic stage are mostly distributed 8~14 cm above the ground. Thus irrigation water should be kept up to the depth of over 15 cm to protect young ears. This deep water irrigation has been found to be effective to prevent sterility outbreak, but it is necessary to build a high ridge for keeping the depth of 15 cm water irrigation. This is only one method to prevent sterility in case of unseasonable low temperature. It is expected in future that some effective techniques of preventing sterility outbreak will be developed.

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