ISSN 1807-1929
Revista Brasileira de Engenharia Agrícola e Ambiental
Brazilian Journal of Agricultural and Environmental Engineering
v.26, n.12, p.947-952, 2022
Campina Grande, PB – http://www.agriambi.com.br – http://www.scielo.br/rbeaa

DOI: http://dx.doi.org/10.1590/1807-1929/agriambi.v26n12p947-952

Cold stress reduces rice grain yield in temperate conditions[1]

Estresse por baixas temperaturas reduz as produtividades do arroz em condições de clima temperado

Rasim Unan[2*], Temel Genctan[3] & Rafael M. Pedroso[4]

[1] Research developed at Trakya Agricultural Research Institute, Edirne, Turkey
[2] University of California/Department of Plant Sciences, Davis, California, USA
[3] Namik Kemal University/Department of Field Crops, Tekirdag, Turkey
[4] Oregon State University/Department of Horticulture, Corvallis, Oregon, USA

HIGHLIGHTS:
- *Cold stress causes grain yield losses of up to 38.6%.*
- *Cold stress is more damaging at the germination and seedling stages than at the booting stage in temperate regions.*
- *For economical grain yield, minimum of 60.8-79.6 plants m^{-2} and optimum of 132.3 plants m^{-2} are required.*

ABSTRACT: This study aimed to evaluate the cold stress effect in natural field conditions by employing different rice sowing timings over three years, to identify national varieties tolerant to cold stress at the vegetative and generative stages, and to propose a data-derived solution regarding termination and resowing due to cold stress conditions. Early, regular, and late sowing dates were employed to capture natural cold stress conditions in a 3-year-long experiment. Sowing dates resulting in the greatest yields fluctuated from regular to late sowing dates, according to the stress duration in cold stress years. Yield losses resulting from cold stress ranged from 0.810 to 2.740 t ha^{-1} and reached 38.6%. 'Halilbey,' 'Pasali,' and 'Mevlutbey' were found to be most cold-tolerant varieties. Grain yield was correlated with plant number; the critical minimum level was between 60.8 and 79.6 plants m^{-2} and the optimum was 132.3 plants m^{-2} for economical yield. Cold stress negatively affected rice plant density, and plant densities below the critical minimum plant warranted crop termination and resowing, depending on application costs. Cold stress had a far more devastating effect on germination and seedling stages than on later development stages in temperate conditions.

Key words: cold tolerance, farmer application, sowing date

RESUMO: O arroz é originário de climas tropicais e, quando cultivado em regiões temperadas, pode ter sua produtividade reduzida por exposição ao estresse por frio. Este estudo buscou avaliar as perdas produtivas associadas ao frio em diversas variedades de arroz e a tomada de decisão pela resemeadura da lavoura. A semeadura foi realizada em época precoce, regular ou tardia visando capturar a condição natural de ocorrência de estresse pelo frio em experimento com duração de três anos. As datas de semeadura que obtiveram os maiores rendimentos variaram entre data normal a tardia de acordo com a duração do estresse por frio nos anos com sua ocorrência. Perdas produtivas ocasionadas por condições de estresse por frio variaram entre 0.81 a 2.74 t ha^{-1}, atingindo até 38.6%; Halilbey, Pasali e Mevlutbey foram as variedades mais tolerantes ao frio. O rendimento de grãos correlacionou-se ao número de plantas; uma densidade mínimo 60.8-79.6, e ótimo 132.3 plantas m^{-2} foi necessária para se atingir o rendimento econômico. Estresse por frio afetou a densidade de plantas de arroz negativamente, e densidades ≤ 60.8-79.6 plantas m^{-2} indicariam necessidade de eliminação da cultura e sua resemeadura dependendo dos custos de aplicação. O estresse pelo frio ocasionou maiores perdas nos estádios de germinação e plântula em relação a sua ocorrência em estádios mais avançados de desenvolvimento.

Palavras-chave: tolerância ao frio, aplicação pelo agricultor, época de semeadura

• Ref. 264691 – Received 04 Jun, 2022
* Corresponding author - E-mail: rasimunan@hotmail.com
• Accepted 08 July, 2022 • Published 20 July, 2022
Editors: Ítalo Herbet Lucena Cavalcante & Hans Raj Gheyi

Introduction

Rice (*Oryza sativa* L.) is a major staple food and feeds almost 4 billion people daily. This crop originated in warm tropical climates; thus, when grown in temperate regions, rice can experience negative effects from cold stress. Even though reports in the literature suggest that the booting stage is rice's most sensitive period for cold stress occurrence (Xiao et al., 2018), the effects of early cold damage on rice yields are also pronounced, as determined in studies conducted in Turkey (Unan, 2016). One of the main reasons is the high probability of exposure to cold stress in May, which corresponds to rice's sowing period, but the chances for exposure to damaging cold conditions are lower in July-August, which is when the booting stage typically occurs.

The optimum temperature for rice farming ranges between 25 and 35°C (Biswas et al., 2019a). Seed germination is significantly impaired at temperatures below 10°C, and vital activities in the seedling stage are negatively affected at 13-18°C (Bodapati et al., 2005; Biswas et al., 2019b). Cold stress causes poor germination at the germination growth stage; the stress factor lead to seedling injury or poor stand establishment at the seedling growth stage (Sharma et al., 2021). It can also lead to pollen cell sterility, causing self-pollination failures in the booting growth stage in rice (Shakiba et al., 2017; Alemayehu et al., 2021).

To evaluate cold stress occurring throughout the year, sowing in different periods, such as early and late sowing dates, is a frequently used method. In addition, it is known that the reactions of varieties to cold stress differ (Cruz et al., 2010).

This study aimed to (i) evaluate the cold stress effect in natural field conditions by employing different rice sowing timings over three years to experience cold stress; (ii) to identify national varieties tolerant to cold stress at the vegetative and reproductive stages; (iii) and to propose a data-derived solution regarding termination and resowing due to cold stress conditions.

Material and Methods

In this study, 13 rice varieties with different properties were used as materials: 'Kiziltan,' 'Pasali,' 'Tosyagunesi,' 'Duragan,' 'Halilbey,' 'Edirne,' 'Osmancik-97,' 'Tunca,' 'Aromatik-1,' 'Hamzadere,' 'Mevlutbey,' 'IR50' (cold-sensitive check), and 'HSC55' (cold-tolerant check). The varieties are of japonica origin from Turkey, except for the check varieties. The varieties widely cultivated in the ecological zone were utilized as certified seeds with uniform characteristics, and the seeds had a moisture content of 14% at sowing. The experiment was carried out in the experimental field of the Thrace Agricultural Research Institute located in the center of Edirne province in 2013, 2014, and 2015. The soil type was clay and had a neutral pH (pH 7.25-7.53). The nutrient values were as follows: 5.0-16.8 g kg^{-1} organic C, P_2O_5 8.9-27.8 mg kg^{-1} (Olsen P), and 550.0-720.0 mg kg^{-1} extractable K. The soil had a sandy loam texture, with the following physical characteristics: sand 468-604 g kg^{-1}; silt 202-271 g kg^{-1}; clay 125-315 g kg^{-1}; $CaCO_3$ 9-10 g kg^{-1}; and electrical conductivity of saturation extract 0.10-0.18 dS m^{-1}

Climate data (i.e., minimum, average, and maximum temperature) were recorded at the meteorology regional directorate. Low temperatures are a major factor affecting the growth, development, and productivity of rice in its geographical distribution. Cold stress mediates a series of physiological and metabolite changes (Liu et al., 2018). The research location had a daily average temperature between 13.3 and 27.0°C, relative air humidity of 46.4-79.7%, and rainfall of 0-105.4 mm m^{-2} during the rice-growing period from May to October from 2013 to 2015 (Table 1). When the temperature data were analyzed, cold stress was not experienced in 2013. However, it was experienced in early and regular sowing periods in 2014 and in the early sowing period in 2015. Therefore, to better capture the effects of cold stress on the yields of 13 rice varieties, only experimental data from 2014 and 2015 (cold stress years) were evaluated.

The experiment had a randomized block design with three replicates. To better capture cold stress effects under natural conditions, three sowing dates were utilized: early (30 April), regular (20 May), and late (10 June) between 2013 and 2015. The plot sowing size was 4 × 5 = 20 m^2, and the harvest area corresponded to 15.75 m^2 upon removal of the border area. Typically, 500 seeds m^{-2} are sown. The seeds were soaked in water for 48 hours for pre-germination (Unan et al., 2022), and seeding was performed in wet soil conditions. Pre-germinated seeds were directly sown by hand in the flooded field. Maintenance, fertilization, and irrigation operations were carried out as standard rice production systems, such as with 0.18 t ha^{-1} N and continuous flooding (Surek, 2002). Harvest was done by hand, and the material was threshed with a plot threshing machine. The yield data were calculated and corrected for 14% grain moisture content.

The number of plants (crop density) was counted on a 0.25 m^2 area by employing a hoop with an inside diameter of 0.56 m, and presented on a per square meter basis. Empty (blank) and filled spikelets were separated and counted to determine spikelet sterility percentages, which were calculated by dividing the number of sterile spikelets by the total spikelets and expressed as percentages (IRRI, 2002).

The experiments were conducted over three years. Data from the second and third experimental years were evaluated as cold stress-inducing years, since cold stress was not experienced in

Table 1. Minimum, maximum, and average temperatures in 2013, 2014, 2015, and long term

Month	Mean minimum temperature (°C)				Mean maximum temperature (°C)				Average monthly temperature (°C)			
	2013	2014	2015	Long term	2013	2014	2015	Long term	2013	2014	2015	Long term
April	8.9	9.0	6.9	7.2	21.8	20.1	19.3	19.3	15.3	14.1	12.7	12.9
May	14.1	12.5	14.4	11.6	29.2	25.0	28.0	24.8	21.6	18.6	20.7	18.2
June	16.3	16.4	16.2	15.4	30.4	28.7	28.9	29.2	23.2	22.3	22.6	22.4
July	17.9	18.3	18.2	17.3	33.0	31.9	34.5	31.7	25.9	25.3	27.1	24.7
August	19.0	18.7	19.2	17.1	34.2	32.8	34.4	31.6	27.0	25.6	26.5	24.3
September	13.9	14.5	16.3	13.3	28.7	26.5	29.8	27.2	21.2	19.6	22.4	19.8
October	7.1	9.3	10.5	9.1	20.7	19.8	19.6	20.5	13.3	15.4	14.3	14.2

the first year, which allowed for the highest yields to be obtained in the early sowing date treatment.

Data were analyzed using analysis of variance (SAS Institute, 1992), and the means of treatment were compared based on the least significant difference test (LSD) at a 0.05 probability level.

Results and Discussion

Climate data analysis indicated that cold stress conditions did not occur in the first year (2013); therefore, only data collected in the second and third experimental years (2014 and 2015, respectively) were evaluated to uncover the impacts of cold stress conditions on rice growth and development. Furthermore, the early sowing date in 2013, the late sowing date in 2014, and the regular sowing date in 2015 had the highest grain yields.

In 2014, the average daily temperatures of early, regular, and late sowing dates were 22.5, 22.8, 23.8°C, and rice grain yields were 4.216, 4.471, and 5.285 t ha^{-1}, respectively. The increase in grain yields was accompanied by an increase in temperature. Early sowing dates led to yields that were 5.7 and 20.2% lower than those recorded for regular and late rice sowing, respectively. When the early sowing date was preferred, it had 0.255 and 1.069 t ha^{-1} lower grain yields compared to the regular and late sowing dates, respectively (Table 2).

Rice yield equaled 4.346, 7.083, and 5.985 t ha^{-1} grain when rice was seeded on early, regular, and late sowing dates, respectively, in 2015. The early sowing date yield, which was the lowest, was 38.6 and 27.4% lower compared to the regular and late sowing date yields, respectively. When the early sowing date was preferred, it was 2.737 and 1.639 t ha^{-1} lower compared to the regular and late sowing dates, respectively.

In the three-year study, grain yields ranged between 3.248 and 9.610 t ha^{-1}. Early, regular, and late sowing date yields were between 4.216 and 8.021, 4.471 and 7.083, and 5.285 and 5.985 t ha^{-1}, respectively. When the country-wide yield was examined, the yields in 2014 were very low, while 2013 entered statistical records for its high yields. Country statistics and research data showed similarities. Late sowing has a stable yield, but it usually has a lower yield than other dates (Sharifi, 2010; Stout et al., 2011; Van Oort et al., 2016). Yield losses depend on cold stress, which is correlated with the number of plants per square meter rather than spikelet sterility.

When considering long-term regional climatic conditions, regular sowing dates, such as May, have less risk in terms of cold stress and mean yield than other sowing times in temperate regions. An early sowing date might be favorable in years when cold stress is not experienced. A regular sowing date is effective in normal seasonal conditions. Late sowing might be suitable when extreme and long cold stress conditions occur. The most stable yields were calculated in late sowing periods, but the yields were lower in late sowing periods than in normal conditions due to the shortening of the vegetation period.

Early, regular, and late sowing dates displayed plant density average numbers of 32.4, 67.0, and 110.6 plants m^{-2} in 2014 (Tables 2 and 3). Compared to early sowing dates, plant survival rate was two-fold higher in regular sowing dates and 3.5-fold higher in late sowing dates. The number of plants per square meter had a significant correlation with climatic conditions, given that sowing was carried out in the same norm. After exposure to cold stress, the non-germinated seeds or non-vital seedlings had significantly lower productivity. The minimum germination temperature of rice is 13°C, and the optimum temperature for rice cultivation is between 25 and 35°C in temperate regions (Ghadirnezhad & Fallah, 2014). Rice plant density was below the critical 60.8 plants m^{-2} in early and late plantings, the greatest impact of which is thought to be cold damage. Cold damage was seen only in the early sowing period, but the regular and late sowing dates were not affected by the cold in 2013. Cold damage affected the number of plants per square meter, and it decreased below the critical number of 60.8 plants in the early sowing date. There was a significant correlation between yield and the number of plants per square meter. The increase in the number of plants per square meter led to increased productivity. One of the most important factors causing a decrease in yield and the number of plants per square meter was cold stress.

Regression analysis revealed a quadratic relationship between the number of plants per m^{-2} (crop density) and grain yield (Figure 1).

The mean yield (5.743 t ha^{-1}) was calculated at a crop density of 79.6 plants m^{-2}. Yields were significantly affected by plant density. Based on the field removal and resowing limits, 60.8 plants m^{-2} was calculated.

Counce (1987) stated that the results obtained should be 120-150 plants m^{-2} for high yield. Baloch et al. (2002) found an average number of plants per square meter of 114-169 plants. Hamid et al. (2015) reported the number of plants per square meter as 149-156 plants, but beyond these results, our study indicated a minimum of 60.8-79.6 plants m^{-2}, and maximum of 132.3 plant m^{-2} required for economical grain yield to be attained according to the equation in Figure 1.

Early, regular, and late sowing dates had a spikelet sterility of 9.6, 10.7, and 12.7, respectively, in 2014. The results are very similar across sowing dates and next year's average result. Early, regular, and late sowing dates had a spikelet sterility of 8.0, 14.0, and 15.6, respectively, in 2015 (Table 3). Sterility was lowest at the early date, but yield was not positively affected at this sowing date. The correlation between spikelet sterility and yield was positive and had a high correlation coefficient; however, a

Table 2. Effect of sowing date on rice grain yield (t ha^{-1}), plant number (number m^{-2}), and spikelet sterility (%) in different years

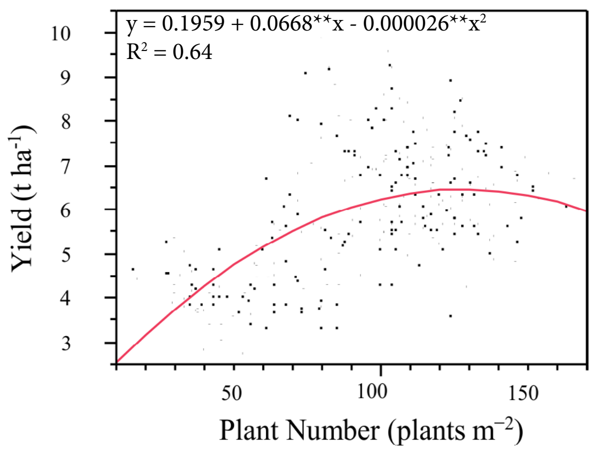
Years	Grain yield (t ha^{-1})			Plant number (number m^{-2})			Spikelet sterility (%)		
	Early	Regular	Late	Early	Regular	Late	Early	Regular	Late
2013	8.021a**	6.674 b	5.646 c	91.1 b	117.7 a	112.1a	12.1	10.6	11.7
	LSD = 69.2; CV = 8.6%			LSD = 13.9; CV = 11.4%			LSD = ns; CV = 12.4%		
2014	4.216 b	4.471 b	5.285 a	32.4 c	67.0 b	110.6a	9.6 c	10.7 b	12.7 a
	LSD = 59.8; CV = 8.9%			LSD = 13.2; CV = 18.5%			LSD = 0.93; CV = 14.9%		
2015	4.346 c	7.083 b	5.985 a	59.1 c	108.4 b	121.9 a	8.0 b	14.0 a	15.6 a
	LSD = 67.1; CV = 9.0%			LSD = 4.9; CV = 7.8%			LSD = 2.80; CV = 18.4%		

ns, ** - not significant and significant at p < 0.01. The values followed by the same letter indicate no significant difference at 0.05 level of probability. LSD (p < 0.05) - Least significant difference

Table 3. Effect of sowing dates and varieties on plant number per square meter (plant m^{-2})

Varieties	2013			2014			2015		
	Plant number according to sowing date (plant m^{-2})								
	Early sowing date (30 April)	Regular sowing date (20 May)	Late sowing date (10 June)	Early sowing date (30 April)	Regular sowing date (20 May)	Late sowing date (10 June)	Early sowing date (30 April)	Regular sowing date (20 May)	Late sowing date (10 June)
1 Kiziltan	112.0 b A†	105.3 c CD	132.5 a A	39.0 c A	64.7 b BCD	99.3 a CDE	64.3 c A	99.0 b AB	123.0 a ABCD
2 Pasali	95.7 c ABCD	130.7 a AB	119.0 bABC	40.0 c A	101.3 b A	123.3 a ABC	45.3 c A	104.3 b AB	131.7 a ABC
3 T.gunesi	99.3 b ABC	114.7 a BCD	97.8 b DE	36.7 c A	70.0 b BC	100.7 a CDE	47.7 c A	96.7 b AB	151.0 a AB
4 Duragan	86.5 c BCDE	144.0 a A	120.8 b ABC	37.3 c A	62.7 b BCD	145.3 a A	74.0 c A	130.7 b A	155.3 a A
5 Halilbey	92.7 b ABCDE	122.2 a BC	127.0 a A	30.3 c A	62.0 b BCD	110.7 a BCD	59.7 b A	126 a A	115.3 a BCD
6 Edirne	89.5 b BCDE	115.5 a BCD	112.8 a ABCD	33.0 c A	71.7 b BC	105.7 a CDE	49.3 a A	113.3 a A	106.3 a CD
7 Osmancik97	82.5 c CDE	117.3 b BC	125.3 a AB	33.3 b A	38.3 b D	94.0 a DE	65.0 c A	95.3 b AB	124.0 a ABC
8 Tunca	85.7 b BCDE	121.0 a BC	119.8 a ABC	26.7 c A	39.3 b D	102.7 a CDE	67.3 b A	106.7 a AB	105.0 a CD
9 Aromatik	104.7 bAB	129.8 a AB	102.3 b CDE	33.3 c A	82.7 b A B	145.0 a A	64.3 b A	106.0 a AB	113.0 a CD
10 Hamzadere	92.7 c ABCDE	114.7 b BCD	133.3 a A	32.7 c A	64.7 b BCD	133.7 a AB	58.0 b A	115.7 a A	119.7 a ABCD
11 Mevlutbey	93.8 c ABCDE	114.7 a BCD	105.0 b BCDE	27.7 c A	73.3 b BC	103.0 a CDE	66.0 c A	127.0 b A	140.7 a ABC
12 IR50 (S)	75.5 c DE	105.7 a CD	86.5 b EF	18.7 b A	86.3 a AB	94.7 a DE	54.7 b A	76.3 a B	87.3 a D
13 HSC55 (T)	73.5 b E	95.2 a D	75.5 b F	32.7 c A	54.3 b CD	79.3 a E	53.0 b A	112.0 a AB	112.3 a CD
LSD ($p < 0.05$)	LSD (Sow. Date) = 6.96; LSD (Variety) = 20.72			LSD (Sow. Date) = 9.09; LSD (Variety) = 27.02			LSD (Sow. Date) = 12.27; LSD (Variety) = 36.49		
CV (%)	11.4			18.5			7.8		

† - Significant at p < 0.05. The values followed by the same letter indicate no significant difference at 0.05 level of probability; uppercase letters compare in the columns, and lowercase letters compare in the rows for each years; ns - Non-significant at p < 0.05; CV (%) - Coefficient of variation; LSD (p < 0.05) - Least significant difference



** - Significant at p ≤ 0.01

Figure 1. Rice grain yield (kg ha^{-1}) as a function of plant density

negative correlation was expected (Osman et al., 2015). This might have caused the plant number coefficient to repress the yield and sterility. Spikelet sterility has been reported as the main result of cold stress in many papers (Zeng et al., 2017; Unan, 2021), but the sterility effect might also appear during booting if cold damage occurs due to weather conditions in temperate regions.

Cold tolerance evaluations across 13 rice cultivars were discussed in this study. Among these, there were 11 national rice varieties, all of which were of the japonica type, except Aromatic-1. IR50 was employed as a susceptible control, whereas HSC55 was used as a cold-tolerant control. HSC55 had the lowest score, although tolerant, it had a very limited maturity time and lodging problems. IR50 is an indica-type variety, and their yield was low compared to national varieties. Experimental average yield scores ranked between 452.6 and 667.7 t ha^{-1} in all experiment years and sowing dates (Table 4). As there was an interaction effect between variety and year, the years were evaluated separately.

Most countries, such as the USA, Korea, Australia, and Turkey, have screened for cold-tolerant varieties. A national genetic stock with 237 germplasm resources has been screened in Turkey at vegetative and generative stages (Unan, 2016), revealing that 5-11% of the germplasm was nightly susceptible to cold stress.

In 2013, no cold stress occurred. Tunca had the highest yield of 7.596 t ha^{-1} and HSC55 had the lowest yield of 5.23 t ha^{-1}. Variety and sowing time interactions revealed that Tunca had the highest yield at the early sowing date at 9.616 t ha^{-1} with no cold stress in 2013. HSC55 had the lowest grain yield at the late sowing date of 3.248 t ha^{-1}.

In 2014, excessive cold stress was experienced. Mevlutbey had the highest yield (5.766 t ha^{-1}), and HSC55 had the lowest yield (3.733 t ha^{-1}). Variety and sowing date interactions revealed that Mevlutbey had the highest yield at the early sowing date at 726.7 t ha^{-1}. HSC55 had the lowest grain yield at the early sowing date, at 358.2 t ha^{-1}.

In terms of the 2015 cold stress year, Mevlutbey had the highest yield of 6.900 t ha^{-1}, and HSC55 had the lowest yield of 4.630 t ha^{-1}. Variety and sowing date interactions were insignificant in 2015.

According to the evaluation of variety and variety × sowing time interaction, Halilbey, Pasali, and Mevlutbey were the most cold tolerant; Edirne and Kiziltan were the most cold susceptible national rice varieties in all experimental years. The selection and development of cold-tolerant varieties and cultural management are important requirements for maintaining consistently high yields under low-temperature stress in temperate regions (Glaszmann et al., 1990). The cultural practice of using cold-tolerant varieties is an effective method (Biswas et al., 2019a).

Farmers use cold-tolerant varieties to protect against cold damage, as this causes the termination of plants in the germination or seedling stages, and seeds have to be resown due to the lack of germination, low germination, or late germination. In some locations, farmers can sow rice seeds in very early periods, such as April 15, to be able to offer crops to the market early and to avoid precipitation at harvest time, depending on changing rainfall trends (Dinh & Dang,

Table 4. Effect of sowing dates and varieties on rice grain yield (t ha^{-1})

Varieties	2013			2014			2015		
				Yields according to sowing date (t ha^{-1})					
	Early sowing date (30 April)	Regular sowing date (20 May)	Late sowing date (10 June)	Early sowing date (30 April)	Regular sowing date (20 May)	Late sowing date (10 June)	Early sowing date (30 April)	Regular sowing date (20 May)	Late sowing date (10 June)
1 Kiziltan	8.035 a BC†	7.034 b ABC	5.622 c BCD	3.318 b E	4.210 a EF	4.236 a GH	4.235 BCD	6.581 CDE	5.129 EF
2 Pasali	8.007 a BC	7.741 a A	7.000 b A	4.645 b AB	4.436 b DEF	5.730 a BCD	4.301 BCD	7.100 BCD	6.161 ABC
3 T.gunesi	7.711 a BCD	6.479 b BCD	5.948 c BCD	4.418 b ABC	4.018 c EF	4.980 a EF	3.722 CD	7.450 ABC	5.807 BCDE
4 Duragan	7.816 a BC	6.137 b CD	6.306 b ABCD	4.190 b BCD	3.856 c A	5.225 a CDE	4.400 BCD	6.487 DE	6.133 ABC
5 Halilbey	7.656 a BCD	6.020 b CD	6.501 c AB	4.580 b AB	5.379 a BC	5.290 a CDE	4.720 B	7.800 AB	6.625 AB
6 Edirne	7.165 a CD	5.827 b D	5.894 b BCD	3.741 c CDE	4.409 b DEF	4.990 a DEF	3.745 CD	6.985 BCD	5.569 CDF
7 Osmancik97	7.970 a BC	6.488 b BCD	5.539 c BCD	3.812 c E	3.694 b F	4.875 a EFG	4.422 BCD	7.254 ABCD	6.700 AB
8 Tunca	9.610 a A	7.718 b A	5.460 c CD	4.470 b ABC	4.652 b CDE	5.732 a BCD	4.841 B	7.078 BCD	6.267 ABC
9 Aromatik	8.246 a B	6.967 b ABC	5.337 c D	4.408 c ABC	4.980 b CD	5.844 a BC	4.569 BC	7.140 ABCD	6.067 AD
10 Hamzadere	8.011 a BC	6.678 b BCD	6.378 b ABC	4.136 b BCD	3.905 b F	6.280 a B	4.556 BC	7.296 ABCD	6.751 A
11 Mevlutbey	8.649 a AB	7.221 b AB	6.263 c ABCD	4.989 b A	5.041 b BCD	7.267 a A	5.898 A	8.046 A	6.757 A
12 IR50 (S)	8.672 a AB	6.736 b ABCD	3.909 c E	4.522 b AB	5.773 a B	4.411 b FGH	3.588 D	7.168 ABCD	5.144 DEF
13 HSC55 (T)	6.730 a D	5.713 b D	3.248 c E	3.582 b DE	3.772 ab F	3.847 a H	3.497 D	5.696 E	4.696 F
LSD ($p < 0.05$)	LSD (Sow. Date) = 0.347; LSD (Variety) = 1.033			LSD (Sow. Date) = 0.249; LSD (Variety) = 0.743			LSD (Sow. Date) = ns; LSD (Variety) = 0.931		
CV (%)	8.6			8.9			9.0		

† - Significant at p < 0.05. The values followed by the same letter indicate no significant difference at 0.05 level of probability; uppercase letters compare in the columns, and lowercase letters compare in the rows for each years; ns - Non-significant at p < 0.05; CV% - Coefficient of variation; LSD (p < 0.05) - Least significant difference

2022). These farmers tend to terminate and resow their fields if the conditions are not good. Although it is rare, exposure to cold stress can occur during the normal sowing periods of May. The last time cold conditions were experienced, they caused extensive damage and warranted the resowing of 1000 ha in the Bafra plain between 15 and 30 May in 2020. Termination and resowing processes include one or two rotavator applications, irrigation, seed purchase, and sowing. The cost of the operations is, on average, equivalent to 0.50-0.70 t ha^{-1} rice product. In the following section, the suitability of comparing the sowing periods based on cold stress and average conditions is discussed. The year and sowing periods when stress conditions did not occur were not considered.

The termination and resowing applications were beneficial to late sowing instead of early and regular sowing dates because of cold stress, but the regular sowing period was not profitable compared to early sowing because both sowing dates had cold stress in 2014. When 0.60 t ha^{-1} of crop was calculated as the cost of field termination and resowing, the late sowing date used more product compared to the early and normal sowing periods (0.47 and 0.21 t ha^{-1}, respectively; Table 4).

The termination and resowing applications were profitable for regular and late sowing dates instead of early sowing dates due to cold stress in 2015. The regular sowing date used more product (2.14 t ha^{-1}) compared to the early sowing date, and the late sowing date used more product (1.04 t ha^{-1}) compared to the early sowing date.

The evaluations revealed that it is suitable to choose the sowing date according to long-term climatic conditions. Termination and resowing applications were affected by the cold. Cold stress damage in the early growth stages in rice fields causes delayed germination, in some cases up to 20 days; in other cases, germination is reduced, is not uniform, or occurs with excessive weeds. In this case, the farmers have to decide whether to continue with the existing field or terminate and resow the field. The existing plant number per square meter is one of the most important factors that can affect this decision. It might be a sufficient reason for termination and resowing application in the field if the cold stress effect is maintained 20-30 days after sowing, and in the case of uneven germination, it was calculated as 60.8 plants m^{-2}. A uniform rice field has 100-150 plants m^{-2} (Counce, 1987; Baloch et al., 2002). A minimum of 79.6 plants m^{-2} was found in this study to achieve an average yield. Since densities below this threshold are expected to result in massive yield losses, farmers should evaluate termination and resowing costs between 60.8 and 79.6 plants m^{-2} within their own conditions. In cases where it is under the limit, terminating the crop and resowing it are expected to be more beneficial than allowing the crop to produce due to low yields.

Conclusions

1. Rice yield was significantly affected by the sowing date, and yield losses of up to 38.6% were measured due to cold stress conditions. Moreover, the yield fluctuations result was experienced at different sowing dates, even in the same growing season.

2. Periodic cold stress most affected the early development stage of plants in a temperate climate. Cold stress occurring during the germination and seedling stages had a far more devastating effect than when it occurred at the booting stage.

3. A minimum of 60.8-79.6 plants and an optimum of 132.3 plants per square meter are required for effective grain yield. If there is less than the minimum threshold of plants per square meter in the field due to cold stress occurring in the early and regular sowing periods, effective termination and resowing is recommended.

Acknowledgments

This research was funded by Republic of Turkey Ministry of Agriculture and Forestry, General Directorate of Agricultural Research and Policies (Project no: TAGEM/TA/03/03/06/00).

Literature Cited

Alemayehu, H. A.; Dumbuya, G.; Hasan, M.; Tadesse, T.; Nakajyo, S.; Fujioka, T.; Abe, A.; Matsunami, M.; Shimono, H. Genotypic variation in cold tolerance of 18 Ethiopian rice cultivars in relation to their reproductive morphology. Field Crops Research, v.262, p.1-9, 2021. https://doi.org/10.1016/j.fcr.2020.108042

Baloch, A. W.; Soomro, A. M.; Javed, M. A.; Ahmed, M.; Bughio, H. R.; Bughio, M. S.; Mastoi, N. N. Optimum plant density for high yield in rice (*Oryza sativa* L.). Asian Journal of Plant Science, v.1, p.25-27, 2002.

Biswas, P. S.; Khatun, E.; Anisuzzaman, M. Molecular and phenotypic characterization for cold tolerance in rice (*Oryza sativa* L.). Bangladesh Rice Journal, v.23, p.1-15, 2019b. https://doi.org/10.3329/brj.v23i2.48243

Biswas, P. S.; Rashid, M. M.; Khatun, H.; Yasmeen, R.; Biswas, J. K. Scope and progress of rice research harnessing cold tolerance. In: Hasanuzzaman, M. M.; Fujita, N. K.; Biswas, J.K. (eds.) Advances in rice research for abiotic stress tolerance. Amsterdam: Elsevier, 2019a. p.225-264. https://doi.org/10.1016/B978-0-12-814332-2.00011-3

Bodapati, N.; Gunawardena, T.; Fukai, S. Increasing cold tolerance in rice by selecting for high polyamine and gibberellic acid content, 2005.

Counce, P. A. Asymptotic and parabolic yield and linear nutrient content responses to rice population density. Agronomy Journal, v.79, p.864-869, 1987. https://doi.org/10.2134/agronj1987.00021962007900050022x

Cruz, R. P. da; Duarte, I. T. de L.; Cabreira, C. Inheritance of rice cold tolerance at the seedling stage. Scientia Agricola, v.67, p.669-674, 2010. https://doi.org/10.1590/S0103-90162010000600008

Dinh, T. K. H.; Dang, T. A. Potential risks of climate variability on rice cultivation regions in the Mekong Delta, Vietnam. Revista Brasileira de Engenharia Agrícola e Ambiental, v.26, p.348-355, 2022. http://dx.doi.org/10.1590/1807-1929/agriambi.v26n5p348-355

Ghadirnezhad, R.; Fallah, A. Temperature effect on yield and yield components of different rice cultivars in flowering stage. International Journal of Agronomy, v.2014, p.1-4, 2014. http://dx.doi.org/10.1155/2014/846707

Glaszmann, J. C.; Kaw, R. N.; Khush, G. S. Genetic divergence among cold tolerant rices (*Oryza sativa* L.). Euphytica, v.45, p.95-104, 1990. https://doi.org/10.1007/BF00033276

Hamid, A.; Ullah, M. J.; Haque, M. M.; Mollah, M. F. H.; Rahman, M. M. Improving grain yield of indigenous rice in tidal floodplain of Southern Bangladesh: Effect of seedling age and transplanting method. Agricultural Sciences, v.6, p.1538-1546, 2015. https://doi.org/10.4236/as.2015.612147

IRRI - International Rice Research Institute. Standard evaluation system for rice. Manila: International Rice Research Institute, 2002. 57p.

Liu, C. T.; Wang, W.; Mao, B. G.; Chu, C. C. Cold stress tolerance in rice: physiological changes, molecular mechanism, and future prospects. Hereditas, v.40, p.171-185, 2018. https://doi.org/10.16288/j.yczz.18-007

Osman, K. A.; Mustafa, A. M.; Elsheikh, Y. M. A.; Idris, A. E. Influence of different sowing dates on growth and yield of direct seeded rice in semi-arid zone Sudan. International Journal of Agronomy and Agricultural Research, v.6, p.38-48, 2015.

SAS Institute. SAS user's guide: Statistics, 4.ed. Cary: SAS Institute, 1992.

Shakiba, E.; Edwards, J. D.; Jodari, F.; Duke, S. E.; Baldo, A. M.; Korniliev, P.; McCouch, S. R.; Eizenga, G. C. Genetic architecture of cold tolerance in rice (*Oryza sativa*) determined through high resolution genome-wide analysis. PLoS ONE, v.12, p.1-22, 2017. https://doi.org/10.1371/journal.pone.0172133

Sharifi, P. Evaluation on sixty-eight rice germplasms in cold tolerance at germination stage. Rice Science, v.17, p.77-81, 2010. https://doi.org/10.1016/S1672-6308(08)60107-9

Sharma, N.; Reinke, R.; Sacks, E. J. Comparison of methods to evaluate rice (*Oryza sativa*) germplasm for tolerance to low temperature at the seedling stage. Agronomy, v.11, p.1-17, 2021. https://doi.org/10.3390/agronomy11020385

Stout, M. J.; Hummel, N. A.; Frey, M. J.; Rice, W. C. The impact of planting date on management of the rice water weevil in Louisiana rice. The Open Entomology Journal, v.5, p.1-9, 2011. https://doi.org/10.2174/1874407901105010001

Surek, H. Rice agriculture. Istanbul: Hasad Publication, 2002. 239p.

Unan, R. Determination of effect of cold stress on yield and quality parameters and evaluation of cold tolerant genotypes by morphological and molecular methods in rice. Tekirdağ: Namik Kemal University, Science Institute, 2016. 303p. PhD thesis

Unan, R. Development of japonica type cytoplasmic male sterile (CMS) rice lines for commercial hybrid rice in Mediterranean ecological condition. Turkish Journal of Field Crops, v.26, p.111-116, 2021. https://doi.org/10.17557/tjfc.650908

Unan, R.; Deligoz, I.; Al-Khatib. K.; Mennan, H. Protocol for ethyl methanesulphonate (EMS) mutagenesis application in rice. Open Research Europe, v.1, p.1-23, 2022. https://doi.org/10.12688/openreseurope.13317.3

van Oort, P. A. J.; Balde, A.; Diagne, M.; Dingkuhn, M.; Manneh, B.; Muller, B.; Sow, A.; Stuerz, S. Intensification of an irrigated rice system in Senegal: Crop rotations, climate risks, sowing dates and varietal adaptation options. European Journal of Agronomy, v.80, p.168-181, 2016. https://doi.org/10.1016/j.eja.2016.06.012

Xiao, N.; Gao, Y.; Qian, H.; Gao, Q.; Wu, Y.; Zhang, D.; Zhang, X.; Yu, L.; Li, Y.; Pan, C.; Liu, G. Identification of genes related to cold tolerance and a functional allele that confers cold tolerance. Plant Physiology, v.177, p.1108-1123, 2018. https://doi.org/10.1104/pp.18.00209

Zeng, Y.; Zhang, Y.; Xiang, J.; Uphoff, N. T.; Pan, X.; Zhu, D. Effects of low temperature stress on spikelet-related parameters during anthesis in Indica-Japonica hybrid rice. Frontiers in Plant Science, v.8, p.1-11, 2017. https://doi.org/10.3389/fpls.2017.01350