Original Research Article

CHILLING RESISTANCE AND PHYSIO -CHEMICAL CHANGES DURING COLD ACCLIMATION IN FOXTAIL MILLET GENOTYPES

ABSTRACT

Millets are known as poor man crops, because they grow well in rain-fed or dry land conditions with insufficient soil fertility and water. Food shortages could become more severe in the coming years as the world population rises and arable land is depleted. As a result, various strategies are needed to ensure food security. Millets are also notable for their short growth season. They can reach maturity in as little as 85 days after being sowing. Apart from high temperatures and scarcity of water, low temperature is a major environmental factor that can limiting the plant anabolic, metabolic processes and yield globally. Temperature is an extremely important growth limiting factor because it regulates plant physiological and biochemical activity throughout the growth cycle. Low temperatures may have an effect on plant processes such as photosynthesis, respiration, water absorption, chlorophyll stability, and yield. Temperature stress, particularly high or low temperatures, can disrupt plant metabolism and shorten the time of distinct plant growth phases. Plants' responses to low temperature exposure can have a significant impact on various growth parameters such as leaf area, leaf area index, crop growth rate, relative water content, photosynthetic efficiency, days to 50% flowering, number of tillers, number of grains per tiller, total dry matter production, and yield.

Plant genotypes have been tested for a variety of physiological and biochemical characteristics in order to generate cold tolerance through altering metabolism in response to cold stress. The present study was conducted to determine the low temperature effect on growth analysis and yield of tenai genotypes screening at Agricultural College and Research Institute, Vazhavachanur, Tiruvannamali district and Jawathu hill region of Vellore district.

Key words: Foxtail millet, Leaf area, Specific leaf weight, Crop growth rate, Photosynthetic efficiency and Productivity

INTRODUCTION

Temperature stress is the most vital environmental factor limiting plant growth, physiological and biochemical processes, and, ultimately, productivity worldwide. All plant species require the optimal temperature to develop and complete their life cycle. Apart from other environmental stresses, low temperature is an important environmental factor in limiting plant survival, metabolism, and productivity. Low temperature affect all the physiological processes like, photosynthesis, chlorophyll content, respiration, crop growth rate and total dry matter production of plants (Rahul kumar *et al*., 2018)

Chilling injury is defined as an injury produced by a temperature drop below 15°C but above the freezing point. Chilling injury is characterised by rapid wilting of the leaves and the formation of water-soaked areas. Small millets are drought tolerant in general. It is now a highly popular meal all around the world. The hill people cultivate a vast amount of small millets. Even though it is a drought-tolerant crop, it does not tolerate cold temperatures. Foxtail millet, in particular, decreases growth and development at low temperatures. When compared with conventional cultivation, total dry matter production is very low at cold temperature. As a result, yield loss is severe. When compared to coarse cereals like wheat and rice, small millets are extremely high in protein, fibre, and minerals. Therefore, millets are now being pronounced as "Miracle grains and nutria-cereals". Millets contain many nutritional and health benefits, when compare to other crops. Plant species can sustain their response to non-freezing temperatures by sustaining molecular and physiological modifications during cold acclimation. (Hsieh et al. 2004; Zhu et al. 2007).

Millets and particularly small millets are in a situation of crisis in India. The period between 1961 and 2009 saw a dramatic decrease in cultivated area under millets (80% for small millets, 46% for finger millet); a 76% decrease in total production of small millets and a steep fall in overall millets consumption. The change in climatic conditions, water scarcity, increasing world population, rising food prices, and other socioeconomic impacts are expected to generate a great threat to agriculture and food security worldwide, especially for the poorest people. Apart from water scarcity, low temperatures reduce yield in small millets, particularly foxtail millet. If the temperature is too low (below 15 °C) during the vegetative and panicle initiation periods, the leaves will show indications of withering, chlorosis, and necrosis (Ruelland and Zachowski 2010), as well as damage to cell membrane structures and lipid metabolism (Uemura and Steponkus 1999; Matteucci *et al*. 2011). Cold exposure has a significant impact on photosynthetic efficiency and production at the physiological level ((Ruelland and Zachowski, 2010). Chilling-sensitive plants minimise their growth at several physiological phases, which is particularly pronounced in susceptible species and variations compared to tolerant genotypes (Rab, A. and Saltveit, 1996; Ting *et al*., 2012; Venema *et al*., 1999). Therefore, there is an urgent need of scientist and researcher should focus more on sustained production of foxtail millet under low temperature condition.

MATERIALS AND METHOD

A field experiment on foxtail millet was conducted with 7 genotypes with two locations viz., Agricultural College and Research Institute, Vazhavachanur, Tiruvannamali district and Jawathu hills at Tiruvannamali district. The cardinal temperature of foxtail millet is 9.3°C for base, 37.0°C for optimum and 46.0°C ceiling temperature (Kamkar *et al.*, 2006).The experiment was laid out with factorial randomized block design with three replications. During *Rabi'*2020 and 2021, day and night temperatures in Tiruvannamali district range from 12°C to 24 °C. Furthermore, the temperature at Jawathu Hill varies between 9 and 20 °C during the day and night. Low temperatures of 10 to 25°C generally cause damage to the tropical and subtropical plants. It reduces growth, produces surface leaf lesions, necrosis, chlorosis, internal stem discolouration, increased susceptibility to decay, loss of vigour, and finally cell tissue collapse.

Treatment details

Factor	-	Two
Factor A	-	Genotypes
Levels	-	Seven
Factor B	-	Locations
Levels	-	Two (Agricultural College and Research Institute, Vazhavachanur, Tiruvannamali district and Jawadhu hills)

Varietal Details

Raising 6 advanced cultures each in Tenai with a check variety. Advanced cultures are TNSi 337, TNSi 354, TNSi 356, TNSi 375, TNSi 376, TNSi 379 with check variety of Tenai ATL 1.

All agronomic practices are considered as normal for all the treatments except those which were under study. Each entry was represented by 3 rows with 22.5 x 10 cm spacing and need based recommended doses of fertilizers and plant protection measures.

Observations on plant height (cm), Number of productive tillers hill$^{-1,}$, days to 50% flowering, leaf area (cm2 plant-1), crop growth rate (g m^{-2} day-^{1}), specific leaf weight t (mg /cm^{2}), chlorophyll content (mg/g), 1000 grain weight (gm) and yield/ plant (g) observations were recorded on randomly chosen plants per genotype per plot in each replication.

Specific Leaf Weight (SLW)

Specific Leaf Weight (SLW) was calculated by using the formula of Pearce *et al.* (1968) and expressed in mg cm^{-2}.

$$SLW = \frac{\text{Leaf dry weight per plant (mg)}}{\text{Leaf area per plant } (cm^2)}$$

Crop Growth Rate (CGR)

The Crop Growth Rate (CGR) was estimated by using the formula of Watson (1956) and expressed in g m^{-2} day^{-1}.

$$CGR = \frac{W_2 - W_1}{\rho\,(t_2 - t_1)}$$

Where,

W_1 and W_2 = Whole plant dry weights (g) at time t_1 and t_2 respectively.

t_2 and t_1 = Time of sampling (days)

ρ = Ground area occupied by plant (m^2)

Total Dry Matter Production (TDMP)

Plant samples were first shade dried and then oven dried at 70 °C for 24 hours. The dry weight of whole plant including the seeds was taken and expressed in g $plant^{-1}$.

Chlorophyll content

Contents of fractions of 'a', 'b' and total chlorophyll were estimated in a fully expanded young leaf at the specified time intervals and expressed in mg g^{-1} fresh weight (Yoshida *et al.*, 1971).

$$\text{Chlorophyll 'a'} = \frac{(12.7 \times \text{O.D. at } 663) - (2.69 \times \text{O.D. at } 645)}{W} \times V$$

$$\text{Chlorophyll 'b'} = \frac{(22.9 \times \text{O.D. at } 645) - (4.68 \times \text{O.D. at } 663)}{W} \times V$$

Total chlorophyll = Chl 'a' + Chl 'b'

Amount of chlorophyll present in leaves was also qualitatively measured by a non-destructive method using SPAD-502 Chlorophyll Meter (Minolta Co., Japan) as suggested by Gratani (1992).

RESULTS AND DISCUSSION

Currently, various climate changes are causing yield losses in all agricultural crops. It has the ability for affecting plant physiological and biochemical processes. Although temperature and water scarcity are important factors, low temperatures cause greater yield loss in small millets, particularly in foxtail millet. Small millets tend to be 80-85 day crops. It can germinate in 5-7 days, followed by tillering and panicle initiation 35-45 days after sowing. If chilling impact is detected during the flower initiation period, the flowering time can be extended. That is, the entire growing period is extended by 10 to 15 days. As a result, plant growth and development, photosynthetic efficiency, dry matter production and grain yield will be reduced.

Genotypes TNSi 375, TNSi 376 , TNSi 337 and ATL 1 had maximum no of tillers, leaf area at 12 to 24 °C, therefore these genotypes is good for *Rabi'* cultivation, since it has low temperature tolerance. They also that the specific leaf area, specific leaf weight & crop growth rate were similar in those genotypes at 12°C as well as 24°C. The maximum leaf area (323.6) and crop growth rate (4.93) was recorded in TNSi 375. These genotypes exhibited tolerance for chilling injury for growth and development period. Satahe and Hayase (1970) reported that sterility of the spikelets caused by cool temperature. Critical stages for cold damage including active tillering, panicle initiation and grain filling stages. Since, the most sensitive stage for chilling harm is the flowering stage, which occurs 10-15 days prior to grain filling stages. The cold stress was assessed in flower initiation period, genotypes performed with least percentage of chlorophyll reduction (3.565) and average yield of (1397) is TNSi 375 genotype and the most sensitive genotype is TNSi 375 at Tiruvannamali and Jawathu hills. Low temperature stress can affect plant photosynthesis and reduce light utilisation [Glaszmann et al., 1990; Lightner et al., 1997].Chilling causes changes in glucose levels, which are associated with decreased respiration, photosynthesis, and carbohydrate metabolism enzyme activity (Ebrahim *et al*., 1992). Plant species native to tropical and subtropical places often exhibit injury signs at temperatures below 12 degrees Celsius (Esmaili and Salehi, 2012). Chilling stress frequently causes decreased leaf area, crop growth rate and photosynthetic efficiency and chlorosis, cellular membrane damage, and oxidative stress in plants (Suzuki and Mittler, 2006). Photosynthetic pigment content will be reduced to 92%, 74%, and 45% of ideal levels for Latitude 40, and 88%, 51%, and 30% of optimum levels for Latitude-22, respectively, at chilling and freezing temperatures (Shuangming Li *et al*, 2018). The grain maize varieties, low temperature causes seed rotting, chlorosis and necrosis (Dolstra & Miedema, 1986). Low temperature stress causes the limitations of light interception in the plant canopy, that leads to reduce the photosynthesis (Glaszmann, 2020 ; Lightner, 1997). Plants possess several kinds of efficient processes that allow them to adapt to adverse situations in order to survive with low temperatures (Bressan *et al.*, 2009; Liu *et al.*, 2012). This adaptive process includes a variety of biochemical and physiological changes, such as prolonging the flower initiation period, lowering dry matter production and efficiency of photosynthesis, and finally yield loss. Sarkar et al., (2013) revealed that low temperature stress is one of the most critical environmental stresses affecting plant growth and development in rice. Chilling temperatures have an effect on plant growth and development in temperate zones, leading in an extended flowering period, direct harm to floral abortion, or delayed maturation. Even a slight temperature drop, which produced no obvious damage to chilling-sensitive plants, but reduced their yield.

Table 1: Effect of low temperature on Plant height (cm), Number of productive tillers hill^{-1}, Leaf Area (cm^{2} plant^{-1}), Specific Leaf Area (cm^{2} g^{-1}), Specific Leaf Weight (mg cm^{-2}), Crop Growth Rate (g m^{-2} day^{-1}), Total chlorophyll content (mg g^{-1}), Total Chlorophyll content (mg g^{-1}), 1000 grain weight (gm) and Grain yield (kg/ha) of foxtail millet genotypes at Tiruvannamai district.

Treatments	Plant height (cm)	Number of productive tillers hill^{-1}	Leaf Area (cm^{2} plant^{-1})	Specific Leaf Area (cm^{2} g^{-1})	Specific Leaf Weight (mg cm^{-2})	Crop Growth Rate (g m^{-2} day^{-1})	Total chlorophyll content (mg g^{-1})	1000 grain weight (gm)	Grain yield (kg/ha)
TNSi 337	86	3.2	316.4	253.8	3.90	4.20	2.606	3.29	1233
TNSi 354	82	3.6	217.4	90.5	7.5	4.82	1.368	3.20	1005
TNSi 356	87	3.4	256.0	143.8	7.00	4.74	1.397	3.56	962
TNSi 375	93	5.2	323.6	180.3	8.30	4.93	3.565	3.66	1397
TNSi 376	81	4.8	222.3	121.7	8.20	4.68	3.226	3.48	1275
TNSi 379	97	3.2	186.9	124.3	8.00	4.90	2.957	2.84	943
ATL 1	95	3.7	244.3	142.2	6.80	5.12	2.829	2.91	1290
SEd	2.51	0.05	32.98	14.61	0.37	0.36	0.09	0.068	69.43
CD (P=0.05)	7.74	0.16	101.62	45.02	1.14	1.11	0.26	0.150	152.95

Table 2: Effect of low temperature on Plant height (cm), Number of productive tillers hill^{-1},

Leaf Area (cm^2 $plant^{-1}$), Specific Leaf Area (cm^2 g^{-1}), Specific Leaf Weight (mg cm^{-2}), Crop Growth Rate (g m^{-2} day^{-1}), Total chlorophyll content (mg g^{-1}), Total Chlorophyll content (mg g^{-1}), 1000 grain weight (gm) and Grain yield (kg/ha) of foxtail millet genotypes at Jawathu hill

Treatments	Plant height (cm)	Number of productive tillers $hill^{-1}$	Leaf Area (cm^2 $plant^{-1}$)	Specific Leaf Area (cm^2 g^{-1})	Specific Leaf Weight (mg cm^{-2})	Crop Growth Rate (g m^{-2} day^{-1})	Total chlorophyll content (mg g^{-1})	1000 grain weight (gm)	Grain yield (kg/ha)
TNSi 337	66	3.0	329.1	140.0	6.1	3.63	2.981	3.26	1200
TNSi 354	61	3.0	236.2	147.6	6.8	3.51	2.957	3.28	997
TNSi 356	72	3.2	245.1	155.4	6.4	3.61	3.047	3.36	948
TNSi 375	80	4.5	341.1	132.4	7.6	3.91	3.157	3.39	1230
TNSi 376	73	4.1	310.1	137.8	7.3	3.68	3.023	3.32	1255
TNSi 379	41	3.0	233.2	155.1	6.4	3.50	2.976	3.16	1125
ATL 1	65	3.2	317.0	135.5	7.4	3.55	2.658	3.34	1150
SEd	7.29	0.06	43.68	3.60	0.165	0.310	0.227	0.201	62.43
CD (P=0.05)	16.07	0.19	96.24	11.10	0.363	0.683	0.499	0.443	137.53

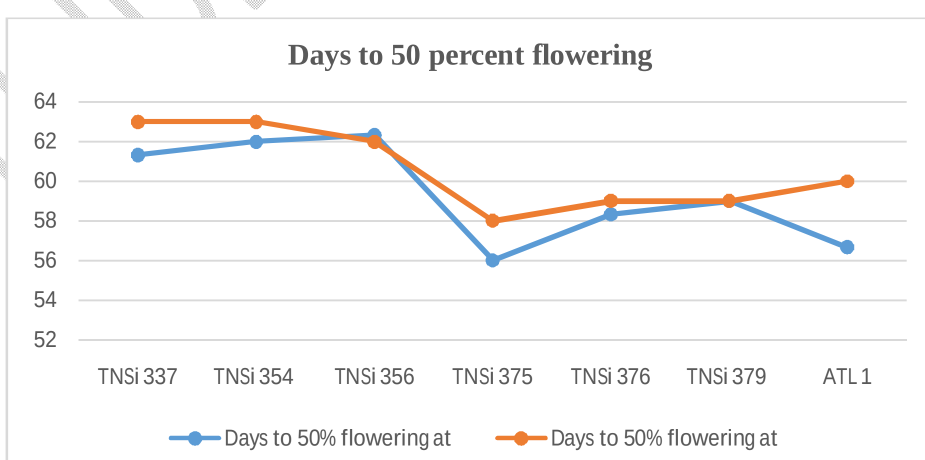

Fig.1. Effect of low temperature on Days to 50% flowering of foxtail millet genotypes at Tiruvannamali and Jawathu hill.

CONCLUSION AND FUTURE OUTLOOK:

Apart from high temperature, low temperatute also caused plant development. It leads, to reduce the water absorption, photosynthesis, respiration, mineral absorption and different physio-chemical process. It is the complex function over low temperature and crop duration. Besides, low temperature causes extension of flower initiation period and finally duration also extended. This will leads to reduce the crop growth rate and total dry matter production. As results indicated, low temperature stress caused a certain degree of physiological damage. Based on the study, TNSi 375 and TNSi 375 may be further advanced and released as a variety for commercial exploitation after proper evaluation.

REFERENCES:

1. Dolstra, O. & P. Miedema, 1986. Breeding for improved vegetative growth at low temperature in maize (Zea mays L.). In: O. Dolstra & P. Miedema (Eds): Breeding of silage maize. Pudoc, Wageningen, p. 61 -70 .
2. Ebrahim, M.K.H., Vogg, G., Osman, M.N.E.H. and Komor, E., Photosynthetic performance and adaptation of sugarcane at suboptimal temperatures. Journal of Plant Physiology, 153(5–6): 587–592 (1998).
3. Esmaili S, Salehi H. Effects of temperature and photoperiod on postponing bermudagrass (Cynodon dactylon [L.] Pers.) turf dormancy. J Plant Physiol. 2012; 169:851–858. https://doi.org/10.1016/j.jplph. 2012.01.022 PMID: 22465814
4. Hsieh TH, Lee JT, Yang PT, Chiu LH, Charng YY, Wang YC, Chan MT (2004) Heterology expression of the Arabidopsis C-repeat/ dehydration response element binding factor 1 gene confers elevated tolerance to chilling and oxidative stresses in transgenic tomato. (vol 129, pg 1086, 2002). Plant Physiol 135:1145–1155.
5. J. C. Glaszmann, R. N. Kaw, and G. S. Khush, “Genetic divergence among cold tolerant rices (Oryza sativa L.),” Euphytica, vol. 45, no. 2, pp. 95–104, 1990.
6. J. C. Glaszmann, R. N. Kaw, and G. S. Khush, “Genetic divergence among cold tolerant rices (Oryza sativa L.),” Euphytica, vol. 45, no. 2, pp. 95–104, 1990.

7. J.Wu, J. Lightner, N.Warwick, and J. Browse, “Low-temperature damage and subsequent recovery of fab1 mutant arabidopsis exposed to 2∘ C,” Plant Physiology, vol. 113, no. 2, pp. 347–356, 1997
8. J.Wu, J. Lightner, N.Warwick, and J. Browse, “Low-temperature damage and subsequent recovery of fab1 mutant arabidopsis exposed to 2∘ C,” Plant Physiology, vol. 113, no. 2, pp. 347–356, 1997
9. Kamkar. B, A. Koocheki, M. Nassiri Mahallati, P. Revani Moghaddam. 2006. Cardinal temperatures for three millet specieses. Asian J Plant Sci. Vol. 5 (2). 316-319.
10. Matteucci M, D’Angeli S, Errico S, Lamanna R, Perrotta G, Altamura MM (2011) Cold affects the transcription of fatty acid desaturases and oil quality in the fruit of Olea europaea L. genotypes with different cold hardiness. J Exp Bot 62:3403–3420.
11. R. Bressan, H. Bohnert, and J.-K. Zhu, “Perspective: Abiotic stress tolerance: from gene discovery in model organisms to crop improvement,” Molecular Plant, vol. 2, no. 1, pp. 1–2, 2009.
12. Rab, A. and Saltveit, M.E., Differential chilling sensitivity in cucumber (Cucumis sativus) seedlings, Physiologia Plantarum, 96(3): 375–382 (1996a).
13. Ruelland E, Zachowski A (2010) How plants sense temperature. Environ Exp Bot 69:225–232.
14. Sarkar BC, Haque MM, Bashar MA, Roy B, Rahman MS. Physiological responses of rice sedling towards screening out cold tolerant rice cultivars in Northwest Bangaldesh. Asian Journal of Experimental Biological Sciences. 2013; 4(4):623-628.
15. Satake T, Hayase H. Male sterility caused by cooling treatment at the young microspore stage in rice plants. Estimation of pollen developmental stage and the most sensitive stage to coolness. Proceedings of the Crop Science Society of Japan. 1970; 39:468-473.
16. Shuangming Li, Yong Yang, Qiang Zhang, Ningfang Liu1 , Qingguo Xu , Longxing Hu. 2018. Differential physiological and metabolic response to low temperature in two zoysiagrass genotypes native to high and low latitude. Plos one. 1-20.
17. Suzuki N, Mittler R. Reactive oxygen species and temperature stresses: A delicate balance between signaling and destruction. Physiol Plant. 2006; 126:45–51. https://doi.org/10.1111/j.0031-9317.2005. 00582.x

18. Ting, C.S., Owens, T.G. and Wolfe, D.W., Seedling growth and chilling stress effect on photosynthesis in chillingsensitive and chilling ISSN 1392-3196 Zemdirbyste Agriculture, 99(2): (2012).
19. Uemura M, Steponkus PL (1999) Cold acclimation in plants: relationship between the lipid composition and the cryostability of the plasma membrane. J Plant Res 112:245–254.
20. Venema, J.H., Posthumus, F., de Vries, M. and van Hasselt, P.R., Differential response of domestic and wild Lycopersicon species to chilling under low light: growth, carbohydrate content, photosynthesis and the xanthophyll cycle, Physiologia Plantarum, 105(1): 81–88 (1999).
21. W. Y. Liu, Y. F. Zhang, and D. X. Zhang, "Study on cold resistant genes in plants," Journal of Shanxi Datong Unirersity, vol. 28, no. 6, pp. 52–55, 2012.
22. Zhu JH, Dong CH, Zhu JK (2007) Interplay between cold-responsive gene regulation, metabolism and RNA processing during plant cold acclimation. Curr Opin Plant Biol 10:290–295.
23. Pearce, R.B., R.H. Brown and R.E. Balaster. 1968. Photosynthesis of alfalfa leaves as influenced by environment. **Crop Sci., 36:** 677-680.
24. Rahul Kumar , P. C. Chaurasiya, R. N. Singh and Satyapal Singh (2018). A review report: Low temperature stress for crop production. Int. J. Pure App. Biosci. 6 (2): 575-598.
25. Yoshida, S.D., A. Foron and J.H. Cock. 1971. Laboratory Manual for Physiological Studies of Rice. IRRI, Philippines. pp.36-37.
26. Watson, D.J. 1956. Comparative physiological studies on the growth of field crops. I. Variation in net assimilation rate and leaf area between species and varieties and within and between years. **Ann. Bot., 11:** 41-46.