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## Foliar application of salicylic acid and calcium enhance morpho-physiological and yield contributing characters of tomato (*Lycopersicon esculentum* L.) by attenuating late planting induced chilling stress

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**ABSTRACT**

Now-a-days chilling stress has gained global concerned as it poses a grate economic loss in Agricultural sector. To arrest this loss an experiment was conducted to find out the role of exogenous foliar application of salicylic acid (SA) and calcium (Ca<sup>2+</sup>) on the changes of morpho-physiology and fruit yield of tomato under chilling injury. The experiment was laid out in two factors, (a) three different times of transplanting: T<sub>1</sub> = first/early transplanting time (1<sup>st</sup> November, 2016), T<sub>2</sub> = second transplanting time (16<sup>th</sup> November, 2016), T<sub>3</sub> = third transplanting time (1<sup>st</sup> December 2016) and (b) six different combination of salicylic acid (SA) and calcium (Ca<sup>2+</sup>) viz. A<sub>0</sub> = 0 mM of SA and 0 mM Ca<sup>2+</sup>, A<sub>1</sub> = 0.25 mM SA and 0 mM Ca<sup>2+</sup>, A<sub>2</sub> = 0 mM SA and 5 mM Ca<sup>2+</sup>, A<sub>3</sub> = 0.25 mM SA and 5 mM Ca<sup>2+</sup>, A<sub>4</sub> = 0 mM of SA and 10 mM Ca<sup>2+</sup> and A<sub>5</sub> = 0.25 mM SA and 10 mM Ca<sup>2+</sup> under RCBD (Randomized Complete Block Design) with three replications. Although the first/early transplanting time (T<sub>1</sub>) showed increased morpho-physiological characters such as plant height, number of branches plant<sup>-1</sup> and SPAD value of leaf as well as yield contributing characters such as number of flower clusters plant<sup>-1</sup>, number of flowers plant<sup>-1</sup>, number of fruits plant<sup>-1</sup>, fruit length, fruit diameter and yield compared to late transplanting induced cold injury, which was attenuated by exogenous foliar application of both salicylic acid and calcium at different combinations. Among the treatment combinations 0.25 mM SA along with 5 mM Ca<sup>2+</sup> performed better in all aspects.

**Keywords:** Calcium, chilling stress, morpho-physiological characters, salicylic acid, SPAD, yield



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## 1. INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is one of the most popular and cultivated horticultural species in the world including Bangladesh (Mohammad et al, 2014; Kerketta et al, 2018) because of its nutritive and phytochemical properties (Olaniyi et al, 2010; Shankar et al, 2013; Talens et al, 2016). It is an abundant source of vitamins, minerals, antioxidants and dietary fibers (Hedges and Lister, 2005; Frusciante et al, 2007; Olives et al, 2008) that reduces the prevalence of chronic diseases such as certain types of cancer, arteriosclerosis, cataract formation and as well as cardiovascular diseases (Di Mascio et al, 1989; Di Mascio Pandey et al, 1995; Giovannucci, 1999).

In Bangladesh, tomato has great demand throughout the year but its production is mainly focused during the winter season especially in early November whereas late planting results in lower growth and yield with higher insect and disease infestation (Hossain et al, 2014; Biswas et al, 2017; Ali et al, 2020; Hossain, 2021). The most favorable night temperature for tomato fruit setting ranges between 15-20 °C (Went, 1944) which only obtain during early November planting but after mid and late November planting receives a lower night temperature (around 5-10 °C) during fruit setting which exhibits a significant drop on yield of tomato (Li-yun et al, 2017) because low temperature impedes the development of male and female parts of the flowers (Kurosaki and Yumoto, 2003; Ohnishi et al, 2010) degrades pollen viability and germination (Charles and Harris, 1972) and alters various physiological and biochemical processes (Hussain et al, 2018).

To date several mechanisms are developed to attenuate low temperature induced chilling stress along with the improved growth and yield. Application of different Plant Growth Regulators (PGRs) such as auxin (Rahman, 2013; Shibasaki et al, 2009; Bielach et al, 2017), cytokinin (Veselov et al, 2017; Liu et al, 2020), Gibberellic acid (GA) (Ding et al, 2015; Yang et al, 2016), salicylic acid (SA) (Mutlu et al, 2013, 2016; Ijaz et al, 2014), Abscisic acid (ABA) (Janowiak et al., 2002, 2003), organic solute such as proline (Saghfi and Eivazi, 2014; Vera-Hernández et al, 2018; Masouleh et al, 2019), glycinebetaine (Nayyar et al, 2005; Rasheed et al, 2010) and inorganic minerals such as Calcium (Ca) (Yang et al, 2010; Furuya et al, 2013; Yuan et al, 2018), Magnesium (Mg) (Hussain et al, 2018), Silicon (Si) (Moradtalab et al, 2018), Potassium (K) (Hasanuzzaman et al, 2018) reduces the detrimental effect of chilling stress and improve the morpho-physiological parameters and yield of crops. Among these, applications of SA and/or Ca are the most using technique to mitigate the detrimental effect of chilling stress on growth and yield performance of crops due to their easy accessibility and economic viability.

Salicylic acid is a phenolic PGR (Plant Growth Regulator) found in plants with roles in plant growth, development, photosynthesis, transpiration, endogenous signal transduction, ion uptake and transport (Fariduddin et al, 2003; Yildirim and Dursun, 2008; Yildirim et al, 2008; Vlot et al, 2009). Besides SA acts as an antioxidant (Eraslan et al, 2007; Chen et al, 2016) that protect the plants against chilling stress by regulating the quantities of ROS (Reactive Oxygen Species) (Xia et al, 2009; Zhang et al 2011; Luo et al, 2012; Miura, K. and Tada, Y., 2014). On the other hand, besides functioning as an essential plant nutrient, Ca also plays a significant role in the structure of cell walls and cell membranes, plant growth and fruit development (Kadir, 2004; Hirschi, 2004; Thor, 2019; Singh, 2020) In addition Ca helps in mitigating diverse environmental stresses in plants (Waraich et al, 2012; Ibrahim et al, 2016; Wilkins et al, 2016; Roy et al, 2019; Malik et al, 2020).

Many studies have been accomplished to evaluate the ameliorative effect of SA or Ca in chilling stress on crops however there is insufficient information regarding combined application of SA and Ca in chilling stress extenuation on tomato. Therefore, considering the above facts, current study was planned to inspect the effects of combined foliar application of SA and Ca on morpho-physiological and yield attributes of tomato under chilling environment.

## 2. MATERIALS AND METHODS

### Location and climatic conditions of the experimental site

The experiment was carried out at the field laboratory of the Department of Agricultural Botany, Sher-e-Bangla Agricultural University, Dhaka, during the period from November, 2016 to February, 2017. The experimental area was in the subtropical climatic zone (23.81° N to 90.41° E), under Modhupur Tract AEZ (Agro-Ecological Zone) distinguished by low rainfall, humidity and temperature and shortday period during Rabi season (October to March).

### Plant materials and raising of seedlings

BARI tomato 1 (Manik) was used in the experiment. It is a high yielding variety with resistant to bacterial wilt. Seeds were collected from Bangladesh Agriculture Research Institute, Gazipur. Well drained and sandy loam land was selected for seed bed which was 3m<sup>2</sup>. Three seed beds were prepared with 50 cm spacing among them for seedling raising. Seed were grown separately in different seedbed for each treatment. Weeding and irrigation were done when required.

**Treatments of the experiment**

The two factorial experiments was as follows,

Factor A: Three different levels of transplanting dates

T<sub>1</sub>: 1<sup>st</sup> November (Early transplanting)

T<sub>2</sub>: 16<sup>th</sup> November (Late transplanting)

T<sub>3</sub>: 1<sup>st</sup> December (Very late transplanting)

Factor B: six different combinations of SA and Ca<sup>2+</sup>

A<sub>0</sub> = 0 mM of SA and 0 mM Ca<sup>2+</sup>,

A<sub>1</sub> = 0.25 mM SA and 0 mM Ca<sup>2+</sup>,

A<sub>2</sub> = 0 mM SA and 5 mM Ca<sup>2+</sup>,

A<sub>3</sub> = 0.25 mM SA and 5 mM Ca<sup>2+</sup>,

A<sub>4</sub> = 0 mM of SA and 10 mM Ca<sup>2+</sup> and

A<sub>5</sub> = 0.25 mM SA and 10 mM Ca<sup>2+</sup>

**Experimental layout and design**

The lay out of this experiment was RCBD with three replications. The experiment consists of 18 treatment combinations with 54 plots. The size of each plot was 1.8m × 1.5m.

**Land preparation and transplanting of seedlings**

The experimental land was ploughed several times with power tiller for good tilth then removed weeds and debris of the previous crops. The total land received similar amount of fertilizer and then the land was divided into 54 plots with a spacing 50cm among them. 30 days old healthy and uniform seedlings were shifted in the main plots on 1<sup>st</sup> November, 16<sup>th</sup> November and 1<sup>st</sup> December.

**Intercultural operations and application of foliar treatments**

Intercultural operations were accomplished when required and different compositions of SA and Ca<sup>2+</sup> were sprayed exogenously at 15, 30, and 45 DAT (Days After Transplanting) by a hand sprayer.

**Data collection**

Data on the following parameters were noted from the sample plants when required.

**Plant height**

Plant height was measured at 20, 40 and 60 DAT from root tip to shoot tip and expressed in cm.

**Number of branches plant<sup>-1</sup>**

The number of branches was counted manually from the sample plants at harvesting.

**SPAD value**

SPAD values were calculated by 502 plus chlorophyll meter from the sample plants at 40 DAT.

**Number of flower clusters plant<sup>-1</sup>**

The number of flower clusters plant<sup>-1</sup> was calculated manually from the sample plants and recorded at the time of final harvest.

**Number of flowers plant<sup>-1</sup>**

The number of flower plant<sup>-1</sup> was calculated manually from the sample plants and recorded at the time of final harvest. The formula was,

Number of flowers plant<sup>-1</sup> = Number of flowers per cluster × Number of flower cluster per plant

**Number of fruits plant<sup>-1</sup>**

The number of flower clusters plant<sup>-1</sup> was calculated manually from the sample plants and recorded at the time of final harvest.

**Fruit diameter (cm) and fruit length (cm)**

Fruit diameter was measured by a slide calipers at the middle position of samples and their average was recorded. The fruit length was measured with a measuring tape from the neck to the bottom of sample fruits and their average was noted.

**Yield in (kg plot<sup>-1</sup>) and (t ha<sup>-1</sup>)**

A digital balance was used to take the weight of fruits per plot from the period of first to final harvests and the yield per hectare in ton was calculated by following formula: Fruit yield per hectare (t) = Fruit yield per plot (kg)×10000/ Area of plot(m<sup>2</sup>)×1000

**Statistical analysis**

All the data obtained were statistically analyzed by using Minitab software to observe the significant differences at 5% level of probability. The mean values of all the characters were calculated and factorial analysis of variance was performed.

**3. RESULTS****Plant height (cm)**

Plant height at different DAT and foliar doses (SA + Ca<sup>2+</sup>) was found statistically significant (Table 1). At 60 DAT the plant height ranged between 55.08 cm to 75.66 cm. The tallest plant (75.66 cm) was found on 1<sup>st</sup> November transplanting with combined foliar application of 0.25 mM SA and 5 mM Ca<sup>2+</sup> (T<sub>1</sub>A<sub>3</sub>) which do not differ significantly on late transplanting at 16<sup>th</sup> November with combined foliar application of 0.25 mM SA and 5 mM Ca<sup>2+</sup> (T<sub>2</sub>A<sub>3</sub>) and 1<sup>st</sup> December transplanting with combined foliar application of 0.25 mM SA and 5 mM Ca<sup>2+</sup> (T<sub>3</sub>A<sub>3</sub>). The shortest plant (55.08 cm) was observed at T<sub>3</sub>A<sub>0</sub> (1<sup>st</sup> December transplanting with no foliar treatments).

**Table 1** Effect of combined foliar application of SA and Ca<sup>2+</sup> on plant height at different transplanting time.

Treatment combinations	Plant height (cm) at different days after transplanting (DAT)		
	20 DAT	40 DAT	60 DAT
T <sub>1</sub> A <sub>0</sub>	19.35 <sup>ef</sup>	33.46 <sup>defgh</sup>	64.06 <sup>ef</sup>
T <sub>1</sub> A <sub>1</sub>	22.87 <sup>ab</sup>	38.22 <sup>bc</sup>	73.41 <sup>ab</sup>
T <sub>1</sub> A <sub>2</sub>	22.12 <sup>bc</sup>	35.71 <sup>cdef</sup>	68.26 <sup>cd</sup>
T <sub>1</sub> A <sub>3</sub>	24.03 <sup>a</sup>	45.61 <sup>a</sup>	75.66 <sup>a</sup>
T <sub>1</sub> A <sub>4</sub>	21.14 <sup>cd</sup>	35.39 <sup>cdef</sup>	67.53 <sup>de</sup>
T <sub>1</sub> A <sub>5</sub>	22.53 <sup>abc</sup>	37.82 <sup>bcd</sup>	72.41 <sup>abc</sup>
T <sub>2</sub> A <sub>0</sub>	18.37 <sup>fg</sup>	29.36 <sup>hi</sup>	57.17 <sup>gh</sup>
T <sub>2</sub> A <sub>1</sub>	21.56 <sup>bcd</sup>	33.89 <sup>cdefg</sup>	69.85 <sup>bcd</sup>
T <sub>2</sub> A <sub>2</sub>	19.08 <sup>ef</sup>	32.23 <sup>fgh</sup>	63.39 <sup>ef</sup>
T <sub>2</sub> A <sub>3</sub>	23.11 <sup>ab</sup>	41.80 <sup>ab</sup>	73.44 <sup>ab</sup>
T <sub>2</sub> A <sub>4</sub>	18.64 <sup>efg</sup>	30.72 <sup>ghi</sup>	58.10 <sup>gh</sup>
T <sub>2</sub> A <sub>5</sub>	20.11 <sup>de</sup>	33.02 <sup>efgh</sup>	69.19 <sup>cd</sup>
T <sub>3</sub> A <sub>0</sub>	15.77 <sup>i</sup>	22.00 <sup>j</sup>	55.08 <sup>h</sup>
T <sub>3</sub> A <sub>1</sub>	18.18 <sup>fg</sup>	31.55 <sup>fgh</sup>	69.34 <sup>bcd</sup>
T <sub>3</sub> A <sub>2</sub>	17.21 <sup>ghi</sup>	29.22 <sup>hi</sup>	61.03 <sup>fg</sup>
T <sub>3</sub> A <sub>3</sub>	20.98 <sup>cd</sup>	37.45 <sup>bcde</sup>	72.13 <sup>abc</sup>
T <sub>3</sub> A <sub>4</sub>	16.56 <sup>hi</sup>	26.50 <sup>j</sup>	56.04 <sup>h</sup>
T <sub>3</sub> A <sub>5</sub>	17.98 <sup>fgh</sup>	30.43 <sup>ghi</sup>	62.36 <sup>f</sup>
LSD (0.05)	1.585	4.449	4.198
Level of signification	NS	*	*
CV (%)	4.85%	8.09%	4.88%

Here, T<sub>1</sub>=first transplanting time (1<sup>st</sup> November); T<sub>2</sub>=second transplanting time (16<sup>th</sup> November); T<sub>3</sub>=third transplanting time (1<sup>st</sup> December) and A<sub>0</sub>=0 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>1</sub>=0.25 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>2</sub>=0.25 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>3</sub>=0.25 mM SA + 10 mM Ca<sup>2+</sup>; A<sub>4</sub>=0.25 mM SA + 10 mM Ca<sup>2+</sup>; A<sub>5</sub>=0.25 mM SA + 10 mM Ca<sup>2+</sup>. Besides, CV= Co-efficient of variance; LSD= Least significant Difference and \*= Significant at 5% level. Values marked with the same letter within the columns do not differ significantly @ 5% level of confidence.

**SPAD value**

SPAD value of leaves of different transplanting time with different foliar doses (SA + Ca<sup>2+</sup>) was found statistically significant at 40 DAT (Table 2). The values ranged between from 52.60 to 61.63. The highest SPAD value (61.63) was obtained from the treatment combination of 1st November transplanting with SA 0.25 mM + Ca<sup>2+</sup> 5 mM (T<sub>1</sub>A<sub>3</sub>) that do not differ significantly at 16<sup>th</sup> November transplanting with same foliar treatment (T<sub>2</sub>A<sub>3</sub>). The lowest SPAD value (52.60) was obtained from very late transplanting (1<sup>st</sup> December) with no foliar treatments.

**Table 2** Effect of combined foliar application of SA and Ca<sup>2+</sup> on SPAD value at 40 DAT

Treatment combinations	SPAD value
T <sub>1</sub> A <sub>0</sub>	58.50 <sup>abc</sup>
T <sub>1</sub> A <sub>1</sub>	60.43 <sup>ab</sup>
T <sub>1</sub> A <sub>2</sub>	59.32 <sup>abc</sup>
T <sub>1</sub> A <sub>3</sub>	61.63 <sup>a</sup>
T <sub>1</sub> A <sub>4</sub>	59.27 <sup>abc</sup>
T <sub>1</sub> A <sub>5</sub>	60.33 <sup>ab</sup>
T <sub>2</sub> A <sub>0</sub>	54.43 <sup>efg</sup>
T <sub>2</sub> A <sub>1</sub>	57.11 <sup>cde</sup>
T <sub>2</sub> A <sub>2</sub>	56.41 <sup>cdef</sup>
T <sub>2</sub> A <sub>3</sub>	58.94 <sup>abcd</sup>
T <sub>2</sub> A <sub>4</sub>	55.13 <sup>defg</sup>
T <sub>2</sub> A <sub>5</sub>	56.28 <sup>cdef</sup>
T <sub>3</sub> A <sub>0</sub>	52.60 <sup>g</sup>
T <sub>3</sub> A <sub>1</sub>	55.27 <sup>defg</sup>
T <sub>3</sub> A <sub>2</sub>	53.89 <sup>fg</sup>
T <sub>3</sub> A <sub>3</sub>	56.33 <sup>cdef</sup>
T <sub>3</sub> A <sub>4</sub>	53.70 <sup>fg</sup>
T <sub>3</sub> A <sub>5</sub>	53.91 <sup>fg</sup>
LSD (0.05)	3.202
Level of signification	*
CV (%)	3.44

Here, T<sub>1</sub>=first transplanting time (1<sup>st</sup> November); T<sub>2</sub>=second transplanting time (16<sup>th</sup> November); T<sub>3</sub>=third transplanting time (1<sup>st</sup> December) and A<sub>0</sub>=0 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>1</sub>=0.25 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>2</sub>=0 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>3</sub>=0.25 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>4</sub>=0 mM SA + 10 mM Ca<sup>2+</sup>; A<sub>5</sub>=0.25 mM SA + 10 mM Ca<sup>2+</sup>. Besides, CV= Co-efficient of variance; LSD= Least significant Difference and \*= Significant at 5% level. Values marked with the same letter within the columns do not differ significantly @ 5% level of confidence.

**Number of branches plant<sup>-1</sup>**

Different transplanting time and foliar doses (SA + Ca<sup>2+</sup>) significantly influenced the number of branches plant<sup>-1</sup> at harvest. Number of branches plant<sup>-1</sup> varies from 4.22 to 10. Plants that transplanted at 1<sup>st</sup> November and received 0.25 mM SA and 5 mM Ca<sup>2+</sup> (T<sub>1</sub>A<sub>3</sub>) foliar treatment produced higher number of branches (10) which was not differ significantly with the branch number of plants that are transplanted lately at 16<sup>th</sup> November and received 0.25 mM SA and 5 mM Ca<sup>2+</sup> (T<sub>2</sub>A<sub>3</sub>). The lowest branch number plant<sup>-1</sup> was found in very late transplanting at 1<sup>st</sup> December with no foliar application.

**Number of flower clusters plant<sup>-1</sup>**

The interaction effect of various transplanting time and different combination of SA and Ca<sup>2+</sup> on number of flower clusters plant<sup>-1</sup> varied significantly that ranged from 5.8 to 24.44. The lowest number of flower clusters plant<sup>-1</sup> (5.8) was found at 1<sup>st</sup> December transplanting with no foliar application. In contrast November 1<sup>st</sup> transplanting with 0.25 mM SA + 5 mM Ca<sup>2+</sup> foliar treatment

revealed the maximum number of flower clusters plant<sup>-1</sup> (24.44) which was statistically similar to T<sub>2</sub>A<sub>3</sub> treatment (16<sup>th</sup> November transplanting with 0.25 mM SA + 5 mM Ca<sup>2+</sup> foliar treatment).

### Number of flowers plant<sup>-1</sup>

Significant variation was found on the number of flowers plant<sup>-1</sup> through the collaborative effect of different transplanting time and composition of SA and Ca<sup>2+</sup>. The number of flowers plant<sup>-1</sup> ranged from 118.6 to 180.8. The number of flowers plant<sup>-1</sup> was the highest (180.8) at 1<sup>st</sup> November transplanting with 0.25 mM SA + 5 mM Ca<sup>2+</sup> (T<sub>1</sub>A<sub>3</sub>) that does not differ with the transplanting time at 16<sup>th</sup> November with same foliar application (T<sub>2</sub>A<sub>3</sub>). Usually the lowest number of flowers plant<sup>-1</sup> was observed at 1<sup>st</sup> December transplanting with no foliar treatments (T<sub>3</sub>A<sub>0</sub>).

**Table 3** Effect of combined foliar application of SA and Ca<sup>2+</sup> on number of flower clusters plant<sup>-1</sup> at harvesting

Treatment combinations	Number of branches plant <sup>-1</sup>	Number of flower clusters plant <sup>-1</sup>	Number of flowers plant <sup>-1</sup>
T <sub>1</sub> A <sub>0</sub>	8.22 <sup>bcd</sup>	10.56 <sup>ghij</sup>	132.2 <sup>cdefg</sup>
T <sub>1</sub> A <sub>1</sub>	9.55 <sup>ab</sup>	19.78 <sup>bc</sup>	151.0 <sup>c</sup>
T <sub>1</sub> A <sub>2</sub>	8.77 <sup>abc</sup>	17.44 <sup>cde</sup>	144.6 <sup>bc</sup>
T <sub>1</sub> A <sub>3</sub>	10.00 <sup>a</sup>	24.44 <sup>a</sup>	180.8 <sup>a</sup>
T <sub>1</sub> A <sub>4</sub>	8.66 <sup>abc</sup>	13.78 <sup>fg</sup>	137.0 <sup>bcdef</sup>
T <sub>1</sub> A <sub>5</sub>	9.22 <sup>ab</sup>	18.56 <sup>bcd</sup>	146.8 <sup>bc</sup>
T <sub>2</sub> A <sub>0</sub>	7.00 <sup>def</sup>	8.66 <sup>ijk</sup>	124.4 <sup>fg</sup>
T <sub>2</sub> A <sub>1</sub>	8.33 <sup>bcd</sup>	15.67 <sup>def</sup>	146.2 <sup>bc</sup>
T <sub>2</sub> A <sub>2</sub>	7.44 <sup>cde</sup>	12.41 <sup>fgh</sup>	137.0 <sup>bcdef</sup>
T <sub>2</sub> A <sub>3</sub>	9.11 <sup>ab</sup>	21.44 <sup>ab</sup>	173.0 <sup>ab</sup>
T <sub>2</sub> A <sub>4</sub>	7.22 <sup>de</sup>	11.22 <sup>ghi</sup>	133.1 <sup>cdefg</sup>
T <sub>2</sub> A <sub>5</sub>	7.22 <sup>de</sup>	14.89 <sup>ef</sup>	143.9 <sup>bcd</sup>
T <sub>3</sub> A <sub>0</sub>	4.22 <sup>i</sup>	5.81 <sup>k</sup>	118.6 <sup>g</sup>
T <sub>3</sub> A <sub>1</sub>	5.66 <sup>fgh</sup>	12.78 <sup>fgh</sup>	140.6 <sup>bcde</sup>
T <sub>3</sub> A <sub>2</sub>	5.55 <sup>ghi</sup>	9.33 <sup>hijk</sup>	128.7 <sup>defg</sup>
T <sub>3</sub> A <sub>3</sub>	6.22 <sup>efg</sup>	15.45 <sup>def</sup>	150.4 <sup>b</sup>
T <sub>3</sub> A <sub>4</sub>	4.77 <sup>hi</sup>	7.11 <sup>jk</sup>	126.8 <sup>efg</sup>
T <sub>3</sub> A <sub>5</sub>	5.22 <sup>ghi</sup>	10.55 <sup>ghij</sup>	133.2 <sup>cdefg</sup>
LSD (0.05)	1.411	3.618	15.3
Level of signification	*	*	*
CV (%)	11.71	15.91	6.06

Here, T<sub>1</sub>=first transplanting time (1<sup>st</sup> November); T<sub>2</sub>=second transplanting time (16<sup>th</sup> November); T<sub>3</sub>=third transplanting time (1<sup>st</sup> December) and A<sub>0</sub>=0 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>1</sub>=0.25 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>2</sub>=0 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>3</sub>=0.25 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>4</sub>=0 mM SA + 10 mM Ca<sup>2+</sup>; A<sub>5</sub>=0.25 mM SA + 10 mM Ca<sup>2+</sup>. Besides, CV= Co-efficient of variance; LSD= Least significant Difference and \*= Significant at 5% level. Values noted with the same letter within the columns do not differ significantly @ 5% level of confidence.

### Number of fruits plant<sup>-1</sup>

Number of fruits plant<sup>-1</sup> varied significantly with the interaction effect of different transplanting time and foliar application of SA and Ca<sup>2+</sup> (Table 4). Number of fruits plant<sup>-1</sup> at various treatment combinations ranged from 56 to 110.5. The highest number of fruits plant<sup>-1</sup> (110.5) was found at T<sub>1</sub>A<sub>3</sub> treatment i.e. 1<sup>st</sup> November transplanting along with 0.25 mM SA + 5 mM Ca<sup>2+</sup> foliage treatment. The number of fruits plant<sup>-1</sup> do not differ significantly on late transplanting at 16<sup>th</sup> November with combined foliar application of 0.25 mM SA and 5 mM Ca<sup>2+</sup> (T<sub>2</sub>A<sub>3</sub>) and 1<sup>st</sup> December transplanting with combined foliar application of 0.25 mM SA and 5 mM Ca<sup>2+</sup> (T<sub>3</sub>A<sub>3</sub>) respectively. On the other hand the lowest number of fruits plant<sup>-1</sup> (56) was attained from control treatment with 1<sup>st</sup> December transplanting (T<sub>3</sub>A<sub>0</sub>).



**Fruit diameter (cm)**

Interaction between various transplanting times and foliage treatment on fruit diameter was statistically significant. The highest fruit diameter (5.92 cm) was observed from 1<sup>st</sup> November transplanting with 0.25 mM SA + 5 mM Ca<sup>2+</sup> foliage treatment (T<sub>1</sub>A<sub>3</sub>) which was statistically identical with late transplanting at 16<sup>th</sup> November with T<sub>2</sub>A<sub>3</sub> 0.25 mM SA + 5 mM Ca<sup>2+</sup> foliage treatment (T<sub>2</sub>A<sub>3</sub>) and very late transplanting at 1<sup>st</sup> December with 0.25 mM SA + 5 mM Ca<sup>2+</sup> foliage application (T<sub>3</sub>A<sub>3</sub>). The lowest fruit diameter (5.04 cm) was found at 1<sup>st</sup> December transplanting with no foliage application (T<sub>3</sub>A<sub>0</sub>).

**Fruit length (cm)**

Interaction between different transplanting time and compositions of SA and Ca<sup>2+</sup> showed significant variation on fruit length (Table 4) which as ranged from 5.48 to 6.70. The highest fruit length (6.70) was observed from the T<sub>1</sub>A<sub>3</sub> treatment which was statistically identical with T<sub>2</sub>A<sub>3</sub> (6.58) whereas, the lowest fruit length (5.48) was observed from T<sub>3</sub>A<sub>0</sub> treatment.

**Table 4** Effect of combined foliar application of SA and Ca<sup>2+</sup> on number of fruits plant<sup>-1</sup> at harvesting

Treatment combinations	Number of fruits plant <sup>-1</sup>	Fruit diameter (cm)	Fruit length (cm)
T <sub>1</sub> A <sub>0</sub>	68.89 <sup>efghi</sup>	5.17 <sup>def</sup>	5.73 <sup>g</sup>
T <sub>1</sub> A <sub>1</sub>	91.56 <sup>bc</sup>	5.61 <sup>abc</sup>	6.49 <sup>bc</sup>
T <sub>1</sub> A <sub>2</sub>	82.57 <sup>bcde</sup>	5.46 <sup>bcdef</sup>	6.30 <sup>cde</sup>
T <sub>1</sub> A <sub>3</sub>	110.5 <sup>a</sup>	5.92 <sup>a</sup>	6.70 <sup>a</sup>
T <sub>1</sub> A <sub>4</sub>	78.11 <sup>cdefg</sup>	5.40 <sup>bcdef</sup>	6.11 <sup>ef</sup>
T <sub>1</sub> A <sub>5</sub>	86.66 <sup>bcd</sup>	5.50 <sup>abcde</sup>	6.40 <sup>bc</sup>
T <sub>2</sub> A <sub>0</sub>	63.22 <sup>hi</sup>	5.11 <sup>ef</sup>	5.65 <sup>gh</sup>
T <sub>2</sub> A <sub>1</sub>	89.66 <sup>bcd</sup>	5.55 <sup>abcd</sup>	6.39 <sup>bcd</sup>
T <sub>2</sub> A <sub>2</sub>	80.22 <sup>cdef</sup>	5.39 <sup>bcdef</sup>	6.16 <sup>ef</sup>
T <sub>2</sub> A <sub>3</sub>	96.44 <sup>ab</sup>	5.70 <sup>ab</sup>	6.58 <sup>ab</sup>
T <sub>2</sub> A <sub>4</sub>	75.67 <sup>defgh</sup>	5.23 <sup>cdef</sup>	5.81 <sup>g</sup>
T <sub>2</sub> A <sub>5</sub>	87.44 <sup>bcd</sup>	5.47 <sup>bcdef</sup>	6.22 <sup>def</sup>
T <sub>3</sub> A <sub>0</sub>	56.00 <sup>i</sup>	5.04 <sup>f</sup>	5.48 <sup>h</sup>
T <sub>3</sub> A <sub>1</sub>	82.33 <sup>bcde</sup>	5.39 <sup>bcdef</sup>	6.12 <sup>ef</sup>
T <sub>3</sub> A <sub>2</sub>	67.56 <sup>fghi</sup>	5.26 <sup>cdef</sup>	5.83 <sup>g</sup>
T <sub>3</sub> A <sub>3</sub>	91.33 <sup>bc</sup>	5.65 <sup>abc</sup>	6.26 <sup>de</sup>
T <sub>3</sub> A <sub>4</sub>	64.78 <sup>ghi</sup>	5.15 <sup>def</sup>	5.77 <sup>g</sup>
T <sub>3</sub> A <sub>5</sub>	78.00 <sup>cdefg</sup>	5.28 <sup>bcdef</sup>	6.03 <sup>f</sup>
LSD (0.05)	14.42	0.4302	0.1938
Level of signification	*	*	*
CV (%)	10.92	4.86	1.94

Here, T<sub>1</sub>=first transplanting time (1<sup>st</sup> November); T<sub>2</sub>=second transplanting time (16<sup>th</sup> November); T<sub>3</sub>=third transplanting time (1<sup>st</sup> December) and A<sub>0</sub>=0 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>1</sub>=0.25 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>2</sub>=0 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>3</sub>=0.25 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>4</sub>=0 mM SA + 10 mM Ca<sup>2+</sup>; A<sub>5</sub>=0.25 mM SA + 10 mM Ca<sup>2+</sup>. Besides, CV= Co-efficient of variance; LSD= Least significant Difference and \*= Significant at 5% level. Values noted with the identical letter within the columns do not differ significantly @ 5% level of confidence.

**Yield in (kg plot<sup>-1</sup>) and (t ha<sup>-1</sup>)**

The interaction effect between transplanting date and different composition of SA and Ca<sup>2+</sup> showed a significant effect on fruit yield of tomato (Table 5). The highest yield (23.39 kg plot<sup>-1</sup>) and (86.62 t ha<sup>-1</sup>) were recorded from the T<sub>1</sub>A<sub>3</sub> treatment which was statistically similar with the values (19.94 kg plot<sup>-1</sup> and 73.84 t ha<sup>-1</sup> respectively) of late transplanting at 16<sup>th</sup> November with 0.25 mM SA + 5 mM Ca<sup>2+</sup> treatment. In contrast, the lowest (9.88 kg plot<sup>-1</sup>) and (36.59 t ha<sup>-1</sup>) yield was observed from T<sub>3</sub>A<sub>0</sub> treatment (1<sup>st</sup> December transplanting with 0 mM SA + 0 mM Ca<sup>2+</sup>).

**Table 5** Effect of combined foliar application of SA and Ca<sup>2+</sup> on yield at harvesting

Treatment combinations	Yield kg plot <sup>-1</sup>	Yield t ha <sup>-1</sup>
T <sub>1</sub> A <sub>0</sub>	14.44 <sup>def</sup>	53.49 <sup>def</sup>
T <sub>1</sub> A <sub>1</sub>	19.90 <sup>b</sup>	73.72 <sup>b</sup>
T <sub>1</sub> A <sub>2</sub>	16.50 <sup>cd</sup>	61.10 <sup>cd</sup>
T <sub>1</sub> A <sub>3</sub>	23.39 <sup>a</sup>	86.62 <sup>a</sup>
T <sub>1</sub> A <sub>4</sub>	15.60 <sup>cde</sup>	57.77 <sup>cde</sup>
T <sub>1</sub> A <sub>5</sub>	17.83 <sup>bc</sup>	66.05 <sup>bc</sup>
T <sub>2</sub> A <sub>0</sub>	12.96 <sup>efg</sup>	47.99 <sup>efg</sup>
T <sub>2</sub> A <sub>1</sub>	17.99 <sup>bc</sup>	66.64 <sup>bc</sup>
T <sub>2</sub> A <sub>2</sub>	14.77 <sup>cdef</sup>	54.71 <sup>cdef</sup>
T <sub>2</sub> A <sub>3</sub>	19.94 <sup>b</sup>	73.84 <sup>b</sup>
T <sub>2</sub> A <sub>4</sub>	13.09 <sup>efg</sup>	48.49 <sup>efg</sup>
T <sub>2</sub> A <sub>5</sub>	15.52 <sup>cde</sup>	57.47 <sup>cde</sup>
T <sub>3</sub> A <sub>0</sub>	9.88 <sup>g</sup>	36.59 <sup>g</sup>
T <sub>3</sub> A <sub>1</sub>	12.59 <sup>efg</sup>	46.63 <sup>efg</sup>
T <sub>3</sub> A <sub>2</sub>	12.09 <sup>fg</sup>	44.78 <sup>fg</sup>
T <sub>3</sub> A <sub>3</sub>	15.46 <sup>cde</sup>	57.26 <sup>cde</sup>
T <sub>3</sub> A <sub>4</sub>	11.85 <sup>fg</sup>	43.89 <sup>fg</sup>
T <sub>3</sub> A <sub>5</sub>	12.04 <sup>fg</sup>	44.59 <sup>fg</sup>
LSD (0.05)	3.248	12.03
Level of signification	*	*
CV (%)	12.94	12.94

Here, T<sub>1</sub>=first transplanting time (1<sup>st</sup> November); T<sub>2</sub>=second transplanting time (16<sup>th</sup> November); T<sub>3</sub>=third transplanting time (1<sup>st</sup> December) and A<sub>0</sub>=0 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>1</sub>=0.25 mM SA + 0 mM Ca<sup>2+</sup>; A<sub>2</sub>=0 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>3</sub>=0.25 mM SA + 5 mM Ca<sup>2+</sup>; A<sub>4</sub>=0 mM SA + 10 mM Ca<sup>2+</sup>; A<sub>5</sub>=0.25 mM SA + 10 mM Ca<sup>2+</sup>. Besides, CV= Co-efficient of variance; LSD= Least significant Difference and \*= Significant at 5% level. Values noted with the same letter within the columns do not differ significantly @ 5% level of confidence.

#### 4. DISCUSSION

Planting/transplanting time and temperature are considered as the key determinants of growth, productivity and yield of crops. So, suitable planting/transplanting time is indispensable for improved crop production. Therefore, the best transplanting time of tomato for advanced growth and higher yield in Bangladesh is between 1<sup>st</sup> to 7<sup>th</sup> November (Biswas et al, 2017; Ali et al, 2020) but decreases trend is observed with decreasing temperature because of late planting (Lawahori et al, 1963; Islam et al, 2017; Ali et al, 2020). According to Kumar et al, (2018) tropical plants are more susceptible to chilling stress, being a tropical plant, late transplanting of tomato observes a lower night temperature (around 5-10 °C) that impede the development of male and female flowers parts, pollen viability, germination and finally fruit setting (Kurosaki and Yumoto, 2003; Ohnishi et al, 2010; Li-yun et al, 2017). Chilling stress alters plant metabolisms by forming ice within the plant cells which cause dehydration inside the plant. This adverse condition lowers the rate of water and nutrient uptake leading to cell desiccation and starvation and finally degrades both crop quality and quantity (Miura and Tada, 2014). Again chilling stress reduces N uptake and partitioning in shoots (Kumar et al, 2018) that decreases plant vegetative growth and thus yield. Besides, hyper osmotic stress due to chilling stress opens various alternative pathway (Dwivedi and Raghavendra, 2004), leading to cause oxidative damage in plants (Imahori et al, 2008; Abdel Kader et al, 2011).

In this study, the detrimental effect of late transplanting induced chilling stress was attenuated by combined application of SA and Ca. SA has been known to play diverse positive functions during plant acclimation to chilling stresses (Ahanger et al, 2004; Hernández et al, 2017). It controls plant performance including advanced growth, nutrient absorption, protein synthesis, cell division and differentiation that allied to an increase yield (El Tayeb and Ahmed, 2010; Askari and Ehsanzadeh, 2015). Besides, SA regulates ionic balance, photosynthetic activity and ROS detoxification that bestow normal physiological and biochemical behaves



(Ahmad et al, 2017; Nooren et al, 2017; Nie et al, 2018). We found similar consequences in winter wheat (Ta,sgin et al, 2003), green bell pepper (Fung et al, 2004), bamboo shoot (Luo et al., 2012), mustard (Setia et al, 2006), and cucumber (Zhang et al, 2011).

Besides an essential plant nutrient, Ca acts as a signaling molecule which regulates different physiological and biochemical processes in plants that influences both growth and yield in responses to chilling stress (Palta, 2000; Mahmood-ur-Rahman et al, 2019). Generally, plants escape low temperature stress through maintaining high leaf water potential by closing their stomata (Wilkinson et al, 2001). Foliar application of Ca provokes stomatal closure and protects the leaves from dehydration at chilling environment (Waraich et al., 2012).

Again Ca plays an important role in the maintenance of cell structure and it triggers the plasma membrane ATPase which is involved to pump back the nutrients that were lost in cell damage (Waraich et al., 2012). Ca also performances a role as calmodulin which regulates the plant metabolic activities and enhances the plant growth under low temperature stress (Waraich et al., 2011, 2012). Our recent study also reveals that high concentration (10 mM) of Ca<sup>2+</sup> application had less performance than low concentration (5 mM) application. It is assumed that ABA induced stomatal closure is partially mediated by Ca<sup>2+</sup> (Wilkinson et al, 2001), so surplus application of Ca<sup>2+</sup> leads stomatal closure by keeping the guard cell turgid (Agurla et al, 2018) that hinders photosynthesis, carbon and N partitioning and finally decrease growth and yield.

## 5. CONCLUSION

Late planting induced chilling stress mediated growth and yield reduction could be regained through a foliar application at 0. 25 mM SA + 5 mM Ca<sup>2+</sup> concentrations.

### Conflict of interest

The authors declare that they have no conflict of interest.

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### Ethical approval

This article does not contain any studies with human participants performed by any of the authors.

### Data and materials availability

All data associated with this study are present in the paper.

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