

## Original Research Article

### **Synthetic PGR's modify Phenology, Stress Tolerance and Mean Productivity in wilted and cold stressed chickpea (*Cicer arietinum* L.)**

#### **ABSTRACT**

**Comparative** study of four synthetic PGRs—Abscisic Acid (ABA), Naphthyl Acetic Acid (NAA), Salicylic Acid (SA), and Fusaric Acid (FA) — was done as pot experiment against artificially inoculated *Fusarium oxysporum* (wilt pathogen) and cold exposure in four chickpea varieties. This experiment was done to know about phenophasic response to mitigate stress tolerance for achieving maximum mean productivity. The results showed that spraying plants with ABA @ 5 and 2 ppm were effective in extending the vegetative growth period by delaying flowering, allowing flowering to avoid the negative effects of wilt and cold, thus facilitating stress tolerance. This resulted in reduced percentage of wilt and cold incidence as compared to all other treatments, leading to higher mean productivity. However, Fusaric acid (FA), a fungal toxin responsible for early in flowering that allowed flower to coincide with wilt and cold occurrence timing. Flowering is also very sensitive to cold stress therefore occurrence of wilt at seedling and cold at flowering accelerated the incidence of wilt and cold, which resulted in lower mean productivity after applying Fusaric acid (FA) @ 10 and 20ppm.

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**Key point-** Phenology, Abscisic Acid (ABA), Wilt Incidence, Cold Incidence, Mean Productivity.

**Introduction** - Chickpea (*Cicer arietinum* L.) is the third most important winter-season grain legume crop after dry beans and field peas, and it is an important component of crop rotation worldwide. Chickpeas are currently farmed on around 13.8 million ha of land worldwide, producing 13.65 million tons and yielding 9.83 q ha<sup>-1</sup> (citation). In India, it is grown on around 10.5 million ha, producing 11.15 million tons and 10.56 q ha<sup>-1</sup> productivity. The total area of chickpea in Madhya Pradesh is 3.5 million ha, with a production of 4.59 million tons and a productivity of 10.82 q ha<sup>-1</sup> (2). Chickpea seeds contain a large amount of protein (220 g kg<sup>-1</sup>), total carbohydrate (670 g kg<sup>-1</sup>), starch (470 g kg<sup>-1</sup>) and fat (50 g kg<sup>-1</sup>). As a result, they play a significant role in the nutritional system of humans (10). When chickpeas experience cold stress during the reproductive phase, especially during flowering and pod formation, abortion of flowers, poor pod set, infertile pods, and abortion of pods are found (14). Temperatures up to 15 °C have been demonstrated to cause flower and pod abortion, and temperatures lower than 10°C at

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flowering can reduce grain yield by 15-20% (12). In addition to this, chickpea production is severely curtailed by Fusarium wilt caused by *Fusarium Oxysporum* in most chickpea growing areas of the world. It is considered a major exotic disease of and annual chickpea yield losses from Fusarium wilt vary from 10–15% but can result in total loss of the crop under specific conditions (6).

Knowledge about the timing of the phenological events and their variability can provide valuable information for the planning, organizing, and timely execution of our farm activities. This knowledge is also helpful in making adjustments to farm activity to save our farm's produce in response to predictable climate change (21). The phenological stages of chickpea growth may be broadly classified as emergence, flowering, pod initiation, pod set, seed maturation, and physiological maturity. Being indeterminate, these stages occur simultaneously in different parts of the plant along with vegetative growth. Flowering is considered the critical stage because the environmental conditions that prevail at flowering and the duration of the reproductive phase determine the percentage of fruit set and the final yield (20) (Ahmed 2019; Alyafei et al., 2022). Plant hormones are generally organic compounds that are responsible for modifying the developmental pattern and yield response of crops by preventing fruit and flower drop for a longer period of time (Hassan et al., 2023; Kaur et al., 2023). Plants have developed complex mechanisms to detect external signals and can trigger an optimal response against stress conditions with the support of PGRs that mainly control the defensive responses of plants through synergistic and antagonistic activities called signaling crosstalk (16) (Khalil et al., 2022). PGRs like of Fusaric Acid, NAA, ABA, Salicylic acid also play an important role during stress conditions because they act as thermo protectants, reactive oxygen scavengers, improve photosynthesis, accumulate stress proteins, and perform many other regulatory functions related to metabolism (19). Therefore present investigations are aimed to study the Influence of different phenophases on productivity in response to PGRs under wilt and cold stressed chickpea (*Cicer arietinum L.*).

**Materials and methods** - The studies were carried out at the Herbal Garden Department of Plant Physiology, JNKVV, Jabalpur (MP) in the years 2020-2021 and 2021-2022. Crop was planted in pots in factorial completely randomized design with four varieties: V<sub>1</sub> (JG74, susceptible to wilt), V<sub>2</sub> (JG11, susceptible to cold), V<sub>3</sub> (RAJAS, resistance to wilt), and V<sub>4</sub> (PBG5, tolerant to cold), as well as PGRs, Abscisic Acid (ABA), Naphthyl Acetic Acid (NAA), Salicylic Acid (SA), and Fusaric Acid (FA) at various concentrations. Spraying was performed two times, once during the early seedling stage and again at the flower initiation stage.

#### Method of wilt and cold application

**Comment [ZA4]:** 1. Alyafei, M. A.S, Al Dakheel, A., Almoosa, M., Ahmed, Z.F.R. (2022). Artificial pollination of date palm (*Phoenix dactylifera L.*) using a platform mounted robotic pollination system (Drone). HortScience, 57(10):1298–1305.

**Comment [ZA5]:** 2. Ahmed, Z.F.R (2019). Flowering, fruiting of two table olive cultivars "Olea europaea L." grown in Southern Egypt. Egyptian Journal of Horticulture, 46(1), pp.145-153.

**Comment [ZA6]:** 3. Hassan, F.A., Alshamsi, A.S.M., Alyafei, M.A.S., Kurup, S., Al Busaidi, N. and Ahmed, Z.F.R. (2023). Enhancing germination of ghaf seeds (*Prosopis cineraria L.*) using sulfuric acid scarification and cytokinin. Acta Hort. 1365, 39-44

**Comment [ZA7]:** 4. Kaur, N., Shahwar, D., Hassan, F.E. and Ahmed, Z.F.R. (2023). Antioxidant and antibacterial activities of date palm fruit (*Phoenix dactylifera L.*) in response to postharvest application with natural elicitors. Acta Hort. 1364, 187-194

#### Comment [ZA8]:

**Comment [ZA9]:** Khalil, H; ZFR Ahmed (2022) Salicylic Acid Mitigates the Adverse Effects of Salinity Stress on Pomegranate (*Punica granatum L.* cv. Wonderful) by Activating the Antioxidant Enzymatic Mechanism, Protecting Morphological Structure, and Regulating Lipid Peroxidation Level. Horticulturae. 8(5), 375.

When crops reach 20 DAS stage, an equal amount of wilt affected soil and maize flour containing Fusarium spores is applied to each pot, and when plants exhibited wilting like symptoms, the first foliar spray of different PGRs was done as per treatment details. Cold treatment was artificially done at floral initiation stage. For this, at midnight during the cold hour, an equal amount of crushed ice was applied to each pot. After applying wilt and cold treatments, a second spraying of plant was done.

**Phenological studies** - The phenological development of the crop was monitored by visual observations at 2-day intervals for all the crop seasons, beginning from the onset of flowering until maturity.

**Wilt incidence (%)** - The number of plants that showed visible observations such as wilting, chlorosis, and browning of the vascular system after the application of wilt inoculum in pot was counted. It was carried out at 30 DAS, and the % wilt incidence was estimated using the Mayee and Datar (1986) (11) formula.

**Cold incidence (%)** - It was carried out at 48 DAS, and the cold % incidence was calculated using the method proposed by Mayee and Datar (1986) (11), with appropriate modifications.

**Mean productivity** - Mean Productivity was calculated by using formula -

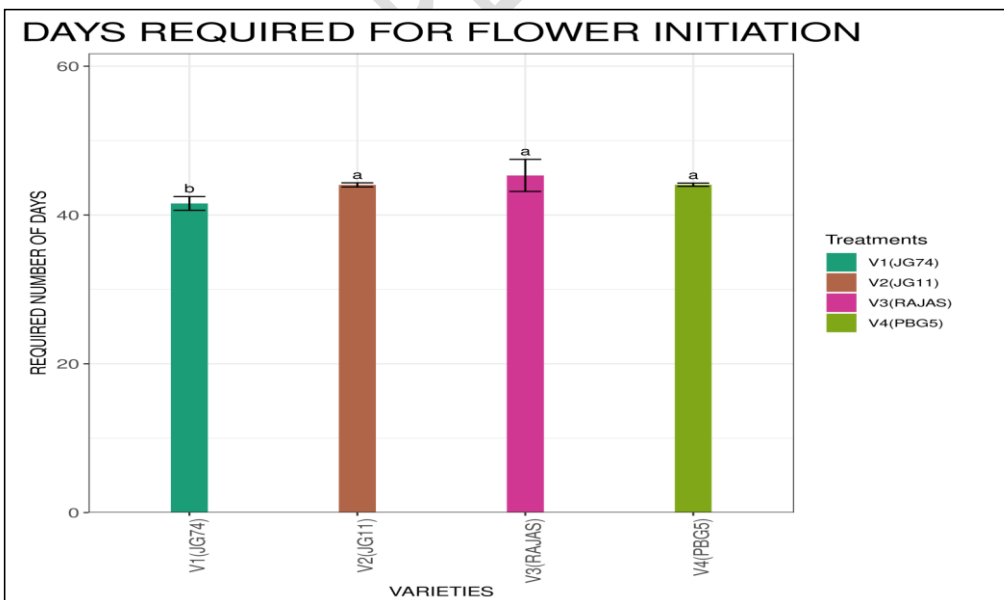
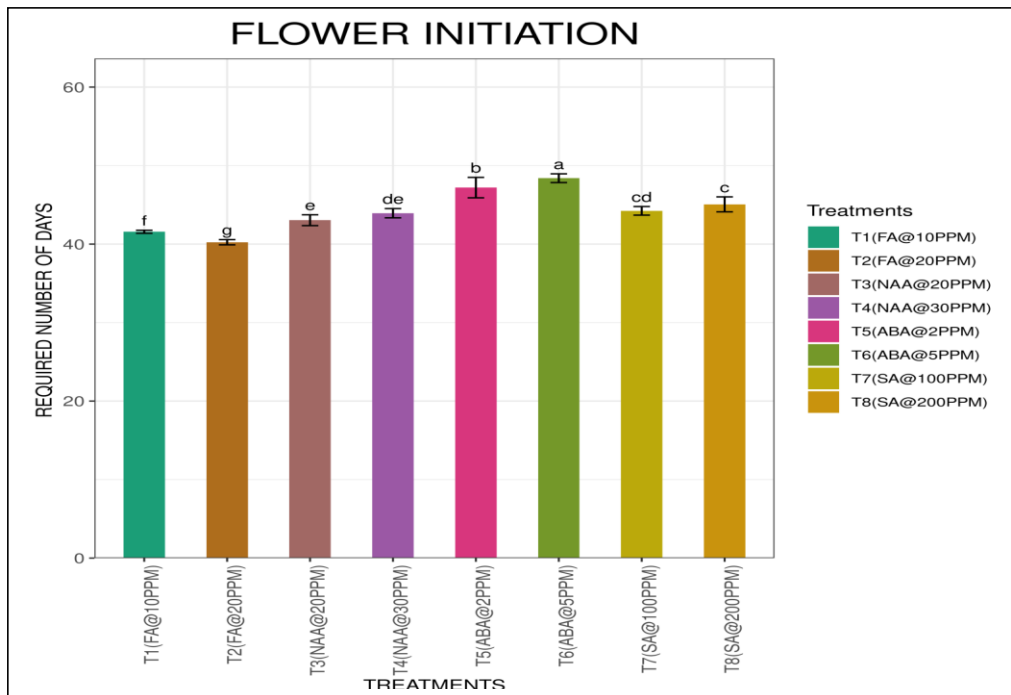
$$MP = (Y_{pi} + Y_{si}) / 2$$

$Y_{si}$  and  $Y_{pi}$  are the mean grain yields of individual treatments under stressed and non-stressed conditions.

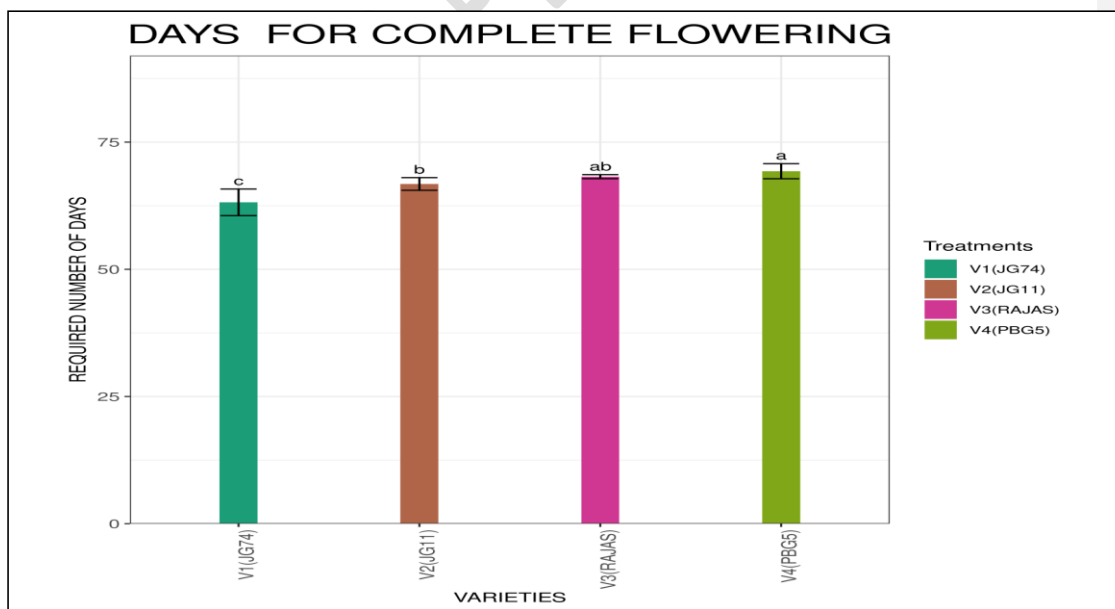
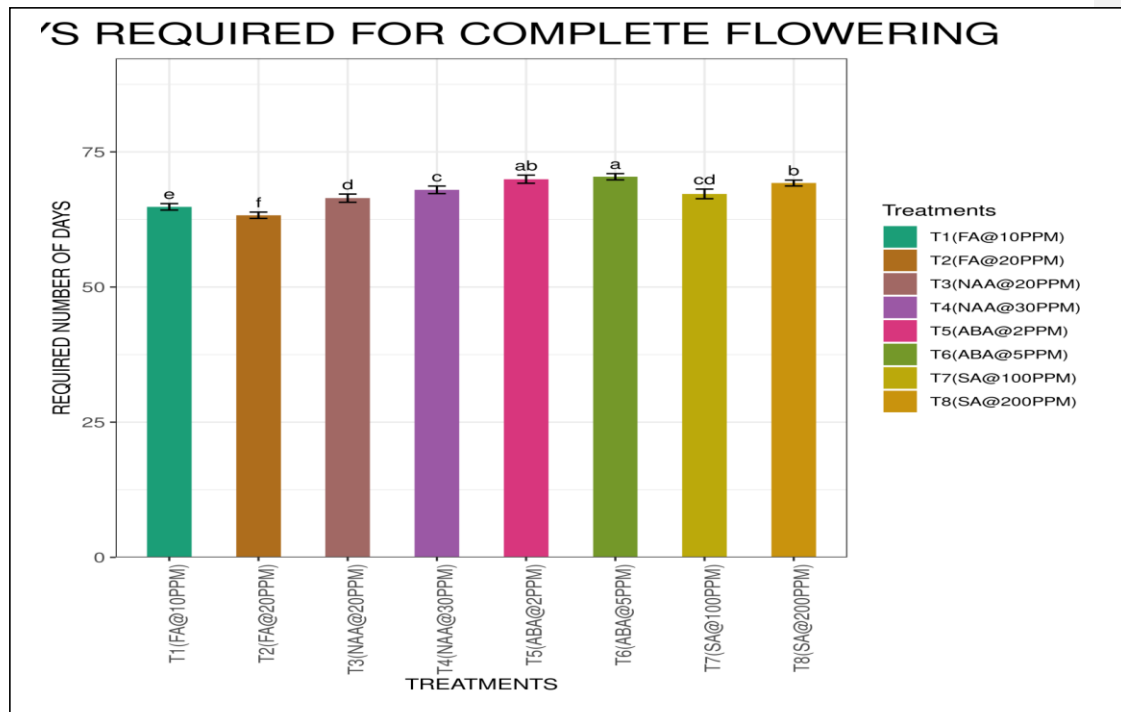
**Statistical analysis** - The data were statistically analyzed through completely randomized design given by Fisher (1955) (7) and comparison of mean was performed on the basis of least significant difference test (LSD) according to method given by Gopinath *et al.*, (2021) (9).

**Result and discussion** – Plant phenology is a critical component of crop adaptation, especially under environmental conditions that don't allow crop growth for unlimited periods (5).

**FIGURE 1– Effects of PGRs and Varieties on number of days required for flower initiation in wilt and cold stressed chickpea.**



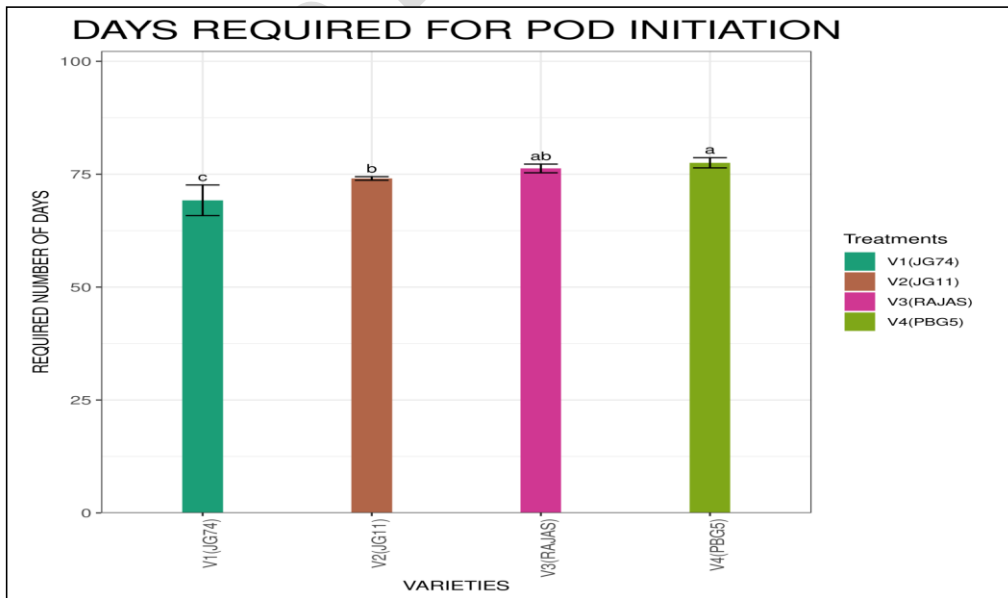
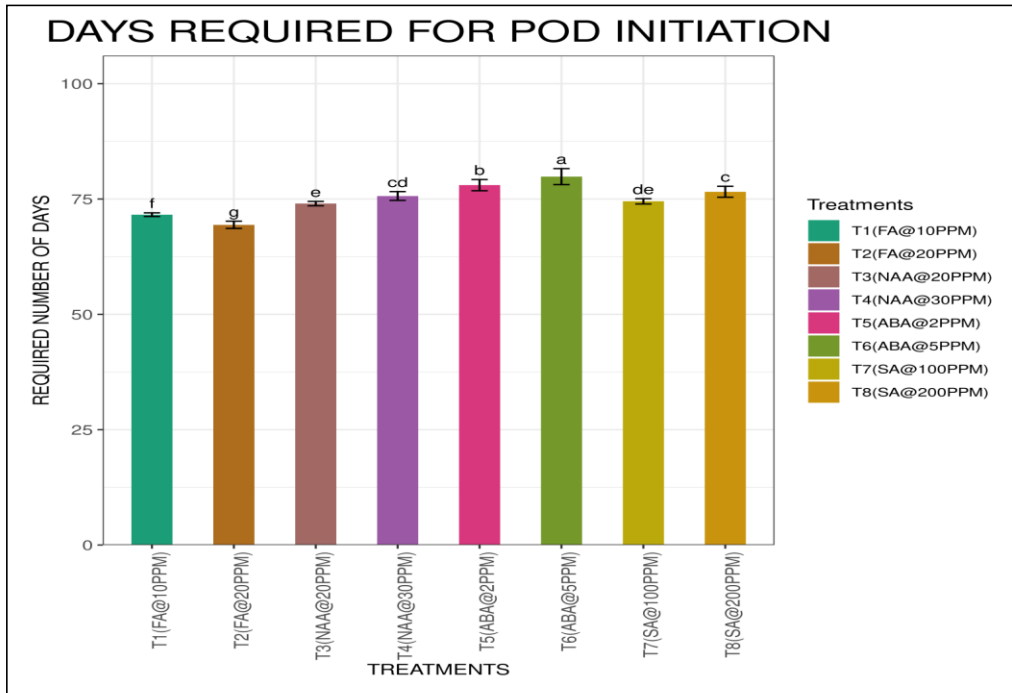
**FIGURE 2– Effects of PGRs and Varieties on number of days required for complete flowering in wilt and cold stressed chickpea.**



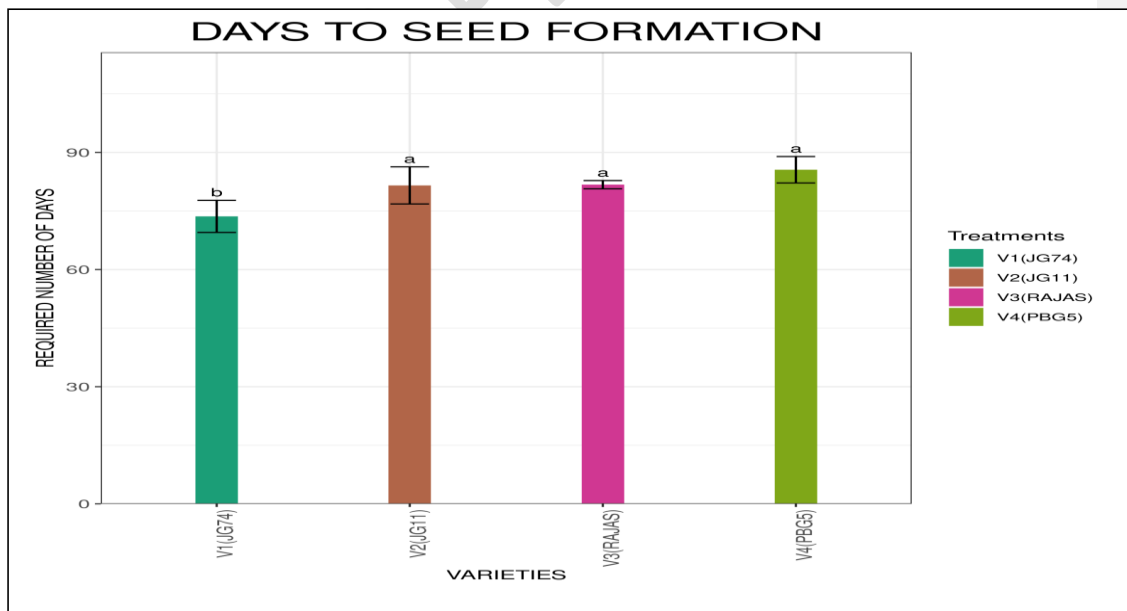
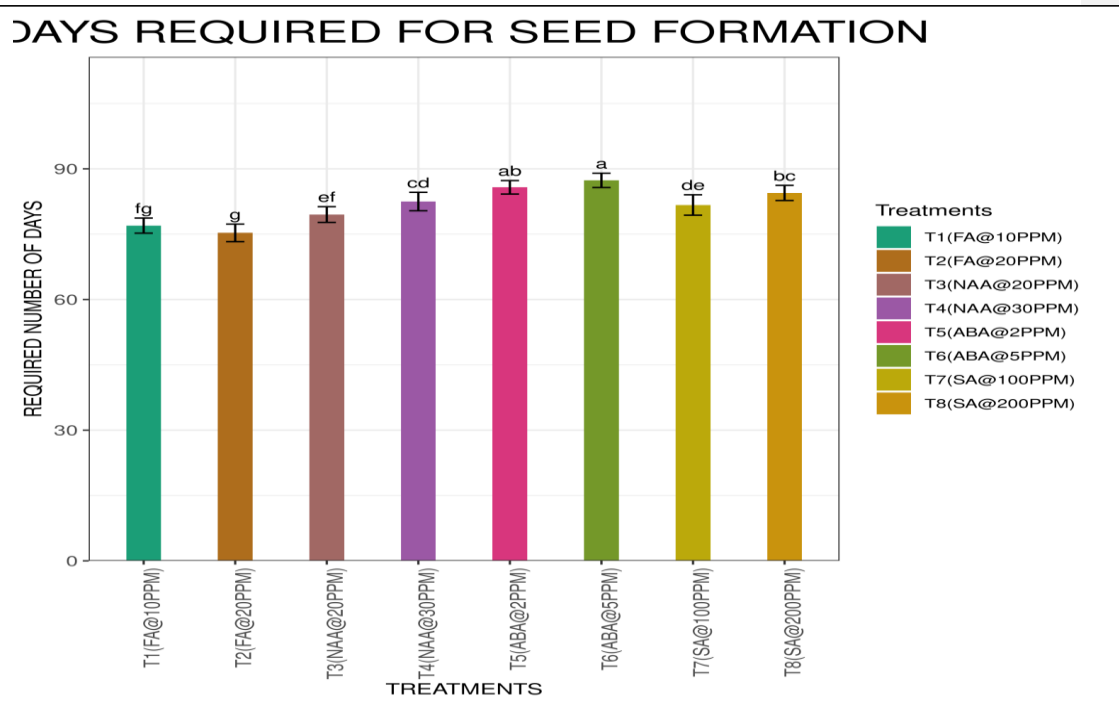
From the pooled data obtained from year (2020-2021 and 2021-2022), it was found that hormonal treatment differed significantly ( $p > 0.05$ ) for days required to flower initiation and completion of flowering. Higher number of days required for flowering (48.39 & 47.20DAS) and complete flower formation (70.40 & 69.95DAS) was take placed under the treatment of ABA @ 5 PPM and ABA @ 2 PPM. Result revealed that application of ABA are responsible for delaying in flowering which caused higher the duration of vegetative growth, resulted in flowering do not coincide with the impact of cold treatment provided to the crop at 42 DAS. Hence flowers are protected from the incidence of cold shock and less damaged by its harmful effect. Higher the duration of vegetative growth also responsible for acquirement of more amount of photo assimilated that was utilized by the crop plant during its reproductive growth period (1). The earliest flowering (40.24 & 41.57DAS) and complete flower formation (63.28 & 64.83DAS) occurred under fusaric acid treatment @ 20 ppm and 10 ppm. It could be due to fusaric acid is a fungal toxin responsible for causing wilt in treated plants, or whenever plants suffer from abnormalities caused by any internal or external factors, they try to complete their life cycle in a shorter period of time (3) resulting in earlier flowering under fusaric acid application. Variety V<sub>1</sub> (JG74) had the earliest flower initiation and completion of flowering, taking (41.55 & 63.17 days) respectively, but variety V<sub>4</sub> (PBG5) had a higher number of days required for flower initiation and completion of flowering. Our findings are according to Veeramani and Sendhilvel (2020) (25), who find that variety with wilt incidence (17.20%) needed significantly shorter time to achieve 50% flowering, whereas the variety with incidence (11.50%) took longer time to achieve 50% flowering.

Treatments of ABA @ 5 ppm and ABA @ 2 ppm required more days for pod initiation (79.86 DAS) and seed development (87.35 DAS). Fusaric acid treatment resulted in the shortest number of days for pod initiation (79.86 DAS) and seed development (87.35 DAS). Higher number of days required under ABA application may be due to this hormone's appeared in alleviating the damage caused by stress to ensure the normal growth of plants at later stages of development (26). Fusaric acid, on the other hand, was responsible for boosting the damage caused by wilt incidence, resulting in plant reduced reproductive growth and, ultimately, a reduction in the number of days required under fusaric acid application (18). Similarly V<sub>4</sub> (PBG5) required more number of days for pod initiation (79.86 DAS) and seed formations (87.35 DAS) due to its normal growth period resulted from its tolerance capacity to nullifying adverse impact imposed by wilt and cold, whereas V<sub>1</sub> (JG74) have requirement of less number of days for pod initiation (69.20 DAS) and seed formations (73.61 DAS) might be due to changes in growth period imposed by wilt and cold.

**FIGURE 3– Effects of PGRs and Varieties on number of days required for pod initiation in wilt and cold stressed chickpea.**



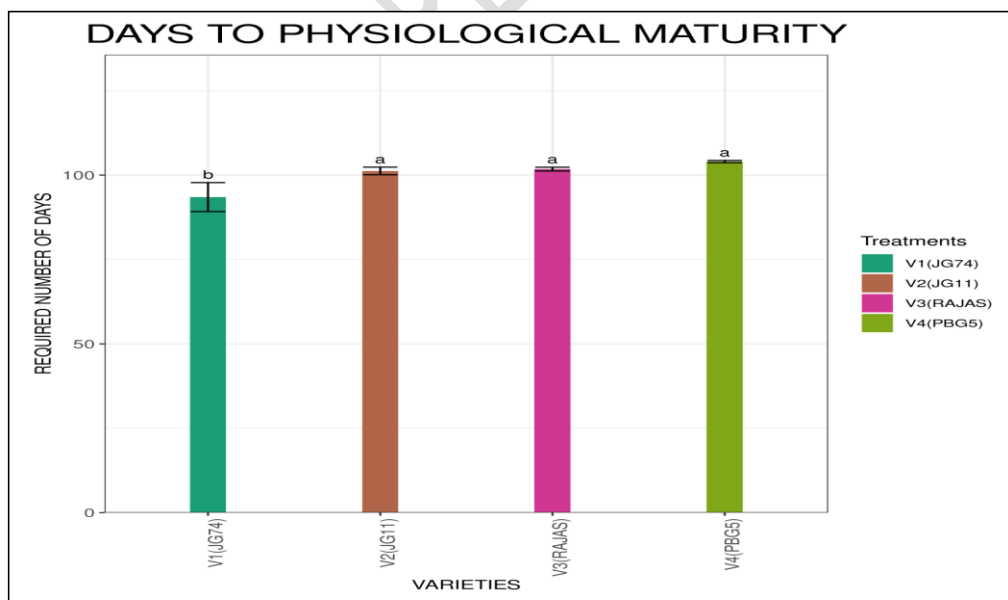
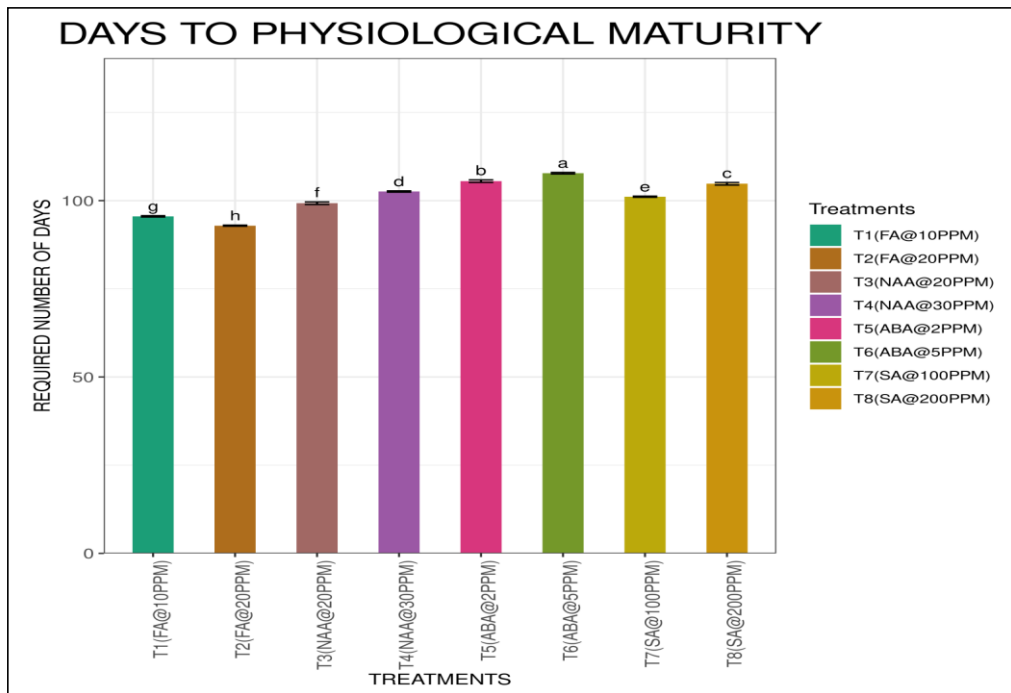
**FIGURE 4– Effects of PGRs and Varieties on number of days required for seed formation in wilt and cold stressed chickpea.**



The process of seed development begins with ovule fertilization and continues through physiological maturity, which is characterized by maximal dry matter content when plant-seed translocation finishes (4). According to Samarah and Yahya (2008) (17), chickpea seeds reached physiological maturity when the pods turned yellow, but germination and vigor peaked beyond that point, when the pods turned brown. The treatment of ABA @ 5 ppm and ABA @ 2 ppm resulted in a larger number of days required for physiological maturity (107.78 & 105.53 DAS) and harvestable maturity (119.96 & 116.89 DAS). Whereas Fusaric acid treatment @ 20 ppm and @ 10 ppm resulted in a shorter time to acquire physiological (92.90 & 95.54 DAS) and harvestable maturity (101.70 & 104.50 DAS). Our findings are in accordance with Qi and Zhang (2020) (13), who reported that when crop plants suffer from abnormalities caused by any external or internal factors, they lower their growing period and try to complete their life cycle in a shorter period by maximizing the utilization of available resources. Genotypes V<sub>4</sub> (PBG5) required more days for physiological maturity (104.01 DAS) and harvestable maturity (115.60 DAS), whereas V<sub>1</sub> (JG74) required the least number of days (93.50 DAS) and (101.28 DAS) for these traits. The variation in days required for physiological maturity and harvestable maturity among genotypes is related to differences in stress tolerance and susceptibility features. Genotype V<sub>4</sub> (PBG5), which can withstand in stress (wilt and cold), requires the most days for physiological maturity and harvestable maturity, whereas susceptible genotypes V<sub>1</sub> (JG 74) require the least (25).

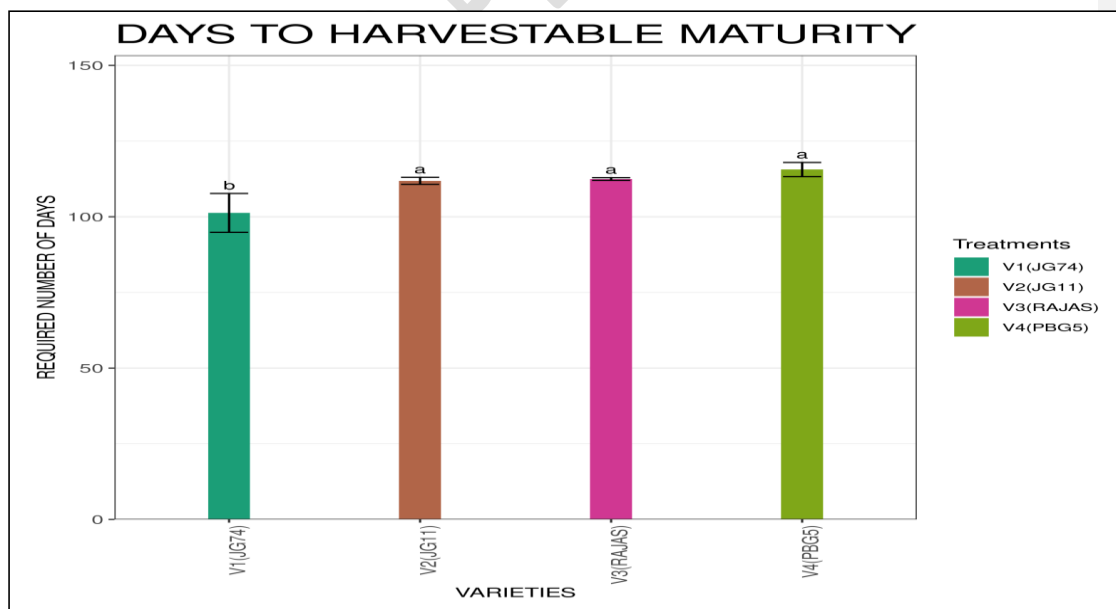
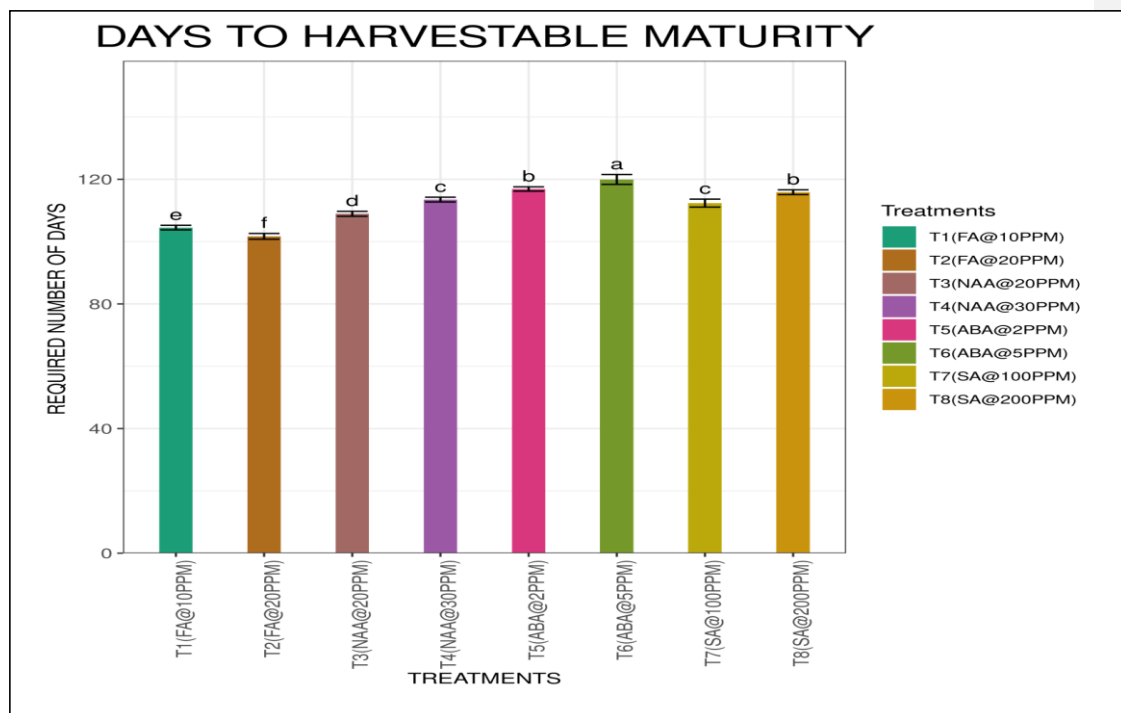
Pooled data from both years (2020-2021 and 2021-2022) shown that treatments with FA @ 20ppm, and FA @ 10ppm exhibited the highest disease incidence of 31.25%, and 30.71%, respectively. Fusaric acid is a well-known fungal toxin which leads to chickpea death (8). Foliar sprays of FA in wilt contaminated pots accelerated wilting, resulting in the chickpea crop dying quickly due to the synergistic effects of FA with wilt inoculums. Increased occurrence of disease incidence under FA @ 20 ppm and FA @ 10 ppm made plants more prone to various types of stresses due to changes in crop plant structure and functional activities (15). As a result, cold incidence (23.96% and 22.29%) was also shown to be higher under FA @ 20 ppm and FA @ 10 ppm. Plants' incapacity to engage osmoregulation mechanisms during their active stage of development might contribute to their sensitivity to these treatments (20).

**FIGURE 5– Effects of PGRs and Varieties on number of days required for physiological maturity in wilt and cold stressed chickpea.**



**FIGURE 6– Effects of PGRs and Varieties on number of days required for harvestable maturity in wilt and cold stressed chickpea.**

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After artificial wilt inoculation, plants receiving ABA @ 5 and ABA @ 2 ppm were the least wilted (18.75 and 20.21 %). It might be due to ABA is a major PGR responsible for activating defensive mechanisms in infected plants through up regulation of ABA-dependent transcription pathways (26) or enhancement of tolerance responsive gene expression, both of which are important for providing crop plant tolerance against potential stress damage (24) (Hussien et al., 2018). Cold incidence percentage of 16.46% and 17.29% were also found to be lower under both treatments. The possible explanation for this result is that in response to wilt stress, plants biosynthesize several types of tiny molecules, known as stress proteins or heat-shock proteins (Hsp), which may act as stress mitigating agents by maintaining water potential in cell sap and creating a barrier against membrane disruption and solute leakage, thereby offering strengths to the plant to cope with stress by maintaining intracellular activity (22). Wilt incidence was most prevalent (36.67%) in JG 74 (wilt susceptible variety. PBG 5, which is tolerant to cold and resistant to wilt, had the lowest (18.72%) incidence of wilt. Similarly higher cold incidence (25.83%) was also noted in JG 74 as a result of wilt pathogen disturbances in crop plant structure and functional activities (15). PBG 5 cold resistance cultivars had a lower cold incidence (15.52%) than all other cultivars used in the studies.

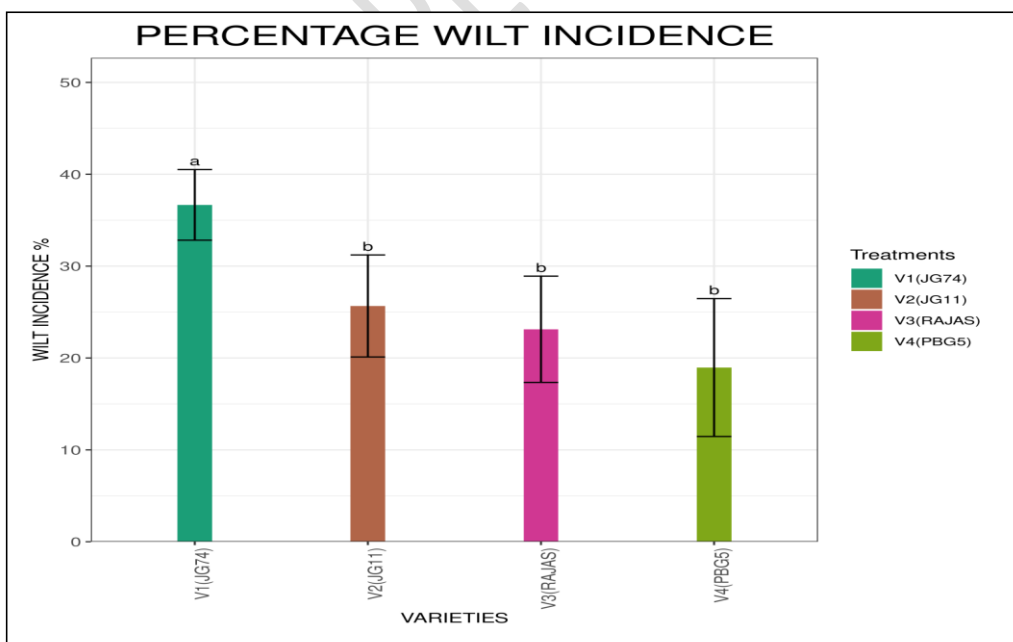
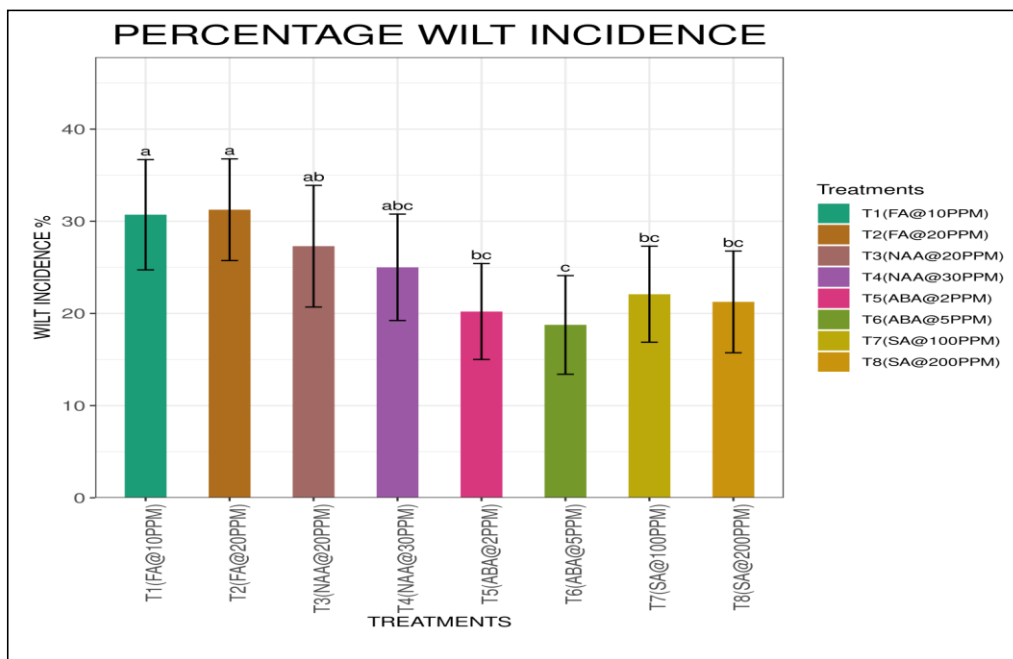
Plant sprayed with of ABA @ 5 ppm and ABA @ 2 ppm was able to maintained stress tolerance response under artificial exposure of wilt and cold and exhibited its highest mean productivity (12.73 & 12.33 g plant<sup>-1</sup>). Similarly variety PBG 5 conferred its highest mean productivity (12.50 g plant<sup>-1</sup>. Being fungal toxin in nature spraying of FA @ 20 ppm and 10 ppm, severe death of plant was occurred which resulted in lower the plant productivity under these treatments. Genotype JG 74 also had a lower (10.40 g plant<sup>-1</sup>) mean productivity due to the hindrance caused by stress conditions.

## Conclusion

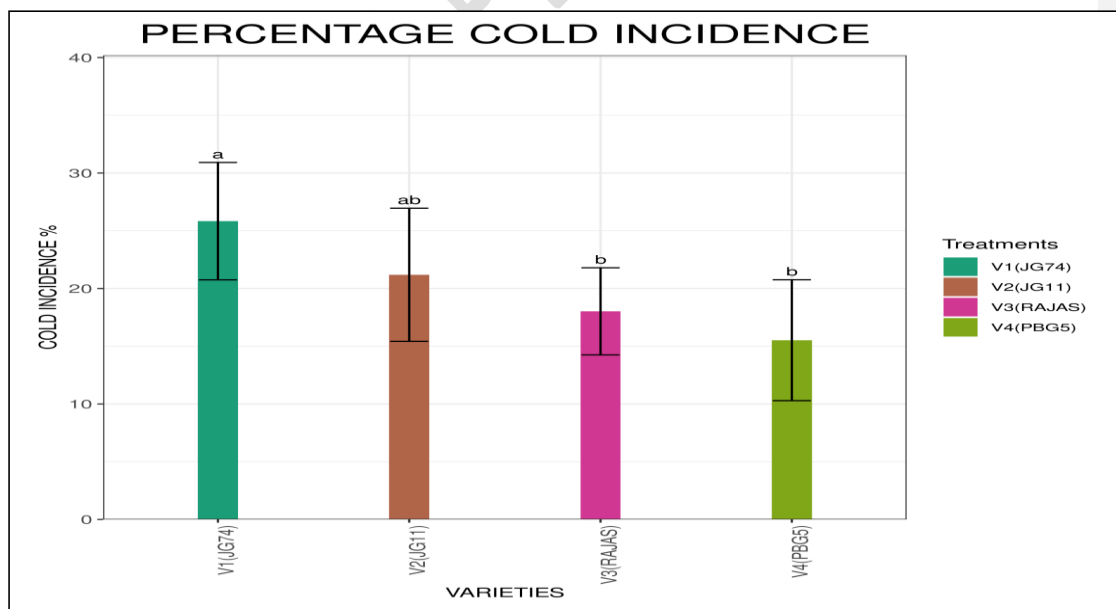
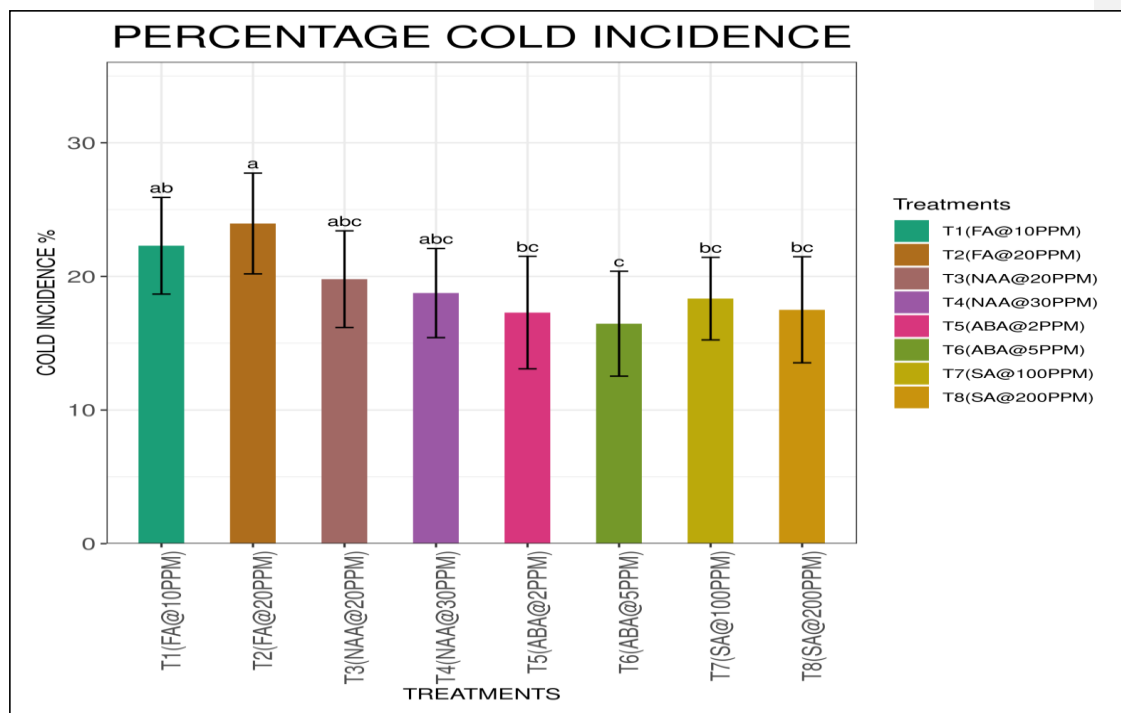
Plant sprayed with ABA @ 5 ppm and ABA @ 2 ppm effectively manage the damaged caused by the wilt at early stage and cold at later therefor provided tolerance to crop to withstand under unfavorable conditions and minimize the yield loss due to impact of wilt and cold. Similar variety PBG 5 performed well under both (wilt and cold) adverse induced conditions and exhibits its maximum tolerance efficiency in comparison to others three genotypes. Whereas due to synergistic effect of FA with artificially wilt inoculum maximum disease (wilt) as well cold incidence was found under the application of FA @ 20 ppm.

**Comment [ZA11]:** Hussien, M.A. and Ahmed, Z.F.R. (2018). Plant extracts treatment along with flower bud thinning promotes cactus pear production and fruit quality. Acta Hort. 1216, 127-134

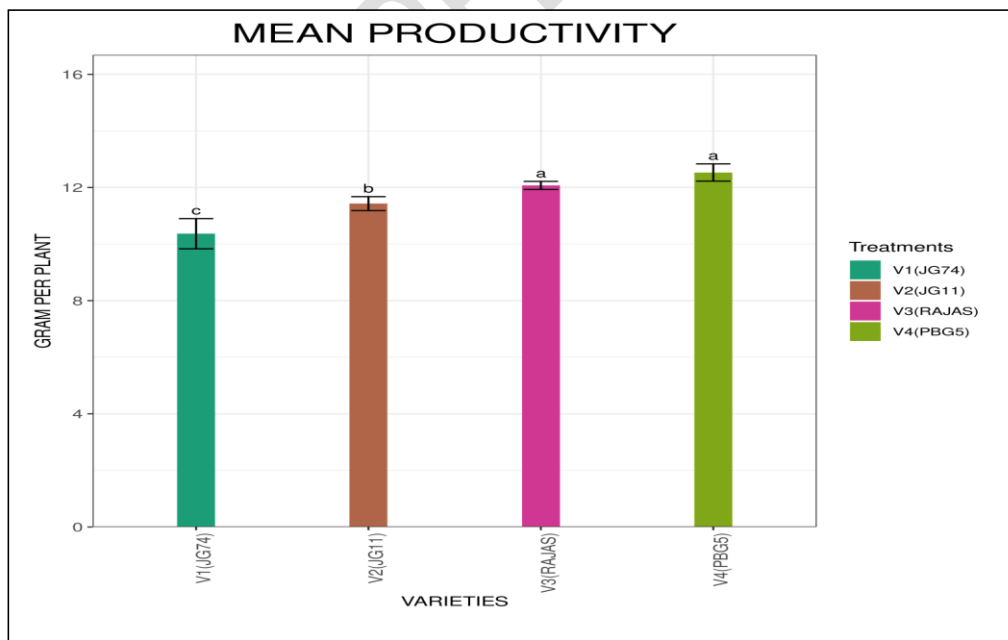
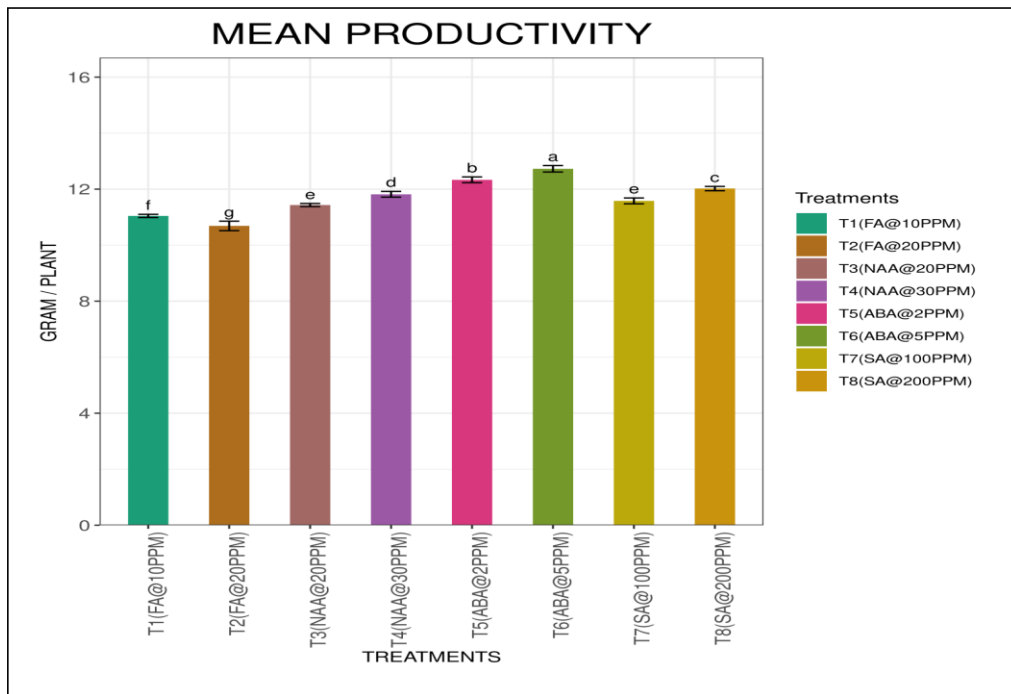
**FIGURE 7– Effects of PGRs and Varieties on percentage wilt incidence in wilt and cold stressed chickpea.**



**FIGURE 8– Effects of PGRs and Varieties on percentage cold incidence in wilt and cold stressed chickpea.**



**FIGURE 9– Effects of PGRs and Varieties on mean productivity in wilt and cold stressed chickpea.**



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