



Exogenous anti-gibberellin agents for biennial bearing management in Balady mandarin trees

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Abstract

Exogenous application of growth regulators often induces physiological changes by altering endogenous hormone levels, thereby affecting plant productivity. Consequently, this trial aimed to explore the impacts of anti-gibberellin (A-GA) agents, using both abscisic acid (ABA) and ethephon (ETH) at 50 ppm alongside a control, before the Off-year season (at flower bud induction period) twice during the last week of December and January on fruit yield, cumulative impacts on the following On-year and biennial bearing (BB) of Balady mandarin trees. Regarding the two Off-years, ETH effectively decreased endogenous GA content in March by 28.26% and 29.78%, while it increased the C/N ratio by 16.67% and 14.29%, the number of fruits by 55.18% and 50.26%, and fruit yield by 50% and 45.45%. Also, ABA decreased endogenous GA content in March by 8.46% and 16.10%, while it increased the number of fruits by 87.4% and 52.8% and increased fruit yield by 75% and 45.45%. ABA increased Off-year yield by 12.48% relative to ETH. Concerning the following two On-years, higher endogenous GA content was observed in the previous September (Off year) in A-GA treatments, resulting in a decrease in mandarin fruit yield. ETH decreased the number of fruits by 17.37% and 24.1%, resulting in increases of 3.8% and 16% in fruit weight. Also, ABA decreased the number of fruits by 22.94% and 6.97%, resulting in increases of 10.34% and 10.71% in fruit weight. Furthermore, ETH decreased BB by 39% and 60% and ABA decreased BB by 52.44% and 38.24%. Using A-GA agents prior to the off year during the flower bud induction period alleviates BB in Balady mandarin trees.

Key words: *Citrus aurantium*, ethephon, abscisic acid, TSS/ acid ratio, yield, alternate bearing.

Introduction

Increasing agricultural production represents important part of Sustainable Development Goals agenda particularly the food security sector and sustainable land management. Thus, there is an urgent requirement to incorporate innovative methods in agriculture to increase crop productivity (Elbaalawy *et al.* 2023). Recently, the mandarin cultivation area in Egypt accounts for about a quarter of total citrus production and is regarded as the second-most-exported fruit after oranges (Mohamed *et al.*, 2019).

Biennial bearing (BB) phenomenon is the tendency of fruit trees to produce a heavy crop in one year (On-year) followed by a minor crop the following year (Off-year). Heavy yield usually reduces flowering intensity in the following year (Monselise and Goldschmidt, 1982). It can impact on the tree's health through limb breakage and the tree's dieback or death (Kihara *et al.*, 1995). BB is common in many fruit species such as olive (Bonyanpour *et al.*, 2017), mango (Singh *et al.*, 2025) and mandarin (Shaban *et al.*, 2025a,b). In BB mandarin varieties, the yield of heavy crops during On-year is high, but with small fruit size and low quality (El-Sayed *et al.*, 2017; Haim *et al.*, 2021; Griebeler *et al.*, 2023; Jangid *et al.*, 2023). Balady mandarin is known for its tendency to BB. Large harvests of mandarins (On-year) resulted in smaller, unmarketable fruits of little commercial value, delayed maturity, and a worsening of the fruit's exterior color (Shaban *et al.*, 2025a; Mahmoud and Abo-Eid, 2019).

The hormonal hypothesis that explains BB suggests that during the On-year fruit growth, different substances, such as gibberellic acid (GA) is produced which is recognized as a flowering inhibitor

in citrus (Goldschmidt *et al.*, 1997). High levels of GA released by seeds during the On-year may reduce flower development during the next Off-year and thus be the main cause of BB (Jan *et al.*, 2022; Shaban *et al.*, 2025b).

Management of BB presents a significant challenge for citrus producers, as it involves understanding and addressing hormonal control of this phenomenon which is considered a key regulatory process involved in BB cycle. GA content was higher in olive, lychee and mango during On-year compared to Off-year (Baktir *et al.*, 2004; Ulger *et al.*, 2004; Pal and Ram, 1978; Chen, 1990). Higher endogenous GA stimulate vegetative growth via blocking flower bud induction, whereas fruit removal, seed destruction, or application of anti-gibberellin (A-GA) chemicals stimulated the buds to differentiate into flowers (Jan *et al.*, 2022; Andreini and Bartolini, 2008).

Ethephon is a molecule that remains stable in low-pH solutions, but it breaks down at higher pH in plant tissues, releasing ethylene (ETHL) (Ferrara *et al.*, 2016). ETHL regulates the transition from vegetative to reproductive tissues (Martínez *et al.*, 2022). ETH applications reduced yield variation between years when applied to Delicious apple cv. during On-years (Bukovac *et al.*, 2006). Flower bud development occurs where higher level of ETHL is present, indicating ETHL's florigenic activity (McArtney *et al.*, 2013). Recently, modulation of growth and maturation of sexual organs and petals up to anthesis are linked to ETHL levels in flower buds (Martínez *et al.*, 2022). Ethephon ETH applications have been reported to stimulate flower bud formation (Meland and Kaiser, 2011; McArtney *et al.*, 2013; Hladnik and Stopar,

2021). Exogenous application of ETH to apple trees in On-year enhanced flowering in the following Off-year (Campbell and Kalcsits, 2024).

Abscisic acid (ABA) is a key regulator of BB in pistachio (Kafkas *et al.*, 2020). Higher levels of inhibitors like ABA and lower levels of GA are vital for a florigenous shoot and promote flowering (Bajpai and Bajpai, 2021). ABA-related compounds are likely to be the most influential in promoting flower bud initiation in apple (Milyaev *et al.*, 2022). Valencia orange trees, during the On-year, contain more of *t*-ABA (2-transabscisic acid) than those in Off-year trees (Jones *et al.*, 1976). Also, low GA content and high ABA content stimulated flower bud formation in olives (Baktir *et al.*, 2004).

Therefore, this study aimed to investigate the impacts of exogenous A-GA compounds (Ethylene and Abscisic acid) prior to Off-year as a tool for mitigating BB in Balady mandarin and monitoring their influence on yield components of the trees during the following On-year.

Material and methods

Experimental site: The present study was carried out during two Off-year seasons (2021, 2022) and the following two On-year seasons (2022, 2023) at El-Beheira Governorate, Egypt. Trees were grown in sandy soil under a drip irrigation system.

The soil had a pH of 7.64 and an electrical conductivity (EC) of 300 $\mu\text{S}/\text{cm}$ (equivalent to 0.3 dS/m) in a 1:2.5 soil–water suspension. The concentrations of soluble anions and cations (meq L^{-1}) were as follows: chloride (Cl^{-})-2, sulfate (SO_4^{2-})-6.4, carbonate (CO_3^{2-})-0, bicarbonate (HCO_3^{-})-2, sodium (Na^{+})-1.8, potassium (K^{+})-0.3, magnesium (Mg^{2+})-1, and calcium (Ca^{2+})-4.

The irrigation water had a pH of 7.7 and an electrical conductivity of 0.81 dS/m . The concentrations of soluble anions and cations (meq L^{-1}) were as follows: chloride (Cl^{-})-3.8, sulfate (SO_4^{2-})-0.3, carbonate (CO_3^{2-})-0, bicarbonate (HCO_3^{-})-4.0, sodium (Na^{+})-2.6, potassium (K^{+})-0.3, magnesium (Mg^{2+})-2.0, and calcium (Ca^{2+})-3.2. The micronutrient concentrations (mg L^{-1}) were: iron (Fe)-0.001, zinc (Zn)-0.001, manganese (Mn)-0.001, and copper (Cu)-0.001.

First season 2021	Off year 2021 (Tree A) Anti-gibberellins treatments	
Second season 2022	Off year 2022 (Tree B) Anti-GA treatments	On year 2022 (Tree A), following yield components
Third season 2023	On year 2023 (Tree B) Following yield components	

Fig.1. Anti-gibberellin treatments for alleviating biennial bearing in Balady mandarin trees. Two Off-year (2021 and 2022) and the following two On-year (2022 and 2023).

Plant material and treatments: Twelve-years-old Balady mandarin (*Citrus reticulata* Blanco) trees budded on the rootstock sour orange (*Citrus aurantium* L.) and planted at 4 × 4 m apart were used in this study. This experiment began in September 2021 and ended in December 2023. The treatments were carried out only at the end of the On-year season for the tree which will be in the following Off-year season. For the cumulative impacts in the following two On-year seasons, fruit yield and BB were recorded. Foliar spraying of ABA (Abscisic 99%; CAS NO.: 14375-45-2,

Bloom Tech Changzhou, China) and ETH (Ripex; Ethephon 48%, Shoura Chemicals, China) at 50 ppm was carried out two times with 30 days interval before Off-year flowering (the last week of January and February), using an average of 5 liters of solution per tree each time (Fig. 1) in presence of 0.1% Tween 20 as a surfactant. There was no visual symptom after GA sprays. All trees (36 trees) received the recommended horticultural practices (fertilizers, irrigation, pruning, weed control, pest control, etc.) according to the Agriculture Ministry. Each treatment included 12 trees presented in 3 replicates.

GA content: GA samples were taken according to (Shaban *et al.*, 2025a,b) from treated trees (leaves) at the middle of March (flowering stage), June and September in for both Off-year seasons. GA in mandarin leaves was analyzed using the method described by Fales *et al.* (1973) and Furniss (1989). Freeze-dried leaf samples were extracted in methanol (80%) containing butylated hydroxytoluene as an antioxidant at 4 °C in the dark. GA content was quantified and peaks were identified using ATI Unicam Gas–Liquid Chromatography, 610 Series, equipped with a flame ionization detector. The chromatographic parameters consisted of a C18 column (100mm x 4.6mm), a detection wavelength (206 nm), an injection volume (20 μL), and a column temperature set (35 °C). The mobile phase was composed of methanol-water and acetic acid (0.5%), with a flow rate maintained at 1 mL min^{-1} . GA (5 mg) was mixed in methanol (50 mL) to prepare a stock solution (100 ppm). This was diluted to form a range of known concentrations (0.5, 1, 5, 10, 20, 40, and 80 ppm). A calibration curve is created by plotting peak area against standard concentration. GA concentration ($\mu\text{g g}^{-1}$ FW) in leaf samples was assessed using a calibration curve based on the corresponding peak area (Zhang *et al.*, 2017; Caboni *et al.*, 2023).

C/N ratio: Leaf samples were collected after maturation of young shoots in July. From each tree, eight leaves were gathered (two leaves from each tree side), resulting in a total of 96 leaves for each replicate. It was then dried (70°C) for a constant weight (72 hr) and blended to form a powder. A sample (0.2g) was taken for determining the C/N ratio for each replicate (three samples per treatment). Leaf carbon content was determined according to Jackson (2005) and total nitrogen using the Kjeldahl method as described in AOAC (1995). C/N ratio was calculated as leaf carbon content divided by leaf nitrogen content.

Fruit yield: At harvest time, the yield of each tree was determined as number and weight (kg) of fruits per tree during the Off-year. Fruit weight was recorded using digital sensitive balance.

Total soluble solids: Fruits were cut in half and carefully hand-squeezed in a commercial juicer. The content of total soluble solids (TSS) was determined through a hand refractometer (model MASTER-53 S; Atago Co. Ltd., Tokyo, Japan) and expressed as °Brix.

Titrateable acidity (TA): It was determined by titrating 10 mL juice to the end point (first stable pink color) of using 0.1 N NaOH and two points of 1% phenolphthalein (1g phenolphthalein per 100 mL of 90% ethanol). Finally, it was expressed as a percentage of citric acid. Once the TSS and TA contents had been assessed, the ripening index was calculated as the TSS/TA ratio. TA determination performed as per Ranganna (1986).

Total sugars content (TSC): The phenol–sulfuric acid method was used to determine TSC (DuBois, 1956). Fruit pulp samples (0.25 g) were homogenized in 20 mL of ethanol (70%) and then filtered. The filtrates (1 mL) were treated with 1 mL of phenol (5%) and then 5 mL of H₂SO₄ (98%). After 1 h, the absorbance of the cold and colored solutions was read (490 nm) using a UV/Vis spectrophotometer (UNICO S2100, Cole Parmer Instruments, Chicago, IL, USA). A standard curve was generated using a standard glucose solution, and TSC content was expressed as mg glucose equivalents per g fresh weight.

Cumulative impact during the following On-year season

Fruit yield: In the following On-year, number of fruits, fruit weight, and total yield were recorded to study the cumulative impact of the previous treatments on the following On-year on the same treated trees in the previous Off-year.

Biennial bearing (BB) index: It was determined by the difference between On-year and Off-year yields of a tree divided by their sum (Jangid *et al.*, 2023). For example, for the Off-year 2021 and the following On-year 2022, the following equation was used:

$$BB (\%) = \frac{\text{On-year yield (2022)} - \text{Off-year yield (2021)}}{\text{Off-year yield (2021)} + \text{On-year yield (2022)}} \times 100$$

Statistical analysis: The experiment was arranged in a complete randomized design with three replicates, each consisting of four trees. The experimental unit consisted of one tree from each treatment, with two trees left as boundaries before and after each treated tree. Results were subjected to variance analysis (ANOVA) using the general linear model (GLM) procedure—SAS software Version 9.0 (SAS Institute Inc., Cary, NC, USA). Conducting mean comparisons between treatments using Duncan's multiple range test (Duncan, 1955) at a significance level of $p < 0.05$.

Results and discussion

GA content: In March, at the flowering stage during Off-year trees, the endogenous GA level was significantly affected by A-GA treatments (Table 1). ETH treatment recorded the lowest significant values (7.03 & 6.67 $\mu\text{g g}^{-1}$) which were lower than control (9.80 & 9.5 $\mu\text{g g}^{-1}$) with a percentage of 28.26% and 29.78% followed by ABA treatment which recorded a reduction in GA at March (8.98 & 7.97 $\mu\text{g g}^{-1}$) with a percentage of 8.46% and 16.10%, for the first and second Off-years, respectively.

In June, the endogenous GA level was affected by A-GA treatments (Table 1). ETH treatment resulted in the lowest significant endogenous GA values (7 & 7.03 $\mu\text{g g}^{-1}$), which were lower than control (8.43 & 7.67 $\mu\text{g g}^{-1}$) with a percentage of 16.96 and 8.34% for the first and second Off-year seasons, respectively. At the same time, exogenous ABA recorded a reduction in endogenous GA in June, with a percentage of 5.45% in the first Off-year only.

Table 1. Effect of anti-gibberellin treatments on gibberellin content ($\mu\text{g g}^{-1}$ FW) during March, June and September of Balady mandarin trees during two off-year seasons (2021&2022).

Treatments	GA3 in March		GA3 in June		GA3 in September	
	2021	2022	2021	2022	2021	2022
50 ppm ABA	8.97b	7.97b	7.97a	7.97a	7.80a	6.67a
50 ppm ETH	7.03c	6.67c	7.00b	7.03b	7.97a	7.97b
Control	9.80a	9.50a	8.43a	7.67ab	7.30b	6.63c

In September, the endogenous GA level was significantly affected by A-GA treatments (Table 1). Endogenous GA increased significantly compared to the control. ABA increased GA with a percentage of 6.8 and 30.77% and ETH increased GA with a percentage of 9.17 and 20.21%, for the first and second Off-year seasons, respectively. The increase in GA may be due to an increase in fruit number, which contains more seeds, a source of GA.

The reduction in endogenous GA levels during Off-year was more pronounced in A-GA treatments in March, reflecting the impact of A-GA application on lowering GA levels at the time of flowering, which helps increase fruit set. On the other hand, increased endogenous GA levels during the Off-year was affected by A-GA treatments in September, which was resulted from an increased number of GA suppliers (seeds). Higher GA levels in seeds during the On-year can inhibit flower development in the following Off-year, causing biennial bearing (Jan *et al.*, 2022; Shaban *et al.*, 2025b). Since increased GA enhances vegetative growth (Ali *et al.*, 2025) and inhibits flower bud induction, while removing fruit or applying A-GA encourages flowering (Jan *et al.*, 2022; Andreini and Bartolini, 2008). Using A-GA (ABA or ETH) during the Off-year promotes more yield in Balady mandarin fruits compared to the control. GA production gradually increased in September as the fruit developed since Balady mandarin fruit contains around 25 seeds (Shaban *et al.*, 2025b), which serve as an endogenous source of GA.

Flowering in citrus results from short generative shoots with low endogenous GA levels (Martínez-Alcántara *et al.*, 2015). GA application significantly reduced the number of flowers in citrus and decreased flowering intensity in Montenegrina mandarin (Griebeler *et al.*, 2021). Exogenous GA inhibited flowering in citrus (Garmendia *et al.*, 2019). GA at 21 ppm decreased the number of flowers in Tahiti acid lime (Pereira *et al.*, 2014). Spraying GA during the fall/winter period inhibited flower induction (Monselise and Goldschmidt, 1982; Martínez-Fuentes *et al.*, 2004). GA can control the time and intensity of citrus flowering (Agustí *et al.*, 2022). More recently, GA at 1000 ppm during flowering decreased fruit set in the Balady mandarin tree (Shaban *et al.*, 2025a).

A-GA substances can induce citrus flowering (Huchche & Ladaniya, 2014). The application of ABA at flower induction increased the flowering of Satsuma mandarin (García-Luis *et al.*, 1989). Also, ETH can promote plant flowering (Sanyal *et al.*, 1998). ETH inhibit enzyme activation by inhibiting the production and function of GA and GA derivatives, especially during the early phenological period (Wang *et al.*, 2014). In this regard, Shaban *et al.* (2025a) found a reduction in endogenous GA levels in Balady mandarin trees treated with methionine and potassium, resulting in an earlier harvesting date in the On-year season. Shaban *et al.* (2025b) reported that reduced number of seeds per mandarin fruit decreased endogenous GA levels, which in turn helped regulate BB in Balady mandarin.

C/N ratio: C/N ratio was influenced by A-GA treatments (Fig. 2). ETH increased C/N ratio significantly compared with the control. This increase was higher than the control, with percentages of 16.67% and 14.29% for the first and second Off-year seasons, respectively. Meanwhile, there were no differences between ABA

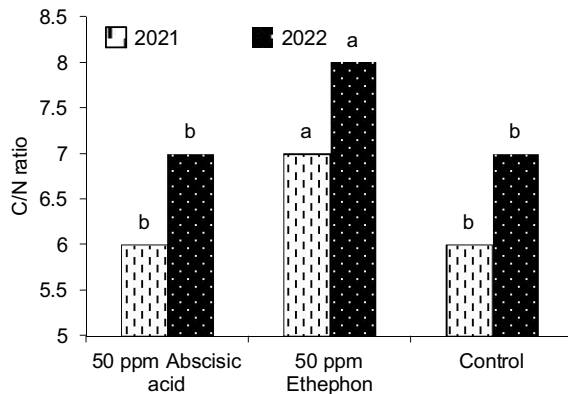


Fig. 2. Effect of anti-gibberellin treatments (50 ppm abscisic acid or 50 ppm ethephon) on C/N ratio of Balady mandarin trees during two off-year seasons (2021 & 2022).

and control treatments in C/N ratio.

Exogenous application of ETH surpassed ABA treatment in increasing the leaf C/N ratio. This may be due to ABA stimulating the root system by increasing cytokinin production, which may compete with leaves for carbohydrate consumption, reduce carbohydrate availability, and then decrease the C/N ratio in leaves. Additionally, the application of shoot-ABA increases basipetal auxin transport by 114%, thereby enhancing root cell elongation by 56% (Xie *et al.*, 2020). Research suggests that a high ratio of ABA to GA promotes tuber development (Chen *et al.*, 2022). Also, ETH may maintain high photosynthetic products that are not consumed in producing more vegetative or root growth, which leads to an increase in leaf C/N ratio. Moreover, high fruit yield led to a lower carbon-to-nitrogen (C/N) ratio, resulting in decreased flower initiation (Koshita *et al.*, 1999; García-Luis *et al.*, 1989).

Fruit yield: The Number of fruits was affected by A-GA treatments (Table 2). Exogenous ABA recorded the highest increase in number of fruits (428 & 479) with a percentage of increase reaching about 87.4 and 52.8% followed by ETH, which increased fruit number (354.3 & 471.3) by a higher percentage by 55.18 and 50.26% compared to the control (228.3 & 313.67) for the first and second Off-years, respectively.

Table 2. Effect of anti-gibberellin treatments on the number of fruits, Fruit weight (g) and fruit yield (kg per tree) of Balady mandarin trees during two off-year seasons (2021&2022)

Treatments	Fruit number		Fruit weight		Fruit yield	
	2021	2022	2021	2022	2021	2022
50 ppm ABA	428.0a	479.33a	163.3a	166.7a	70.0a	80a
50 ppm ETH	354.3ab	471.33a	175.0a	175.0a	60.0ab	80a
Control	228.3b	313.67b	175.0a	175.0a	40.0b	55b

Fruit weight was not affected by A-GA treatments (Table 2). This may be due to the fact that during the Off-year the trees had very little fruit load, with no competition for photosynthetic products, resulting in more harmonious size and weight. In this respect, On-year trees had a greater fruit number with low fruit weight (Monselise and Goldschmidt, 1982; Shaban *et al.*, 2025a,b).

An increasing number of fruits in A-GA treatments during Off-year resulted from decreasing endogenous GA at flowering time, which supported more fruit set and a larger number of fruits. The

superiority of exogenous ABA over exogenous ETH in increasing the number of fruits clarified why the C/N ratio was lower in the ABA treatment than in the ETH treatment. This was due to greater carbohydrate consumption by more fruits, which decreased the C/N ratio. In the ABA treatment, maintaining C/N ratio resulted in values that were the same as the control. The higher fruit number in ABA treatments in the first Off-year season than in ETH may be due to ABA stimulating root growth and restricting vegetative growth, which provides more carbohydrates for supporting fruit setting. During the off-Year, the number of fruits was higher in the ABA-treated tree than in the ETH-treated tree, leading to an increase in GA during September and a decrease in the C/N ratio compared to ETH.

Fruit yield was significantly affected by A-GA treatments (Table 2). Since exogenous ABA recorded the highest increase in fruit yield with a percentage of 75% and 45.45% followed by ETH, which increased fruit yield with a percentage of 50 and 45.45% for the first and second Off-year seasons, respectively. In contrast, increasing fruit yield was recorded due to GA at 100ppm on onion plants (Farad *et al.*, 2025).

Higher endogenous gibberellin levels in Off-year trees may negatively affect flowering, whereas A-GA treatments decreased endogenous GA content and increased fruiting (number of fruits) and yield. A low C/N ratio in Off-year trees prevented increases in fruit set and fruit number. A low C/N ratio in Off-year trees prevents increases in fruit weight. Also, increasing fruit number in A-GAs treatments with a relative C/N ratio prevents an increase in fruit weight. ABA enhanced the Off-year yield more than ETH may be attributed to ABA treatment stimulating more fruit set, which was reflected in increased fruit number per tree and then fruit yield. The high number of fruits in the ABA treatment resulted in a higher C/N ratio during fruit growth, which lowered the C/N ratio and produced more GA from developed fruits. Since Heavy fruit yield caused a lower C/N ratio (Koshita *et al.*, 1999; García-Luis *et al.*, 1989; Monselise, 1986).

Fruit characteristics in Off-year

Fruit TSS, acidity and TSS/acid ratio: Fruit TSS content was not affected by A-GA treatments (Table 3), while a slight increase was recorded in the second season. Neither fruit acidity content nor TSS/acid ratio was affected by A-GA treatments (Table 3). There is a slight increase in fruit acidity in the second season. These results were in agreement with Ferrara *et al.* (2016), who found that ETH did not affect pH, SSC and TA of table grapes. In contrast, increasing fruit quality was recorded due to GA at 100ppm on onion plants (Farad *et al.*, 2025). The lack of significant differences between A-GA and control treatments in chemical fruit characteristics may be attributed to the low fruit load in the Off-year, which did not require more nutrients or a

Table 3. Effect of anti-gibberellin treatments on fruit TSS content (Brix), Fruit titratable acidity and fruit TSS/acid ratio of Balady mandarin trees during two off-year seasons (2022&2023).

Treatments	Fruit TSS		Fruit acidity		Fruit yield	
	2021	2022	2021	2022	2021	2022
50 ppm ABA	12.10a	12.20a	1.00a	1.03a	12.18a	11.83a
50 ppm ETH	12.13a	12.07a	1.03a	1.03a	11.77a	11.70a
Control	12.13a	11.93a	0.98a	1.03a	12.33a	11.57a

higher C/N ratio. Also, it may be due to increased vegetative growth during Off-year, as they consume more carbohydrates than fruit does. A higher reduction in GA level during the flowering stage in ETH compared to ABA led to a lower yield than in low-GA ABA treatments. This means that plants need a specific GA concentration, balanced with other growth substances, to achieve an acceptable fruit yield.

Cumulative impact during the following On-year season

C/N ratio: The cumulative impact of A-GA on the following On-year season was clear: the C/N ratio significantly increased in ETH by 6% and 14% for the first and second On-year seasons, respectively (Fig. 3). ABA increased the C/N ratio by 5.71% in the first On-year season only. Increasing C/N ratio in A-GA treatments may be due to relative high fruit yield in the previous Off-year which led to moderate fruit load in the following On-year which did not need for more consume of C/N ratio, while in control trees had a little fruit yield in the previous Off-year which resulted in huge fruit yield in the following On-year which need for more carbohydrates and consumption more photosynthetic products and C/N ratio. This also explained why the C/N ratio was higher during the On-year in the ETH treatment than in the ABA treatment. Due to ABA treatment, a higher number of fruits and yield in the previous Off-year than with ETH treatment.

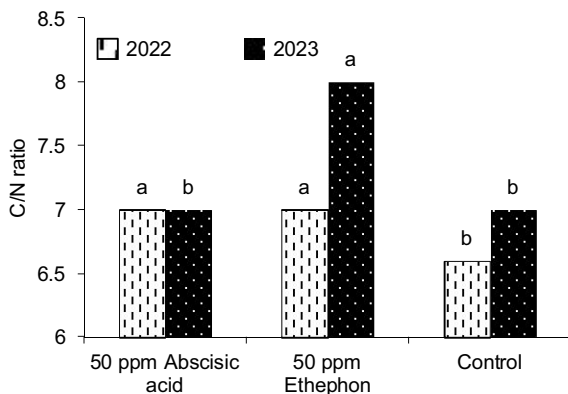


Fig. 3. Effect of cumulative impact of anti-gibberellin treatments (50 ppm abscisic acid or 50 ppm ethephon) on C/N ratio of Balady mandarin trees during the following two On-year seasons (2022 & 2023).

Fruit yield: The cumulative impact of A-GA on the following On-year cleared that the number of fruits was significantly decreased in ETH treatment with a percentage of 17.37 and 24.1% followed by ABA treatment, which decreased fruit weight with a percentage of 22.9 and 6.98% compared to the control for the first and second On-year seasons, respectively (Table 4).

The cumulative impact of A-GA on the following On-year seasons cleared that, fruit weight increased significantly in ABA with a percentage of 10.34 and 10.71% followed by ETH, which increased fruit weight with a percentage of 3.8 and 16%

Table 4. Effect of cumulative impact of anti-gibberellin treatments on the number of fruits per tree, Fruit weight (g) and fruit yield (kg per tree) of Balady mandarin trees during the following two On-year seasons (2022 & 2023)

Treatments	Fruit number		Fruit weight		Fruit yield	
	2022	2023	2022	2023	2022	2023
50 ppm ABA	829.33b	929.00a	145a	140.00a	120b	130a
50 ppm ETH	889.33b	758.00b	135b	145.00a	120b	110a
Control	1076.33a	998.67a	130b	125.00b	140a	125a

compared to the control for the first and second On-year seasons, respectively.

The cumulative impact of A-GA on the following On-year showed a significant reduction in fruit yield in the ETH treatment, with percentages of 14.29% and 12% for the first and second On-year seasons, respectively, compared to the control. Also, ABA decreased fruit weight by 14.28 in the first On-year season.

The findings indicated that, in the following On-year seasons, fruit yield was decreased with a significant differences in 2022 On-year. This reduction was offset by increases in fruit weight of 10.34% and 10.71% for ABA and 3.8% and 16% in the ETH treatment compared to the control in the first and second On-years, respectively.

Biennial bearing (BB) index: AB was affected by all treatments, with a significant effect for the ETH treatment (Fig. 4). Exogenous ETH decreased AB by 39% and 60% in the first and second seasons, respectively, compared to the control. Also, exogenous ABA decreased AB by 52.44% and 38.24% compared to the control, respectively, with significant values in the first season. Improving fruit yield by increasing fruit number during Off-year seasons led to a decrease in AB. During the Off-year, higher GA content was recorded in September in anti-gibberellin treatments, leading to a regulated (decreased) mandarin fruit yield in the following On-year.

In this regard, Shaban *et al.* (2025a) found a reduction in AB in Balady mandarin trees treated with methionine and potassium due to early harvest in the On-year season. Also, Shaban *et al.* (2025b) found that reduced GA supply in Balady mandarin and reduces endogenous GA levels.

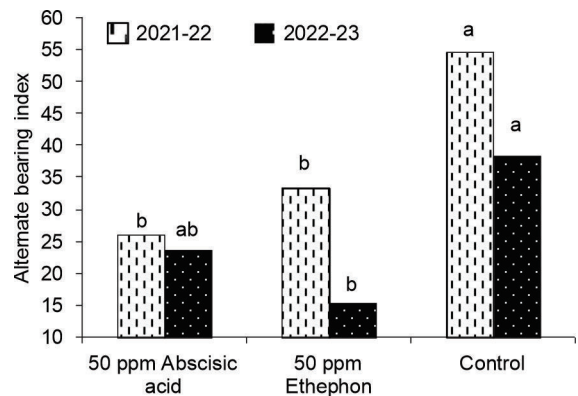


Fig. 4. Effect of anti-gibberellin treatments (50 ppm abscisic acid or 50 ppm ethephon) on biennial bearing index of Balady mandarin tree.

Exogenous application of anti-gibberellin agents, ABA and ETH on Balady mandarin prior to the Off-year season at 50 ppm twice in the last week of December and January resulted in a significant reduction in endogenous GA levels in March during flowering time. Exogenous application of anti-gibberellin agents, ABA and ETH on Balady mandarin prior to the Off-year season at 50 ppm twice in the last week of December and January resulted in a significant reduction in endogenous GA levels in March during flowering time. In comparison with control, in the Off-year, exogenous ETH improved the number of fruits by 55.18% & 50.26% and enhanced yield by 50% & 45.45%, while exogenous ABA treatment increased the number of fruits by 87.4% & 52.8%, along with improved yield by 75% and 45.45%. Regarding the following two On-year seasons, ETH reduced number of fruits by 17.37% and 24.1%, while ABA reduced

number of fruits by 22.94% and 6.97% in the first and second seasons, respectively. ABA being more effective in increasing Off-year yield by 12.48%, than ETH, Both anti-gibberellin agents effectively reduced biennial bearing, while ETH decreased AB by 4.2% more than ABA.

Competing interests: The authors declare no conflict of interest..

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