

The Effects of Silver Nitrate Applications on the Flower Quantity of Cucumbers (*Cucumis sativus* L.)

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Abstract

This study was conducted to investigate the effects of silver nitrate (AgNO_3) on the flower quantity of cucumbers. The seeds used in this study, which was carried out in a plastic greenhouse, in the Gazi village of Antalya Province (Turkey) the during spring and autumn 2005 breeding periods, were 'Mostar F₁' (designated as 'GND1') and 'Vesco Seeds Beith Alpha F₁ (26.50 F₁)', designated as 'GND2' and those are the types having common production. The silver nitrate application was performed by the method of spraying on the growth tips of plants and 0, 250, 500, 750, and 1000 ppm silver nitrate doses were administered. The research was conducted with 4 repetitions having 5 plants in each repetition according to the Random Parcel Trial Pattern. In order to determine the effects of the applications, the effects of a number of female flowers and male flowers on generative characteristics of planting periods (spring and fall) were identified and the results were statistically evaluated. According to the results obtained in this research, AgNO_3 has led to the formation of male flowers (no male flower formation in control), has increased the number of male flowers, and has led to a decrease in the number of female flowers. The increase in the number of male flowers varied according to the periods (in 'GND2').

Keywords: *Cucumis sativus* L., female flowers, male flowers, silver nitrate

Introduction

Until recent years, Turkey was dependent on foreign countries in terms of seeds as the initial cultivation material both in open and under cover vegetable breeding. Recently, the growing number of Turkish rehabilitation companies has reduced this dependency and many organizations were also encouraged to work on this issue. The operations in many of the rehabilitation companies are in the form of either bringing in the parent lines here from abroad and the production of their hybrids or the performance of their adaptation trials or taking out just a few generations here. A full rehabilitation program still cannot be executed by many companies. Our country is suitable for rehabilitation studies ecologically and in terms of genetic resources; our own seed needs should be ensured through planning many rehabilitation programs and we could even export the seeds to foreign countries (Şensoy, 2004).

In almost all commercial cucumber growing fields in the world, the gynoeocious and monoecious kinds are used. With these kinds, efficiency is directly proportional to the number of female flowers. However, the ecological factors such as light intensity, light duration and temperature can affect the generative structure of cucumbers. For example, while the high temperatures and long days in the cultivation period increase the number of male flowers of cucumbers, they lead to the reduction of the number of female flowers, and hence cause the degradation of efficiency (Cantliffe, 1981).

In the cucumber plants, there are forms of flowers: andromonoecious (the male flowers and the form of male flowers that were transformed into the hermaphrodite flowers), gynomonoecious (the female flowers and the female flowers that were transformed into the hermaphrodite flowers), trimonoecious (the male flowers, female flowers and hermaphrodite flowers), gynomonodioecious (the monoecious and gynoeocious flowering plants in a population), gynodioecious (the female and hermaphrodite flowering plants in a population) and androdioecious (the male and hermaphrodite flowering plants in a population), in addition to the monoecious (monoecious), androecious (male flowers) and gynoeocious (female flowers) flowers (Vural *et al.*, 2000).

The sex appearance of cucumbers is closely connected to their genetic and environmental conditions (Apan, 1974). As a matter of fact, the cucumber plants have been put under stress from changing the environmental conditions of plants and we have attempted to measure changes in their gender appearance in this study.

There are some strict rules to be complied both by the rehabilitator companies developing the seed varieties and as well as the production companies developing the production in the seed breeding business. Among these, protecting the purity of these varieties is an important issue. The rehabilitator must maintain the sustainability of the rehabilitation program to help protect the characteristics of developed varieties. The persons who perform the seed production are required to timely implement primarily the isolation distance, combating and culturing processes,

and to properly apply the applications (Frankel and Glun, 1977). For this reason, this research focuses only on the sex appearance of individual cucumbers obtained as a result of rehabilitation programs affected by the applications, with untouched desired characteristics.

The *Cucurbitaceae* family is a group of vegetables having rather different characteristics from other vegetable families in terms of its fertilization characteristics. Within this group, the *Cucumis sativus* L. (cucumber) species have very variable flower profiles on the basis of types. Some varieties can engender only female flowers, some can engender female and male flowers, or hermaphrodite flowers together with some other types of flowers. In Turkey in the greenhouse cucumber breeding, the varieties with female flowers are much more preferred due to the parthenocarpic (without pollination and fertilization; those that can bear fruit without needing the male flowers) characteristics of the plant and almost all the used cucumber varieties are constituted from F₁ hybrid varieties. However, the engendering of male and female flowers in parent plants that will be used for the rehabilitation of hybrid varieties is an important problem. This is because almost all F₁ hybrid varieties on the market are parthenocarpic, as also requested in their registrations by the ministry, and surely those do not have male flowers. When the need arose to use them as a source of pollen as a parent plant with a female flower, the engendering of male flowers was attempted by applying different chemicals on those plants. AgNO₃ (silver nitrate) is one of these chemicals used in almost every area in agriculture for many important issues, such as to control plant growth and development, to obtain good yields from the plants, to organize the fruit bearing, to ensure a well-developed plant, to get flowers before their time, to delay flowering, to shorten dormant seasons, and to increase flowering, etc. In this study, it was intended to exhibit the effects of different doses of AgNO₃ applications on the gender formation of the cucumber species, to boost male flower formation; to identify which level of silver nitrate application would best boost the formation of male flowers, and to develop practical recommendations on these issues.

Materials and methods

This study was carried out in the iron skeleton plastic greenhouse having a North-South direction at Bircan Tarim Turizm Tic. Ith. Ihr. ve San. Ltd. Şti. (Turkey), with dimensions of 42 m in width, a length of 54 m, a side height of 2.5 m, and a roof height of 4 m, in the Gazi village of Antalya Province during the spring and autumn 2005 breeding periods.

The seeds used in the trial were 'Mostar F₁' belonging to Bircan Tarim Turizm Tic. Ith. Ihr. ve San. Ltd. Şti. and 'Vesco Seeds Beith Alpha F₁ (26.50 F₁)' belonging to Su Tarim Tic. Ltd. Şti. Those are the types having common production.

In the study, the production method with seedlings was preferred. This meant that the first the seedlings were obtained then switched to the production phase. A 3:1 ratio mixture of peat and perlite was used as the seedlings' soil mortar. The seedlings were grown in vials and 54 (9x6) sized vials were used for this purpose.

Among the used seed types, 'Mostar F₁' belonging to Bircan Tarim Turizm Tic. Ith. Ihr. ve San. Ltd. Şti., was designated as 'GND1' and 'Vesco Seeds Beith Alpha F₁ (26.50 F₁)', belonging to Su Tarim Tic. Ltd. Şti., were designated as 'GND2' to provide convenience and practicality in practice.

The silver nitrate application was performed by the method of spraying on the growth tips of plants and 250, 500, 750, and 1000 ppm silver nitrate doses were administered. Also, the AgNO₃ application was conducted early in the morning (before sunrise) to avoid the sunburn of the plants. The irrigation was conducted in the form of drip irrigation and the necessary fertilization was applied in the form of fertigation.

Results

The obtained data was assessed according to the method of two-factor change analysis, which contains two factors: the amount of silver nitrate and the plantation season. The differences between applications were decided on the basis of the Duncan multiple comparison test ($P < 0.01$), according to importance at the 0.01 level. The statistical analyses were conducted with the SPSS 13.0 package program. No comparisons of types have been performed.

The Effects of AgNO₃ on the number of male flowers

The male flowers were observed only in AgNO₃ applied plants during both the spring and fall seasons.

It was determined that the number of male flowers also increased depending on the increased AgNO₃ doses applied on the 'GND1' variety in the spring season, and the differences between those increases were determined to be statistically significant at the 1% level (Tab. 1).

The AgNO₃ applications increased the number of male flowers on the 'GND1' variety in the spring at a statistically significant level ($P < 0.01$) (Tab. 2).

Tab. 1. The number of male flowers of the AgNO₃ applied 'GND1' variety in the spring season

The applied silver nitrate amounts (ppm)	20 May	27 May	03 June	26 June	03 July	10 July
0 ppm	0.00d*	0.00e	0.00e	0.00e	0.00e	0.00e
250 ppm	5.96c	12.88d	16.46d	19.58d	27.92d	37.54d
500 ppm	13.71b	37.46c	42.54c	50.04c	64.21c	79.75c
750 ppm	14.67b	72.88b	81.04b	90.04b	113.25b	133.92b
1000 ppm	17.25a	82.92a	94.38a	111.67a	147.67a	178.54a

* The differences between values shown with different letters are statistically significant (Duncan multiple comparison test $P < 0.01$).

Tab. 2. The number of male flowers of the AgNO₃ applied 'GND2' variety in the spring season

The applied silver nitrate amounts (ppm)	20 May	27 May	03 June	26 June	03 July	10 July
0 ppm	0.00d*	0.00e	0.00e	0.00e	0.00e	0.00e
250 ppm	6.46c	10.13d	11.42d	13.33d	16.00d	18.29d
500 ppm	6.92c	12.46c	14.54c	19.50c	25.17c	29.04c
750 ppm	8.33b	17.29b	22.46b	28.13b	42.42b	52.79b
1000 ppm	12.75a	26.79a	34.08a	42.58a	61.59a	78.13a

* The differences between values shown with different letters are statistically significant (Duncan multiple comparison test $P < 0.01$).

Tab. 3. The number of male flowers of the AgNO₃ applied 'GND1' variety in the fall season

The applied silver nitrate amounts (ppm)	22 Oct	29 Oct	05 Nov	28 Nov	05 Dec	12 Dec
0 ppm	0.00e*	0.00e	0.00d	0.00e	0.00e	0.00e
250 ppm	10.25d	21.46d	30.04cd	35.67d	48.00d	60.63d
500 ppm	16.58c	42.67c	52.92bc	62.42c	80.59c	99.13c
750 ppm	18.67b	79.13b	92.29ab	103.58b	130.79b	154.46b
1000 ppm	22.25a	92.79a	109.34a	130.63a	170.63a	205.50a

* The differences between values shown with different letters are statistically significant (Duncan multiple comparison test $P < 0.01$).

Tab. 4. The number of male flowers of the AgNO₃ applied 'GND2' variety in the fall season

The applied silver nitrate amounts (ppm)	22 Oct	29 Oct	05 Nov	28 Nov	05 Dec	12 Dec
0 ppm	0.00d*	0.00e	0.00e	0.00e	0.00e	0.00e
250 ppm	8.13c	13.67d	17.63d	21.54d	25.21d	28.21d
500 ppm	9.00c	16.63c	21.71c	28.67c	36.25c	41.13c
750 ppm	11.25b	22.96b	32.38b	40.04b	57.59b	69.83b
1000 ppm	15.75a	36.25a	47.17a	58.67a	82.25a	101.79a

* The differences between values shown with different letters are statistically significant (Duncan multiple comparison test $P < 0.01$).

Tab. 5. The number of female flowers of the AgNO₃ applied 'GND1' variety in the spring season

The applied silver nitrate amounts (ppm)	20 May	27 May	03 June	26 June	03 July	10 July
0 ppm	10.92a	12.83a	14.83a	18.58a	20.83a	22.83a
250 ppm	9.92b	10.92b	11.92b	13.92b	14.92b	15.92b
500 ppm	7.88c	8.88c	9.88c	11.88c	12.88c	13.88c
750 ppm	4.88d	5.88d	6.88d	8.88d	9.88d	10.88d
1000 ppm	1.88e	2.88e	3.88e	5.88e	6.88e	7.88e

* The differences between values shown with different letters are statistically significant (Duncan multiple comparison test $P < 0.01$).

It was determined that the number of male flowers increased depending on the increased AgNO₃ doses applied

Tab. 6. The number of female flowers of the AgNO₃ applied 'GND2' variety in the spring season

The applied silver nitrate amounts (ppm)	20 May	27 May	03 June	26 June	03 July	10 July
0 ppm	16.00a*	18.00a	20.00a	23.75a	25.50a	27.50a
250 ppm	15.00b	16.00b	17.00b	19.00b	20.00b	21.00b
500 ppm	13.00c	14.00c	15.00c	16.92c	17.92c	18.92c
750 ppm	10.00d	11.00d	12.00d	14.00d	15.00d	16.00d
1000 ppm	7.00e	8.00e	9.00e	11.00e	12.00e	13.00e

* The differences between values shown with different letters are statistically significant (Duncan multiple comparison test $P < 0.01$).

Tab. 7. The number of female flowers of the AgNO₃ applied 'GND1' variety in the fall season

The applied silver nitrate amounts (ppm)	22 Oct	29 Oct	05 Nov	28 Nov	05 Dec	12 Dec
0 ppm	8.92a*	10.83a	12.83a	15.83a	17.83a	19.54a
250 ppm	7.92b	8.92b	9.92b	10.92b	11.92b	12.92b
500 ppm	5.88c	6.88c	7.88c	8.88c	9.88c	10.88c
750 ppm	2.88d	3.88d	4.88d	5.88d	6.88d	7.88d
1000 ppm	1.54e	2.54e	3.54e	4.54e	4.79e	5.17e

* The differences between values shown with different letters are statistically significant (Duncan multiple comparison test $P < 0.01$).

Tab. 8. The number of female flowers of the AgNO₃ applied 'GND2' variety in the fall season

The applied silver nitrate amounts (ppm)	22 Oct	29 Oct	05 Nov	28 Nov	05 Dec	12 Dec
0 ppm	14.00a*	16.00a	18.00a	20.75a	22.50a	23.50a
250 ppm	13.00b	14.00b	15.00b	16.00b	17.00b	18.00b
500 ppm	11.00c	12.00c	13.00c	14.00c	15.00c	16.00c
750 ppm	8.00d	9.00d	10.00d	11.00d	12.00d	13.00d
1000 ppm	3.50e	5.75e	6.75e	8.00e	8.42e	8.58e

* The differences between values shown with different letters are statistically significant (Duncan multiple comparison test $P < 0.01$).

to the 'GND1' variety in the fall season, and the differences between those increases were determined to be statistically significant ($P < 0.01$) (Tab. 3).

It was determined that the number of male flowers again increased depending on the increased AgNO₃ doses applied to the 'GND2' variety as happened on the 'GND1' variety in the fall season, and the differences between those applications were determined to be statistically significant at the 1% level (Tab. 4).

The effect of applied AgNO₃ on the number of male flowers on 'GND1' and 'GND2' varieties between seasons was determined to be statistically insignificant level on the 'GND1' variety and significant on the 'GND2' variety ($P < 0.01$) (Tab. 1, Tab. 2, Tab. 3, Tab. 4).

The Effects of AgNO₃ on the number of female flowers

The female flowers were observed both in the control group and AgNO₃ applied plants during both the fall and spring seasons.

The differences between the number of female flowers having applied different AgNO₃ doses to the 'GND1' variety in the spring season were determined to be statistically significant ($P < 0.01$) (Tab. 5).

It was determined that the number of female flowers again decreased depending on the increased AgNO₃ doses applied to the 'GND2' variety in the spring season, as happened in the fall season, and the differences between those decreases were determined to be statistically significant ($P < 0.01$) (Tab. 6).

It was determined that the number of female flowers decreased depending on the increased AgNO₃ doses applied to the 'GND1' variety in the fall season, and the differences between those decreases were determined to be statistically significant ($P < 0.01$) (Tab. 7).

It was determined that the number of female flowers decreased depending on the increased AgNO₃ doses applied to the 'GND2' variety in the fall season, and the differences between those decreases were determined to be statistically significant at the 1% level (Tab. 8).

The effects of applied AgNO₃ on the number of male flowers on the 'GND1' and 'GND2' varieties were determined to be statistically significant in terms of differences between seasons ($P < 0.01$) (Tab. 5, Tab. 6, Tab. 7, Tab. 8).

Discussion

Since the varieties used in the study are parthenocarpic, the male flowers were observed only on AgNO₃ applied plants both in the fall and spring seasons.

It was determined that the number of male flowers also increased depending on the increased AgNO₃ doses applied to the 'GND1' variety in the spring season, and the differences between those increases were determined to be statistically significant at the 1% level. The AgNO₃ applications increased the number of male flowers on the 'GND1' variety in the same season at a statistically significant level ($P < 0.01$).

It was determined that the number of male flowers increased depending on the increased AgNO₃ doses applied to the 'GND1' variety in the fall season and the differences between those increases were determined to be statistically significant ($P < 0.01$). It was determined that the number of male flowers again increased depending on the increased AgNO₃ doses applied to the 'GND2' variety, as happened on the 'GND1' variety in the same season, and the differences between those applications were determined to be statistically significant at the 1% level.

In the measurements, the highest number of male flowers was determined as 1000 ppm, and this was followed by doses of 750 ppm, 500 ppm and 250 ppm AgNO₃ applications, respectively.

It was determined that the number of male flowers increased depending on the increased AgNO₃ doses applied

to the 'GND1' and 'GND2' varieties in the fall and spring seasons, and the differences between those increases were determined to be statistically significant ($P < 0.01$). This situation complies with the findings regarding similar opinions that the AgNO₃ applications cause an increase in the number of male flowers in cucumbers (Beyer, 1976; Den Nijs and Visser, 1980; Anonymous, 1987; Robinson and Walters, 1997; Hallidri, 2004).

The effects of applied AgNO₃ on the number of male flowers on the 'GND1' and 'GND2' varieties between seasons were determined to be statistically insignificant on the 'GND1' variety and significant on the 'GND2' variety.

In the cucumber plants of the 'GND1' and 'GND2' varieties exposed to silver nitrate applications, the number of male flowers also increased together with the increasing silver nitrate concentration. The increase in the fall planting period was more than the increase in the spring planting period. The greater increase in the number of male flowers during the fall season can be explained by the fact that the planting period was during the warmer months or the length of the days.

Since More and Munger (1986) determined that the light period after the AgNO₃ application also affected the formation of male flowers and that the formation of male flowers decreased in the long (15-20 hours) light periods, the results of the aforementioned research are consistent with the results obtained in our study.

In the literature, it has been reported that morphological and physiological changes that occur in plants differ depending on air temperature (Krug and Liebig, 1989; Gosselin and Trudel, 1986; Ney and Turc, 1993; Reath and Wittwer, 1952).

The control plants were in the first ranking in both periods in terms of the obtained number of female flowers. In the silver nitrate applied plants, it was observed that the decrease in the number of female flowers with the increase of the applied AgNO₃ doses was statistically significant ($P < 0.01$). The planting periods were also found to be effective in this decrease detected in the number of female flowers. Despite the dominant effects of the solutions containing of silver nitrate concentrations, the number of female flowers was greater in the spring planting period compared to the fall planting period.

The result that the determined decrease in the number of female flowers in the 'GND1' and 'GND2' varieties with the increasing doses of AgNO₃ in fall and spring was statistically significant ($P < 0.01$) is consistent with the results of Takahashi and Jaffe's (1984) study intended to determine the role of the hormone-controlled sex formation in cucumbers.

The reason for the decrease in the number of female flowers can be connected to the inhibition of signal production of the internal ethylene responsible for the development of the female flowers by applied AgNO₃ in cucumber plants. Prodanovic and Stankovic (2002), who performed research similar to this study with the purpose of identifying the possibilities of optimal sex changes, investigated the effectiveness of silver nitrate solutions ap-

plied in different concentrations (0.01%, 0.02%, 0.03%, 0.04%) and planting seasons (spring, summer) for the determination of gender in two kinds of cucumbers in their study. They identified that the number of male flowers increased as the silver nitrate concentration increased, and that this increase was more significant in the first planting season than the second planting season; that the silver nitrate decreased the number of female flowers compared to the control group in both of the planting seasons; that the optimal sex change was achieved with 0.02% silver nitrate solution, while this effect was occurring with 0.03% silver nitrate solution in the period with a longer daylight period. In addition, the researchers concluded that the sex determination within the limits determined by genotype was affected by environmental conditions; that the internal ethylene responsible for the determination of female sex in the gynoecious cucumber varieties is blocked by the Ag⁺ ion effect (ethylene action blocker) and that the silver ions cause the male flower development; that the silver nitrate application needs to be observed in the seed production and selection processes of gynoecious cucumber varieties; that the silver to obtain male flowers can mask the sex; that some plants, which will serve as pollen donors, can be treated with silver nitrate in this study.

The silver nitrate application did not lead to the formation of hermaphrodite flowers having both male and female characteristics in cucumber varieties in both cultivation periods.

In this study, similar to Hallidri's study (2004), the effects of the silver nitrate concentration were examined in determining the gender in parthenocarpic and gynoecious cucumbers; it was observed that the formations of male flowers were connected to the silver nitrate concentrations; that the silver nitrate applications with low concentrations were insufficient to engender male flowers; that the highest number of male flowers were observed on the plants sprayed with 1000 ppm silver nitrate solutions; that the weak plants recovered within 7 to 10 days after they had been sprayed with 750 and 1000 ppm silver nitrate solutions.

The research, which was intended to quantify the effects of different concentrations of silver nitrate applications and planting periods in the determination of sex of the 'GND1' and 'GND2' varieties having the AgNO₃ applications in both plantings periods, shows the importance of the determination of appropriate silver nitrate doses.

This research can be applied to the large-scale commercial seed production of gynoecious x gynoecious cucumber hybrids by selecting the most appropriate silver nitrate concentrations and by obtaining male parents that will be used as the pollen sources from gynoecious types appropriate to provide male flowers. The 1000 ppm AgNO₃ application is recommended considering the number of male flowers in the cases when it will be used in the production of F₁ cucumber seeds. In light of the results obtained, this research has proved to be guidance to develop practical recommendations on the basis of hybrid cucumber seeds production and for subsequent studies.

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