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# INFLUENCE OF PACLOBUTRAZOL AND CYCOCEL ON GROWTH, FRUITFULNESS AND PHYSIOLOGICAL CHARACTERISTICS OF OLIVE (*OLEA EUROPAEA* L.)

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## Abstract

The olive (*Olea europaea* L.) is an evergreen tree, but postulates chilling for fruiting. Foliar application of growth retardants such as paclobutrazol and cycocel have successfully been tried to increase the productivity of fruit trees by promoting flowering, fruiting and by ensuring optimum use of available water in the plant system. Use of paclobutrazol and cycocel has also been advocated in drought prone areas to increase the degree of fruitfulness in olive. Paclobutrazol and cycocel have extensively been used to regulate cropping of subtropical fruit crops like mango and litchi, which are more prone to alternate bearing. But these chemicals have not yet been tried extensively for crop regulation in olive. Keeping this in view, a field experiment was carried out in the olive experimental orchards of the Department of Fruit Science, Dr. Y S Parmar University of Horticulture & Forestry, Nauni, Solan (H.P.), India; to infer the influence of growth retardants on growth, flowering and physiological traits in olive. The experiment consists of different treatments *i.e.* paclobutrazol and cycocel in 500, 1000, 2000 ppm concentrations were given to olive trees. Paclobutrazol application of 2000 ppm significantly reduced vegetative growth *i.e.* shoot extension growth (4.60 cm), relative growth rate (0.043 $cm^{-1}$ $cm^{-1}$ $month^{-1}$), leaf area (4.15 $cm^2$) and increased bloom intensity (0.55%), proportion of perfect flowers (57.00 %) and fruit set (6.33%) in olive. Furthermore, it also significantly decreased photosynthetic rate (5.52 µ mol $m^{-2}$ $s^{-1}$), transpiration rate (0.031 m mol $m^{-2}$ $s^{-1}$), stomatal conductance (0.148 mol $m^{-2}$ $s^{-1}$) and increased amino acid content in leaf *i.e.* Asparagine content (1.76 mg/g), Glutamine content (1.76 mg/g) and Tryptophan content (0.145 mg/g). The present investigation has evinced that application of 2000 ppm paclobutrazol application was most efficacious in reducing vegetative growth, which in turn enhanced bloom intensity, perfect flowers and fruit set in olive. This treatment also reduced the photosynthetic rate, transpiration rate, stomatal size and stomatal conductance in olive.

***Key words :*** Olive, paclobutrazol, cycocel, stomatal density, photosynthesis, transpiration.

## Introduction

Olives are mostly grown for their oil, which is extracted from its fruits. Olive oil possesses numerous biological and medicinal values. In Himachal Pradesh, olives are grown on a limited scale in Kullu, Shimla, Solan and Sirmour districts. Olive trees have been designated as a drought tolerant plant. The growth flush of olive trees in these areas is confined to a very short period of 2-3 months due to the occurrence of monsoon rains. These areas experience mild and inadequate winter rains, which resulted in insufficient chilling of olive trees. One of the major concerns of olive growers in sub-tropical areas of monsoon type of climate is that yields are often irregular and uneconomical (Bartolini and Fabbri, 1994). A comprehensive knowledge about water use, transpiration and photosynthetic rates as a function of environmental conditions is crucial for optimizing orchard management in relation [1]to the efficient use of available soil water (Diaz *et al*., 2004). Soil and foliar application of growth retardants such as paclobutrazol and cycocel have successfully been tried to increase the productivity of olive trees by enhancing their flowering, fruiting and by ensuring optimum use of available water in the plant system. Use of paclobutrazol and cycocel has also been advocated in drough[t] prone areas to increase the degree of fruitfulness in olive. Paclobutrazol has been reported to reduce vegetative flush and increase the proportion of flower bearing shoots (Lal *et al*., 2000). Application of

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these growth retardants have ensured early and profuse flowering in mango in the following year. Cycocel application at 1000 ppm in late September also resulted in enhanced flowering in Kagzi lime (Desai *et al*., 1982). Paclobutrazol and cycocel have extensively been used to regulate cropping of subtropical fruit crops like mango and litchi, which are more prone to alternate bearing, but these chemicals have not yet been tried for crop regulation in olive. Therefore, present studies were proposed to regulate and improve the productivity of olive trees by appropriate manipulation of their physiological status. The investigation was, therefore, undertaken to study the influence of growth retardants on growth, flowering and physiological characteristics of olive trees.

## Materials and Methods

The present investigation was conducted at the experimental orchard of Department of Fruit Science, Dr. Y.S. Parmar University of Horticulture and Forestry, Nauni, Solan (30°20' North latitude, 77°11' East longitude, altitude 1240 amsl) and falls under under the mid hill zone of Himachal Pradesh, where summers are moderately hot (35-38°C). The winter season is experienced between the months of December to February. However, maximum rainfall is received during the month of June to September, whereas, light winter showers are experienced during the months of January to March. The average annual rainfall of the area ranges from 1000-1300 mm. The trial was laid out with seven treatments in a randomized block design and replicated thrice. Uniform, healthy and disease free trees of 17 years of age were selected. The treatments studied in this experiment were $T_1$- Paclobutrazol 500 ppm, $T_2$- Paclobutrazol 1000 ppm, $T_3$- Paclobutrazol 2000 ppm, $T_4$- Cycocel 500, $T_5$- Cycocel 1000 ppm, $T_6$- Cycocel 2000 ppm and control (no application). Growth retardants were applied in the first week of November. Twenty-five healthy uniform shoots of previous season's growth were selected on all sides of the tree covering entire periphery to record observation of shoot extension growth, relative growth rate, leaf area and stomatal density. The length of each shoot was measured at the beginning and end of growing season to determine shoot extension growth. Relative growth rate was determined by the recording new shoot extension growth occurred by the entire growing season time to time. Leaf area was determined by using leaf area meter, LI-COR Model-3100. Stomatal density and stomatal size were done with the method suggested by Beakbane and Majumdar. The stomata size was recorded with the help of Leica Stereoscopic Microscope. The blooming intensity of experimental trees was recorded in accordance with formulae suggested by Westwood. For the ascertaining proportion of perfect flowers, twenty-five inflorescences have sampled all sides of the experimental trees. The individual flowers of these inflorescences were carefully examined to determine their sex *i.e.* perfect and staminate on the basis of presence or absence of a fully developed and functional pistil. The observations on the fruit set were recorded at two weeks after petal fall and then were again confirmed at six weeks after full bloom so as to allow sufficient time for the abscission of unfertilized parthenocarpic fruits. Transpiration rate, photosynthetic and stomatal conductance were recorded through LICOR 6200. Amino acids were estimated by the method suggested by Sharma (2002).

## Results and Discussion

Results of the study revealed that the effect of the treatments like paclobutrazol and cycocel of different concentrations showed significant effects on growth flowering and physiological characteristics of olive. It is evident from the data depicted in table 1 that growth retardants have shown a significant effect in decreasing shoot extension growth of experimental trees and this effect was proportional to their concentration used. Therefore, the maximum decrease in shoot extension growth in comparison to control was observed in 2000 ppm paclobutrazol (4.97 cm), which was statistically at par with cycocel (4.98 cm) at 2000 ppm. The treatments comprising of paclobutrazol at 500 ppm and cycocel at 500 ppm were also statistically at par with one another. It was also apparent from the above results that paclobutrazol and cycocel have shown more or less identical trend on the reduction of shoot extension. It is evident from data that the extent of decrease in relative growth rate of olive was inversely proportional to the concentration of growth retardants used. Therefore, among different treatments, the maximum and significant decrease in relative growth rate over control was observed in 2000 ppm paclobutrazol (0.043 $cm^{-1}$ $cm^{-1}$ $month^{-1}$) followed by 2000 ppm cycocel (0.045 $cm^{-1}$ $cm^{-1}$ $month^{-1}$), but former treatment was significantly different than the latter. The maximum decrease (4.14 $cm^2$) in leaf area was recorded in paclobutrazol 2000 ppm, which was significantly higher than rest of the treatments whereas, minimum reduction (5.36 $cm^2$) in leaf area was recorded under control. This reduction in shoot extension growth, relative growth and leaf area of treated trees might be attributed to the fact that paclobutrazol acted as a potential inhibitor of gibberellins biosynthesis and thus, has caused a sharp decrease in the shoot extension growth, relative growth rate and leaf area of olive trees

**Table 1 :** Effect of different concentrations of paclobutrazol and cycocel on growth parameters of olive.

| Cultivars | Growth (cm) | Relative growth rate ($cm^{-1}$ $cm^{-1}$ $month^{-1}$) | Leaf area ($cm^2$) |
|---|---|---|---|
| $T_1$ (Paclobutrazol 500 ppm) | 5.95 | 0.059 | 4.69 |
| $T_2$ (Paclobutrazol 1000 ppm) | 5.37 | 0.053 | 4.49 |
| $T_3$ (Paclobutrazol 2000 ppm) | 4.60 | 0.043 | 4.15 |
| $T_4$ (Cycocel 500 ppm) | 6.01 | 0.060 | 4.63 |
| $T_5$ (Cycocel 1000 ppm) | 5.35 | 0.054 | 4.43 |
| $T_6$ (Cycocel 2000 ppm) | 4.74 | 0.045 | 4.80 |
| $T_7$ (Control) | 6.67 | 0.067 | 5.36 |
| $CD_{0.05}$ | 0.30 | 0.002 | 0.38 |

**Table 2 :** Effect of different concentrations of paclobutrazol and cycocel on stomatal characteristics of olive

| Cultivars | Stomatal size (µm) | | Stomatal density (No.) |
|---|---|---|---|
| | Length | Breadth | |
| $T_1$ (Paclobutrazol 500 ppm) | 15.06 | 9.56 | 79.17 |
| $T_2$ (Paclobutrazol 1000 ppm) | 13.54 | 9.17 | 78.00 |
| $T_3$ (Paclobutrazol 2000 ppm) | 11.21 | 8.52 | 77.67 |
| $T_4$ (Cycocel 500 ppm) | 15.72 | 9.83 | 78.83 |
| $T_5$ (Cycocel 1000 ppm) | 14.19 | 9.39 | 77.83 |
| $T_6$ (Cycocel 2000 ppm) | 13.10 | 9.03 | 77.50 |
| $T_7$ (Control) | 16.59 | 10.27 | 76.67 |
| $CD_{0.05}$ | 2.45 | 0.68 | 1.83 |

by decreasing the rates of cell division and cell elongation (Rademacher, 1991). Antognozzi *et al*. (1989) reported that foliar application of paclobutrazol at different concentrations (1000, 2500 or 5000 ppm) reduced tree height, shoot extension growth, internodal length and leaf area of the olive trees, thus confirmed our findings. Similar results were also reported by Wang *et al*. (1997). There is a considerable evidence to show that application of paclobutrazol or cycocel reduces vegetative growth in terms of shoot elongation, leaf area and relative growth rate in many fruits crops by interrupting GA biosynthesis (Singh and Singh, 2003).

The data presented in table 2 revealed that the increase in growth retardants application decreased the stomatal size and increased stomatal density of olive. However, the minimum value of stomatal size (11.21µm) in length and (8.52 µm) in breadth was recorded in 2000 ppm which was statistically at par with cycocel 2000 ppm and paclobutrazol at 1000 ppm. Maximum stomatal density (79.17) was observed in paclobutrazol at 500 ppm, which was statistically at par with cycocel at 500 ppm (78.83). All treatments of paclobutrazol and cycocel used were statistically at par with one another but they were superior to control, which possessed a minimum value of stomatal density (76.67). Paclobutrazol and cycocel at 2000 ppm resulted in a maximum increase in stomatal density of treated plants but they caused a simultaneous decrease in the stomatal size of treated leaves. The increase in stomatal density might be due to the fact that with the reduction of stomatal size a simultaneous increase in the number of stomata per unit area of the leaf occurred. These findings are in line with the findings of Sharma (1998), who also reported that higher stomatal density following paclobutrazol application in apple and almond, respectively. Paclobutrazol is a potent inhibitor of gibberellic acid biosynthesis and its action might be mediated through abscisic acid produced within the leaf mesophyll tissues and translocated to the stomata.

The perusal of the data given in fig. 1 revealed that different treatments have failed to show their discernible influence on the time and duration of flowering in olive. However, flowering began one day earlier among different treatments as olive compared to control. A similar trend was also found at the time of full bloom and cessation of the flowering period. It was also apparent from above that slight shift in the time of full bloom was observed in all treatments except cycocel at 500 ppm. These results are in conformity with those of Salazor and Vazaquez (1997) in mango. Proetti and Tombesia (1996) also observed that paclobutrazol applications markedly increased the extent of flowering in olive might be due to the fact that paclobutrazol and cycocel reduces vegetative growth which results in reduced competition of vegetative parts for soil moisture and nutrients, thus increased availability of metabolites assimilation to floral buds, which ultimately might have enhanced the time and duration of flowering.

It is evident from table 3 that bloom intensity of olive trees was significantly influenced by the different treatments. Among different treatments applied, maximum bloom intensity (0.55%) was recorded in paclobutrazol at 2000 ppm, which is statistically at par with cycocel at 2000 ppm (0.54%). However, minimum bloom intensity (0.48%) was observed in control, which was much lower than all other treatments. A perusal of data embodied in table 1, the maximum proportion of perfect flowers (57.00%) was observed in 2000 ppm paclobutrazol, which was significantly higher than in remaining treatments. Among other treatments, paclobutrazol at 1000 ppm and 500 ppm and cycocel at

**Table 3 :** Effect of different concentrations of paclobutrazol and cycocel on flowering parameters of olive.

| Treatments | Bloom Intensity (%) | Proportion of perfect flower (%) | Fruit set (%) |
|---|---|---|---|
| $T_1$ (Paclobutrazol 500 ppm) | 0.53 | 53.09 | 4.38 |
| $T_2$ (Paclobutrazol 1000 ppm) | 0.52 | 54.46 | 5.20 |
| $T_3$ (Paclobutrazol 2000 ppm) | 0.55 | 57.00 | 6.33 |
| $T_4$ (Cycocel 500 ppm) | 0.53 | 50.75 | 4.48 |
| $T_5$ (Cycocel 1000 ppm) | 0.54 | 53.68 | 5.01 |
| $T_6$ (Cycocel 2000 ppm) | 0.48 | 54.49 | 5.98 |
| $T_7$ (Control) | 0.68 | 48.62 | 3.40 |
| $CD_{0.05}$ | 0.01 | 0.089 | 0.09 |

**Table 4 :** Effect of different concentrations of paclobutrazol and cycocel on physiological characteristics of olive.

| Treatments | Photosynthetic rate (μ mol $m^{-2}$ $s^{-1}$) | Transpiration rate (m mol $m^{-2}$ $s^{-1}$) | Stomatal conductance (mol $m^{-2}$ $s^{-1}$) |
|---|---|---|---|
| $T_1$ (Paclobutrazol 500 ppm) | 6.60 | 0.044 | 0.271 |
| $T_2$ (Paclobutrazol 1000 ppm) | 6.04 | 0.034 | 0.179 |
| $T_3$ (Paclobutrazol 2000 ppm) | 5.52 | 0.031 | 0.148 |
| $T_4$ (Cycocel 500 ppm) | 6.74 | 0.049 | 0.224 |
| $T_5$ (Cycocel 1000 ppm) | 6.30 | 0.040 | 0.192 |
| $T_6$ (Cycocel 2000 ppm) | 5.71 | 0.034 | 0.156 |
| $T_7$ (Control) | 7.33 | 0.058 | 0.279 |
| $CD_{0.05}$ | 0.12 | 0.005 | 0.009 |

**Table 5 :** Effect of different concentrations of paclobutrazol and cycocel on amino acid contents of olive.

| Treatments | Asparagine content (mg/g) | Glutamine content (mg/g) | Tryptophan content (mg/g) |
|---|---|---|---|
| $T_1$ (Paclobutrazol 500 ppm) | 1.72 | 1.72 | 0.129 |
| $T_2$ (Paclobutrazol 1000 ppm) | 1.74 | 1.73 | 0.139 |
| $T_3$ (Paclobutrazol 2000 ppm) | 1.76 | 1.76 | 0.145 |
| $T_4$ (Cycocel 500 ppm) | 1.71 | 1.70 | 0.129 |
| $T_5$ (Cycocel 1000 ppm) | 1.73 | 1.74 | 0.135 |
| $T_6$ (Cycocel 2000 ppm) | 1.74 | 1.75 | 0.140 |
| $T_7$ (Control) | 1.70 | 1.70 | 0.125 |
| $CD_{0.05}$ | 0.014 | 0.02 | 0.02 |

1000 and 2000 ppm were statistically at par with one another in respect of their proportion of perfect flowers, whereas their minimal (48.62%) proportion of perfect flowers was found under control. It is quite conspicuous from the data that fruit set of olive was significantly influenced by foliar application of growth retardants. Among different treatments, maximum fruit set (6.33%) was obtained in paclobutrazol at 2000 ppm, which was significantly higher than in other treatments except for cycocel at 2000 ppm. But fruit set in 2000 ppm paclobutrazol and 2000 ppm cycoccl was statistically at par with each other. All treatments of growth retardants were statistically superior to control in their fruit set. This might be attributed to their interference with gibberellic acid biosynthesis, which in turn creates favourable conditions for flower bud differentiation and subsequent development of floral organs (Lal *et al*., 2000). Faizan *et al*. (2000) also observed higher fruit set in litchi following application of paclobutrazol and thus supported our findings. Similar results were also by Porlingis *et al.* (1999), who observed olive trees treated with paclobutrazol at 500 mg per plant showed a higher fruit set (23.1%) than the control trees (12.6%). Increase in fruit set after application of paclobutrazol was also reported by Proietti and Tombesia (1996).

Application of growth retardants has significantly influenced the plant physiological processes in olive as evident from the data given in table 4. A significant decrease in the rate of photosynthesis of olive trees was observed, when compared to control, where maximum photosynthetic (7.33 μ mol $m^{-2}$ $s^{-1}$) rate was observed. Among other growth retardant treatments, the maximum value of photosynthesis (6.74 μ mol $m^{-2}$ $s^{-1}$) was observed in cycocel 500 ppm, which was significantly higher than paclobutrazol at the same concentration. The decrease in the rate of photosynthesis was proportional to the concentration of growth retardants used and the minimum values of photosynthesis were found in treatments, wherein maximum concentrations of these growth retardants were used. Antognozzi *et al*. (1989) also reported reduced rate of photosynthesis in olive trees given four sequential treatments with paclobutrazol. This decrease in photosynthetic rate might be attributed to alteration in leaf orientation, which become much less erect during daylight hours and thus may reduce light absorption and consequently reduced rate of photosynthesis occurred (Sharma, 1998). The other reasons for paclobutrazol induced reduction in photosynthesis could be due to its decreasing effect on leaf area, which resulted in reduced photosynthetic surface per plant. Minimum transpiration rate (0.031 μ mol $m^{-2}$ $s^{-1}$) was recorded in paclobutrazol

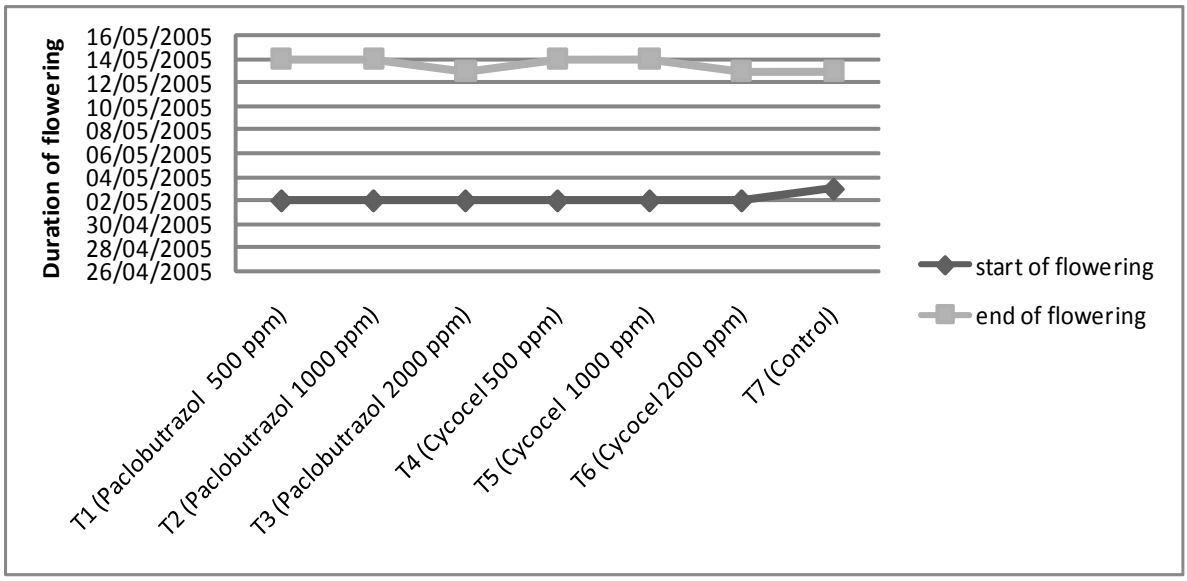


**Fig. 1 :** Effect of different concentrations of paclobutrazol and cycocel on time and duration of flowering.

at 2000 ppm, which was statistically at par with treatments of paclobutrazol and cycocel 1000 ppm. Similarly, treatments comprising of paclobutrazol at 500 ppm was statistically at par with cycocel 500 ppm. Furthermore, the minimum value (0.148 mol $m^{-2}$ $s^{-1}$) of stomatal conductance was found in paclobutrazol at 2000 ppm. However, this treatment was statistically at par with cycocel at 2000 ppm. But other two treatments of these growth retardants have shown significant differences among themselves. However, much higher values of Stomatal conductance were observed in lowest concentrations of these two growth retardants. Highest value (0.279 mol $m^{-2}$ $s^{-1}$) of stomatal conductance was recorded in control. The decreasing effect of paclobutrazol on transpiration rate and stomatal conductance was relatively higher than that of cycocel. This decrease in the rate of transpiration and stomatal conductance following application of growth retardants might possibly be attributed to the consequence of partial closure of stomata of the treated plants (Bora and Mathur, 1989).

The data presented in table 5 revealed, significant differences in amino acids after growth retardants application in olive trees. Among different treatments, maximum value of asparagine (1.76 mg/g), glutamine (1.76 mg/g) and tryptophane (0.145 mg/g) contents were observed in 2000 ppm paclobutrazol, which were statistically superior to all the remaining treatments, whereas treatments comprising of paclobutrazol at 1000 ppm, cycocel at 2000 ppm and 1000 ppm were statistically at par with one another and were superior to all the remaining treatments. Minimum values were observed under control. These results are in conformity with Thakur (1998), who also reported that total amino acids were significantly higher following paclobutrazol treatment in olive trees, which were under water stress. Similar observations were also reported by Chaudhury and Gupta (1996). These amino acids might have acted like messengers evoking the process of floral initiation by increasing availability of essential metabolites at the growing apices of fruit trees (Proietti and Tombesia, 1996). These retardants also caused a shift in autumn-winter dormancy (starting earlier and finishing earlier) and an increase in RNA : DNA ratio in the overwintering buds of the treated plants. The increase in total amino acids might have caused a simultaneous increase in free amino acids (Kuryata and Sogur, 1994).

Therefore, it can be concluded from the field investigation that application of paclobutrazol at 2000 ppm was more effective in controlling vegetative growth, which in turn stimulate flower bud formation steerages to enhanced flowering and higher fruit set in olive. This treatment also reduced photosynthetic rate, transpiration rate, stomatal size and stomatal conductance, which helps plant to perform better under drought prone areas.

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