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Effect of Growth Regulators on the Dormancy of Mulberry (*Morus alba* L.) Winter Buds in Taegu, Korea

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大邱地方에서의 뽕나무 休眠打破를 爲한 生長調節劑 處理 效果

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Summary

These experiments were carried out to define the rest period of mulberry by treating growth regulators in Taegu, Korea. Results obtained were as follows:

It was recognized that the depth of rest in Taegu, Korea, was not deeper than that in Tokyo and Kagoshima, Japan.

The rest of mulberry was begun at the end of September, subsequently became deeper through the first October into the late October and then turned gradually into quiescence by the beginning of November.

Buds sprayed by gibberellic acid (GA_3) 10ppm and urea 0.5% were promoted to sprout, while naphthalene acetic acid (NAA) 0.02% inhibited strongly bud sprouting and abscisic acid (ABA) 20ppm had no effect on the rest of mulberry.

Gibberellic acid 10ppm enhanced the rate of green color of bud after incubation for 10 days at 30°C.

By the portion of mulberry stems, the depth of rest was different that the middle buds were less dormant than those lower.

The optimal time required for the mulberry winter bud break is 15 days incubation at 30°C as treated with GA_3.

Introduction

In 1986, when frost damage experiments were conducted on 17 November with mulberry stems collected from the Cheonbuk Provincial Silkworm Egg Production Station in Cheonjoo, most of the cuttings were sprouted under the controlled condition keeping at 30°C. But so far, we have understood that it is the deep rest period of mulberry during November in Korea (Kim, 1987). Therefore, these experiments were conducted to confirm the mulberry rest period.

Beyond mulberry, most of the perennial woody plant undergo a period of rest when the apical bud ceased to grow (Delvin and William, 1984; Hazama and Naoi, 1965; Krishnamoothy, 1981; Malcolm, 1986; Nicolas and Louis, 1966; Salisbury and Ross, 1975; Vegis, 1963), for giving an ecological advantage to the plant to survive and withstand unfavo-

rable conditions during the winter.

Both Wareing & Phyllips (1978) and Gregory (1987) reported that, bud development had three phases of dormancy, namely, summer dormancy or ectodormancy, rest or endodormancy and quiescence or ecodormancy. Similar result was indicated by Kent (1987) and Loyed (1987).

Physiologically, the mulberry and a number of woody species in temperate zones should require a certain amount of chilling periods in winter to break the dormancy (Westwood and Bjornstad, 1968). In areas with mild winter (Wainwright *et al.*), the chilling requirement for many fruit trees is often not met resulting in poor bud break and delayed foliation. This problem has been solved by application of artificial dormancy breaking treatments which compensate for the lack of chilling. However, this chilling requirement has been reported to induce an increase in the concentration of gibberellic acid which is a factor of breaking dormancy (Danvta and Lewak, 1978; Frankland and Wareing, 1972). The level of abscisic acid has been shown to diminish in seeds and embryos of different species during stratification (Martin *et al.*, 1969; Sondheimer *et al.*). Latter, Kaminski (1971), Janna & Wareing (1968) established that abscisic acid is an antagonist of gibberellic acid. Similar result was confirmed by Lin & Boe (1972).

Therefore, several investigations have been reported in a number of plant species about rest and effect of chemicals on rest of winter buds, but relatively little attention has been given to mulberry.

In this connection, Hamada (1931) reported that around Tokyo, Japan, which is located at the same latitude (36°N) as Taegu, Korea, the rest began at the end of September, became deeper through late October into late November and then turned gradually into quiescence from the beginning of December. The same results were confirmed in Kagoshima, Japan by Yahiro (1962).

Powell (1987) reported that in woody plant, maximum rest intensity occurs from one week after leaf falls to one month after leaf falls.

Another research (Suzuki *et al.*, 1987) indicated that basal buds of the mulberry were less dormant than those toward the apex.

Yahiro (1968) and Hayashi (1971) pointed out, that content of growth substances in mulberry winter buds under the rest was increased in deeper rest state, while decreased before sprouting because buds were released from the rest stage.

ABA has been known as an important dormancy hormone in the higher plant(Irwin, 1983). However, Ohyama & Oka (1973) emphasized that low concentration of ABA had no effect on breaking of mulberry winter buds.

Studies have been shown that application of naphthalene acetic acid delayed bud break in apple (Elving and Forstry, 1977) and inhibited mulberry bud break (Yahiro, 1965). Similar results have been reported in other *in vitro* study (Oka and ohyama, 1975).

It is important to note some changes in content of nucleoprotein and other biochemical substances in the treated buds with growth regulators in apple, orange, lemon, peach, grape(Bewley and Black, 1982; Champagnat and Come, 1986; Cottignies, 1986; Gregory, 1987; Ko and Kang, 1981; Voyiatzir and Porlingis, 1986) and in mulberry winter bud (Wareing *et al.*, 1968; Yahiro and Hayashi, 1971a, 1971b; Yahiro, 1954) during rest period, rest release and bud sprouting.

Furthermore, in tropical zones, mulberry has no dormant period and continue to grow throughout the year, whereas in temperate regions, mulberry leaves are available for rearing purposes only during May to October. Therefore, by the end of September, there could not be found good quality of soft leaves for the young silkworm. Production of good mulberry leaves is a problem being solved by the inhibition of the onset of the rest by some artificial means that often aid farmers in an economic way. Some of chemicals found useful in the release of dormancy are urea and GA_3 acting as a bud dormancy breaker in fruit plants (Delvin and William, 1984; Fernadez and Martin, 1987; Vegis, 1963), mulberry winter buds (Iwata and Nakagawa, 1972; Ohyama and Oka, 1975; Yahiro, 1965; Yahiro and Hayashi, 1966) and seeds (Judith, 1985).

The present study was performed with an objective to define the rest period of mulberry winter buds,

in Taegu, Korea, and to investigate the effect of some chemicals on the termination of the bud rest.

Materials and Methods

Four years old mulberry trees, Kaeryangppong, *Morus alba* LINN were used in the present study. The experiment was performed in the mulberry field located in Kyungpook National University.

Four chemicals were used namely: gibberellic acid (GA_3) 10ppm, abscisic acid (ABA) 20ppm, α-naphthalene acetic acid (NAA) 0.02% and urea 0.5%. The chemicals were sprayed twice, on September 7 and September 15, 1987 before the bud rest. Two hundred mililiters of the solutions per tree was sprayed on both sides of leaves at each time.

On October 15, 1987, 30 days after chemicals spraying, 3 branches were collected from each treatment, and were divided into 3 portions, upper, middle and lower portion. Mainly, middle and lower portions of branches were used, because upper portions were previously dried and not sprouted normally. Cuttings were 10 to 15cm long with 3 buds. 18 cuttings of each portion were planted on moistened sand.

Cuttings were incubated at 30°C under the dark and proper moisture condition.

Cuttings were sampled every 15 days from October 15, 1987 to January 15, 1988.

The effect of chemical application on the induction of bud sprouting was evaluated at every 10, 15 and 20 days after incubation. The evaluation of bud sprouting followed the bud development stages:

1. Green color of bud.
2. Opening bud.
3. Opened bud without petiole.
4. Opened bud with petiole.

Results and Discussion

Physiological studies on winter buds of mulberry in 1987/88 in Taegu, Korea, and the observations were taken on dormant cuttings after forcing at 30°C at different times during autumn and winter and the result of these studies further illustrated that the mulberry bud entered the rest period (Fig. 1) from

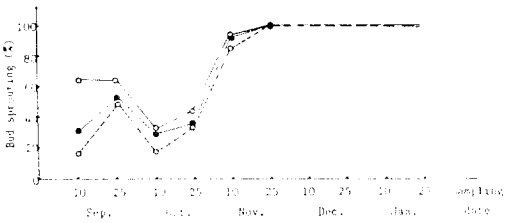

Fig. 1. Percentage of mulberry bud sprouting during the rest period in Taegu, preserved for 20 days at 30°C opening bud (—○—), opened bud without petiole (—●—), opened bud with petiole (···○···).

the late of September to late of October.

By the beginning of November, the mulberry bud was released from the rest and entered the second phase of dormancy, quiescence, imposed by environmental conditions and at last the preparation of bud sprouting became almost complete by the end of November. In case of cuttings sampled on September 10, bud sprouting was bad because of dry condition during incubation.

In earlier studies (Hamada, 1931; Yahiro, 1962), it was indicated that either in Tokyo, Japan, which is located at the same latitude (36°N) as Taegu, Korea or in Kagoshima (32°N), Japan, the rest intensity of mulberry was deep from late October to late November, and turned gradually into quiescence from the beginning of December. It could be said that the depth of rest in Taegu is not deeper than that in Tokyo and Kagoshima. On the other hand, mulberry trees in Korea entered rest state one month earlier than that in Japan. While Powell (1987) reported that the maximum rest intensity of woody plant occured from one week to one month after leaf falls.

In fact, the maximum intensity of the rest was observed around October 10, and the intensity gradually declined until the end of October. The release from this state began by the beginning of November and this period coincided with the chilling temperature(Fig. 2) that enables trees to undergo a period of stratification and is required to resume growth.

By the portion of mulberry stems, the middle part exhibited the best sprouting percentage in comparison to lower portion at the development stage of opening

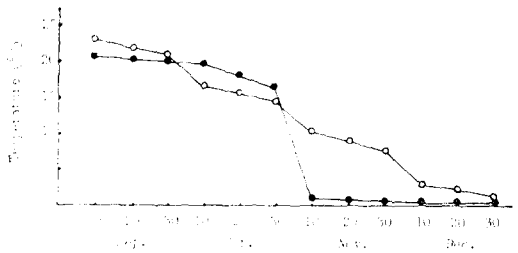

Fig. 2. Comparison of average temperature in 1987 and normals in Taegu(36°N): 1987 (—•—), 1951~1980 (—○—).

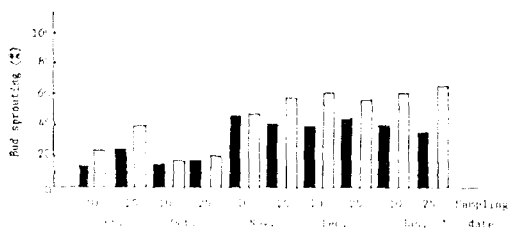

Fig. 3. Percentage of bud break of the different portion of stem at the stage of opening bud, preserved for 20 days at 30°C: low portion of stem ■, middle portion of stem □.

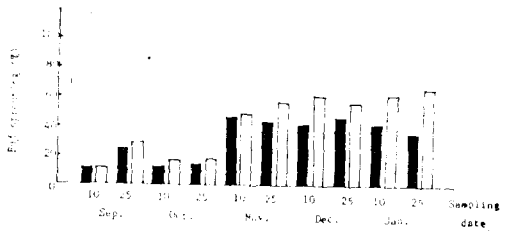

Fig. 4. Percentage of bud break of the different portion of stem at the stage of opened bud without petiole, preserved for 20 days at 30°C: low portion of stem ■, middle portion of stem □.

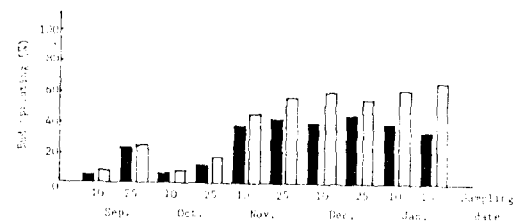

Fig. 5. Percentage of bud break of the different portion of stem at the stage of opened bud with petiole, preserved for 20 days at 30°C: low portion of stem ■, middle portion of stem □.

bud (Fig. 3), opened bud without petiole (Fig. 4) and opened bud with petiole (Fig. 5). On the other hand, the middle buds of stem were less dormant than those lower.

Table 1 shows the effect of chemical application on the percentage of mulberry bud break at the stage of green color of buds after 10 days incubation at 30°C. However, the data expressed in this table represetns an average stage of all samples inves tigated. The bud development beyond stage 2 which corresponed to opening bud, was defined as bud break. So, by this table, all the treatments were appeared still under dormancy. In the mean time, GA_3 seems to enhance the rate of bud burst more than the other

Table 1. Effects of chemical application on mulberry bud break at the stage of green color of bud preserved for 10 days at 30°C

Treatments	Sampling date													
	Oct. 15, 87		Oct. 30, 87		Nov. 15, 87		Nov. 30, 87		Dec. 15, 87		Dec. 30, 87		Jan. 15, 88	
	Green color of bud													
	%	stage*	%	stage	%	stage	%	stage	%	stage	%	stage	%	stage
Control	6	0	9	0.1	0	0	2	0	0	0	10	0	9	0.1
GA_3 10ppm	8	0	34	0.5	38	0.6	71	1.6	87	1.6	94	2	88	1.7
Urea 0.5%	7	0.1	11	0.1	24	0.3	55	1	76	1.5	94	1.7	21	0.4
NAA 0.02%	0	0	0	0	0	0	0	0	0	0	20	0.9	0	0
ABA 20ppm	0	0	2	0	0	0	0	0	0	0	20	0.7	9	0.2

* Represent an average stage of all investigated samples. The bud development beyond stage 2 is defined as bud break.

Table 2. Effects of chemical application on mulberry bud break at the stage of green color of bud preserved for 15 days at 30°C

Treatments	Sampling date													
	Oct. 15, 87		Oct. 30, 87		Nov. 15, 87		Nov. 30, 87		Dec. 15, 87		Dec. 30, 87		Jan. 15, 88	
	Green color of bud													
	%	stage*	%	stage	%	stage	%	stage	%	stage	%	stage	%	stage
Control	28	0.8	47	1.2	76	2.5	85	2.7	100	5.5	100	5.3	96	4.9
GA_3 10ppm	65	2.6	69	2.9	90	3	95	4	96	5.2	94	5.1	100	5.4
Urea 0.5%	30	0.8	55	2	84	2.6	92	4	94	5.2	94	4.8	94	4.6
NAA 0.02%	0	0	1	0	16	0.3	20	0.4	42	0.8	100	5.3	80	4.2
ABA 20ppm	29	0.6	32	0.7	79	3.1	70	2	87	4.4	87	5	93	4.3

* Represents an average stage of all investigated samples. The bud development beyond stage 2 is defined as bud break.

Table 3. Effects of chemical applicationmulberry bud break at the stage of green color of bud preserved for 20 days at 30°C

Treatments	Sampling date													
	Oct. 15, 87		Oct. 30, 87		Nov. 15, 87		Nov. 30, 87		Dec. 15, 87		Dec. 30, 87		Jan. 15, 88	
	Green color of bud													
	%	stage*	%	stage	%	stage	%	stage	%	stage	%	stage	%	stage
Control	28	0.8	50	1.8	82	2.6	85	2.7	100	5.5	100	5.3	96	4.9
GA_3 10ppm	65	2.6	72	3	93	3.7	98	4.1	96	5.2	94	5.1	100	5.4
Urea 0.5%	30	0.8	55	2	90	3.2	95	4.1	94	5.2	94	4.8	94	4.6
NAA 0.02%	0	0	5	0	29	0.8	64	2.2	79	2.9	100	5.3	80	4.2
ABA 20ppm	29	0.6	44	1.3	79	3.1	80	3	95	4.4	97	5	93	4.3

* Represents an average stage of all investigated samples. The bud development beyond stage 2 is defined as bud break.

treatments. But after 15 days incubation at 30°C (Table 2), GA_3 increased the bud break during the rest period, October, and enhanced the bud development to some extent during November and December. While Urea behaved similarly to GA_3, and other treatments were still in rest state in October. But from the beginning of November, all treatments showed bud break except NAA. Similar tendency is represented in Table 3.

However, the optimal time required for the mulberry winter bud break is 15 days incubation at 30°C as treated with gibberellic acid.

Fig. 6 shows the effect of chemical application on the percentage of mulberry bud break after 15 days incubation at 30°C. In this figure, we can discuss mainly that treatment of vegetative cuttings with GA_3 and Urea produced higher percentage of sprouting than that of control. This result confirms the findings of Yahiro & Hayashi (1966) and Yahiro (1965) that 10ppm of GA_3 is the mulbery bud dormancy breaker, and appeared to be more effective than urea in overcoming the rest of mulberry winter bud. Similar results were reported in other *in vitro* studies by Ohyama (1975).

Treatment of cuttings with NAA did not increase the rate of bud sprouting. NAA application showed to inhibit strongly the mulberry bud break. This result is in agreement with the work of Yahiro (1965). Whereas ABA treatment and control behaved similarly, it could be said that ABA was whithout effect in this experiment. Ineffectiveness of ABA can be illustrated that the concentration of 20ppm is not

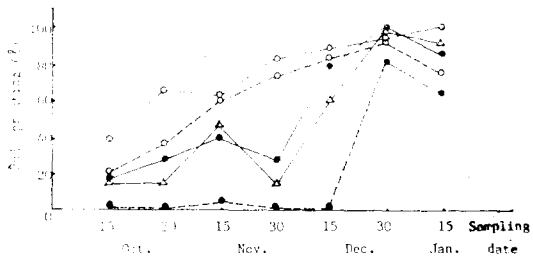

Fig. 6. Effects of chemical application on the percentage of mulberry bud break at the stage of opening bud and opened bud without petiole, preserved for 15 days at 30°C: Control (—•—), GA 10ppm (—○—), Urea 0.5% (···○···), NAA 0.02% (--•--), ABA 20ppm (—△—).

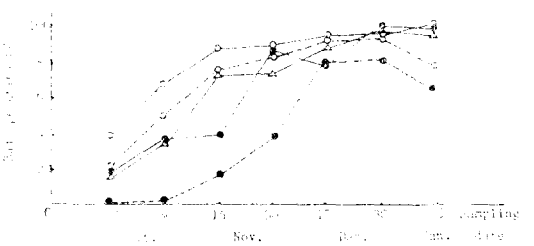

Fig. 7. Effects of chemical application on the percentage of mulberry bud break at the stage of opening bud and opened bud without petiole, preserved for 20 days at 30°C: Control (—•—), GA 10ppm (—○—) Urea 0.5% (--○--), NAA 0.02% (--•--), ABA 20ppm (—△—).

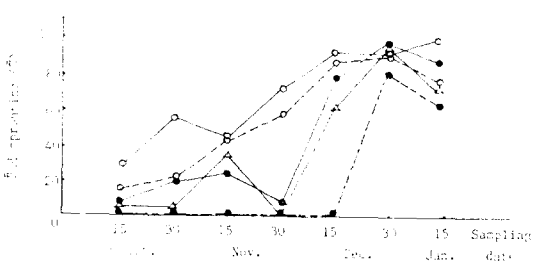

Fig. 8. Effects of chemical application on the percentage of mulberry bud break at the stage of opened bud with petiole, preserved for 15 days at 30°C: Control (—•—), GA 10ppm (—○—), Urea 0.5% (--○--) NAA 0.02% (--•--), ABA 20ppm (—△—).

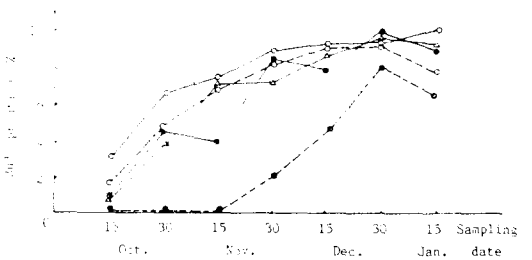

Fig. 9. Effects of chemical application on the percentage of mulberry bud break at the stage of opened bud with petiole, preserved for 20 days at 30°C: Control (—•—), GA 10ppm (—○—), Urea 0.5% (--○--) NAA 0.02% (--•--), ABA 20ppm (—△—).

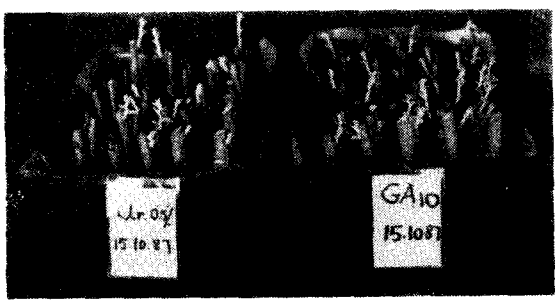

Fig. 10. Sprouting of dormant vegetative cuttings sampled on October 15th, treated with GA_3 10ppm and urea 0.5%.

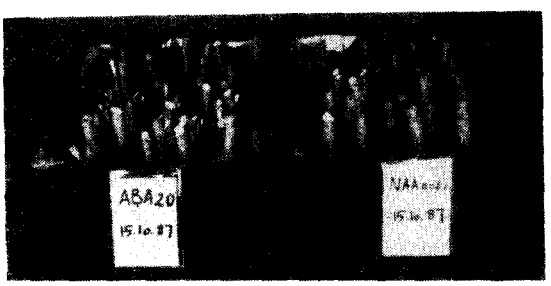

Fig. 11. Sprouting of dormant vegetative cuttings sampled on October 15th, treated with ABA 20ppm and NAA 0.02%.

strong enough to induce bud rest. As Ohyama & Oka (1975) reported that lower concentration of ABA had no effect on the mulberry bud break. While higher concentration was effective on inducing mulberry bud rest.

The effectiveness of chemical treatment on the percentage of bud break could be searched after 20

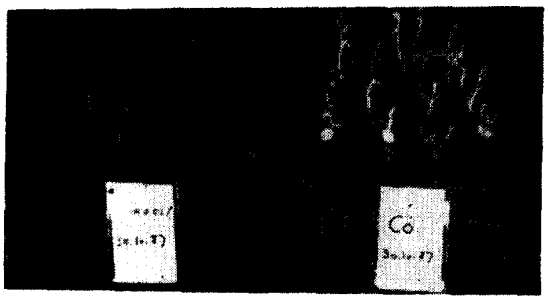

Fig. 12. Sprouting of dormant vegetative cuttings sample on October 30th, treated with ABA 20ppm and control.

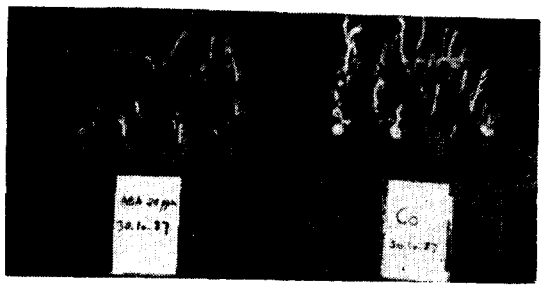

Fig. 14. Sprouting of dormant vegetative cuttings sample on October 30th, treated with NAA 0.02% and control.

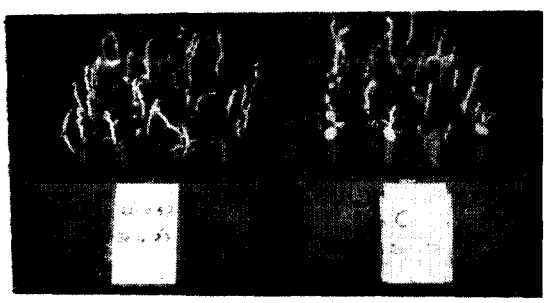

Fig. 13. Sprouting of dormant vegetative cuttings sample on October 30th, treated with urea 0.5% and control.

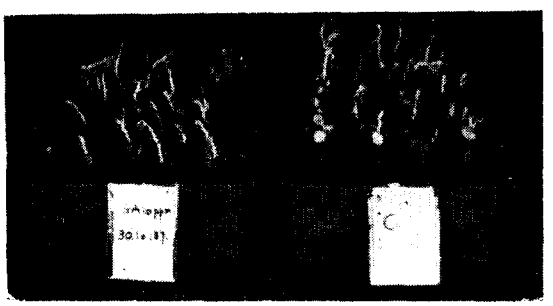

Fig. 15. Sprouting of dormant vegetative cuttings sampled on October 30th, treated with GA_3 10ppm and control.

days incubation (Fig. 7). All the treatments showed higher sprouting rate except NAA treatment and it appeared that GA_3 successfully broke the rest of mulberry winter bud.

Fig. 8 shows the effect of chemical application on the percentage of mulberry bud break after 15 days at 30°C at the stage of opened bud with petiole. In this figure, most of the sprouted buds reported by Fig. 6 attained the stage of opened bud with petiole. While after 20 days incubation (Fig. 9), GA_3 and urea were more effective on bud development comparing to ABA and control treatment. That is to say, GA_3 enabled bud development to reach the stage of opened bud with petiole. Whereas NAA treatment was still in deep rest period.

摘　要

本 實驗은 韓國 大邱地方에 있어서의 뽕나무의 休眠期間 및 休眠의 깊이를 알아보기 위하여 生長促進劑 및 抑制劑를 經時的으로 處理하였으며, 그 結果는 다음과 같다.

大邱地方에서는 日本 東京地方에서보다 自發休眠은 弱하게 나타났으나, 他發休眠은 强하게 나타났다.

弱休眠이 始作되는 時期도 日本과는 다르게 9月 下旬부터 始作되었으며, 10月 上旬부터 10月 下旬까지는 깊은 休眠狀態를 維持하다가 11月 初旬부터는 벌써 冬芽의 自發休眠이 覺醒되어 冬眠期(他發休眠)에 들어갔다.

休眠前 9月 初旬에 지베렐린, 아브시스酸, 나프탈렌酸 및 尿素를 잎에 處理한 結果, 지베렐린 10ppm을 處理한 뽕나무는 休眠이 크게 打破되었으며, 尿素 0.5%도 지베렐린보다는 낮으나 休眠을 打破시키는 效果가 있었다. 나프탈렌酸 0.02%는 休眠을 크게 促進시켰으며, 아브시스酸 20ppm은 休眠을 促進하는 效果가 없었다. 特히, 지베렐린을 處理한 揷穗는 다른 處理에 比해 發芽가 가장 빨랐다.

뽕가지의 部位에 따른 休眠의 强度는 가지의 中間部位가 가지의 上部와 下部에 比해 休眠이 弱했다.

지베렐린을 處理한 다음 揷穗를 30°C下에서 休眠打破를 調査할 때 가장 適當한 處理期間은 15日이었다.

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