THE EFFECTS OF GAMMA IRRADIATION ON POLLEN VIABILTY AND HAPLOID PLANT FORMATION IN SNAKE CUCUMBER (*Cucumis melo* L. *var. flexuosus* Naud.)

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Abstract

In this research, the effects of gamma irradiation (250, 300, 350 Gy) on *in vitro* pollen viability and haploid embryo stimulation in inbred snake cucumber named line 6 were investigated. *In vitro* pollen viability was higher in pollens irradiated with 250 and 300 Gy doses (28.8 % and 28.0 % respectively) than with 350 Gy (19.4 %). Pollen viability decreased by the increase of the pollen age besides the doses.

According to the fruit set and parthenogenetic embryo formation ratios, 300 and 350 Gy doses seemed better than 250 Gy dose.

Keywords : Snake cucumber, gamma irradiation, haploidy, pollen viability

1. Introduction

Doubled haploid plants used in breeding programs can be obtained after pollination with X and γ ray irradiated pollen by inhibition of double fertilization in embryo sac in *Cucurbitaceae*. Parthenogenetic haploid plants were previously obtained by embryo culture in melon (Sauton 1988, Gürsöz 1990, Yanmaz et al. 1996), cucumber (Çaglar 1995), watermelon (Sari and Abak, 1994). In this research, the effects of gamma irradiation on pollen viability and haploid embryo formation in snake cucumber were investigated.

2. Material and methods

Snake cucumber named line 6 obtained from our selection project, was used in the experiment. Seeds were sown in soil bloks on first of April and seedlings which have 2 to 3 leaves were planted in an unheated glasshouse by 70x50 cm row spacing.

The day before anthesis, male flowers were collected prior to irradiation with a Cesium 137 source at Turkish Atomic Energy Authority Lalahan Nuclear Research Institute of Animal Health. Petals were removed. 250, 300 and 350 Gy doses were used to determine optimum irradiation doses.

Female flowers were pollinated by irradiated male flowers at anthesis and 2 to 3 male flower buds were used per female flower. After pollination, female flowers were covered with paper bags to avoid pollen contamination. Paper bags were removed after fruit set. Fruits were harvested 3 to 4 weeks after pollination. Surface disinfection was performed with 96 % ethylalcohol. Seeds were opened under binocular microscope and then embryos were cultivated in petri dishes containing E20 A media supplemented with

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0.01 mg/l IAA (Gürsöz 1990). Total number of seeds and embryos per fruit were determined during cultivation stage. Cultures were placed in a climatic room set at 25° C and 16 hours photoperiod. Explants with 2 to 3 leaves were transferred to the same media one month intervals in 16x2 cm tubes.

As a second experiment, in order to determine optimum irradiation doses and pollen age on germination ability of pollen, 1-2 and 3 days aged irradiated and non irradiated pollen were germinated. Culture media for pollen viability tests were composed at 1% agar, 15 % sucrose and 5 mg/l H_3BO_3 . Counts were made 24, 48, 72, 96 and 120 hours after sowing.

4. Results

4.1. The effects of irradiation on pollen viability

Pollen viability decreased by the increase of irradiation doses and pollen age (Table 1 and Figure 1). The percentage of germination was generally higher in one day old pollen than 2 and 3 day-old ones.

Decrease in pollen viability was higher in pollen irradiated by 350 Gy dose than 300 and 250 Gy doses irradiated pollen. The germination percentage of non irradiated pollen was 81.1 %, while germinability was 34.86 % in 250 Gy, 23.92 % in 300 Gy and 38.22 % in 350 Gy doses (Figure 1).

4.2. The effects of irradiation on haploid embryo formation

Different irradiation doses affected the number of embryo, rate of embryo formation and embryo types (Table 2). No fruit was obtained from pollination by 250 Gy irradiated pollen. 300 Gy dose was found more effective than 350 Gy dose for induction of parthenogenetic embryo formation. Globular and heart-shaped embryo rates were 17.39 % and 23.91 % respectively after pollination with 300 Gy irradiated pollen. Rate of necrotic embryo was high with both doses. No heart-shaped embryo and plantlet were obtained from 350 Gy irradiation dose. Only 8 plantlets were gained from 300 Gy irradiation dose. But none of them survived.

5. Discussion

Pollination by γ-irradiated pollen can induce haploid embryo formation in snake cucumber similar to melon and cucumber. In present study 300 Gy dose seemed more effective than the other doses. Although pollen viability was high in 350 Gy irradiation dose, no heart-shaped embryo were obtained and no plantlet developed from those embryos. Necrotic embryo rate was found high with both irradiation dose. According to Fassuliotis and Nelson (1986), necrotic embryo rate increases in late harvested fruit. It will be worthwile to study on the effect of harvest time on embryo types in the future.

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Table 1. Percentage of pollen germination at different irradiation doses and pollen age

Treatment	Hours					Mean
	Age	24	48	72	96	120	
Nonirradiated	1	82.97	84.02	81.33	77.42	79.77	81.10 a
	2	84.15	86.59	72.56	66.64	62.52	74.49 a
	3	72.29	71.51	55.68	50.07	16.59	53.23 b
250 Gy	1	52.73	38.02	34.45	24.28	23.81	34.86 a
	2	37.62	35.69	28.10	25.65	20.48	27.51 a
	3	34.66	27.10	21.94	19.75	16.36	23.97 a
300 Gy	1	48.73	30.57	19.14	12.14	9.00	23.92 a
	2	46.59	44.41	43.78	42.45	36.56	42.76 b
	3	31.81	23.95	11.64	10.65	8.65	17.34 c
350 Gy	1	49.47	43.55	36.40	32.05	29.63	38.22 a
	2	19.96	12.00	8.29	8.11	3.19	10.11 b
	3	14.81	15.53	11.27	7.43	0.00	9.81 b

Table 2. Embryo types and embryo rate of plants obtained with different γ - irradiated pollen

Obtantion	Irradiation Doses (Gy)		
	250	300	350
Number of fruit	-	4	2
Number of seed	-	1015	231
Number of embryos	-	46	26
Rate of embryo formation	-	4.53	11.26
Rate of globular embryo	-	17.39	42.31
Rate of necrose embryo	-	58.70	57.69
Rate of heartshaped embryo	-	23.91	0.00
Number of plantlet	-	8.00	0.00
Rate of plantlet formation	-	17.39	0.00

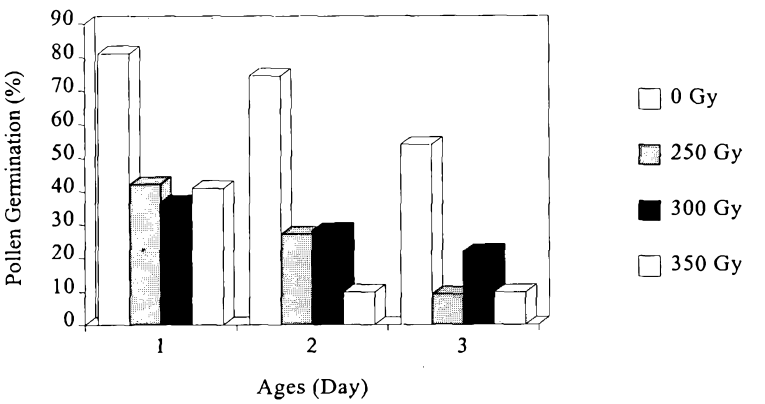

Figure 1. The percentage of germination of pollen irradiated with different doses of γ - ray at different ages