Biosciences, Biotechnology Research Asia Vol. **3**(2), 335-340 (2005)

EFFECT OF HERBICIDE GLYPHOSATE ON POLLEN GERMINATION *IN VITRO* AND POLLEN TUBE LENGTH OF *Hibiscus cannabinus* Linn.

Sanjay Kamble I.

Department of Botany, Phulsing Naik Mahavidyalaya, Pusad, District Yavatmal (India)

(Received: August 04, 2005; Accepted: October 14, 2005)

ABSTRACT

The germination of pollen grains in various concentrations of herbicide by adopting "Hanging drop technique" (Vasil, 1960). Effect of herbicide on pollen germination and pollen tube length was studied. Glyphosate inhibited the pollen germination and pollen tube growth at higher concentrations.

The percentage of pollen germination was decreased from 29.29, 23.74, 11.49, 3.94 and 0.0 at 20, 40, 60, 80 and 100 ppm of glyphosate, respectively. Similarly, the pollen tube length was also declined and which was recorded from 373.7, 212.1, 80.8, 40.4 and 0.0 microns of Glyphosate at 20, 40, 60, 80 and 100 ppm of glyphosate, respectively.

The bursting of pollen tubes at their tips were observed at higher concentrations of the herbicide.

Key words: Herbicide, Glyphosate, pollen grain, *Hibiscus cannabinus* Linn.

INTRODUCTION

The pollen grains have been reported to be most sensitive to herbicidal action. The present investigation deals with the effect of glyphosate on pollen tube length and percentage of pollen germination in different formulation of herbicide.

MATERIAL AND METHODS

The pollen grains of *Hibiscus cannabinus* Linn. were collected at the time of anthesis, during the month of September to December 1996 and 1997 and experiments were conducted at room temperature. A large number of pollen grains of *H. cannabinus* were allowed to germinate in control medium *i.e.* tap water, distilled water and different concentrations of sucrose solution were used as medium separately. The medium that gives the maximum germination percentage and pollen tube length was used for preparation of solution of glyphosate.

Stock solution of 1000 ppm of Glyphosate were prepared by using tap water. Anther was collected from the weeds growing in fields. Pollen grains were collected on a cavity slide by taping the anther. The "Hanging drop" technique (Vasil 1960) was adopted for pollen germination and pollen tube growth experiment. The germination percentage of pollen grains and pollen tube length were calculated with respective concentrations of freshly prepared aqueous solution. The germination of pollen grains and pollen tube growth at different solution was studied from morning 7'O Clock onwards.

RESULTS AND DISCUSSION

The germination percentage of pollen grains and pollen tube length of *H. cannabinus* in tap water were 38.34 with 464.6 microns and 32.31 in distilled water with 300 microns, respectively. In sucrose solution it was 11.0, 19.5, 21.8, 35.52 and 25.2 at 100, 200, 300, 400 and 500 ppm with pollen

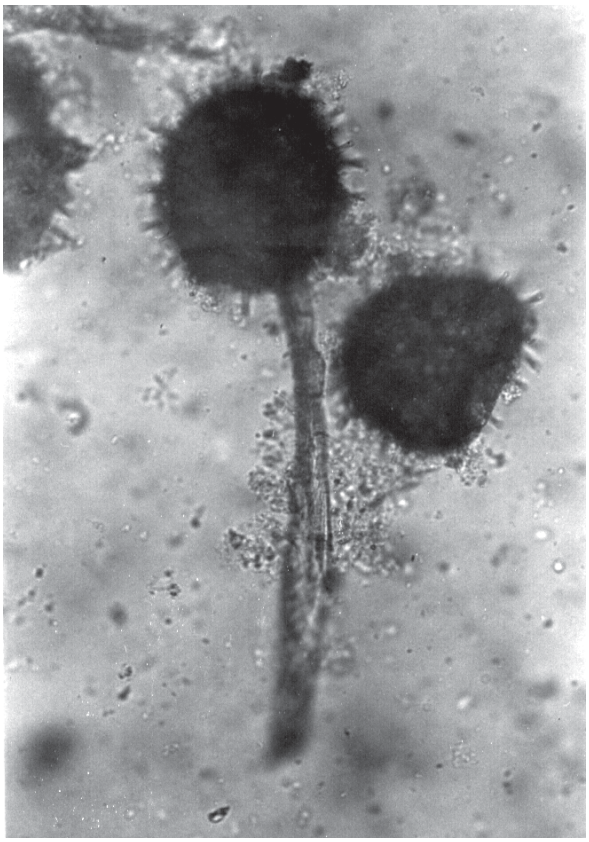

Fig. - 1: Control - showing pollen tube length. x = 102.5

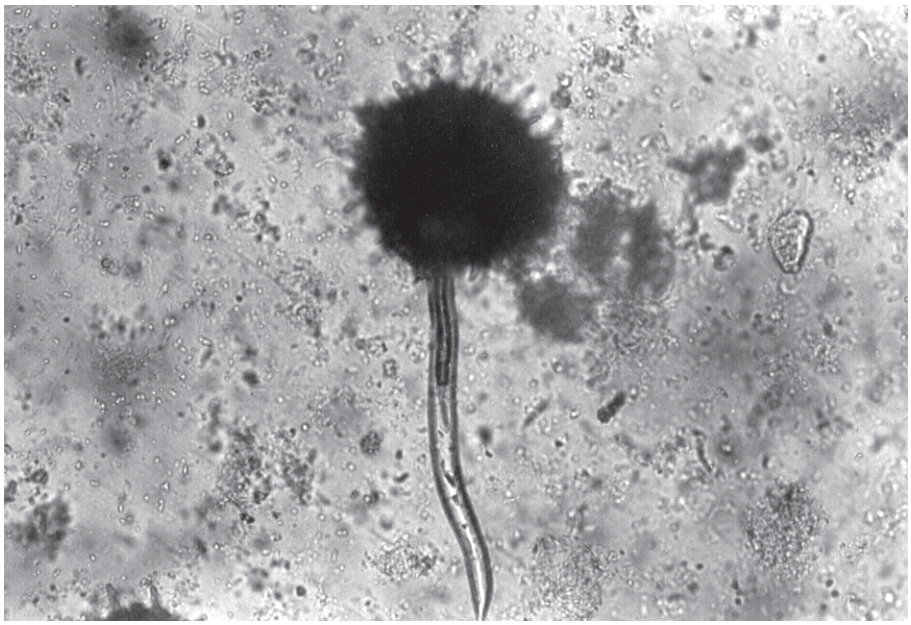

Fig. - 2: Showing pollen tube length at 80 ppm solution of glyphosate. x = 408.2

Table - 1: Effect of herbicide on pollen germination *in vitro* and pollen tube length and various concentrations of medium and glyphosate

Medium / herbicide	Conc. (ppm)	Total no. of pollen studies	Total no. of pollen germination	% of pollen germination	Pollen tube length	Abnormal character	Time / Month
Tap water	-	625	239	38.24	646.6	-	7.00 am to
Tap water - Ab. Al.	-	394	150	38.07	434.3	-	onwards in the month of
Distilled	-	443	150	32.31	300	F.B.	Sep. to Dec. 1996 & 1997
Sucrose	100	100	11	11.0	202	-	
	200	130	15	19.5	272.7	-	
	300	124	17	21.8	300	F.B.	
	400	96	37	35.52	212.1	M.B.	
	500	140	18	25.02	161.6	M.B.	
Glyphosate	20	256	75	29.29	373.7	-	
	40	278	66	23.74	212.1	-	
	60	261	30	11.49	80.8	R.B.	
	80	304	12	3.94	40.4	F.B.	
	100	217	0	0	-	-	

R.B. - Rare bursting, F.B. - Few bursting, M.B. - Maximum bursting, Ab. Al. - Absolute alcohol

tube length were 202, 272.7, 303, 212.1 and 161.6 microns, respectively. In absolute alcohol-tap water, there was 38.07 percentage of pollen germination and 434.3 microns tube length. The tap water in which maximum pollen germination was 38.24 and have maximum pollen tube length *i.e.* 464.6 microns was observed. Therefore, tap water was considered as a control and stock solution of the herbicide was prepared by using tap water as a medium (Table 1, Figs. 1, 3 and 4).

The germination percentage at 20, 40, 60, 80 and 100 ppm was 29.29, 37.74, 11.49, 3.94 and 0.0 and the pollen tube length was 373.7, 212.1, 80.8, 40.4 and 0.0 micros, respectively. The step fall in the percentage of pollen germination was observed at 20 to 80 ppm and at 100 ppm it was zero. Similarly, the steep fall in the pollen tube length was observed in the same concentrations as compared to control. Percentage of pollen germination and pollen tube length were decreased with increase in the concentrations of herbicide (Table - 1, FIgs. 2, 5 and 6).

The bursting of pollen tube at there tips occurred in medium but maximum bursting occurred at higher concentrations of glyphosate. Abnormality like curvature of pollen tube was also occurred at higher concentrations of this herbicide.

DISCUSSION

In the present investigation, the percentage of pollen germination was found to be maximum in tap water as compared to distilled water and different concentrations of sucrose solution. Therefore, tap water considered as control media for further investigation. It was observed that drop of alcohol present in water did not change the percentage of pollen germination and growth of the pollen tube.

The pollen germination was maximum in control *i.e.* in tap water. Banerji and Ganguli (1937) in *Eichhornia crassipes* reported maximum pollen germination in tap water. Similar observations were recorded by Srinivasu (1987) in *Parthenium*

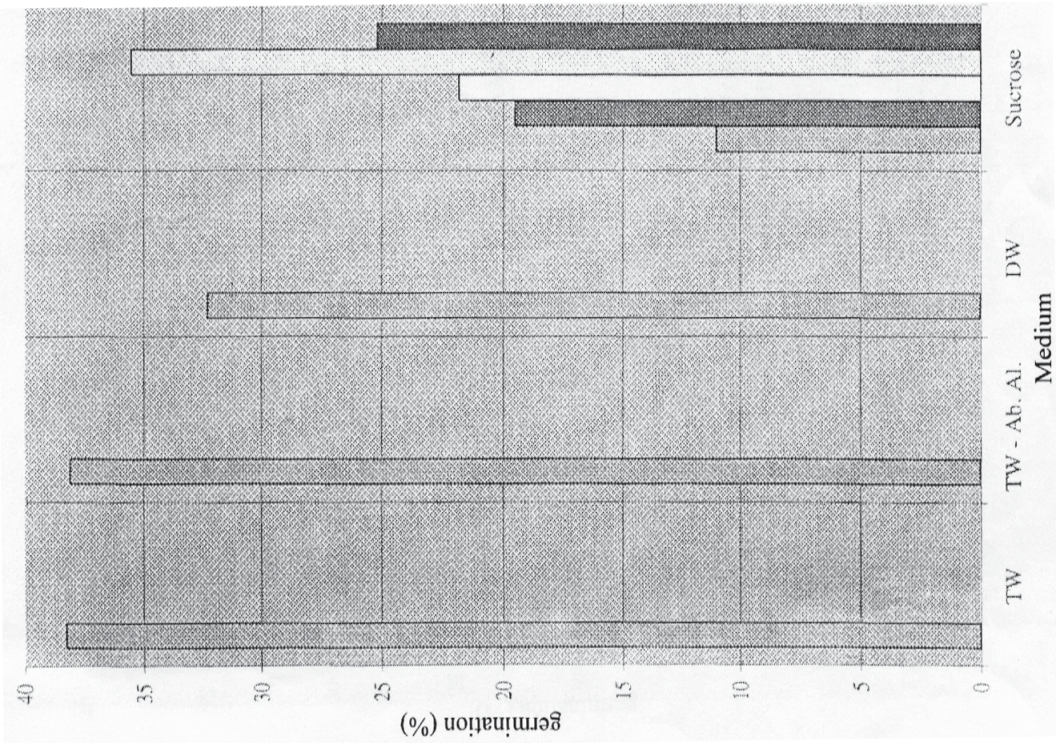

Fig. - 3: Graph showing the percentage of pollen grain germination in tap water (TW), tap water-absolute alcohol (TW-Ab. Al.), distilled water (DW) and different concentrations of sucrose solution

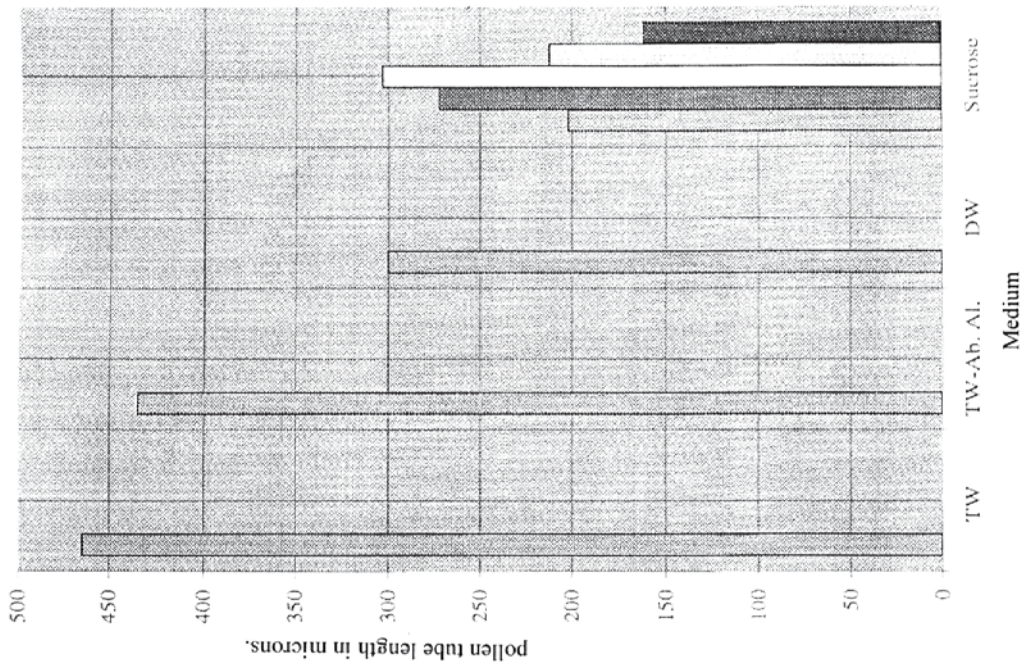

Fig. - 4: Graph showing length of pollen tube in different mediums and in various concentrations of sucrose solution

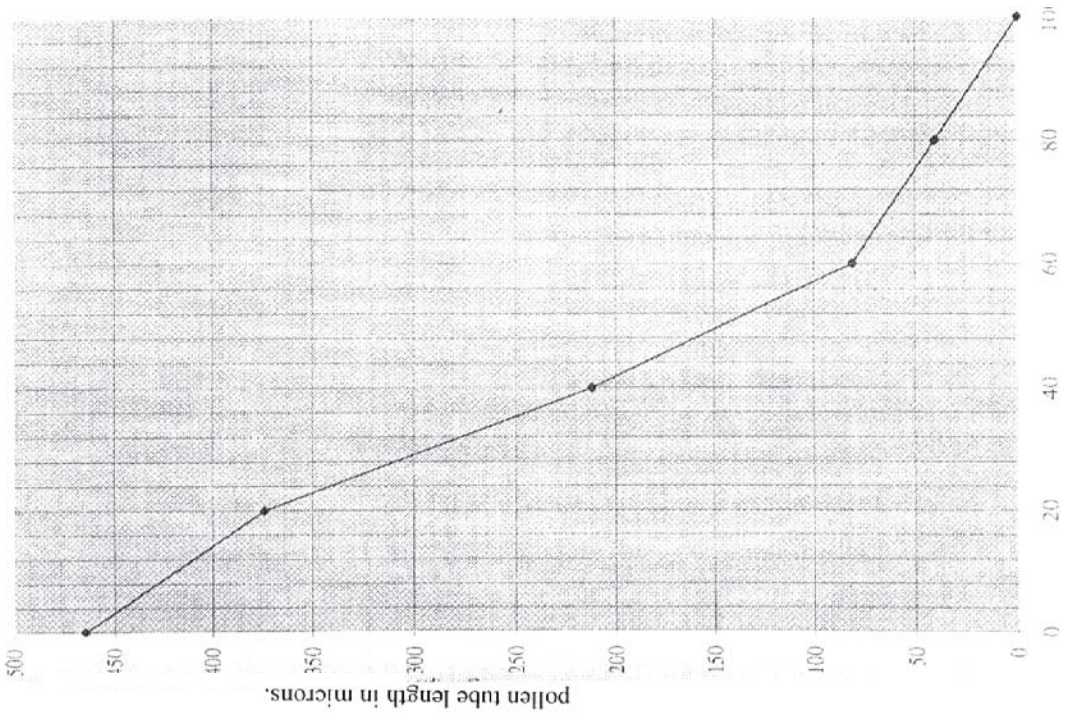

Fig. - 5: Graph showing the percentage of pollen grain germination in different concentrations of glyphosate.

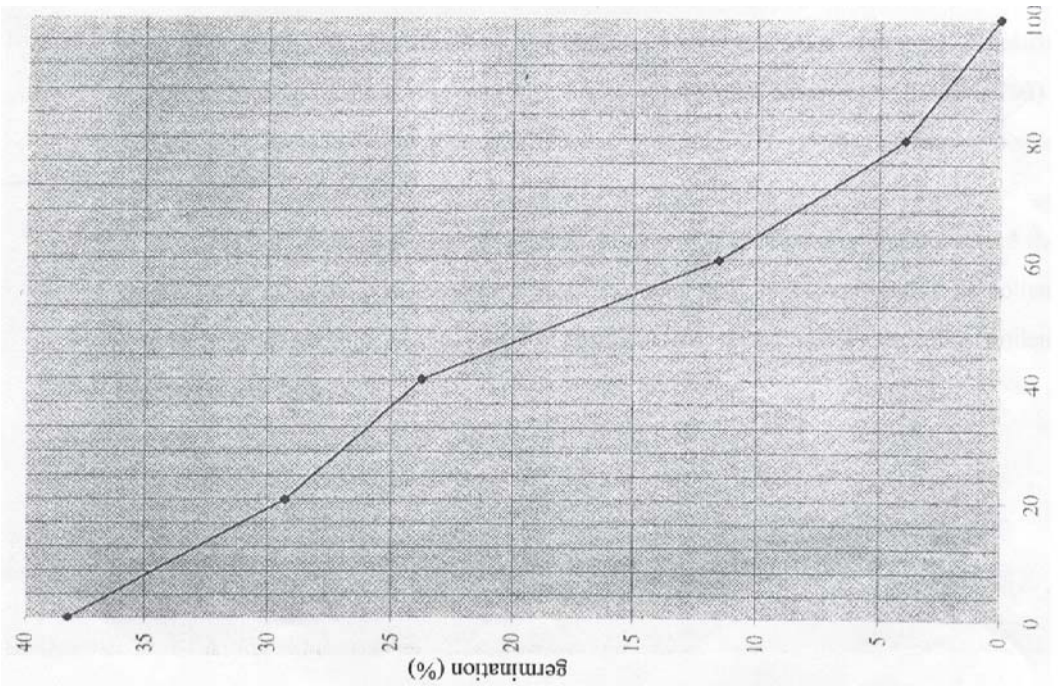

Fig. - 6: Graph showing pollen tube length in different concentrations of glyphosate.

hysterophorus, Jain (1993) in *Chenopodium album* and Gopal, (1993) in *Medicago sativa*.

The germination percentage of pollen grains was decreased gradullay due to the application of glyphosate. It was 38.24, 29.24, 23.24, 11.49, 3.94 and 0.0 percent at control, 20, 40, 60, 80 and 100 ppm respectively.

It was cleared that the percentage of pollen germination was zero at 100 ppm. So this concentration considered to be lethal dose. Workers like Jain (1993) in *Chenopodium album,* Bobde (1993) in *Crotolaria juncea* and Kulkarni (1998) in *Crotolaria medicaginea* var. *Luxurians* reported reduction of percentage of pollen germination due to application of glyphosate.

In present study, pollen tube length also decreased as the concentrations of glyphosate increased. It was 464.6, 373.7, 212.1, 80.8, 40.4 and 0.0 microns at control, 20, 40, 60, 80 and 100 ppm, respectively. An abnormalities like bursting of pollen tubes were occurred due to application of glyphosate. Similar results were observed by Bobde (1993) in *Crotolaria juncea* Jain (1993) in *Chenopodium album* and Kulkarni (1998) in *Crotolaria medicaginea* var. *Luxurians*.

This herbicide inhibited the percentage of pollen germination and decreased the pollen tube length. From foregoing discussion, it was concluded that glyphosate inhibit the pollen tube growth with bursting which enhance the change of fertilization by revealing the moments of pollen tube through the stigma and style.

REFERENCES

1. Banerji, J. and Ganguli, H.C. Spermatogenesis in *Eichhornia crassipes* Solms. *J. Indian. Bot. Soc.,* **16,** 289-296 (1937)
2. Bobde, S.N. Comparative effects of herbicide on *Crotolaria juncea* Linn. *Ph.D. Thesis,* Nagpur Univ., Nagpur (1993)
3. Gopal, K.R. Herbicidal effect on cytomorphology of weed *Medicago sativa* Linn. *Ph.D. Thesis,* Nagpur Univ., Nagpur (1993)
4. Jain, S.B. Cytomorphological effect of weedicide on weed *Chenopodium album*. *Ph. D. Thesis*, Nagpur Univ., Nagpur (1993)
5. Kulkarni, G.B. Effect of agro-chemicals on *Crotolaria midicaginea* var *Luxurians*. *Ph.D. Thesis,* Dr. BabaSaheb Ambedkar Marathwada Univ., Aurangabad (1998)
6. Srinivasu, T. Effect of weedicide on weed *Parthenium hysterophorus* Linn. *Ph.D. Thesis,* Nagpur Univ., Nagpur (1986)