Chapter VI

PHOTOSYNTHESIS

A. Introduction :

Photosynthesis is an indispensable event of the green plants which is singularly responsible for the sustenance of entire auxotrophic and heterotrophic world. The ingenity of the process lies in utilization of most easily and abundently available energy of solar radiation. The continuous inexhausteble flow of energy is required to sustain the biological world which is only possible through solar radiations. The green plants fix the atmospheric carbon dioxide by utilizing the solar energy harnessed by the chlorophyll and split water molecule and keep the continuous flow of electrons which in turn utilized for the fixation of CO_2. Thus the potential source of energy to all auxotrophs comes from the sugar synthesized by the autotrophs the green plants. The process of sugar synthesis takes place in the tiny semiautonomcus cell organelle the chloroplasts.

It is of long realization by man that it must be his endeavour to keep the production of food material to the highest level, so that he can feed, his growing population. The productivity of plants is primarily determined by efficiency of photosynthesizing system. Although the mechanism of photosynthesis includes several biochemical processes such as photochemical energy flow through chlorophyll, ability to fix CO_2 ability of stomata to allow free diffusion of

atmospheric CO_2, adequate availability of water and sunlight, optimum temperature etc. by and large, the process is integrated one and all these events are complementary to each other. Even the stimulation of one is enough to contribute for the stimulation of entire process. Increased efficiency of photosynthesis, therefore, can be achieved if one could bring about by modifying one of the factors to function more efficiently. Such type of change can be achieved by mutation and one can induce mutation and isolate the mutants, which may be efficient in photochemical energy generation or enzyme mechanics etc.

In the present investigation therefore, the effect of both chemical mutagen EMS as well as γ-radiations on the photosynthetic machinery has been studied in the M_1 stage of C.tinctorius Var. N 62-8.

B. Material and methods :

Seedlings raised from the mutagen treated seeds are used for the study. The healthy leaves of seven week old seedlings were randomly sampled from each of the treatment pot. Known size leaf discs were punched, quickly weighed and kept on moist filter paper covered with glass petridish under the light. The entire experiment was carried at 10 A.M. So that the stomata were maximally open in the plant. The known

number of leaf discs were exposed to $NaH^{14}CO_3$ in tris-HCl buffer by the method of Hegde and Patil (1982). The specific activity of $NaH^{14}CO_3$ is 48.3 mCi/m mole obtained from Bhabha Atomic Research Centre, Bombay. The entire experiment was carried out in natural light at 10 A.M. at the temperature of 28°C. Among the treated plants where steady state product of 30 minutes exposure has been studied. In control, 0.4 M EMS treated and 10 Kr r-irradiated ones the rate of $^{14}CO_2$ incorporation has also been studied for 5 sec. The reaction was terminated after feeding, by means of 80 % boiling ethanol. The tissue was homogenized by 80 % ethanol and filtered. The filtrate was measured and activity counted. The residue was hydrolyzed with 1 N HCl in 15 lbs. pressure for 30 min. filtered and activity was counted in the filtrate so as to compute for the total activity in the tissue. The activity was counted in soluble and isoluble fraction under proportional counting system (ECIL).

C. Results and discussion :

The rate of incorporation in the steady state products of 30 minutes of the mutagen treated plants as well as the control is given in the Table 6.1. The rate of incorporation scored in leaf discs of the control, 0.4 M EMS treated and 10 Kr r-irradiated plants after 5 seconds exposure is given in the table 6.2. It can be seen clearly that the rate of incorporation in 5 sec. exposure in 0.4 M EMS treated one is

Table 6.1 : Effect of EMS and γ-Radiation on the rate of $^{14}CO_2$ incorporation in Carthamus tinctorius (L.) Var. N 62-8.

Treatment	Time of exposure	$^{14}CO_2$ fixation*		
		g^{-1} fresh weight	mg^{-1} chlorophyll	cm^{-2} area
Control	30 min.	11.19×10^{-6}	0.530×10^{-6}	3.620×10^{-6}
0.2 M EMS	,,	12.22×10^{-6}	0.534×10^{-6}	3.633×10^{-6}
0.25 M EMS	,,	12.93×10^{-6}	0.562×10^{-6}	3.645×10^{-6}
0.3 M EMS	,,	19.45×10^{-6}	1.062×10^{-6}	7.140×10^{-6}
0.35 M EMS	,,	41.85×10^{-6}	2.370×10^{-6}	16.958×10^{-6}
0.4 M EMS	,,	18.41×10^{-6}	0.875×10^{-6}	7.078×10^{-6}
Control	,,	7.32×10^{-6}	0.513×10^{-6}	3.306×10^{-6}
1 Kr	,,	19.57×10^{-6}	0.672×10^{-6}	6.346×10^{-6}
2.5 Kr	,,	19.63×10^{-6}	0.735×10^{-6}	6.641×10^{-6}
5 Kr	,,	21.04×10^{-6}	0.738×10^{-6}	6.673×10^{-6}
7.5 Kr	,,	21.34×10^{-6}	0.750×10^{-6}	7.148×10^{-6}
10 Kr	,,	30.43×10^{-6}	1.317×10^{-6}	10.339×10^{-6}

* Values expressed in mg of $^{14}CO_2$.

Table 6.2 : EFFECT OF EMS and γ-RADIATION ON THE RATE OF $^{14}CO_2$ INCORPORATION IN CARTHAMUS TINCTORIUS (L.) VAR. N 62-8.

Treatment	Time of exposure	$^{14}CO_2$ fixation*		
		g^{-1} fresh weight	mg^{-1} chlorophylls	cm^{-2} area
Control	5 sec.	2.16×10^{-6}	0.103×10^{-6}	0.737×10^{-6}
0.4 M EMS	,,	3.97×10^{-6}	0.151×10^{-6}	1.521×10^{-6}
10 Kr	,,	3.99×10^{-6}	0.430×10^{-6}	2.720×10^{-6}

*Values expressed in mg of $^{14}CO_2$.

double the rate of the control, while in case of 10 Kr irradiated plants it is almost 3 fold higher than that of the control. This reflects that both EMS and γ-radiation stimulate the rate of incorporation. However, irradiation has greater effect than that of EMS. If the rate of incorporation in steady state is taken in to consideration, it can be seen that with increasing concentration of EMS there is increase in the rate of incorporation but at 0.35 M there is steap rise in the rate of $^{14}CO_2$ incorporation whereas with increasing dose of radiation increased rate of incorporation is gradual. At 10 Kr the activity counted per unit area of leaf is almost 2 fold the activity of control.

Roy and Clark (1970) have studied the CO_2 fixation and translocation for photoassimilates in Vicia faba following X-ray irradiation and have shown that there is 50 % reduction in accumulation of biomass and also shown that assimilation of $^{14}CO_2$ by irradiated plants during 15 min. periods was less than that in control. Nanda and Misra (1977) studied the effect of radiation and chemical mutagen in rice and have isolated large number of chloroplast mutants, which affected in chlorophyll content and chloroplast development upon studying the photosynthetic efficiency of certain mutants using $^{14}CO_2$ lebelling technique, showed that there is reduction in photosynthetic efficiency in most cases, however, in some increased activity was noted. Based on this study it is speculated that

efficiency of enzyme catalyzing CO_2 fixation located in the chloroplast must have been affected by mutagens.

Contrary to the above findings, in the present investigation stimulatory effect of both EMS and γ-radiation has been obtained. Although at low concentration of EMS stimulatory effect is seen, it is to certain extent erratic, but in the irradiated plants, there is steady increase with increasing dose. Since the doses chosen here is lower, it is quite probable that some sort of a modification either in photochemical energy flow mechanism or in the enzymatic machinary must have been brought, if persistant it may be useful.