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INFLUENCE OF GOAL ON NUCLEIC ACIDS CONTENT IN SEEDLINGS OF *Medicago sativa* L.

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Abstract:

Effect of herbicide GOAL (2'-chloro-4'-trifluro-methyl phenyl 3-ethoxy 4-nitrophenyl ether) on nucleic acid of treated seedling of Medicago sativa Linn. was studied. The seeds of Medicago sativa Linn. were treated with various concentrations of Goal for 24 hours and germinated under laboratory conditions. The treated seedlings were used to study percentage of nucleic acids.

As the concentration increased gradually the percentage of nucleic acids decreased. In the present study the DNA per seedling was found to be 6.29 x 10^{-5} to 5.63 x 10^{-5} at the concentrations 500 to 2500 ppm respectively, against control 9.17 x 10^{-4}. Similarly, RNA per seedling was found to be 8.28 x 10^{-5} to 7.72 x 10^{-5} at 500 to 2500 ppm respectively, as against 9.94 x 10^{-4} in control. The lethal dose was found to be 2600 ppm for nucleic acids.

Index Terms -Medicago sativa, GOAL, DNA, RNA

Introduction:

Weeds form a serious negative factor in crop production and are responsible for marked losses in crop yields. A large majority of the weeds found in the country can be kept in check. But the persistence of weeds will not necessarily pose a very serious problem if control measures are applied in time. Hence the foremost criterion of a good herbicide should be its capability to kill target plants for a desired period.

In the present study the chemicals used for weed control & which suppress or destroy the growth of weeds, called as herbicide as well as to study the mode of action of herbicides. According to Ashton and Crafts (1973) it comprises the sum total of anatomical, physiological and biochemical responses that make up the total phytotoxic action of the herbicides. These either help in killing the weeds or in inhibiting their growth.

Therefore, it may be used selectively in treatment provided direct contact with the crop is avoided i.e. pre plant, pre-emergence or post emergence. Eg. GOAL (2'-Chloro-4'-trofluoro-methyl Phenyl 3-ethoxy-4-nitrophenyl ether). In the present study the effect of GOAL was studied on *Medicago sativa* Linn.

Research Methodology:

The seeds were treated with various concentrations of GOAL for 24 hours. The treated seeds were washed thoroughly with distilled water and kept for germination in petridishes lined with moistened filter paper under laboratory conditions. Seeds soaked in distilled water were used as control. The treated seeds and untreated seeds were allowed to grow for seven days.

In each sample one gram fresh weight of seedlings were taken for extraction and estimation of nucleic acids. The method suggested by Ogur and Rosen (1949) and Schneider (1945) were adopted for extraction and estimation of nucleic acids.

Extraction and Estimation of Nucleic Acids

The weighed samples were homogenised in 10% Perchloric acid (PCA) at 0C in a glass pastel and quarter and centrifuged each 20C for 15 minutes. The extracts were discarded and resuspended the residue in cold 5% PCA and centrifuged again for 10-15 minutes. The supernatant was discarded and residue was washed sequentially with 70% ethanol, 95% ethanol and finally with boiling ethanol-ether (3:1) in a water bath and then with cold 0.2 NPCA. The residue was suspended with cold 2NPCA and stored at 2-5C fo2 18 hours, solution was centrifuged with the same condition and supernatant was collected. The residue was resuspended with cold 2NPCA, were centrifuged and two supernatants was combined. This supernatant containing RNA fraction was used for quantitative estimation of total RNA.

The residue was suspended with 1NPCA and heated at 7000 for 20 minutes and the solution was centrifuged. The supernatant was collected and residue was suspended with 1NPCA and centrifuged. Both supernatant were combined, which comprises DNA fraction and it was used for estimation of DNA.

The total RNA and DNA extracts were estimated by measuring absorption at 600 and 595 mm, respectively with the help of spectrometer (Ultra-Spec. Model 540). The DNA and RNA content samples were calculated from standard graphs of calf-thymus DNA and Yeast RNA per seedling in a sample was calculated by using the formula:-

The DNA per seedling in sample was calculated by using formula

$$\text{DNA per seedling} = \frac{\text{Total DNA}}{\text{Total number of seedling per sample}} \times 100$$

The RNA per seedling in sample was calculated by using formula

$$\text{RNA per seedling} = \frac{\text{Total RNA}}{\text{Total number of seedling per sample}} \times 100$$

Results and Discussion:

The DNA and RNA content per seedling decreased gradually with an increase in concentration. The reduction in amount of DNA and RNA was accompanied with a depressive action on mitotic activity. The gradual reduction in nucleic acid contents was observed with decrease in the rate of cell division and seedling growth. As suggested by Chrispeels and Hanson (1962) it appears that the basis for the herbicidal action of phenoxy herbicides was associated with renewal of RNA synthesis leading to massive tissue proliferation, disorganised growth and finally death of the soybean plants. These effects may be related to the GOAL induced inhibition of DNA and RNA synthesis. This may be due to the mitotic inhibition by the herbicide attributed to blocking of mitotic cycle during interphase which may results from a prolonged G2 period or the inhibition of nucleic acid synthesis.

Schultz *et al*. (1968) found that RNA and DNA content in maize seedlings was decreased when treated with trifluralin. Similar, findings were observed by Srinivasu (1986) in weed *Parthenium hysterophorus*, Jain (1993) in *Chenopodium album*, Bobde (1993) in *Crotalaria juncea*, Tulankar (1998) in *Cleome viscosa*, Dudhe (2002) in *Hyptis suaveolens*, Taduwadi (2004) in *Cleome viscosa*, Mahakhode (2008) in *Psoralea corylifolia.*

In present study the results obtained after treatment shows that the percentage of DNA per seedling was 6.29 x 10^{-5} and 5.63 x 10^{-5}, 500 and 2500 ppm respectively, as against control it was 9.17 x 10^{-4} (Table 1).

Table: 1- Amount DNA percentage in the seedlings of *Medicago sativa* after treatment of GOAL

Herbicide	concentration in ppm	% of DNA per seedling	Standard error (±)
	Control	9.17×10^{-4}	6.24×10^{-5}
GOAL	500	6.29×10^{-5}	3.79×10^{-5}
	1000	6.22x10-5	3.51x10-5
	1500	5.95x10-5	3.74x10-5
	2000	5.89x10-5	3.79x10-5
	2500	5.63x10-5	3.16x10-5

Similarly, the RNA percentage per seedling decreased as 8.28×10^{-5} and 7.72×10^{-5} at 500 ppm and 2500 ppm respectively was against control it was 9.94×10^{-4} (Table 2).

Table: 2-Amount of RNA percentage in the seedlings of *Medicago sativa* after treatment of GOAL

Herbicide	Concentration in ppm	% of DNA per seedling	Standard error (±)
	Control	9.94 x 10-4	6.44 x 10-5
GOAL	500	8.28 x 10-5	5.26 x 10-5
	1000	8.03 x 10-5	6.12 x 10-5
	1500	7.94 x 10-5	5.37x 10-5
	2000	7.87 x 10-5	4.17 x 10-5
	2500	7.72 x 10-5	3.70 10-5

According to Chen *et al*. (2017) in wheat the protein/RNA ratio was higher than the control whereas in cucumber the ratio was lower. When the protein levels in both species were compared on a per unit RNA basis, there was an inverse relationship. Kamble (2006) found as the concentration of 2,4-D increases the percentage of DNA, RNA and proteins gradually decrease. Mhaiskar (2022) studied that the RNA contents of seedlings decreased gradually with the increased in concentration of herbicides. Mahakhode and Jachak (2015) reported progressive reduction in DNA, RNA and protein content as the concentrations of paraquat increased.

Conclusion:

The herbicide Goal affected the nucleic acid content of seedlings. The DNA and RNA percentage decreased with increase in the concentrations. This may be due to mitotic inhibition by the herbicide attributed to blocking of mitotic cycle during interphase which may results from

the inhibition of nucleic acid synthesis. Therefore, it may be concluded that this herbicide reduced DNA and RNA at all concentrations.

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