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Research Article

Effect of fluoride toxicity on biochemical parameters (chlorophyll, nitrogen, protein and phosphorus) of wheat (*Triticum aestivum* L.)

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ABSTRACT : The effect of sodium fluoride toxicity has been studied on chlorophyll content present in green leaves on 60th day of sowing of wheat varieties *i.e.* WH-711, HD-2932, PBW-502 and DBW-17 for one year. Nitrogen percentage, protein content (%) and phosphorus % of oven dried plant material at harvest was also estimated. Simple randomized block design was followed with four replications and six treatments including control. Five concentrations of sodium fluoride were taken as 10, 25, 50, 100 and 200 ppm and one control. On the basis of experimental findings the results were found significant. The higher concentrations of NaF from 100-200 ppm were found toxic in all the four varieties of wheat.

KEY WORDS : Wheat varieties, Fluoride toxicity, Chlorophyll, Protein content

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INTRODUCTION

Fluoride stimulates the growth and yield of many plant species. Chlorophyll content in green leaves, nitrogen, protein and phosphorus percentage have been studied by Arya *et al.* (1979). Large necrotic markings appeared on plant leaves and growth of the plants was inhibited due to the accumulation of 'F' ions in the soil and surrounding plant roots (Hadjuc (1966). Plants show more susceptibility to fluoride injury from the soil than the atmosphere. Arya (1971) reported worst injury to tomato plants when fluoride entered through the roots in 250 ppm concentration of NaF solution.

Fluoride injury to plants can be divided into two types i.e. chronic and acute. Chronic injury is produced by prolonged exposure to low concentrations of fluoride. Acute injury develops under widely fluctuating fluoride concentrations or brief exposure to a high concentration possible with a lower total fluoride exposure than is required to cause chronic injury.

Chlorophyll is of two types 'a' and 'b'. Chlorophyll 'a' is mainly responsible for photosynthesis and thus productivity. The loss of chlorophyll due to fluoride toxicity resulted into chlorosis of leaves in different plants (Arya 1971, Arya *et al.* 1979, Singh, 1992, Malik, 1997, Arya, 1997 and Kumar 1998). Menser *et al.* (1965) observed the reduction in chlorophyll contents of mutant plants. The fluoride may affect the early stages or pigment synthesis or induce the degration of chloroplast structure.

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EXPERIMENTAL METHODS

Wheat seeds were obtained from I.A.R.I., New Delhi. The varieties were WH-711, HD-2932, PBW-502 and DBW-17. The experiment was sown in Randomized Block Design with four replications. Six concentrations of NaF solutions were sprayed at the interval of 15 days, after one month of seed sowing. The treatments were control 10, 25, 50, 100 and 200 ppm. The methods adopted for the estimation of chlorophyll, nitrogen, protein content and phosphorus percentage are given below.

Estimation of total chlorophyll:

The chlorophyll content in fresh leaves was determined according to Arnon (1949). The procedure for chlorophyll determination was based on the work of Mac Kinney (1941) on the absorption of light by aqueous acetone (80%) extracts of chlorophyll. Organic solvent 4 : 1 Acetone alcohol was used.

0.5 g fresh leaves of control and treated plants were taken with organic solvent in clean specimen tubes. The extracts were centrifuged at 3000 rpm for 15 minutes and each volume was made upto 25 ml of each sample by adding more organic solvent.

Carlzeiss PMQ 2 spectrophotometer was used at Institute of Life Science, J.N.U., New Delhi and the observations of total chlorophyll content were recorded on 645, 652 and 663 wave lengths, respectively.

Total chlorophyll content was calculated by using the following formula (Arnon, 1949).

C = 20.2 D 645 + 8.02 D 663 in mg / g dry weight

Estimation of total nitrogen and protein:

Nitrogen percentage and the amount of protein content synthesized by the plant tissues were determined according to Jackson (1958) and Misra (1968). 500 mg dried and powdered plant material was taken in 50 ml Kjeldahl flasks with 5 ml of H_2SO_4. 0.1 g catalyst mixture of copper sulphate, potash sulphate and salenium dioxide in the ratio of 1 : 8 : 1, respectively was also added.

After digestion, the volumes were made upto 50 ml. Distillation was done in a Markham apparatus as described by Jackson (1958) and Misra (1968).

The nitrogen percentage = (T - B) x 5 x N x 1.4 / S

where,

T = Volume of HCl (Standard Acid used in actual titration)

B = Blank

N = Normality of NaoH = - N / 10

1.4 = Constant (atomic weight of N2)

S = Dry weight of Plant sample in g.

The difference (T - B) was multiplied by 5 because only 10 ml digested material out of 50 ml was distilled.

The protein content was determined by multiplying total nitrogen by 6.25.

Determination of Phosphorus content:

The standard solution was made by primary phosphate standard 50P. as described by Jackson (1958).

500 mg plant material (oven dried) was digested with ternary mixture at 108^{0}C. After cooling sufficiently the digest was diluted upto 100 ml in volumetric flasks. The total phosphorus in plant tissues was determined colorimetrically.

EXPERIMENTAL RESULTS AND ANALYSIS

The effect of fluoride toxicity on chlorophyll, nitrogen, protein percentage and phosphorus content of wheat (*Triticum*

Table 1: Effect of NaF on chlorophyll, nitrogen, protein % and phosphorus % of wheat (*Triticum aestivum* L.)

NaF Concentrations	Chlorophyll content in mg / gm dry weight of wheat varieties			
	WH-711	HD-2932	PBW-502	DBW-17
Control	7.85	7.78	7.51	7.29
10 ppm	7.82	7.75	7.49	7.27
25 ppm	7.80	7.72	7.43	7.23
50 ppm	7.70	7.63	7.38	7.20
100 ppm	7.62	7.56	7.28	7.17
200 ppm	7.45	7.40	7.20	7.14
NaF Concentrations	Nitrogen percentage of wheat varieties			
	WH-711	HD-2932	PBW-502	DBW-17
Control	2.08	1.88	1.88	1.88
10 ppm	2.04	1.87	1.87	1.87
25 ppm	1.94	1.84	1.84	1.84
50 ppm	1.71	1.60	1.60	1.60
100 ppm	1.35	1.20	1.26	1.20
200 ppm	1.24	1.13	1.13	1.13

Table 2 : Effect of NaF on protein content (%) and phosphorus % of wheat (*Triticum aestivum* L.)

NaF Concentrations	Protein percentage of wheat varieties			
	WH-711	HD-2932	PBW-502	DBW-17
Control	13.00	11.75	11.75	11.75
10 ppm	12.75	11.68	11.68	11.68
25 ppm	12.12	11.50	11.50	11.50
50 ppm	10.68	10.00	10.00	10.00
100 ppm	8.43	7.50	7.87	7.50
200 ppm	7.75	7.06	7.06	7.06
NaF Concentrations	Phosphorus percentage of wheat varieties			
	WH-711	HD-2932	PBW-502	DBW-17
Control	1.195	1.194	1.194	1.194
10 ppm	1.193	1.191	1.192	1.191
25 ppm	1.189	1.175	1.188	1.175
50 ppm	1.186	1.172	1.185	1.171
100 ppm	1.184	1.170	1.180	1.168
200 ppm	1.180	1.163	1.176	1.161

aestivum L.) have been presented in Table 1 and 2. The maximum chlorophyll content in green leaves on 60th day was recorded in control and minimum in 200 ppm concentration of sodium fluoride. Similarly, nitrogen, protein percentage and phosphorus content were also affected in 200 ppm NaF dose (Table 1 and 2) which were estimated from oven dried (80^0C) plant material after harvest. The concentration of 'F' ions in 200 ppm reduced chlorophyll content in green leaves of wheat which caused chlorosis and necrosis. Nitrogen and protein percent were also affected due to fluoride toxicity in higher concentrations i.e. 100-200 ppm. The phosphorus content in control and NaF treated plants was found. Maximum phosphorus content was obtained in control and minimum phosphorus percentage in 200 ppm NaF solution was found (Arya, 1971, Arya, 1997, Nimesh, 2001, Rawat 2005, Saini and Singh, 2005, Chaudhry, 2004, Malik and Arya, 2008, Neeru, 2011 and Singh, 2013).

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