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Effects of the Insecticide Carbicron on Meiotic Chromosome and other Morphological Characters of Wheat

Salma, Hossain,

University of Rajshahi

EFFECTS OF THE INSECTICIDE CARBICRON ON MEIOTIC CHROMOSOMES AND OTHER MORPHOLOGICAL CHARACTERS OF WHEAT

A dissertation

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in fulfilment of the requirements

for the degree of

MASTER OF PHILOSOPHY

by

Salma Hossain B.Sc.(Hons), M.Sc.

Cytogenetics Laboratory,

Department of Botany,

University of Rajshahi.

June, 1986.

DECLARATION

I hereby declare that the entire work submitted as the thesis for the degree of Master of Philosophy at the University of Rajshahi is the result of my own investigation.

M A Khaleque 5-8-86

(Dr. M.A.Khaleque)

Supervisor.

Salma Hossain 2.8.86.

(Salma Hossain)

Candidate

Dedicated to the memory of the Late Professor Sultanul Alam under whose supervision the present work was initiated and completed but who could not see the final form of the dissertation.

The author

ABSTRACT

Chromosomal aberrations viz. sticky and contracted chromosomes, bridges, fragments and micro-nuclei were significantly increased in meiocytes of hexaploid and tetraploid wheat when seeds and/or plants were treated with the insecticide, Carbicron - 100. The insecticide treatment also significantly reduced plant height at heading, number of fertile tillers, number of spikelets per ear and number of grains per ear in both the wheat species. In an attempt to determine whether any of the observed cytological and morphological effects can be transmitted to subsequent generations, plants were grown from seeds of the treated plants in the following year. In the selfed progeny of the hexaploid wheat neither any increase in the proportion of chromosomal aberrations nor any significant effect on the morphological characters was detected. In tetraploid wheat a significant reduction in plant height and number of fertile tillers was detected but for chromosomal aberrations and other morphological traits no difference between progeny of treated and untreated families was found.

ACKNOWLEDGEMENT

The author wishes to express her sincere gratitude to the Late Professor Sultanul Alam of the Department of Botany, University of Rajshahi, for his advice and supervision throughout the course of this investigation.

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The Author

Contents

INTRODUCTION

The chromosomes are of vital importance to all plants and animals. The genes, units of inheritance of characters, are integral part of the chromosomes. It is obvious that deviations from the normal chromosomal complement may affect the inheritance of characters of an organism. Many different types of changes in architecture of the chromosomes are known which involve variations both in chromosome numbers and chromosome structure and most of these affect inheritance of characters. Although such changes in chromosome architecture may give rise to variations useful in natural selection and breeding, the majority of changes of this type lead to the deterioration of quality and yield of crop plants. In agriculture, the stability of quality and high yield of a crop is very important. So plant breeders and farmers want that the quality and other agronomic characters of a selected high yielding variety (HYV) of a crop should remain unchanged from year to year. Change in the quality and characteristics of a crop, caused by an alteration in the chromosome architecture, will be inherited by the offspring, thus, affecting crops of future generations as well.

In modern agriculture, even in an under-developed country, like Bangladesh, chemical insecticides and pesticides become a common factor in crop production. Almost every crop in the field comes in contact with these chemicals often on several occasions during its life-time.

The cry for increasing yield of every agricultural commodity demands the use of insecticides to protect the HYV's from the vicious attack of harmful insects. These insecticides, being poisonous and lethal to the insects may also have some harmful effects on the crop plants themselves.

Research related to the insecticide decomposition, absorption, translocation, incorporation, metabolism and soil residues has been emphasized all over the world. However, basic research aimed at a better understanding of the application of the insecticides on chromosomal behaviour of various crops is lacking specially in case of tropical countries.

Transmittable changes in plants induced by insecticides may be caused by an alteration of the genetic architecture. A better understanding of the cytogenetic response of plants to insecticides and their relationships to the performance of these chemicals would help to enhance safe and effective control of insect pests.

The present investigation was undertaken to demonstrate (a) the effect of insecticide on meiotic chromosomes of treated plants; (b) the effects of the treatments on the morphological and yield contributing characters and (c) whether these effects on meiotic chromosomes, and morphological and yield characters are inherited by the offspring of the treated plants.

To achieve these goals, two species of wheat, hexaploid Sonalika and a variety of tetraploid Durum were subjected to different treatments of a systemic insecticide, Carbicron. The effect of these treatments on meiotic chromosomes and on a number of morphological characters were studied. The seeds from treated plants were sown next year to identify any heritable effects of the insecticide treatments of previous year.

REVIEW OF LITERATURE

Chromosomal abnormalities due to treatment of chemicals used as insecticides and herbicides were reported as early as 1950s(Unrau, 1953, 1954; Unrau and Corns, 1950; Brown, 1950, Doxey, 1949; Dunlap, 1951; McIlrath and Ergle, 1953; Sunneson, 1960; Tukey, 1950; Unrau and Larter, 1952; Wuu and Grant, 1966).

Dunlap (1951) had shown that a 'stimulus' producing symptoms of 2, 4-D injury could be transmitted to the next generation via cotton seeds. There is also much evidence that such effects may persist in the vegetative parts of some plants after 2, 4-D treatment (Brown, 1950; McIlrath and Ergle, 1953; Tukey, 1950). Abnormalities induced by 'Dalofin' were found several generations after the herbicide was applied to barley (Sunneson, 1960).

Unrau and his associates had conclusively demonstrated that herbicide 2, 4-dichlorophenoxy acetic acid (2, 4-D) was responsible for inducing chromosomal aberrations in both meiotic and mitotic cells of barley and wheat (Unrau, 1953, 1954; Unrau & Corns, 1950). They reported that the herbicide might result in heritable changesin some morphological characters. Unrau and Larter (1952) also reported high percentage of pollen mother cells (PMC) of wheat and barley to be affected after spraying the plants with 2, 4-D before microsporogenesis.

Another phenomenon of the coalescence of chromosomes into masses of various numbers and sizes during meiosis in barley plants originating from the seeds treated with a pesticide, Lorox, was reported by Wuu and Grant (1966). In their study, as high as 100% of the pollen mother cells contained chromosome abnormalities. The same authors further reported

76.0 to 98.5% barley pollen mother cells with various types of chromosomal abnormalities when the seeds were treated with another pesticide, Monuron (Wuu & Grant, 1967a). Pesticides were also found to cause chromosomal aberrations in meiotic cells when seeds of _Vicia faba_ and _Gossypium barbadense_ as demonstrated by Amer and his associates (Amer, 1965; Amer and Ali, 1968, 1969, 1974, 1980; Amer and Farah, 1968, 1974, 1975, 1976, 1980). In _Vicia faba_ the induced chromosomal aberrations were stickiness, lagging chromosomes, fragments and anaphase bridges, univalents at diakinesis, disturbed second metaphase and anaphase, micronuclei at first and second anaphase and multi-polar second telophase.

Reddy and Ramanna Rao (1969) studied the cytological effects of two common insecticides 'Dimecron - 100' and 'Rogor - 40' on _Vicia faba_. They reported that both root tip cells in division as well as pollen mother cells exhibited chromosomal aberrations when treated with different doses of these two insecticides.

The cytological effects of the pesticides, 'Menazon', 'Bromuron' and tetrachloro-isophthato nitrite in _Hordeum_ and _Tradescantia_ was reported by Tomkin and Grant (1972). These chromosomal abnormalities were found to be specific and localized.

Ahmed and Grant (1972) also studied the effects of 'Phosdrin' and 'Bladex', two other pesticides on mitotic cells of root tips of _Tradescantia_ and _Vicia faba_. Both of these pesticides produced similar chromosomal abnormalities.

Alam and his associates studied the effects of different insecticides on wheat and observed that various kinds of chromosomal abnormalities were caused by the insecticides, Carbicron, Dimecron and Vapona (Alam _et al._, morphological and yield contributing characters. The need of insecticides in modern agriculture can not be over emphasized. So, it is important to find out whether the increase in the occurance of chromosomal aberrations have any effect on yield and quality of the crop or not.

Plant materials:

A hexaploid wheat (Triticum aestivum var. Sonalika, 2n = 42) and a tetraploid wheat (Triticum durum, 2n = 28) were used as the materials of this study. The pure breeding seeds were obtained from the Cytogenetics Laboratory, Department of Botany, University of Rajshahi.

Insecticide:

The insecticide used is commonly known as 'Carbicron - 100' which was collected from the Thana Agricultural Office, Paba, Rajshahi. The active ingredient of the insecticide is 3-(Dimethoxy-phosphinyloxy) N, N-dimethyl - cis - crotonamide with the emperical formula, $C_8H_{16}O_5PN$ produced by CIBA Agrochemical Division.

This is a systemic insecticide based on enol phosphate dicrotophos. The chemical is taken up by the sprayed plants within a few hours of application. It has been widely used over a broad range of crops as a stomach poison against many sucking, chewing and mining insects.

MATERIALS AND METHODS

MATERIALS

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METHODS

Preparation of chemicals for treatment:

Two different doses (D_1 and D_2) were prepared by mixing 0.2 oz. and 0.4 oz. of Carbicron - 100 with 2.5 gallons of distilled water.

Treatments:

The insecticide was treated to the plant material in three different ways:

(a) Seeds were soaked in the prepared chemical only,

(b) Untreated seedlings were sprayed with the chemical, and

(c) Seedlings from seed-treated materials were again sprayed with the chemical.

In every case controls were kept where seeds were soaked in distilled water or seedlings from untreated seeds were sprayed with distilled water.

The following abbriviations were used to designate the different treatments of each of Sonalika and Durum wheat (Page10):

D_o : Seeds soaked in distilled water and sprayed with distilled water, used as controls.

STD_1 and STD_2 : Seeds soaked in two different doses of insecticides D_1 and D_2 respectively for 12 hours, without any spray.

SPD_1 and SPD_2 : Seedlings from untreated seeds of two weeks old sprayed with the two doses of insecticides D_1 and D_2 respectively.

$STPD_1$ and $STPD_2$: Seedlings from treated seeds (STD_1 & STD_2) were sprayed with the two doses of insecticides D_1 and D_2 respectively.

During 1981-82 winter one row each of D_o, SPD_1, SPD_2; D_o, STD_1, STD_2; D_o, $STPD_1$ and $STPD_2$ plants were planted for both Sonalika and Durum. During 1982-83 winter, the experiment of 1981-82 was repeated along with the selfed seeds of all these treatments from the previous year.

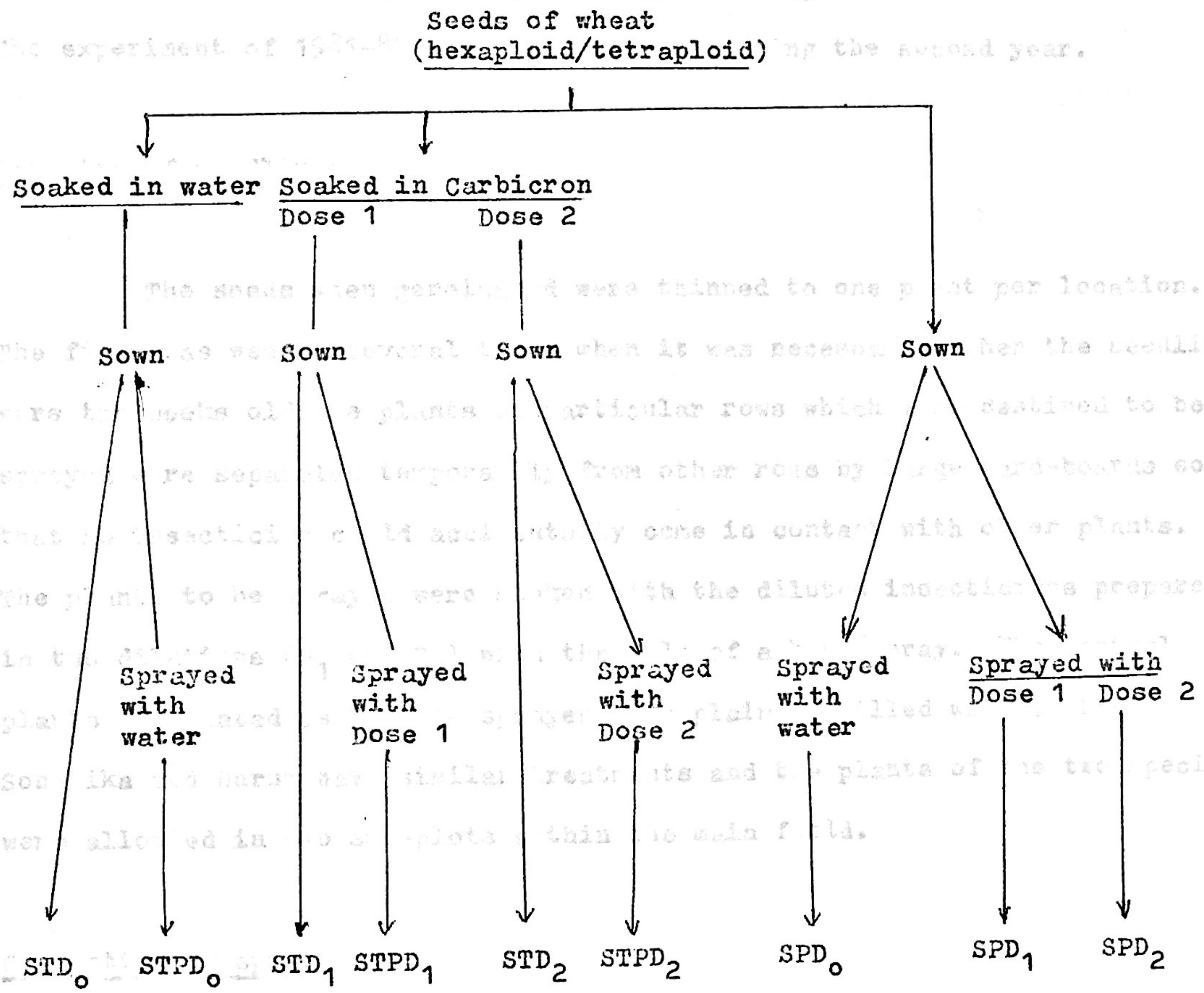
Each of the treatments including the control rows ~~were~~ was assigned to a random code number and this number was always used for reference during the collection of cytological and morphological data. After the tabulation of these data were complete, the code numbers were replaced by the actual treatment names.

Sowing of seeds:

The dry seeds were either soaked in distilled water or in the insecticide of proper concentrations in petri-dishes for 12 hours. Then the solutions were decanted off and the treated seeds were washed thoroughly in tap water. The seeds for control (D_o) were soaked in distilled water for the same period so as the seeds for plants which would be sprayed later.

The field was ploughed repeatedly and pulverized thoroughly. Cow-dung and other fertilizers were added at standard doses. The seeds were sown in rows in a field with randomized block design. The distance between rows was 30 cm. and from plant to plant was 7 cm. Non-experimental rows

Summary of the treatments used in the study



were planted around the experimental field to minimize the edge-effect. The first set of the experiment was carried out during the winter of 1981-82.

The seeds collected from the 1981-82 season were kept separate for each treatment and were grown untreated during the winter of 1982-83. The experiment of 1981-82 was also repeated during the second year.

Spraying of seedlings:

The seeds when germinated were thinned to one plant per location. The field was weeded several times when it was necessary. When the seedlings were two weeks old the plants of particular rows which were destined to be sprayed were separated temporarily from other rows by large hard-boards so that no insecticide could accidentally come in contact with other plants. The plants to be sprayed were soaked with the diluted insecticides prepared in two dilutions (D_1 and D_2) with the help of a hand-spray. The control plants designated as D_o were sprayed with plain distilled water. Both Sonalika and Durum have similar treatments and the plants of the two species were allotted in two sub-plots within the main field.

Collection of spikes:

At the flag leaf stage, young inflorescences from the various treatments and the control plants were collected. The material first collected was checked for the presence of right stages of meiotic division. These collected inflorescences were, immediately fixed in a modified Carnoy's fixative. After 48 hours the materials were transferred to 70% ethanol and stored in a refrigerator for examination.

Preparation of slides:

Temporary slides were prepared by the acetocarmine smear method and the meiotic abnormalities were examined and noted. Cytological screening was made for all the stages of meiosis from Diakinesis to Telophase II. The different types of abnormalities were recorded in a record book. Photographs were taken from the desired preparations (Figs.1 to 6).

Collection of morphological data:

The morphological data on plant height, number of tillers per plant, number of spikelet per ear, number of seeds per ear ~~and weight of seeds per ear~~, were taken from ten randomly selected plants of each treatment and control rows.

RESULTS

The results of the present study are reported under the following heads:

(1) Meiotic Abnormalities of the Hexaploid Wheat of 1981 - 82 and 1982 - 83.

(2) Meiotic Abnormalities of the Tetraploid Wheat of 1981 - 82.

(3) Morphological Data of 1981 - 82 and 1982 - 83.

(4) Performance of the Selfed Progeny of the Insecticide Treated Plants of 1981 - 82 during 1982 - 83.

(5) Correlation Analysis of Grain Data.

(1) Meiotic Abnormalities of the Hexaploid Wheat of 1981-82 and 1982-83

Meiotic studies of the pollen mother cells carried out revealed various types of chromosomal aberrations. Contraction and stickiness of the chromosomes were one of the common effects of the insecticide on the meiotic chromosomes. This contraction and stickiness of the chromosomes was the first noticeable response of the insecticide. The pollen mother cells were found to contain chromosomes with varying degrees of contraction forming masses of chromosomes. In the 1982-83 material, the percentage of PMCs with such contracted chromosome masses were 0.7, 5.9, 6.1, 8.5, 7.8, 8.0 and 9.6 in D_0, STD_1, STD_2, SPD_1, SPD_2, $STPD_1$ and $STPD_2$ respectively. Therefore, this cytological aberration induced by the insecticide was not divergent in case of the different doses and treatments.

The other common and recurrent aberrations observed were chromosome and chromatid fragments, chromosome bridges and micronuclei. A few photographs of different types of abnormalities are included in Figs. 1 to 6. At least five slides per treatment (D_o, STD_1, STD_2, SPD_1, SPD_2, $STPD_1$ and $STPD_2$) were scored and a summary of the occurance of these abnormalities from Metaphase I to Telophase II for 1981 - 82 and 1982 - 83, are shown in Tables 1 & 2 respectively. A total of 12,044 meiocytes were scored in 1981 - 82 and 20,652 meiocytes were examined in 1982 - 83.

In the 1981 - 82 experiment, a total of 11,953 meiocytes with normal chromosomes were scored out of which 7,452 (62.34%) belonged to the different stages of meiosis I, whereas, 4,501 (37.66%) belonged to Meiosis II. On the other hand, of the total abnormal cells examined (1,091), 909 were from stages of Meiosis I (83.33%), whereas, only 182 (16.67%) belonged to Meiosis II.

In the 1982 - 83 experiment, out of 19,170 normal meiocytes scored, 14,431 (75.28%) belonged to Meiosis I and only 4,739 (27.72%) belonged to Meiosis II. Out of the 1982 meiocytes with chromosomal aberrations, 1,179 (79.55%) were in Meiosis I, whereas, only 303 (20.45%) cells belonged to Meiosis II.

When classified according to individual stages i.e., Metaphase, Anaphase, and Telophase, the most frequently encountered one was Metaphase (9,400 i.e., 45.52% of all cells studied). Next frequent was Telophase, 9,306 i.e., 45.06% of all the meiocytes scored. Cells belonged to the Anaphase were the least frequent, 1,946 i.e., only 9.42% of all cells. These figures are from the 1982-83 experiment, similar figures for 1981-82 experiments are 4,674 i.e., 38.81% of Metaphase, 6,043 i.e. 50.17% of Telophase and 2,327 i.e., 19.32% of Anaphase.

Table 1

Summary of meiotic cells studied for chromosomal aberrations in hexaploid wheat after Carbicron treatments in 1981-82 experiment.

Treatment	Division	Cells without aberrations M	A	T	Cells with aberrations M	A	T	Total normal cells	Total abnormal cells	Percentage of abnormal cells
D_o	Meiosis I	745	467	544	69	57	4	3243	132	3.91
	Meiosis II	214	483	790	1	-	1			
STD_1	Meiosis I	626	162	1072	141	54	43	2267	267	10.54
	Meiosis II	222	121	64	13	14	2			
STD_2	Meiosis I	144	100	648	37	40	40	1135	125	9.92
	Meiosis II	89	53	101	-	4	4			
SPD_1	Meiosis I	442	108	220	40	42	14	870	114	11.59
	Meiosis II	44	24	32	11	5	2			
SPD_2	Meiosis I	146	52	433	42	27	21	1494	123	7.61
	Meiosis II	204	126	533	14	8	11			
$STPD_1$	Meiosis I	86	7	193	41	4	25	1346	138	9.30
	Meiosis II	267	156	637	21	10	37			
$STPD_2$	Meiosis I	790	120	347	129	21	18	1598	192	10.73
	Meiosis II	85	59	197	11	3	10			

M = Metaphase;
A = Anaphase;
T = Telophase.

Table 2

Summary of meiotic cells studied for chromosomal aberrations in hexaploid wheat after Carbicron treatments in 1982-83 experiment.

Treatment	Division	Cells without aberrations M	A	T	Cells with aberrations M	A	T	Total normal cells	Total abnormal cells	Percentage of abnormal cells
D_o	Meiosis I	986	127	1066	51	9	23	2815	101	3.46
	Meiosis II	113	15	508	5	3	10			
STD_1	Meiosis I	1229	225	1233	248	63	76	3150	426	11.89
	Meiosis II	145	48	270	19	11	9			
STD_2	Meiosis I	1073	288	915	108	39	32	2846	201	6.60
	Meiosis II	330	71	169	16	1	5			
SPD_1	Meiosis I	773	122	765	84	6	34	2483	174	6.55
	Meiosis II	364	78	381	29	2	19			
SPD_2	Meiosis I	680	131	942	87	14	35	2365	153	6.08
	Meiosis II	257	81	274	11	4	2			
$STPD_1$	Meiosis I	979	187	1239	90	20	37	3406	251	6.86
	Meiosis II	360	123	518	73	5	26			
$STPD_2$	Meiosis I	895	162	414	61	14	48	2105	176	7.72
	Meiosis II	321	86	227	13	11	29			

M = Metaphase;

A = Anaphase;

T = Telophase.

Fig 1 Photomicrograph showing Metaphase I with lagging chromosomes in wheat induced by the insecticide Carbicron.

Fig 2 Photomicrograph showing Metaphase I with lagging chromosomes in wheat induced by the insecticide Carbicron.

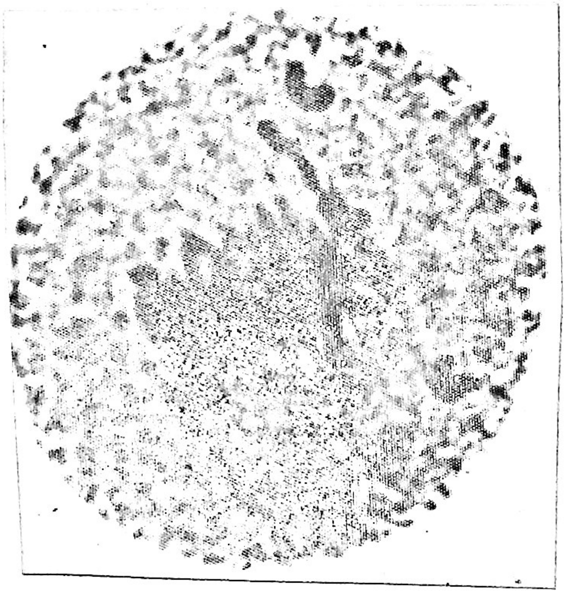


Fig 1

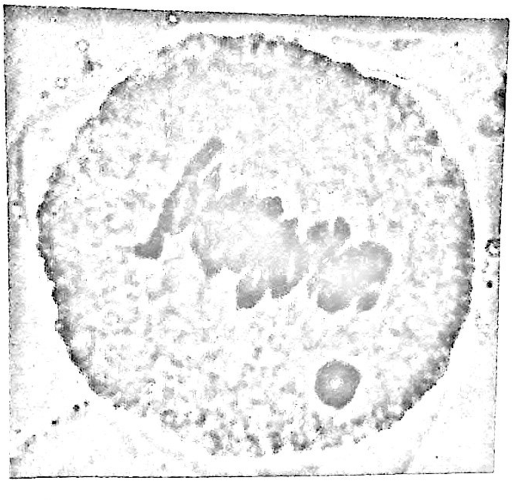


Fig 2

Fig 3 Photomicrograph showing Anaphase I with multiple bridges in meiocytes of wheat induced by the insecticide Carbicron.

Fig 4 Photomicrograph of Anaphase I with multiple bridges in meiocytes of wheat induced by Carbicron.

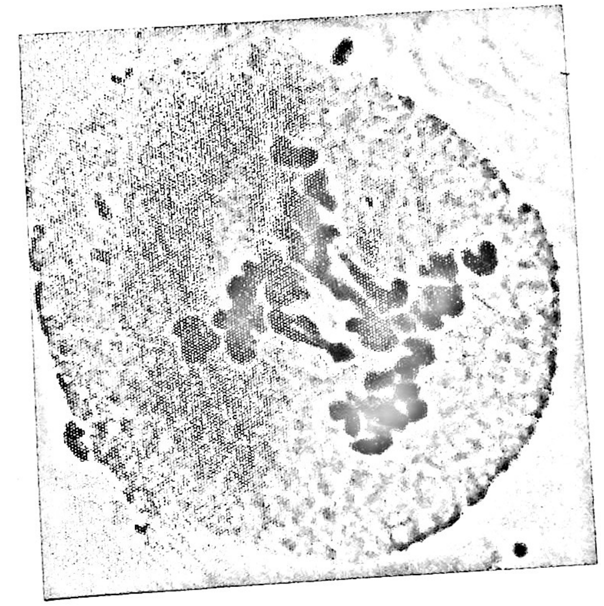


Fig 3

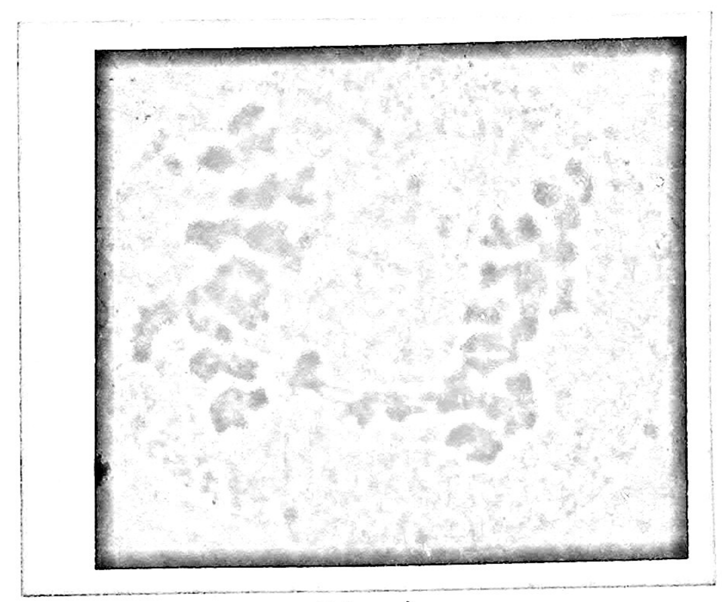


Fig 4

Fig 5 Photomicrograph of Anaphase I with single bridge in wheat meiocytes caused by Carbicron.

Fig 6 Photomicrograph of Telophase I with lagging chromosomes of wheat in Carbicron treated meiocyte.

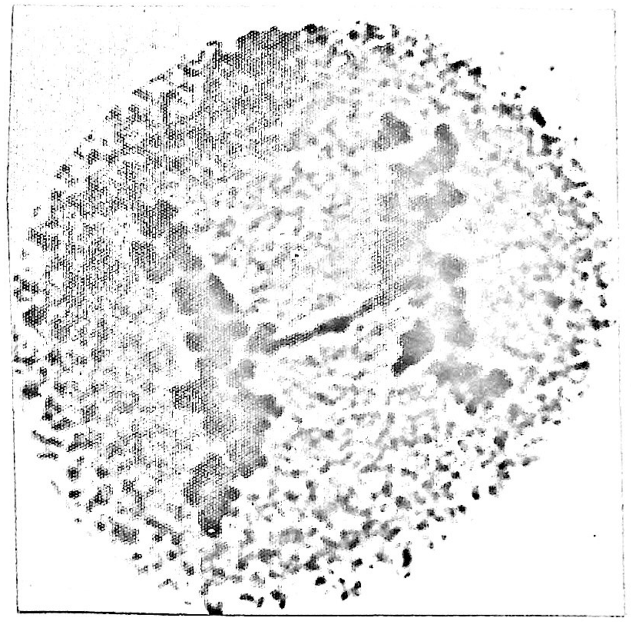


Fig 5

Fig 6

Fig 6 a Photomicrograph of Telophase I with micro-nuclei in wheat meiocytes treated with Carbicron.

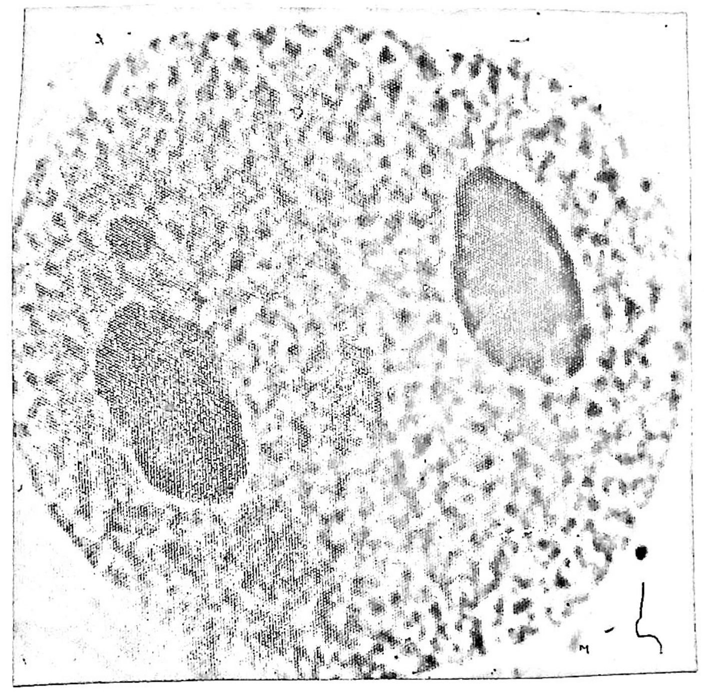


Fig 7

The proportion of the abnormal cells when calculated for individual stages of meiosis, for 1981-82 data, the proportion of meiocytes with aberrations was for Metaphase 570 against 4,104 normal (0.1389), 289 against 2,038 normal (0.1418) for Anaphase and 232 against 5,811 (0.0399) for Telophase; similar data for 1982-83 were for Metaphase 895 abnormal against 8,505 normal meiocytes (0.1052), for Anaphase, 202 against 1744 (0.1158) and for Telophase 385 abnormal against 8,921 normal (0.0432).

When the data in Tables 1 & 2 were summarized for proportion of meiocytes with chromosomal aberrations separately for Meiosis I and II, the following picture emerged. In 1981-82 experiment, the total number of abnormal cells in Meiosis I was 909 against 7,452 normal ones, i.e., the proportion was 0.1220; whereas of Meiosis II, there were 182 abnormal cells and 4,501 normal cells, the proportion being 0.0404. Similar figures for 1982-83 experiment were 1,179 abnormal against 14,431 normal cells in Meiosis I, the proportion being 0.0817 whereas, for Meiosis II, there were 303 abnormal cells and 4,739 normal cells (0.0639).

The different types of chromosomal aberrations observed after the insecticide treatments in hexaploid wheat in the 1981-82 experiment are summarized in Table 3. From the untreated (control) material in all 3,375 meiocytes were scored and 132 were found to contain different types of chromosomal abnormalities. Here the percentage of abnormal cells was 3.91, but in the six treatments (STD_1, STD_2, SPD_1, SPD_2, $STPD_1$ and $STPD_2$) in all 10,969 meiocytes were scored and a total of 959 abnormal cells were detected which gives an overall percentage of 8.74. Thus, it becomes clear that insecticide treatments did increase the number of chromosomal abnormalities in the hexaploid wheat. The overwhelming majority of the chromosomal abnormalities observed were laggards (65.63% of all

Table 3

Different types of chromosomal aberrations observed after Carbicron treatments in hexaploid wheat during 1981 - 82.

Treatment	Chromosomal aberrations				Total normal cells	Total abnormal cells	Total cells studied	Percentage of abnormal cells
	Laggards	Frag-ments	Bridges	Micro-nuclei				
D_0	91	19	21	1	3243	132	3375	3.91
STD_1	205	9	31	22	2267	267	2534	10.54
STD_2	71	7	30	17	1135	125	1260	9.92
SPD_1	56	20	36	2	870	114	984	11.59
SPD_2	63	12	34	14	1494	123	1617	7.61
$STPD_1$	69	3	7	59	1346	138	1484	9.30
$STPD_2$	161	-	13	18	1598	192	1790	10.73

abnormalities), followed by bridges (15.77%) and micronuclei (12.19%). The least frequent abnormality was fragments (6.42%).

Similar results were obtained again in the 1982-83 experiment (Table 4). The percentage of abnormalities in the untreated material was only 3.46 but in the insecticide treated material the overall percentage of the chromosomal abnormalities was 7.78. Once again laggards were the most frequent aberrations observed (72.91% of all aberrations) followed by fragments (14.39%) and micronuclei (8.78%).

The results of Analysis of Variance of the 1981-82 and the 1982-83 data carried out on the percentages of different chromosomal aberrations caused by the various treatments of the insecticide are given in Tables 5 & 6. The percentages were transformed into angles following the angular (Arcsine) transformation procedure developed by Bliss (1937). The transformed data was analysed following randomized design. In both the 1981-82 and the 1982-83 data, the 'Dose' item was highly significant whereas, in the 1982-83 data 'Methods' item was significant at 5% level. Inspection of the treatment means given in Tables 3 & 4 clearly indicates that the difference between the control (D_o) and the Treatment means contributed much to the 'Dose' item.

(2) Meiotic Abnormalities of the Tetraploid Wheat of 1981-82

A variety of tetraploid wheat, Triticum durum ($2n = 4x = 28$) was included in the original experiment to find out whether there is any difference in the response of meiotic chromosomal behaviour to the insecticide treatment associated with chromosome number difference (28 vs. 42).

Table 4

Different types of chromosomal aberrations observed after Carbicron treatment in hexaploid wheat during 1982 - 83.

Treatment	Chromosomal aberrations				Total normal cells	Total abnormal cells	Total cells studied	Percentage of abnormal cells
	Laggards	Frag-ments	Bridges	Micro-nuclei				
D_0	53	25	6	17	2815	101	2916	3.46
STD_1	296	45	17	19	3150	426	3576	11.89
STD_2	163	22	4	12	2846	201	3047	6.60
SPD_1	138	26	1	9	2483	174	2657	6.55
SPD_2	134	12	4	3	2365	153	2518	6.08
$STPD_1$	158	17	15	61	3406	251	3657	6.86
$STPD_2$	137	18	12	9	2105	176	2281	7.72

Table 5

Results of analysis of variance of transformed data (percentage to angular) for meiotic chromosomal abnormalities of hexaploid wheat during 1981 - 82.

Item	SS	df	MS	F	P
Total	1112.21	44			
Replications	30.82	4	7.71	< 1	NS
Treatments	502.66	8			
Methods	0.25	2	0.13	< 1	NS
Doses	479.85	2	239.92	13.26	***
Methods X Doses	22.56	4	5.64	< 1	NS
Error	578.74	32	18.09		

Table 6

Results of analysis of variance of transformed data (percentage to angular) for meiotic chromosomal abnormalities of hexaploid wheat during 1982 - 83.

Item	SS	df	MS	F	P
Total	620.95	44			
Replications	43.81	4	10.95	1.53	NS
Treatments	348.59	8			
Methods	50.66	2	25.33	5.33	*
Doses	271.89	2	135.95	19.04	***
Methods X Doses	26.04	4	6.51	<1	NS
Error	228.35	32	7.14		

Squash preparations of pollen mother cells of tetraploid wheat were studied during the 1981-82 season only. Again, the most common aberration observed was lagging chromosomes, followed by bridges, fragments and micronuclei. A total of 10,048 cells were scored,the details are given in Table 7. Out of 10,048, 6029 (60%) belonged to Meiosis I, whereas 4,019 (40%) belonged to Meiosis II. The number of cells with aberrations scored in Meiosis I stages was 362 out of a total of 527 i.e. 68.69%. The number of normal cells belonging to Meiosis I was 5,667 out of 10,048 i.e., 59.52%.

The most frequent stage in this study was Telophase, a total of 5,316 out of 10,048 i.e., 52.90% followed by Metaphase, a total of 3,205 out of 10,048 i.e. 31.89%; whereas, the least frequent class was Anaphase, 1527 amongst 10,048 cells i.e., only 15.20%.

The proportion of meiocytes with chromosomal abnormalities was highest in Metaphase, 201 out of 3,205 (0.0627), followed by Telophase, 243 out of 5,316 (0.0457) and Anaphase, 83 out of 1,527 (0.0543).

The percentage of abnormal cells found among 636 untreated (control D_o) meiocytes was 3.30, whereas, the percentage of abnormal cells in the six different treatments were 4.65 to 6.59.

The results of Analysis of Variance carried out on the percentages of chromosomal aberrations in the insecticide treated tetraploid wheat is given in Table 8. In this case the Replications, Dose and Dose X Methods items were significant at 5% level.

Table 7

Summary of meiotic cells screened for chromosomal aberrations in tetraploid wheat after Carbicron treatments in 1981-82.

Treatment	Division	Cells without aberrations M	A	T	Cells with aberrations M	A	T	Total normal cells	Total abnormal cells	Percentage of abnormal cells
D_o	Meiosis I	151	150	267	22	21	3	1582	54	3.30
	Meiosis II	136	366	512	0	4	4			
STD_1	Meiosis I	226	87	595	32	10	13	1249	88	6.58
	Meiosis II	161	121	59	18	9	6			
STD_2	Meiosis I	142	86	398	10	7	18	1048	60	5.42
	Meiosis II	151	83	188	13	1	11			
SPD_1	Meiosis I	426	77	373	23	3	20	1042	53	4.84
	Meiosis II	51	15	100	1	-	6			
SPD_2	Meiosis I	186	43	378	11	4	16	1312	64	4.65
	Meiosis II	222	94	389	7	6	20			
$STPD_1$	Meiosis I	253	67	697	16	2	48	1785	102	5.41
	Meiosis II	343	77	348	14	2	20			
$STPD_2$	Meiosis I	443	123	499	31	10	42	1503	106	6.59
	Meiosis II	113	55	270	3	4	16			

M = Metaphase;

A = Anaphase;

T = Telophase.

Table 8

Results of analysis of variance of transformed data (percentages to angular) for meiotic chromosomal aberrations in Carbicron treated tetraploid wheat during 1981 - 82.

Item	SS	df	MS	F	P
Total	446.60	44			
Replications	89.57	4	22.39	3.47	5%
Treatments	150.56	8			
Methods	11.81	2	5.91	<1.00	NS
Doses	53.04	2	26.52	4.11	5%
Methods X Doses	85.71	4	21.43	3.32	5%
Error	206.47	32	6.45		

(3) Morphological Data of 1981 - 82 and 1982 - 83

For collection of morphological data, ten randomly chosen plants from each of the 'Treatments' including the 'control' row were scored. The morphological characters scored were plant height, number of tillers per plant, number of spikelets per ear and number of grains per ear. The data were analysed using a Replicated Two-way Analysis of Variance Model. The Replications item was first separated with 9 degrees of freedom, then the total SS was partitioned into a Treatments item with 8 degrees of freedom, and a Residual item with 72 degrees of freedom. The Treatments item was further partitioned into Methods item with 2 degrees of freedom. Doses item was with 2 degrees of freedom and Methods X Doses item was with 4 degrees of freedom. As a Factorial design can not be fitted to the complicated experimental data, this separation of Treatments item into sub-components was not strictly valid, this is because the effects of different types of application i.e., ST, SP and STP can not be separated from the effects of the different doses applied i.e., D_1 and D_2. However, the significance of these items would conclusively indicate the effect of insecticide, which is the main aim of the present research project. Little importance was given to the dose effects or the effects of the different methods of application of the insecticide. A fully factorial experimental design is needed for such a study.

(i) Plant Height at Heading: The height in cm. at heading of Carbicron treated and control plants of hexaploid and tetraploid wheat grown during 1981-82 are given in Tables 9 & 10 respectively. The ranges of plant height of hexaploid wheat for 'control' and 'Treatment' families

Table 9

Plant height (cm) at heading of Carbicron treated and control plants of hexaploid wheat during 1981-82.

Treatment	Dose	Plants 1	2	3	4	5	6	7	8	9	10	Mean
SP	D_o	97	85	87	92	88	89	102	84	97	91	91.2
	D_1	95	99	96	96	91	96	92	89	79	73	90.6
	D_2	81	80	92	85	89	81	87	81	90	79	84.5
ST	D_o	87	88	86	93	114	81	94	89	106	105	94.3
	D_1	85	74	83	83	98	72	82	86	96	94	85.3
	D_2	93	84	86	96	84	88	81	90	93	88	88.3
STP	D_o	102	92	85	87	73	91	80	96	90	80	88.3
	D_1	102	84	90	85	94	77	85	85	78	87	86.7
	D_2	90	91	87	91	96	91	86	87	86	92	89.7

Table 10

Plant height (cm) at heading of Carbicron treated and control plants of tetraploid wheat in 1981-82.

Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
		Plants										
	D_0	91	94	101	102	101	96	96	90	106	96	97.3
SP	D_1	101	101	95	97	90	98	103	97	90	95	96.7
	D_2	94	102	103	99	102	95	95	102	98	97	98.7
	D_0	88	98	99	101	99	82	92	88	84	88	91.9
ST	D_1	101	95	104	101	85	86	81	91	90	81	91.5
	D_2	90	100	91	92	83	80	84	90	81	98	88.9
	D_0	94	98	99	92	98	89	95	109	99	96	96.9
STP	D_1	96	99	91	89	90	91	101	91	90	81	91.9
	D_2	93	85	91	98	92	90	83	82	90	91	89.5

were 73 to 114 cm. and 72 to 102 cm. respectively; whereas that for tetraploid wheat were 82 to 109 cm. for control families and 80 to 104 cm. for treatment families. The mean height of treated plants of both hexaploid and tetraploid wheat were lower than that of the control plants. The results of the Analysis of Variance (Tables 10 & 12) indicated significant Treatment items. The differences between the means for different doses of the treatments indicated a weak trend towards higher dose reducing the height of treated plants.

(ii) Number of Fertile Tillers per Plant: The number of fertile tillers per plant (Tables 13 & 14) exhibited a wide range of variation both in cases of hexaploid and tetraploid wheat. In hexaploid wheat, the range in 'Control' families was 3 to 23; whereas, in treated families it was 3 to 17, in tetraploid wheat these are 6 to 18 and 2 to 18 respectively. In most of the cases of both the species the mean number of tillers was higher in the untreated (control) families than in the Carbicron treated families. The results of the Analysis of Variance (Tables 15 & 16) confirmed this, in both the cases the 'Treatment' items were highly significant, indicating that the insecticide treatments did reduce the number of fertile tillers in both the wheat species.

(iii) Number of Spikelets per Ear of the Main Branch: The number of spikelets per ear of the main branch for hexaploid and tetraploid wheat species are given in Tables 17 & 18 respectively. In hexaploid species, the range for the 'Control' plants was 10 to 22 and for the treated families was 10 to 20; whereas, in the case of tetraploid wheat, these were 16 to 21 and 15 to 20 respectively. In all cases, the mean number

Table 11

Results of analysis of variance of plant height at heading of Carbicron treated and control plants of hexaploid wheat during 1981 - 82.

Item	SS	df	MS	F	P
Total	4944.10	89			
Replications	815.21	9	90.58	<1.93	NS
Treatments	757.00	8	94.63	2.02	*
Methods	17.07	2	8.08	<1.00	NS
Doses	281.27	2	140.64	3.00	10 - 5%
Methods X Doses	458.66	4	114.67	2.45	NS
Error	3371.89	72	46.83		

Table 12

Results of analysis of variance of plant height of Carbicron treated and control plants of tetraploid wheat during 1981 - 82.

Item	SS	df	MS	F	P
Total	3856.90	89			
Replications	524.23	9	58.25	1.8	NS
Treatments	1092.80	8	136.50	4.39	**
Methods	732.80	2	336.40	11.77	***
Doses	140.00	2	70.00	2.25	NS
Methods X Doses	219.20	4	54.80	1.76	NS
Error	2240.67	72	31.12		

Table 13

Number of fertile tillers per plants of Carbicron treated and control plants of hexaploid wheat during 1981 - 82.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
SP	D_0	9	5	15	11	9	5	7	9	13	12	9.5
	D_1	4	15	10	10	10	10	8	8	11	9	9.5
	D_2	6	6	9	8	6	3	6	5	8	6	6.3
ST	D_0	9	7	14	14	12	10	16	13	23	16	13.4
	D_1	4	3	4	5	5	6	8	6	6	10	5.7
	D_2	11	8	10	11	8	12	8	10	11	10	9.9
STP	D_0	16	15	5	8	3	6	3	10	7	4	7.7
	D_1	8	13	9	8	5	4	4	7	5	10	7.3
	D_2	12	7	17	10	9	12	5	5	6	10	9.3

Table 14

Number of tillers (fertile) per plant of Carbicron treated and control plants of tetraploid wheat during 1981 - 82.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	14	11	9	13	18	9	14	8	11	18	12.5
SP	D_1	17	10	7	14	6	11	13	12	11	9	11.0
	D_2	7	12	18	8	13	11	14	13	17	10	12.3
	D_o	12	15	12	13	13	5	16	10	10	9	11.5
ST	D_1	8	9	12	4	4	15	6	6	9	6	7.9
	D_2	6	11	12	11	8	5	4	7	2	5	7.1
	D_o	11	9	6	6	9	9	14	13	9	11	9.7
STP	D_1	9	6	10	6	11	10	9	11	6	7	8.5
	D_2	6	7	11	8	10	9	5	6	7	10	7.9

Table 15

Results of analysis of variance of number of tillers per plant of Carbicron treated and control plants of hexaploid wheat during 1981-82.

Item	SS	df	MS	F	P
Total	1259.60	89			
Replications	101.38	9	11.26	1.11	NS
Treatments	428.80	8	53.60	5.29	***
Methods	40.87	2	20.44	2.02	NS
Doses	111.80	2	55.90	5.52	**
Methods X Doses	276.13	4	69.03	6.81	**
Error	729.42	72	10.13		

Table 16

Results of analysis of variance of number of tillers per plant of Carbicron treated and control plants of tetraploid wheat during 1981 - 82.

Item	SS	df	MS	F	P
Total	1103.16	89			
Replications	26.94	9	2.91	<1.00	NS
Treatments	340.76	8	42.60	4.17	***
Methods	200.83	2	100.41	9.83	***
Doses	89.63	2	44.82	4.39	*
Methods X Doses	50.29	4	12.57	1.23	NS
Error	735.49	72	10.22		

Table 17

Number of spikelets per ear (of the main branch) of Carbicron treated and control plants of hexaploid wheat during 1981 - 82.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	10	14	18	12	20	15	13	19	12	13	14.6
SP	D_1	15	13	15	20	13	13	18	15	15	12	14.9
	D_2	14	12	19	15	14	14	13	11	14	11	13.7
	D_o	13	20	16	12	20	15	19	13	19	12	15.9
ST	D_1	19	19	20	17	12	14	18	13	11	11	15.4
	D_2	18	19	13	13	15	14	16	10	20	12	15.0
	D_o	22	21	14	20	17	16	19	21	13	19	18.2
STP	D_1	15	19	14	15	14	16	14	18	15	13	15.3
	D_2	19	18	10	19	17	18	18	17	17	18	17.2

Table 18

Number of spikelets per ear (of the main branch) of Carbicron treated and control plants of tetraploid wheat during 1981-82.

Treatment	Dose	Plants 1	2	3	4	5	6	7	8	9	10	Mean
SP	0	18	18	18	19	18	18	19	18	18	17	18.1
	1	17	18	18	17	18	18	18	18	17	17	17.6
	2	17	19	18	16	17	16	16	19	15	20	17.3
ST	0	19	17	17	18	18	19	20	16	17	18	17.9
	1	18	19	16	17	17	17	17	16	16	17	17.0
	2	16	17	16	15	15	17	16	15	18	17	16.2
STP	0	18	20	21	20	18	20	19	20	17	20	19.3
	1	18	20	19	18	17	17	17	18	17	19	18.0
	2	15	17	19	15	17	15	17	15	16	16	16.2

of spikelets per ear of the treated families were lower than the 'Control' families. The significant 'Treatment' items (Tables 19 & 20) in both the species indicated a statistically significant negative effect of the insecticide on this trait.

(iv) Number of Grains per Ear: The number of grains per ear of hexaploid and tetraploid wheat are given in Tables 21 & 22 respectively. This character also exhibited a wide range of variation in hexaploid wheat. The range for the 'Control' families was 30 to 69 and for the treated families was 30 to 77, whereas, in the tetraploid species these were 28 to 68 and 34 to 63 grains per ear respectively. In all cases, except ST in both the species, the 'Treatment' means were lower than the 'Control' means. The significant 'Treatment' items in both the species (Tables 23 & 24) indicate a significant negative effect of the insecticide on this important yield contributing trait.

(v) 1982-83 Morphological Data: The morphological characters were scored again from the different insecticide treated and 'Control' plants of 1982-83. The data were analysed and results similar to those during the previous year were noted. Therefore, only the results for the two characters, Number of spikelets per ear (of the main branch) and Number of grains per ear (of the main branch) for hexaploid wheat were presented in details. The mean number of spikelets per ear (Table 25) of the treated families were always smaller than the 'Controls.' The range for the control plants was 15 to 21 and for the treated families was 13 to 20. Thus, a negative effect of the insecticide treatment on number of spikelets per ear was indicated. The results of the Analysis of Variance (Table 26)

Table 19

Results of analysis of variance of number of spikelets per ear of Carbicron treated and control plants of hexaploid wheat during 1981 - 82.

Item	SS	df	MS	F	P
Total	841.96	89			
Replications	81.07	9	9.01	1.06	NS
Treatments	149.96	8	18.75	2.21	*
Methods	94.66	2	47.33	5.58	**
Doses	19.49	2	9.74	1.15	NS
Methods X Doses	35.82	4	8.95	1.05	NS
Error	619.93	72	8.49		

Table 20

Results of Analysis of Variance of number of spikelets per ear of Carbicron treated and control plants of tetraploid wheat during 1981-82.

Item	SS	df	MS	F	P
Total	244.49	89			
Replications	21.82	9	2.42	1.19	NS
Treatments	76.89	8	9.61	4.76	***
Methods	10.70	2	5.35	2.65	NS
Doses	52.30	2	26.15	12.95	***
Methods X Doses	13.89	4	3.47	1.72	NS
Error	145.78	72	2.02		

Table 21

Number of grains per ear of Carbicron treated and control plants of hexaploid wheat during 1981-82.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
SP	D_o	30	49	60	33	51	44	43	42	36	44	43.2
	D_1	47	39	38	69	41	39	56	43	43	36	45.1
	D_2	42	31	47	50	28	37	43	30	40	37	38.5
ST	D_o	32	69	42	36	60	51	53	47	63	31	48.4
	D_1	70	73	77	45	41	48	67	41	31	33	52.6
	D_2	54	60	39	45	50	45	43	40	76	41	49.3
STP	D_o	55	67	42	53	59	52	68	54	41	66	55.7
	D_1	56	73	40	46	42	64	52	64	40	37	51.4
	D_2	59	48	40	47	48	50	52	53	49	50	49.6

Table 22

Number of grains per ear (of the main branch) of Carbicron treated and control plants of tetraploid wheat during 1981-82.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	0	51	68	50	66	58	50	56	59	40	62	56.0
SP	1	55	44	55	56	62	50	49	45	49	58	52.3
	2	44	52	43	48	41	55	53	56	54	63	51.4
	0	48	63	56	47	53	42	47	48	41	28	47.3
ST	1	57	49	52	43	48	35	57	55	52	43	49.1
	2	56	57	61	42	60	51	46	57	42	61	48.3
	0	52	46	42	45	37	62	41	46	31	47	48.9
STP	1	50	40	47	51	55	38	47	48	49	50	47.5
	2	37	38	57	44	35	51	47	45	34	41	42.9

Table 23

Results of Analysis of Variance of number of grains per ear of Carbicron treated and control plants of hexaploid wheat during 1981-82.

Item	SS	df	MS	F	P
Total	12198.70	89			
Replications	1303.07	9	144.79	1.19	NS
Treatments	2177.60	8	272.20	2.25	*
Methods	1652.47	2	826.23	6.82	**
Doses	246.60	2	132.30	1.09	NS
Methods X Doses	260.53	4	65.13	<1	NS
Error	8718.03	72	121.08		

Table 24

Results of Abalysis of Variance of number of grains per ear of Carbicron treated and control plants of tetraploid wheat during 1981-82.

Item	SS	df	MS	F	P
Total	5851.79	89			
Replications	420.68	9	46.74	<1	NS
Treatments	1417.89	8	177.24	3.18	*
Methods	1003.02	2	501.51	8.99	*
Doses	2.82	2	1.41	<1	NS
Methods X Doses	412.05	4	103.01	1.85	NS
Error	4013.22	72	55.74		

Table 25

Number of spikelets per ear (of the main branch) of Carbicron treated and control plants of hexaploid wheat during 1982-83.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	19	21	15	15	20	18	21	20	21	19	19.2
ST	D_1	16	15	14	14	17	16	15	16	14	15	15.2
	D_2	15	15	16	17	16	14	16	16	16	14	15.5
	D_o	16	16	18	16	15	18	19	18	16	19	17.1
SP	D_1	14	14	18	16	18	15	20	16	17	17	16.5
	D_2	15	16	16	19	16	15	16	16	18	18	16.5
	D_o	15	16	18	16	16	16	18	17	16	18	16.6
STP	D_1	13	16	16	15	17	15	15	17	15	17	15.6
	D_2	16	17	14	17	16	18	14	14	16	14	15.6

Table 26

Results of Analysis of Variance of the number of spikelets per ear of Carbicron treated and control plants of hexaploid wheat during 1982-83.

Item	SS	df	MS	F	P
Total	281.96	89			
Replications	17.96	9	1.99	< 1	NS
Treatments	119.16	8	14.89	7.10	***
Methods	10.82	2	5.41	2.69	NS
Doses	66.16	2	33.08	16.44	***
Methods X Doses	42.18	4	10.54	5.24	**
Error	144.84	72	2.01		

confirmed this as the 'Treatment' item was highly significant.

The number of grains per ear of the treated and the control plants of hexaploid wheat exhibited a high degree of variation (Table 27). The range for the control plants was 26 to 52, whereas, that for the treated plants was 16 to 42. The mean number of grains per ear was lower in the treated families than in the 'Controls' except for Sp. The results of the Analysis of Variance for this character (Table 28) indicated highly significant 'Treatments item'.

(4) Performance of the Selfed Progeny of the Insecticide Treated Plants

The progeny of the selfed seeds from the Carbicron treated and control plants of 1981-82 were grown in the same field with the other experimental plants during 1982-83. No insecticides were further applied.

(A) Cytological Studies of Hexaploid Wheat: The heads from a random sample of plants of hexaploid wheat were collected at the flag leaf stage and fixed and preserved for cytological studies. No chromosomal contraction and stickiness was observed in the PMCs. The data collected on meiotic abnormalities were summarized in Table 29. The percentage of chromosomal abnormalities in the selfed progeny of the control plants was 3.71 and that of the selfed progeny of the treated plants ranged from 3.94 to 1.56. Complete cytological study was not carried out on tetraploid wheat due to the lack of time but random samples of PMCs from 'Control' and selfed treated plants indicated no difference in the chromosomal abnormalities observed.

Table 28

Number of grains per ear (of the main branch) of Carbicron treated and control plants of hexaploid wheat during 1982-83.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	41	40	27	24	32	26	30	30	37	31	31.8
SP	D_1	42	35	40	27	37	35	27	38	38	38	35.7
	D_2	16	26	37	41	50	28	34	41	30	31	33.4
	D_o	42	34	33	33	38	36	38	34	31	46	36.5
ST	D_1	33	25	42	40	38	37	35	42	28	38	35.8
	D_2	33	38	37	40	34	22	36	32	23	32	32.7
	D_o	50	52	40	50	44	42	45	52	50	42	46.7
STP	D_1	35	42	37	39	34	36	37	40	29	33	36.2
	D_2	37	31	33	31	28	30	33	29	36	28	31.6

Table 28

Results of Analysis of Variance of the number of grains per ear of Carbicron treated and control plants of tetraploid wheat during 1982-83.

Item	SS	df	MS	F	P
Total	4229.60	89			
Replications	203.60	9	22.62	< 1	NS
Treatments	1681.20	8	210.15	6.45	***
Methods	324.47	2	162.24	4.98	***
Doses	502.87	2	251.44	7.72	***
Methods X Doses	853.86	4	213.46	6.55	***
Error	2344.80	72	32.57		

Table 29

Summary of meiotic cells studied for chromosomal aberrations in hexaploid wheat one generation selfed after Carbicron treatments in 1981-82.

Treatment	Division	Cells without aberrations M	A	T	Cells with aberrations M	A	T	Total normal cells	Total abnormal cells	Percentage of abnormal cells
D_o	Meiosis I	319	238	190	19	22	9	1456	54	3.71
	Meiosis II	211	187	311	0	4	0			
STD_1	Meiosis I	405	95	616	24	3	12	1402	52	3.58
	Meiosis II	112	45	129	7	5	1			
STD_2	Meiosis I	215	121	230	10	9	15	952	39	3.94
	Meiosis II	95	73	218	0	2	3			
SPD_1	Meiosis I	331	87	290	7	0	8	1064	20	1.88
	Meiosis II	121	42	193	0	1	4			
SPD_2	Meiosis I	109	210	93	4	8	0	624	16	2.56
	Meiosis II	49	43	120	0	0	4			
$STPD_1$	Meiosis I	215	33	98	4	2	3	543	18	3.31
	Meiosis II	29	29	139	1	3	5			
$STPD_2$	Meiosis I	205	101	39	4	0	0	512	8	1.56
	Meiosis II	21	58	88	0	2	2			

M = Metaphase;

A = Anaphase;

T = Telophase.

(B) Morphological Studies of Hexaploid Wheat: Plant height, number of fertile tillers per plant, number of spikelets per ear and number of grains per ear of the main branch were scored for both the hexaploid and tetraploid species of wheat. The performance for these characters and the results of the Analysis of Variance are discussed below:

(a) Plant Height: The performance for the selfed progeny for plant height showed no definite trend (Table 30). The range for the 'Control' plants was 68 to 101 and that for the treated plants was 71 to 101. There was no significant difference between the 'Control' means and the 'Treatment' means as indicated from the results of the Analysis of Variance (Table 31). All the Mean Squares were non-significant.

(b) Number of Tiller per Plant: The performance of the selfed families for number of tillers per plant is given in Table 32 and the results of the Analysis of Variance in Table 33. The range of the 'Control' plant was 4 to 14 and for treated plants 2 to 13. Though the 'Treatment' means were slightly smaller than the 'Control' means, this difference was found to be statistically non-significant.

(c) Number of Spikelets per Ear: The performance of the selfed families for number of spikelets per ear is given in Table 34 and the results of the Analysis of Variance in Tablé 35. The range for the 'Control' plants was 15 to 21 and for the treated plants 12 to 23. Here again the mean number of spikelets of the treated families were slightly lower than the 'Controls' except in STP selfed. But these differences were statistically non-significant.

Table 30

Plant height at heading (cm.) of the selfed progeny of Carbicron treated and control plants of 1981-82 hexaploid wheat grown during 1982-83.

Treatment	Dose	Plants 1	2	3	4	5	6	7	8	9	10	Mean
	D_o	81	87	86	83	76	87	86	79	68	81	81.4
SP selfed	D_1	90	88	83	81	79	101	90	81	78	71	84.2
	D_2	76	80	81	89	71	70	71	91	81	83	79.3
	D_o	81	85	88	81	71	71	87	75	92	71	80.2
ST selfed	D_1	71	78	79	75	81	83	74	78	80	76	77.5
	D_2	91	83	91	90	81	77	71	70	79	85	81.8
	D_o	100	85	84	86	87	101	100	76	78	78	87.5
STP selfed	D_1	98	91	92	80	81	72	73	91	74	76	82.8
	D_2	89	77	77	80	78	68	81	81	90	83	80.4

Table 31

Results of Analysis of Variance of plant height at heading of the selfed progeny of Carbicron treated and control plants of 1981-82 of hexaploid wheat grown during 1982-83.

Item	SS	df	MS	F	P
Total	5303.66	79			
Replications	576.10	9	64.01	1.14	NS
Treatments	685.36	8	85.67	1.53	NS
Methods	209.16	2	104.58	1.86	NS
Doses	97.69	2	48.85	< 1	NS
Methods X Doses	378.51	4	94.63	1.69	NS
Error	4042.20	72	56.14		

Table 32

Number of tillers (fertile) per plant of the selfed progeny of Carbicron treated and control plants of 1981-82 hexaploid wheat grown during 1982-83.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
SP selfed	D_o	4	5	11	5	9	10	5	7	6	7	6.9
	D_1	11	4	7	7	5	7	6	5	5	8	6.5
	D_2	7	7	4	8	3	6	2	7	4	3	5.1
ST selfed	D_o	7	14	7	7	5	6	8	5	13	6	7.8
	D_1	5	8	5	9	3	7	8	11	5	7	6.8
	D_2	13	4	8	8	7	5	7	7	5	7	7.1
STP selfed	D_o	10	6	8	5	6	10	5	5	8	6	6.9
	D_1	8	4	8	4	5	3	6	7	6	5	5.6
	D_2	11	6	6	3	5	5	7	11	7	6	6.7

Table 33

Results of Analysis of Variance of number of tillers per plant of the selfed progeny of Carbicron treated and control plants of 1981-82 of hexaploid wheat grown during 1982-83.

Item	SS	df	MS	F	P
Total	493.60	89			
Replications	57.82	9	6.42	1.20	NS
Treatments	51.80	8	6.48	1.21	NS
Methods	18.87	2	9.43	1.77	NS
Doses	16.20	2	8.10	1.52	NS
Methods X Doses	16.73	4	4.18	< 1	NS
Error	383.98	72	5.33		

Table 34

Number of spikelets per ear (of the main branch) of the selfed progeny of Carbicron treated and control plants of 1981-82 hexaploid wheat grown during 1982-83.

Treatment	Dose	Plants 1	2	3	4	5	6	7	8	9	10	Mean
SP selfed	D_o	20	20	17	15	18	17	20	20	16	17	18.0
	D_1	16	14	20	12	17	15	16	14	17	18	15.9
	D_2	17	16	15	17	18	15	17	16	18	18	16.7
ST selfed	D_o	20	16	19	15	18	21	20	18	20	18	18.5
	D_1	16	18	19	19	18	18	19	18	18	15	17.5
	D_2	16	14	19	16	18	23	18	15	14	15	16.8
STP selfed	D_o	20	20	17	15	18	17	20	20	16	17	18.0
	D_1	22	18	21	18	17	17	17	18	13	19	18.0
	D_2	19	16	14	20	17	19	20	20	18	19	18.2

Table 35

Results of Analysis of Variance of the number of spikelets per ear of the selfed progeny of Carbicron treated and control plants of 1981-82 of hexaploid wheat grown during 1982-83.

Item	SS	df	MS	F	P
Total	390.32	89			
Replications	44.10	9	4.90	1.23	NS
Treatments	60.02	8	7.50	1.88	NS
Methods	22.69	2	11.34	2.85	NS
Doses	17.42	2	8.71	2.19	NS
Methods X Doses	19.91	4	4.98	1.22	NS
Error	286.20	72	3.98		

(d) Number of Grains per Ear: The performance of the selfed progeny for this character is given in Table 36 and the results of the Analysis of Variance in Table 37. The range for this character was 30 to 55 for 'Control' plants and 23 to 58 for treated plants.

Though the mean performance for the number of grains per ear within each treatment showed no definite trend, the 'Treatment' means were different. The performance of the SP-selfed and STP-selfed families were often lower than ST-selfed families, but the average number of grains in the 'Control' and treated families were same (40.9 vs. 40.7). The 'Treatment' item was just significant (Table 37).

(C) Morphological Studies of Tetraploid Wheat:

(a) Plant Height: The performance of the selfed progeny for plant height (Table 38) and the results of Analysis of Variance (Table 39) indicated a significant reduction in plant height in the selfed progeny of treated plants in comparison to the selfed progeny of the 'Control' plants of 1981-82. The height of the 'Control' plants exhibited a range of 88 to 105 cm., whereas, that of the treated and selfed progeny was 77 to 101.

(b) Number of Fertile Tillers per Plant: The number of fertile tillers per plant for the selfed progeny are summarized in Table 40. The range for the selfed progeny of the 'Control' plants was 4 to 19, whereas, that for the selfed progeny of the treated plants was 4 to 13. The results of the Analysis of Variance (Table 41) indicated a significant treatment effect.

Table 36

Number of grains per ear (of the main branch) of the selfed progeny of Carbicron treated and control plants of 1981-82 hexaploid wheat grown during 1982-83.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	43	40	42	35	39	40	33	42	38	35	38.7
SP selfed	D_1	45	39	23	46	37	32	42	33	44	40	38.1
	D_2	48	45	40	40	42	48	36	41	37	38	41.5
	D_o	44	39	54	30	49	38	55	50	39	46	44.4
ST selfed	D_1	48	43	47	58	38	46	47	42	40	41	45.0
	D_2	55	43	48	34	35	39	39	48	42	30	41.3
	D_o	34	39	41	34	30	45	34	50	40	50	39.7
STP selfed	D_1	39	49	57	37	44	52	36	36	40	44	43.4
	D_2	42	30	39	36	31	30	39	32	28	42	34.9

Table 37

Results of Analysis of Variance of the number of grains per ear of the selfed progeny of Carbicron treated and control plants of 1981-82 of hexaploid wheat grown during 1982-83.

Item	SS	df	MS	F	P
Total	4113.56	89			
Replications	294.89	9	32.77	< 1	NS
Treatments	858.16	8	107.27	2.61	5%
Methods	350.16	2	175.08	4.26	5%
Doses	130.16	2	65.08	1.58	NS
Methods X Doses	377.84	4	94.46	2.29	NS
Error	2960.57	72	41.12		

Table 38

Plant height at heading (cm.) of the selfed progeny of Carbicron treated and control plants of 1981-82 tetraploid wheat grown during 1982-83.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	97	89	95	95	91	99	95	97	89	94	94.1
SP selfed	D_1	93	91	88	90	87	96	94	92	90	91	91.2
	D_2	91	90	89	81	89	90	91	90	91	89	89.1
	D_o	95	93	102	88	88	89	92	93	95	95	93.0
ST selfed	D_1	98	92	89	95	93	90	85	95	90	90	91.7
	D_2	84	79	91	86	90	81	84	81	89	81	84.6
	D_o	88	100	97	98	105	97	97	103	102	102	98.9
STP selfed	D_1	91	90	80	100	83	101	87	95	88	98	91.3
	D_2	88	85	90	94	87	77	83	88	89	96	87.7

Table 39

Results of Analysis of Variance for plant height at heading of the selfed progeny of Carbicron treated and control plants of 1981-82 of tetraploid wheat grown during 1982-83.

Item	SS	df	MS	F	P
Total	2930.49	89			
Replications	91.60	9	10.10	<1	NS
Treatments	1313.49	8	164.19	7.75	***
Methods	124.69	2	62.34	2.94	NS
Doses	1009.16	2	504.58	23.82	***
Methods X Doses	179.64	4	44.91	2.12	NS
Error	1525.40	72	21.18		

Table 40

Number of fertile tillers per plant of the selfed progeny of Carbicron treated and control plants of 1981-82 tetraploid wheat grown during 1982-83.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	14	6	6	15	10	16	17	4	8	9	10.5
SP selfed	D_1	7	13	7	4	9	5	4	10	9	7	7.5
	D_2	12	7	10	10	7	6	8	7	6	7	8.0
	D_o	6	8	15	6	9	7	8	10	7	8	8.4
ST selfed	D_1	7	12	6	7	11	9	6	7	5	4	7.4
	D_2	5	11	10	6	9	6	4	7	6	10	7.4
	D_o	10	11	8	15	7	12	13	8	13	19	11.6
STP selfed	D_1	11	9	6	9	7	6	9	8	9	11	8.5
	D_2	7	10	6	9	7	6	6	9	6	7	7.3

Table 41

Results of Analysis of Variance for number of fertile tillers per plant of the selfed progeny of Carbicron treated and control plants of 1981-82 of tetraploid wheat grown during 1982-83.

Item	SS	df	MS	F	P
Total	834.49	89			
Replications	30.49	9	3.39	< 1	NS
Treatments	187.29	8	23.41	2.73	*
Methods	30.49	2	15.25	1.78	NS
Doses	124.16	2	62.08	7.25	**
Methods X Doses	32.64	4	8.16	<1	NS
Error	616.71	72	8.56		

(c) Number of Spikelet per Ear: The number of spikelets per ear of the selfed progeny are given in Table 42. The range of the selfed 'Control' plants was 13 to 19, whereas, that of the selfed progeny of the treated plants was 12 to 17. The results of the Analysis of Variance (Table 43) indicated no significant difference among the various progeny families.

(d) Number of Grains per Ear: The performance of the selfed 'Control' and treated plants are shown in Table 44. The range of the selfed 'Control' plants was 20 to 41, whereas, that of the selfed treated plants was 16 to 42. The results of the Analysis of Variance (Table 45) indicated no significant differences.

(5) Correlation Analysis on Grain Data for 1981-82

In cereals, the number of spikelets and number of grains are highly correlated characters. The highly significant correlation observed between these two yield components (Table 46) reflected this association.

A comparison between the correlation coefficients between these two characters in untreated and treated plants was made. If insecticide treatments cause a high proportion of chromosomal aberrations and if these aberrations result in a reduction in the number of grains, the association between these two traits should be affected. However, the results in Table 46 indicate that there is no noticeable effect of insecticide treatment on the observed correlation coefficient.

Table 42

Number of spikelets per ear (of the main branch) of the selfed progeny of Carbicron treated and control plants of 1981-82 tetraploid wheat grown in 1982-83.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	19	18	16	15	17	15	15	14	15	16	16.0
SP selfed	D_1	15	14	15	14	17	15	14	14	16	17	15.1
	D_2	15	15	15	15	14	15	16	14	15	16	15.0
	D_o	14	16	13	14	15	16	16	14	16	15	14.9
ST selfed	D_1	15	15	14	16	15	14	17	15	15	16	15.2
	D_2	13	15	12	13	16	13	15	14	13	15	13.9
	D_o	15	14	16	15	16	14	14	15	15	16	15.0
STP selfed	D_1	14	14	15	14	16	15	14	14	15	14	15.5
	D_2	16	14	14	15	15	14	14	16	15	14	14.7

Table 43

Results of Analysis of Variance for number of spikelets per ear of the selfed progeny of Carbicron treated and control plants of 1981-82 of tetraploid wheat grown in 1982-83.

Item	SS	df	MS	F	P
Total	214.90	89			
Replications	24.68	9	2.74	1.20	NS
Treatments	26.00	8	3.25	1.42	NS
Methods	7.40	2	3.70	1.62	NS
Doses	11.27	2	5.62	2.47	NS
Methods X Doses	7.33	4	1.83	<1	NS
Error	164.23	72	2.28		

Table 44

Number of grains per ear (of the main branch) of the selfed progeny of Carbicron treated and control plants of 1981-82 tetraploid wheat grown in 1982-83.

		Plants										
Treatment	Dose	1	2	3	4	5	6	7	8	9	10	Mean
	D_o	30	31	36	20	33	26	32	33	33	30	30.4
SP selfed	D_1	42	35	40	27	37	35	27	38	38	38	35.7
	D_2	16	26	37	41	50	28	34	41	30	31	33.4
	D_o	41	40	27	24	32	26	30	30	37	31	31.8
ST selfed	D_1	33	25	42	40	38	37	35	42	28	38	35.8
	D_2	33	38	37	40	34	22	36	32	23	32	32.7
	D_o	31	38	28	30	23	38	39	40	34	32	33.3
STP selfed	D_1	35	42	37	39	34	36	37	40	29	33	36.2
	D_2	37	31	33	31	28	30	33	29	36	28	31.6

Table 45

Results of Analysis of Variance for the number of grains per ear of the selfed progeny of Carbicron treated and control plants of 1981-82 tetraploid wheat grown during 1982-83.

Item	SS	df	MS	F	P
Total	2966.10	89			
Replications	197.44	9	21.94	<1	NS
Treatments	341.80	8	42.73	1.27	NS
Methods	4.27	2	2.14	<1	NS
Doses	281.87	2	140.94	4.18	*
Methods X Doses	55.66	4	13.92	<1	NS
Error	2426.86	72	33.71		

Table 46

Results of correlation and regression analysis between number of spikelets per ear with number of grains per ear in untreated (D_o) and treated (D_1) plants of hexaploid wheat during 1981-82.

Species	Correlation coefficient(r) between spikelet and grains.			
	Untreated (D_o)		Treated (D_1)	
	r	P	r	P
Hexaploid	0.789	***	0.909	***

Species	Regression coefficient(b) for grains on spikelet.					
	Untreated (D_o)			Treated (D_1)		
	b	t	P	b	t	P
Hexaploid	2.664	6.796	1%	4.674	11.54	1%

DISCUSSION

(I) Cytological Action of the Insecticide:

The cytological examinations made during 1981-82 on the insecticide treated materials were repeated again in 1982-83; the results obtained in both the years indicated a significant increase in the number of PMCs with meiotic abnormalities over the controls. Similar increase in chromosomal abnormalities in PMCs of the insecticide treated families was also reported by Alam et al. (1981a, 1981b) and Islam (1983) in wheat and barley and by Soliman and Al-Najjar (1980) and Al-Najjar and Soliman (1982) in wheat and two other related species.

Two points emerge from the results of the cytological study: (a) insecticide treatment results in chromosome stickiness and contraction and (b) the abnormalities were more common in early stages of meiosis. The percentage of normal PMCs belonging to Meiosis I in 1981-82 was 62.34 but percentage of PMCs in Meiosis I with aberrations was 83.33. Similar results were also obtained during 1982-83. These indicate that stickiness of chromosomes due to insecticide treatment might have given rise to these abnormalities. Though in majority of the cases of the treated PMCs the chromosomes did separate, in some PMCs the chromosomes due to stickiness failed to undergo normal course of meiotic events. That is why abnormalities were more common in early stages of the cell division. 12.2% abnormal cells belonging to Division I against 4% in Division II were observed in 1981-82 data. Moreover, the majority of abnormalities were lagging chromosomes and chromosome bridges which are often found as a consequence of chromosome stickiness.

Studies of the cytological effects of various mutagens and antibiotics by various workers also reported the formation of different chromosomal abnormalities (Wilson et al., 1950, 1951a, 1951b; Unrau, 1953, 1954; Unrau and Larter, 1952; Wuu and Grant, 1967a, 1967b; Amer, 1965). Tanaka and Sato (1952) concluded that any morphological abnormalities induced either by the action of ionizing radiations or insecticides are superficially alike and cytological disruption is the most fundamental response. As in their experiment with Tradescantia paludosa, the result of the present experiment revealed a wide range of cytological disruption, i.e., contraction of chromosomes and stickiness, fragmentation and lagging of chromosomes, bridge formation, production of micronuclei, etc.

There was little difference in the response of the tetraploid and the hexaploid wheat to the insecticide treatment. Though the percentages of abnormal PMCs were always lower in the 4x species for all treatments than the 6x species, the treatments significantly increased chromosomal aberrations as well.

Although no consistent pattern emerged between the different treatments and the frequency of chromosomal aberrations, plants with chromosomal aberrations occurred in all Carbicron - treated families indicating that the chemical interferred with the meiotic stability regardless of the type of treatment (i.e., seed treatment and/or spray).

It is especially interesting that the seeds treated before germination also showed irregularities in PMCs. Since both normal and abnormal PMCs were observed in the same flower-bud, it is likely that some chemical residue persisted in the plant tissue and caused these aberrations. Because, if the abnormalities were induced immediately after the application of the chemical, an entire inflorescence or a part of the panicle should have been affected similarly. As this was not noticed in the present study, the insecticide apparently remained active several weeks after application and remained in the cells of the embryos, seedlings and in the germ cells —— becoming active in the PMCs prior to or during meiosis. Similar effect of a herbicide, Atrazine, was reported by Liang et al. (1967) on Sorghum.

Although the mechanisms responsible for observed chromosomal abnormalities are not yet clear, chromosomal breakage could be similar to damage caused by ionizing radiation (Doxey, 1949; Unrau and Larter, 1952). Chemical application may affect genetic process of meiocytes, thus, resulting in various aberrations. Numerous biochemical process occuring in a cell may be inhibited, enhanced or altered by a nasty chemical like Carbicron, thus, upset the cell division cycle. Wuu and Grant (1966) suggested that these changes may be alteration in the activities of genes or gene products rather than gene mutations.

(II) Morphological Effects of the Insecticide:

In a large number of the experiments carried out on mutagenic action of chemicals on plants, cytological disruption were accompanied by an induction of gene mutation. The array of the cytological disruptions observed in the present study suggested that the insecticide used had enough potential to induce the rearrangements of genes. Therefore, it seems more likely that changes in the morphological characters would result as a consequence of the treatment.

The four morphological characters plant height, number of tillers, number of spikelets and number of grains studied all indicated a negative effect of the insecticide. These characters in wheat and barley are known to be controlled by polygenes (Fonseca and Patterson, 1968; Carnnell, 1969; Grafius and Okoli, 1974; Rasmusson and Cannell, 1970). Many workers have reported yield reduction and crop injury from misapplication of herbicides (Bingham and Porter, 1961; Drake et al., 1963; Everson and Arle, 1956; Holstun and Bingham, 1960; Porter et al., 1959; Wiese et al., 1964). Johnson (1961) showed that dicryl N - (3, 4-dichlorophenyl)-methyl - acrylamilide delayed maturity in cotton, which could influence fibre quality. Everson and Arle (1956) found that pre-emergence application of Monuron 3-(P-chlorophenyl)-1, 1-dimethylurea at high doses reduced ball weight, fibre length, and fibre coarseness. Scifres and Santelmann (1966) also found that Paraquat (1, 1-dimethyl-4, 4 - dipyridinum cation) did have influence on cotton fibre quality. Santelmann et al. (1966) also detected effect of two herbicides, Prometryne (2, 4-bis(isopropyl amino) - 6 - methyl mercapto - S - triazine) and DSMA (Dismodium Methane Arsonate) on fibre strength of cotton; both of these reduced fibre strength during one year but not in the following year. In the present study, however, the negative effects of the insecticide treatments on the quantitative characters were observed for both 1981-82 and 1982-83. This suggests a significant influence of the insecticide on these characters.

The significant decrease in the three yield components i.e., number of fertile tillers, number of spikelets per ear and number of grains per ear, due to the insecticide treatment is important. This reduction in the mean performance was more spectacular in Dose 2 than in Dose 1. In presence of this insecticide phytoxicity, the standard dose should be determined in the light of the observed results.

(III) Persistence of the Effects of the Insecticide to the Next Generation:

Transmittable changes in plants induced by chemical insecticides may be caused by an alteration of genetic material. It has already been mentioned in the introduction that effect of certain chemical herbicides could be transmitted via seeds (Dunlap, 1951). Plant abnormalities induced by another chemical, Dalapon, were found in several generations after the herbicides was applied in barley (Suneson, 1960). However, Liang et al. (1967) suggested that the chemical residue in soil from previous treatments might have caused crop injury in certain of these cases.

The selfed seeds of the insecticide treated families were sown in different field in 1982-83. However, no significant difference was observed between the selfed progeny of the treated and the non-treated families in respect to the chromosomal aberration in PMCs. Statistical analysis of the morphological and yield characters indicated that the selfed progeny of the treated families did not differ from the control groups for both 6x and 4x wheat. Similar results were obtained by Liang et al. (1967) in Sorghum.

It is questionable whether tetrads or meiocytes containing chromosomal aberrations are actually transmitted to subsequent generations. A large number of second generation plants from seeds of the insecticide treated plants were screened but no significant difference for chromosome abnormalities or other morphological characters were found. Non-viability of the pollen grains containing chromosomal aberrations can be put forward to explain the non-transmission of the abnormalities. Even if, the pollen grains with small aberrations would be vialble, these would be less compegative than the normal pollen tubes during fertilization.

Though the quantitative characters were affected by Carbicron treatment, these effects were not inherited in the next generation plant. This was also perhaps due to non-viability or non-competative pollen grains containing the aberrations.

Experimental evidence and information concerning the response of the crop plants to insecticides is required to ensure their safe and economic use. Data presented here indicate that before the plants response to the chemicals can be fully determined, information must be obtained concerning the cytogenetic action of the chemicals and their persistent effect in plants.

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