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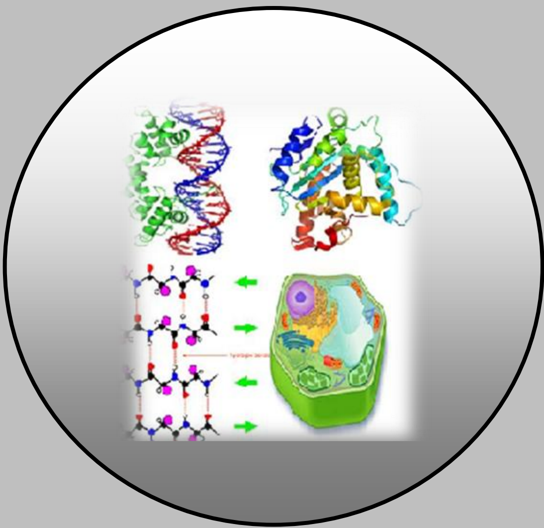

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RESEARCH PAPER

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Diploidization and Fertility Improvement in Highly Advanced Varieties of Autotetraploid Fenugreek

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ABSTRACT

Meiotic analysis of four autotetraploid varieties (genotypes) of autotetraploid fenugreek (Trigonella foenum-graecum) over succeeding generations (C_8, C_{12} and C_{16} generations) indicated that there is gradual decrease in multivalents and univalents with simultaneous increase in bivalents; thus revealing diploidization process. Though autotetraploid Trigonella are vegetatively vigorous and meiotically regular after 16 years selection, the seed yield is still near 50% to their diploid progenitors. Therefore, it may be concluded that poor fertility in autotetraploid T. foenum-graecum is controlled by certain genetical factors which could only be handled by genetic manipulations.
Key words: Trigonella foenum-graecum, Genotypes, Autotetraploid, Diploidization and Meiotic Associations.

INTRODUCTION

Induction of polyploidy by colchicine has been successfully carried out in several ornamentals, grasses, vegetatively propagated plants and also in many legumes Jail et.al. (1974), Rao et al. (1982). The literature of past few decades clearly indicated that cytomorphogenetical studies have been carried out in early generations of induced polyploids and possibility of evolving more vigorous, high yielding varieties has been interpreted. Detailed studies on cytomorphological behavior of very highly evolved and stablilized polyploids are almost lacking. Present investigations have been carried out on four advanced autotetraploid varieties of *Trigonella foenum-graecum* to explore the possibility of evolving and stabilizing more vigorous and high yielding variety of autotetraploids and impact of judicious selections over sixteen generations on vigour, fertility and meiotic behaviour.

MATERIAL AND METHODS

C_8, C_{12} and C_{16} progenies of four autotetraploid varieties or genotypes (Sel. 1, Sel. 2 Sel. 3 & Sel. 4) of *Trigonella foenum-gracecum* which had already been raised in our laboratory, were grown under uniform agroclimatic conditions along with their diploid progenitors over different years. The cytomorphological data were collected during their respective generation. Acetocarmine squash preparations were made to study meiosis. Cytomorphology was studied from 20 randomly selected plants of different genotypes at both ploidy levels and selection for next generation was made accordingly. 400 pmcs were analysed randomly at metaphase 1 and 50 pmcs at anaphase 1 in each genotype in C_8, C_{12} and $_{16}$ generations. Similarly 500 pollen from each plant were scored for pollen fertility in each generation. Cytomorphological data thus obtained was subjected to statistical analysis to calculate actual mean ± standard error values.

RESULTS

Actual mean ± standard error values for chiasma frequency, quadrivalents, trivalents, bivalents and univalents frequencies of different varieties (Sel. 1, Sel.2, Sel.3 and Sel. 4) of *T.foenum-graecum* in C_8, C_{12} and C_{16} generations have been presented in Table 1.

A comparison of mean values of bivalents and pollen fertility between C_8, C_{12} and C_{16} indicated that bivalent differed significantly among all comparison; pollen fertility differed only between C_{12}-C_{16}, C_8-C_{16} but not between C_8-C_{12}, Sel. 3 is an exception where C_{12}-C_{16} also do not differed. Thus in Sel. 3 meiotic stabilization is much faster than rest of the three varieties

Table 1. Actual mean ± standard error for chiasma frequency and different chromosomal associations in C_8, C_{12} and C_{16} of different genotypes of autotetraploid *T. foenum- graecum*.

Varieties and Generations	Chiasma frequency Per Chromosome A.M. ± S.E.		Range and Frequency/ PMC							
			Quadrivalent IV Range A.M. ± S.E.		Trivalents (III) Range A.M. ± S.E.		Bivalents (II) Range A.M. ± S.E.		Univalents (I) Range A.M.± S.E.	
	C_8	0.80±.02	0-3	0.69±.02	0-1	0.20±.02	8-10	13.65±.08	0-4	0.80±.06
Sel. 1	C_{12}	0.78±.01	0-2	0.50±.03	0-1	0.05±.01	9-16	14.26±.14	0-2	0.75±.07
	C_{16}	0.66±.01	0-2	0.15±.01	0-1	0.02±.01	12-16	15.52±.06	0-2	0.23±.03
	C_8	0.80±.01	0-3	1.20±.03	0-2	0.18±02	6-12	73.08±06	0-4	1.26±.06
	C_{12}	0.78±.01	0-3	0.70±.05	0-2	0.13±.05	9-14	13.55±.14	0-4	0.97±.06
Sel. 2	C_{16}	0.68±.01	0-2	0.07±.02	0-1	0.04±.01	12-16	15.21±.06	0-2	0.47±.06
	C_8	0.86± .01	0-3	1.08±.04	0-1	0.12±.03	9-13	13.62±.13	0-4	0.93±.15
	C_{12}	0.85± .01	0-2	0.41±.03	0-1	0.03±.01	9-16	14.33±.18	0-2	0.73±.03
	C_{16}	0.67± .01	0-1	0.18±.02	0-1	0.05±.01	12-16	15.13±.06	0-2	0.47±.08
Sel.3	C_8	0.76±.02	0-2	0.85±.04	0-1	0.14±.04	8-13	13.46±.29	0-4	1.07±.11
	C_{12}	0.74±.01	0-2	0.29±.02	0-1	0.09±.01	9-15	14.68±.13	0-2	0.44±.05
Sel.4	C_{16}	0.65±.01	0-2	0.25±.02	0-1	0.05±.01	12-16	15.03±.09	0-2	0.54±.07

Percentage of equal chromosomal disjunction at anaphase I under succeeding generations have been presented in Table 2. Values showed increased percentage of equal separation with advancement of generations. Pollen fertility percentage has also been presented in the same table. The values showed continuous increase from C_8 to C_{16} generations in all the four genotypes. Morphological paramenters of different genotypes in C_8 and C_{12} and C_{16} generations have been presented in Table 3. Here the data has been presented in terms of percent value of their respective diploid progenitors which gives an idea of percent increase or decrease over diploids. Data shows continuous improvement in vegetative traits and seed fertility also, but after 16 year's selection the seed yield in different genotypes is still near 50% to their diploid progenitors.

Table 2. Percentage of equal chromosomal disjunction at anaphase I and pollen fertility in different varieties of autotetraploid *T. foenum-graecum* under C_8, C_{12} and C_{16} generations.

Verities and generations		% of equal disjunction Actual mean ± standard error	% of pollen fertility Actual mean ± standard error
Sel. 1	C_8	-	81.00±1.31
	C_{12}	58.34±2.43	83.50 ± 0.65
	C_{16}	70.55± 1.94	84.18±1.23
Sel. 2	C_8	-	61.75±1.18
	C_{12}	52.38±2.06	69.25±0.48
	C_{16}	61.37±0.94	77.59±1.69
Sel.3	C_8	-	79.60±1.89
	C_{12}	59.26±1.99	82.32±0.80
	C_{16}	74.84±0.88	82.98±1.26
Sel.4	C_8	-	70.50±1.66
	C_{12}	50.14 ± 3.21	73.22±0.79
	C_{16}	58.26±1.64	78.34±1.12

Table 3. Percent diploid value of fertility parameters of different genotypes of 4x *Trigonella* under succeeding generations.

Parameters	Verities	C_8	C_{12}	C_{16}
Plant height (cm)	Sel.1	136.58	135.29	139.03
	Sel.2	167.82	115.88	127.79
	Sel.3	119.24	127.65	128.27
	Sel.4	117.38	118.55	123.74
Number of branches	Sel.1	68.08	74.42	129.59
	Sel.2	82.66	93.26	135.48
	Sel.3	58.30	60.36	113.18
	Sel.4	54.78	56.93	78.53
Number of pods/plant	Sel.1	65.82	67.85	79.22
	Sel.2	61.76	60.13	74.68
	Sel.3	72.53	75.39	83.23
	Sel.4	60.00	65.48	70.99
Number of seeds/pod	Sel.1	32.75	32.95	33.16
	Sel.2	28.13	30.32	36.79
	Sel.3	34.01	35.14	38.11
	Sel.4	27.47	27.42	28.95
Seed yield/ plant (gm)	Sel.1	-	36.78	37.37
	Sel.2	-	21.67	32.63
	Sel.3	-	40.37	54.88
	Sel.4	-	20.14	31.32

Figure 1. Metaphase I of PMC of diploid *Trigonella*
Figure 2. Metaphase I of PMC of autotetraploid *Trigonella*
Figure 3. Metaphase I of MMC of autotetraploid *Trigonella*

DISCUSSION

A comparative study concerning meiotic behaviour and its correlations with seed fertility under succeeding generations of autotetraploid *T. foenum-graecum* revealed that selection for improved seed fertility rather than vigour and vegetative traits of plants has resulted in progressive improvement of meiotic stability. All the four genotypes followed same pattern of meiotic stabilization and even after 16 year's selection the seed fertility is still near 50% to their diploid progenitors. A survey of literature on autoploids of past 30 years reveled that cytomorphological studies were carried out only in few earlier generations where there is rapid rate of meiotic stabilization and fertility improvement. Data on very highly evolved and stabilized autoploids are however lacking. Our cytomorphological studies on advanced autotetraploid *T. foenum-graecum* indicated that though there is continuous improvement in cytomorpholocial traits but a plateau is formed regarding seed fertility beyond which there is not any marked improvement. Therefore, it may be assigned to certain factors responsible for poor fertility in autotetraploids which is a major drawback for their commercial exploitation as a improved variety. Reports are available regarding genetical control on fertility Hazarica et.al. (1967), Crowley et.al.(1968). Many workers are of the opinion that poor fertility in autotetraploids can be mainly attributed to genetical-physiological disbalance of unexplained nature which are yet to be explored. It may be concluded that poor fertility in autotetraploids cannot be handled by conventional selection methods but certain genetical manipulations are necessary. Keeping it in mind variability in the germ plasm of autotetraploid *T. foenum-graecum* it was exposed to hybridization and mutation. As a result of which seed fertility had reached up to the diploid level in vigorous autotetraploid *Trigonella foenum-graecum* (Raghuvanshi et.al. 1989).

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