

Gamma Radiation Induced Cytomorphological Alternations in *Hyoscyamus muticus* L

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ABSTRACT

The present investigation the effect of Gamma Radiation Induced cytological behaviour during meiosis in Hyoscyamus muticus. Four plants (mutants), namely 2010, 2015-1, 2029-5 and 2062 in S₇ and the one 4046 in S₉ which manifested abnormal chromosomal behaviour during meiosis. At phenotypic level, 2015-1 and 2029-5 were having fringed corolla, while 2010 was a plant with stout buds and twisted (curled) stem and 2062 had persistent style. The mutant 4046 (S₉) was very poor in vigour.

Keywords: Cytomorphological Alternations, Gamma Radiation, Desynapsis

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INTRODUCTION

Hyoscyamus muticus-the Egyptian Henbane is one of the most important medicinal herbs naturally found and produced in Egypt and is a valuable source of alkaloids viz., hyoscyamine, and atropine. Mutation breeding employing gamma radiation was initiated to explore its potential for genetic upgradation, as well as, to elucidate the feasibility of developing translocation tester and multiple interchange stock [1, 2]. The present study deals with the isolation of five induced macromutants, and their cytological behaviour during meiosis.

MATERIAL AND METHODS

Five genotypes, irradiated with 4 doses of gamma rays : $d_1 = 20$ kR, $d_2 = 40$ kR, $d_3 = 60$ kR and $d_4 = 80$ kR in 4000 A Gamma Chamber at CIMAP, Lucknow. The irradiated seeds were grown to raise M1, and subsequently M2 to isolate desirable mutants. In order to determine the nature of chromosomal aberrations due to irradiation, meiotic analysis of selected mutants was performed from their floral buds collected between 0900-1000 hr. The buds were fixed overnight in Carnoy's fixative (6:3:1) and then examined for meiotic associations and anaphase, disjunction following acetocarmine squash technique.

RESULTS AND DISCUSSION

Some of the striking phenodeviants among 51 mutants subjected to cytological analysis for determining the fact whether these phenotypic abnormalities were associated with chromosomal structural changes or with point mutation. Majority of them manifested normal meiotic behaviour with occasional disturbances. However, there were four plants (mutants), namely 2010, 2015-1, 2029-5 and 2062 in S₇ and the one 4046 in S₉ which manifested abnormal chromosomal behaviour during meiosis. At phenotypic level, 2015-1 and 2029-5 were having fringed corolla, while 2010 was a plant with stout buds and twisted (curled) stem and 2062 had persistent style. The mutant 4046 (S₉) was very poor in vigour. Various types of morphological and cytological features of these mutants are summarised in Table 1:

Table 1. Morphological, meiotic features and pollen/seed fertility in five cytological mutant plants.

Sl. No.	Genotype name	Mutant number	Morphological features	Meiotic features	Pollen/seed fertility
1.	S ₇	2010	Curled leaves, stout buds, twisted stem and poor vigour	Desynapsis	75-80% Pollen fertility 50-55% seed fertility
2.	S ₇	2015-1	Dwarf, corolla fringed, poor vigour	Chromosomal stickiness, translocation complex involving 4 bivalent at metaphase	40-45% pollen fertility 15-20% seed fertility
3.	S ₇	2029-5	Dwarf, corolla fringed, poor vigour	Hexavalent translocation chain and multiple interchange involving 6 and 8 bivalent	35-40% pollen fertility 10-12% seed fertility
4.	S ₇	2062	Dwarf, style persistent	Chain and/or interlock quadrivalents	25-30% pollen fertility 5-10% seed fertility
5.	S ₉	4046	Poor vigour, dwarf	Disturbed metaphase orientation leading extrusion of bivalents	60-65% pollen fertility 40-45% seed fertility

H. muticus consists of $2n = 28$ chromosomes which manifest regular anaphase disjunction leading to formation of functional gametes. Although the formation of 14 ring bivalents with distal chiasma is usual feature of this species, a good number of cells (20 to 30%) in the normal untreated material often shows 1-2 pairs of uni- and/or rod bivalents as well. Majority of phenodeviants listed in Table 17 exhibited, by and large, a similar normal meiotic behaviour in their pollen mother cells, thus indicating genic changes rather than chromosomal changes. However, the five mutants mentioned above (2010, 2015-1, 2029-5 and 2062 in S₇ and 4046 in S₉) did exhibit variation in meiotic behaviour of their cells as well as chromosomal structural changes.

The pollen mother cells of mutant No. 2015-1 of S₇ revealed the chromosomes with no sharp ends : there was some sort of attachment among many bivalents. This seems to be due to stickiness of chromosomes at metaphase. This was most prevalent at 20 KR dose of gamma irradiation [3].

In a mutant No. 2010 of S₇ the pollen mother cells exhibited 28 univalents at metaphase in a large number of cells. Apparently, there was either the failure of pairing at zygotene (asynapsis) or the bivalents failed to maintain their pairity, consequently leading to desynaptic state. This again happened most frequently at 20 KR dose. At morphological level, this mutant possessed curled leaves, stout buds and twisted stem coupled with poor vigour.

A phenodeviant No. 4046 (at 40 KR) of S₉ was having poor vigour and considerable degree of sterility. The meiotic analysis of pollen mother cells of this mutant revealed disturbed metaphase orientation and two types of abnormal separation at anaphase in variable frequencies. A number of pollen mother cells in this mutant showed metaphase where certain bivalents (3II) were extruded from their equatorial plate. Some meiocytes manifested precocious anaphase disjunction with lagging chromosome, while some showed multipolar separation leading to the formation of multiple micronuclei at telophase [5].

Three mutant plants in S all at 20 KR dose (viz. No. 2062, 2029-5 and 2015-1) were cytologically interesting as they showed the occurrence of single as well as complex translocations in a number of meiocytes. The plant number 2062 manifested one chain quadrivalent in a few cells, and two interlock quadrivalents and one chain quadrivalent in some other cells. In both cases the frequency was high at diakinesis. On the other hand, the plant No. 2029-5 revealed a hexavalent translocation chain and a multiple interchange involving 6 and 8 bivalents, respectively in different pollen mother cells at metaphase. Similarly, the mutant plant No. 2015-1 also manifested a translocation complex involving four bivalents at metaphase. All these plants were poor in vigour and partially to completely sterile.

Abnormalities in meiotic behavior of five mutants chosen specially for this study were concerned with chromosomal stickiness, desynapsis, abnormal metaphase orientation coupled with disturbed anaphase disjunction and single or multiple translocation chains.

The heterochromatic segments of the chromosomes possess the property of chromosome stickiness [7], a tendency which could be further manifested by gene-environment interactions. Such characteristics have been found mainly to be governed by double recessive genes [8], which in the instant case may have arisen as a consequence of mutation.

The tendency of the formation of a few univalent and/or rod bivalents observed in the control material is indicative of the fact that the distal chiasma localization attendant in the bivalents are of weak nature which may be under genetic control. Even a micromutation for this character may lead to disruption of pairing system resulting into desynaptic mutation [6].

In the present investigation only the symmetrical reciprocal chromosome translocations have been considered which are also known as interchanges. At the meiotic metaphase I, a translocation heterozygote is characterized by quadrivalent formation. The two translocated chromosomes and the two normal chromosomes constituting the quadrivalent may be associated in ring or chain, subject to the number and position of chiasmata [4]. Similarly, the reciprocal translocations involving more than 2 pairs of chromosomes may form multiple associations of chromosomes relative to the number of chromosome pairs involved. Also the magnitude of the multiple interchange may be enlarged by synthesis of complex interchanges by intercrossing between different interchange stocks.

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