

Assessment of Genotoxic Effects of *Nux Vomica* as Homeopathic Drug on Mitotic Chromosomes of *Vicia Faba*

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Extract of *Nux vomica* is used as homeopathic drug to cure various nervous disorders like mental emotion epilepsy, prolepses of the rectum, hydrophobia etc. The given dose of this drug is very small because of its poisonous effect in higher dose. In the present investigation we want to assess toxic effects of *Nux vomica* on somatic chromosomes of *Vicia faba*. Four concentrations 5%, 10%, 20% and 30%, were used for the treatment of root tips of test plant. There were number of abnormalities observed like stickiness at orientation of chromosomes, un-organization of chromosomes, fragmentation during separation of chromatids, multiple chromatin bridges formation etc. These types of aberrations were increased with increased concentration and duration of treatment of this drug.

Keywords: Chromosomal aberrations, Genotoxic, Homeopathic drugs.

Nux vomica is a member of the Logoniaceae family and the medicine is prepared from dried seeds. It's raw seeds are called as Poison-nut due to presence of two alkaloids known as Strychina and Bruccia. Strychina is the principle ingredient. *Nux vomica* is one of the best remedies with which to commence treatment of causes that have been dragged by various medicines to counteract these chronic effects. It is a polycryst remedy with the bulk of symptoms corresponding in similarity with the most common diseases of modern era. It is useful for those persons who lead a sedentary life, doing much work or those who remain under stress from prolonged office work and business worries.

Sir Hahnemann said *Nux vomica* is chiefly successful with persons of an ardent character of an irritable, impatient temperament,

disposed to anger spite or deception. *Nux vomica* play a very useful role to cure thin, spare patient with irritable and nervous temperament who is prone to indigestion and haemorrhoids.

Some researchers believe that *Nux vomica* has anti-inflammatory properties, used to treat conditions that are worsed by inflammation, such as rheumatism as these or haemorrhoids. Some said that *Nux vomica* has antioxidant, that protect against free radical, which are chemicals in your body that can damage your cells.

Due to lot of beneficial uses of *Nux vomica*, there shall be no side effects occurs according to mode of treatment. In the present investigations we have an idea to assess the overall effects of *Nux vomica* in eukaryotic system which is plant system for fast and better observations and their findings we have used.

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We are potentially using *Nux vomica* to cure various kind of diseases and disorders. *Nux vomica* is used as natural extract of seed, so that there is probability to find some abnormal feature or side effects as genotoxic results by the treatment of *Nux vomica* as natural extract (Akinboro and Bakare 2007). *Nux vomica* may shows mitodepressive effects on normal mitotic division (Badr 1983). According to Badr 1983 a mitodepressive and chromotoxic activities were found by two natural herbicide in *A. cepa* another material product extract of *Hibiscus cannabiss* also effects as Cytotoxic activities in *Allium cepa* (Moujir et al 2007).

In the present investigation we were used extract of *Nux vomica* as homeopathic drugs on a text system that was a plant *Vicia faba* to assess various cytotoxic and genotoxic effect of Nux on eukaryotic system.

MATERIAL AND METHOD

Mother tincture of extract of dried seeds of *Nux vomica* were diluted in various concentrate with distilled water. We have used 5%, 10%, 20% and 30% concentrations for root tip treatment of *Vicia faba* (4 hrs., 8 hrs., 12 hrs., 24 hrs). First the seeds of *Vicia faba* were sterilized by using 2% mercuric chloride to 2 min and rinsed with distilled water. Sterilized seeds were soaked in distilled water for 4-5 min, after that the seed were germinated on moist filter paper in sterilized petriplates. When the root tips were 2.3mm long were subjected into 5%, 10%, 20%, 30% solution of *Nux vomica* for 8, 12, and 24hrs treatment. After treatment root tips were washed with water and fixed in Carnoy's solution (IGAA and Ethyl alcohol) for 6-14 yrs followed by transfer into 70% of alcohol at 4C. Slides have been prepared by acetocarmine squash method & observations were taken under 100x with Oil emersion. Pictures were taken by Olympus 3nm camera using Nova Oraplus 21 Din or 125 ASA.

Observations

There were so many structural, behavioural and numerical aberrations were observed in microslides of *Vicia* treated by Nux. Those were as follows:

Mitotic Index

The mitotic index was decreased with increase in concentration (5-10) and duration of treatment (4-24) hrs. The mean present value of mitotic index were decreased from 11.50 to 11.20%.(Table-1)

Condensed to sticky chromatids and chromosomes

The treatment of *Nux vomica* resulted in stickiness of chromosomes at metaphasic plate (Plate-1, fig -A, C, F). This kind of stickiness was observed increasing order in respect to increasing order of duration and concentration. The mean per cent value was higher through 12.13 to 13.93% in *Vicia*. (Table-1, fig-F).

Multipolar Spindles and inactivation

Multipolar spindles were noticed in ascending order in order of conc. and duration of treatment. The mean percentage was 0.52-0.62% (Table-1, fig- E,H,L).

Chromatin bridges at anaphase

It was observed that some chromatin bridges were disturbed the separation of chromatin. This abnormality was increased with increase duration and conc. of treatment of *Nux vomica*.

The mean percentage value was 4.47-5.14 (Table-1, fig- G,H,K, L).

Fragments

Another kind of abnormality observed in *Vicia*, that was fragments of chromatids during anaphasic separation. The heigher value was 4.85 at 12 hrs treatment.(Table-1, Fig-I,J).

Swelled and Stretched Chromosomes

Some swelled and stretched chromosomes were also observed in *Vicia* at higher concentration only (fig-D,F).

Figures Showing various chromosomal aberrations during different stages of mitotic cell division in root tips cells of *Vicia faba*.

RESULT AND DISCUSSION

The present investigation has suggested that *Nux vomica* has potential to damage chromosomes and its content. Various types of cytogenetical abnormalities were appeared by treatment of *Nux vomica* on *Vicia faba*.

The inhibition in MI values are perhaps due to reduction DNA and RNA contents in

dividing root meristem cells as observed by Anuradha (1991), through the pesticides treatment on the basis of cytogenetical, morphological and physiological studies in *Lens* and *Vicia*. Amato (1942, 1952) suggested that reduced MI has resulted due to Prophase poisoning and restriction of the division, Kuras et al (2009) found alkaloids from *Licaria fomentosa* bark were retard on inhibit mitosis and change mitotic phase in *A. cepa*.

In the present investigation it was clearly showed that appearance of sticky chromosomes was the most prominent abnormality observed (Table-1). The differential response of the plant

against drugs may be due to diverse correlation in the heterochromatin and euchromatin contents of the plant chromosomes (Evans, H.J. 1962). Stickiness is the most common abnormality. It is probably due to delay in chromosome movement through drug effect or influenced by gene action (Darlington 1945). In the present study, stickiness of chromosomes may be attributed to the alteration in the chromosomal protein resulting in the physiological changes of surface nucleoprotein configuration or to disturbances in cytochemically balanced reaction, (Salimuddin, B. Ramesh 1994).

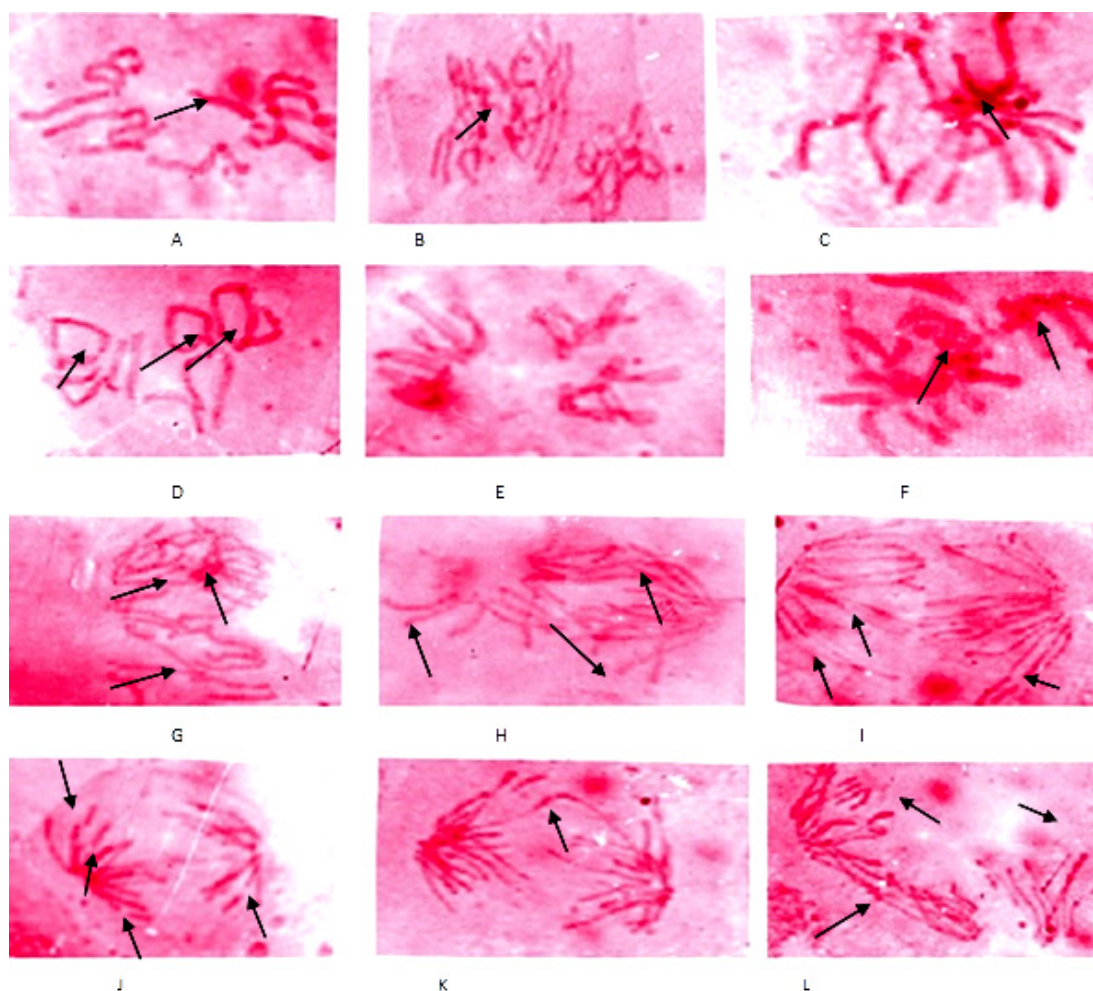


Fig. A-Condensed and sticky chromosomes, B-Unoriented chromosomes, C-clumped and stuck chromosomes, D-swollen and ring chromosomes, E-scattered chromosomes and multipolar spindle, F-Sticky and swollen chromosomes, G-chromatin bridges at anaphase, H-multipolar spindle and chromatin bridges, I-fragmented chromatids, J-chromatin fragmentation during separation, K-chromatin bridges at anaphase, L-multipolar separations of chromatids.

Another significant effect of *Nux vomica* was partial or entire inactivation of spindles leading to scattering of chromosomes, restitution of nucleus and multipolar spindle. The frequency of multi poles were observed in higher conc. of *Nux vomica*. The chromosome movement is adversely affected by inactivation and inhibition of spindle formation, due to inhibited ATP synthesis (Freg Wyssling 1938). The present study reveals that there is a tendency towards the arrest of division at metaphase or lengthened the period of division, similar aberration was also reported by the effect of drug (El-Bayoumi et al 1987).

Chromatin bridges were also observed due to occurrence of chromatin stickiness and subsequent failure of anaphase separation or it may be the result of chromosome breaks and reunion. Grover and Virk (1986). Fragments and breaks of chromatids were seen in some cells, it was may be the result of cytotoxic property of drug disturbed the synthesis and degradation of nucleic acids, proteins, etc. According to Sharma (1985) fragments followed by translocation may lead to heritable phenotypic differences. Kato (1960) observed that the extracts of young pods of *A. fistilerium* induced fragmentation and chromosomal breaks. These breaks are know to produced chromosomal mutations because of deletion and duplication, which lead to cell death Swaminathan, and Natrajan (1956) has fragmentation *A. sativum*. Induced by vegetable oils and edible fats.

Grant W.F. (1978) was reported Laggards same as present observations. It may be attributed by the adhesion of the centromere of a prometaphase chromosomes at the equator. The Lagging chromosomes has been reported earlier by Somashekhar et. al (1984) in *Allium*. The Laggards usually form micro nuclei able to perform DNA synthesis and mitotic condensation with the main nucleic. They can subsequently be integrated into the main nuclei in the first mitosis. Rao et al (1986) in *A. cepa*.

The micro nuclei and multi nuclei were observed in low frequency. They may arise from laggards and chromosomes fragments (Torkoglu et al 2007), or may also be attributed to the multiple spindle, or building of nucleus. Micronuclei are true mutagenic effect. (Kirkla D. 1998).

CONCLUSION

On the basis of observations and results, present investigation revealed that the action of the *Nux vomica* as homeopathy is a very cytogenotoxic with increase in concentrations and duration of treatment. It has suggested that the dose and duration of given drug *Nux vomica* as homeopathy drug should be tested before for it mutagenic and genotoxic properties in various eukaryotic system.

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