



FLOATING PLATFORM *Allium cepa* MODEL FOR TESTING ENVIRONMENTAL POLLUTION

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Abstract

The use of *Allium cepa* root tip models for testing cytogenetic effects of water-soluble substances of health and environmental concern is limited to the laboratory only. It was strongly felt worth modifying the basic protocol in such a way so that it can be easily be used both, in the laboratory as well as in the field with least efforts and expenses. This prudent need-based efforts resulted in the fabrication of a novel modified floating platform model which uses brick shaped fenestrated thermocol floats replacing glass tubes to hold *A. cepa* bulbs to grow roots during exposure while floating on the surface of aquatic test fluid. Floats with *A. cepa* bulbs can also be released in in aquatic medium filled large aquarium, plastic or metal tubs or cemented tanks in the laboratory and on the surface of water bodies like ditches, ponds, lakes, effluent filled water accumulated areas and even in running water of river and stream in the fields as detailed in this communication.

KEYWORDS: *Allium cepa*, Mitotic Effects, Floats, Aquatic Pollutants

Original *Allium cepa* root tip laboratory test was initially introduced in 1938 for testing chromosomal effects of water-soluble substances of environmental and health concern (Levan, 1938), however, standard *Allium cepa* laboratory protocol was published twice at an interval of about ten years by the same author for testing cytological effects of toxic substances (Fiskesjo, 1985 & 1995). During this decade first modified *A. cepa* test was suggested (Rank and Nielsen, 1993). Surprisingly different cytological results of one test chemical were obtained in standard and modified method which could be discussed and explained and it was suggested to test any substance in both models to draw logical and meaningful conclusion (Rathore *et al.*, 2010). Present proposed protocol of *A. cepa* is designed to replace the use of glass tubes for growing *A. cepa* bulbs by using the “floats” in the laboratory as well as in the fields like ponds, ditches and river.

MATERIALS AND METHODS

This is described under separate sub headings for convenience:

Requirements for fabrication: Containers to keep contaminated and uncontaminated aquatic samples like large aquaria, plastic tubs, portable cemented tanks, thermacol sheets, cutter, adhesives, marker pens and nylon threads etc.

Fabrication of “Floats” for holding *A. cepa* bulbs: Thermacol sheets of 2 cm thickness can be cut in brick shaped floats of about 14cm broad and 30 cm long. On each float, circles of 2 to 3 cm diameter (depending

on the widest part of *A. cepa* i.e., onion bulbs to be used to insert) are to be drawn to cut remove rounded thermacol portions leaving clear holes. Each float can have two rows of three holes in each row at equal intervals. Total six holes per float were found enough to keep float with fitted onion bulbs floating on aquatic medium for growth of roots. Components of fabricated model are shown in figures 1 to 4 in plate-1.

Preparation of *A. cepa* i.e., onion bulbs before use: As referred, protocol of Fiskesjo (1995) or Rank and Nielsen modified model (1993), anyone can be followed.

Fixing of *A. cepa* bulbs on floats: Each hole of float is to be used to insert and to fix (hold) only one bulb in it. Total six bulbs can be fixed in six holes per float. Bulbs can be inserted either before or after initial root growth depending on the protocol selected by the investigator.

Use of test samples in the laboratory: One large aquarium, plastic or metal tubs or cemented tanks can be filled with tap water or uncontaminated water for controls and others with aquatic samples for testing morphological changes in the shape, size and color of root tips and cytological i.e., chromosomal (mitotic) effects. Let floats float on the surface of medium.

Use of water bodies i.e., field study: Any natural lakes and ponds or man-made water bodies like ditches, accumulated standing water etc. can be used to put floats fitted with *A. cepa* bulbs for further growth of roots.

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In field water bodies with flow like river and stream or flowing discharge of domestic waste or industrial effluent can also be tested for cytogenetic effects in root tip cells if floats are tied with a thick nylon thread with a firm object near bank at one end and with float at another end. These tied floats can be kept floating at a place for desired days as per need of model to be followed.

Macro (morphological) and Micro (cytological-cytogenetic) observations: Shape of root tips, their color and mean length can be recorded; fixed and stored tips can be used to squash and stain following routine procedures to observed and to record cytogenetic effects if any.

RESULTS AND DISCUSSION

Aim of this communication is to bring first this novel modified model of *Allium cepa* in the knowledge of global scientific community hence pilot results are not given, however, initial laboratory testing of acetaminophen, myrobalan, *Cassia tora*, lead and triphala in floating model gave confirmatory results as already reported by authors team in the past (Rathore and Choubey, 2005; Rathore *et al.*, 2006; Singh *et al.*, 2009; Rathore *et al.*, 2007; Marmat and Rathore, 2013). Field studies are also encouraging and shall be published in future. It is hoped that proposed model shall prove as an asset to environmental biologist to test cytogenetic effects of aquatic media in mitotically dividing cell with least efforts in the laboratory and in the fields too.



Plate 1: Photos showing components of proposed *Allium cepa* floating model

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REFERENCES

Fiskesjo G., 1985. The *Allium* test as a standard in environmental monitoring. *Hereditas*, **102**:99-112.

- Fiskesjo G., 1995. *Allium* Test-In Methods in Molecular biology, 43: In-Vitro toxicity testing protocols, editors-S.O. 'Hare and Atterwill, C.K., Humana Press, Totowa, NJ pp. 119-127.
- Levan A. 1938. The effect of colchicine on root mitoses in *Allium*. *Hereditas*, **24**:471-486.
- Rank J. and Nielsen M.H., 1993. A modified *Allium* test as a tool in the screening of genotoxicity of complex mixtures. *Hereditas*, **118**:49-53.
- Rathore H.S., Rathore M., Panchal S. and Makwana M., 2010. Can genotoxic effect be model dependent in *Allium* test? An Evidence. *Environment Asia*, **3**:29-33.
- Rathore H.S. and Choubey P., 2005. Prevention of acetaminophen-induced mitodepression with myrobalan (fruit of *Terminalia chebula* Retz Combretaceae) in *Allium cepa* model. *Iranian J. Pharmacol and Therap.*, **4**:100-104.
- Rathore H.S., Khare A. and Sharma A., 2006. A study on the cytological effects of myrobalan (fruit of *Terminalia chebula*) in *Allium* test. *Ethnobotanical Leaflets USA*, **10**:92-97.
- Singh M., Solanke P., Rathore H., Sharma A., Makwana M. and Srivastava S., 2009. Influence of decoction of seeds of *Cassia tora* on the genotoxicity of sodium azide and acetaminophen in *Allium cepa* model. *The Internet J. Toxicol.*, **6**:1-6.
- Rathore H.S., Punyasi R., Joshi P., Rathore D. and Bhatnagar D., 2007. Studies on the reversal of lead-induced mitostatic effect in *Allium cepa* root tip cells with myrobalan (fruit of *Terminalia chebula*, Retz Combretaceae). *Internet J. Alt Med.*, **4**:1-10.
- Marmat, Savita and Rathore H.S., 2013. Cytological effects of triphala in *Allium cepa* model. *Internet J. Pharm Tech.*, **5**:5602-5609.