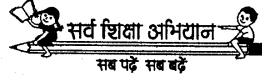


भारत सरकार
विज्ञान और प्रौद्योगिकी मंत्रालय
बायोटेक्नोलॉजी विभाग
GOVERNMENT OF INDIA
MINISTRY OF SCIENCE & TECHNOLOGY
DEPARTMENT OF BIOTECHNOLOGY



ब्लॉक-2,7 वां तल, सी० जी० ओ० कम्प्लेक्स
लोदी रोड, नई दिल्ली-110003
Block-2, 7th Floor C.G.O. Complex
Lodi Road, New Delhi-110003

No.BT/17/02/94-PID

Dated: 24.05.2005

To

M/s. Maharashtra Hybrid Seeds Company Ltd,
Resham Bhawan, 4th Floor,
78, Veer Nariman Road,
Mumbai - 400 020.

Subject: Application submitted for permission to carry out multi-location contained limited field trials on transgenic brinjal containing *cryIAc* gene during Kharif 2005.

Gentlemen,

The Department is to refer to your letter No. SBD/832/2005 dated 05.04.2005 on the above mentioned subject and to inform you that your application was considered by the Review Committee on Genetic Manipulation (RCGM) in its meeting held on 21.04.2005. On the basis of the recommendations of the RCGM, the Department is permitting you to conduct the multi-location field trials on transgenic brinjal (*Solanum melongena*) namely **MHB-11 Bt, MHB-39 Bt and MHB-112 Bt** containing *cryIAc* gene and their non-Bt counter parts for experimental purposes at 15 locations during Kharif-2005, subject to the following objectives and conditions:

- (a) The trials would be conducted on MHB-11 Bt brinjal along with non-Bt brinjal and Mohini hybrid and local checks at **Coimbatore (TN) & Akola (Maharashtra)**; MHB-39 Bt brinjal along with non-Bt brinjal and Pragati hybrid at **Dindigal (TN) & Kolar (Karnataka)** and on MHB-112 Bt brinjal along with non-Bt brinjal and Krishna hybrid at **Karnal (Haryana) & Jaipur (Rajasthan)**. The experiments design would be randomized block design (RBD) with conducted 5 replications. The objectives of the trials is to generation of data on the following:
- To evaluate the field infestation levels of target insect pests viz. Shoot & fruit borer (*Leucinodes orbonalis*), Gram caterpillar/ fruit borer (*Helicoverpa armigera*) and Stem borer (*Euzophera perticella*) on Bt and their corresponding non-Bt brinjal hybrids.
 - To assess the agronomic performance of Bt and corresponding non-Bt brinjal hybrids vis-à-vis local non-Bt checks.
 - Savings in terms of pesticides as well as the yield losses will be calculated at the end of the season.
 - To assess the effect of Bt protein on non-target pests and beneficial insects in case of brinjal crop.
 - Estimation of the levels of expression of Bt. protein in Bt brinjal hybrids fruits at different growth period on regular interval of 15 days. The expression of Bt. protein shall be in milligrams of CryIAc protein per gram fresh weight of the fruit.

Contd..2/-

तार / Telegram : 'BIOTECH' दूरभाष / Telephone : 24363012, 24360899 फैक्स / Fax : 011-24362884

- vi) Generation of baseline susceptibility data on shoot and fruit borer (*Leucinods orbonalis*) and fruit borer (*Helicoverpa armigera*) population collected from the site of trial and rearing them at the premises of the company's bio-assay laboratory at Mahyco Life Science Research Centre, Aurangabad-Jalna Road, Jalna.
- b) The field design of the trials at above-mentioned locations would be the same as was submitted to the Department through your application-dated 05.04.2005. The experimental plot would be measuring 669.60 sq. meters (37.2 x 18.0 meters). An isolation distance of 200 meters from the periphery of the nearest row of transgenic brinjal has to be maintained. You shall not plant any sexually compatible plants to the brinjal in the isolation distance of 200 meters. The experimental layout is annexed at **Annexure-1**. You would be submitting the details of the locations to the Department before undertaking experiments.
- c) You would mount a Notice Board at the site of experiment indicating in the Notice Board the purpose and duration of the experiment as well as the authorization under which the experiments are being conducted.
- d) You would provide with three photographs of experimental site, taken from a distance sufficient to indicate the transgenic plots in a single photograph; such photographs would be taken at three intervals during the season to document the start of the experiments, the mid way of the experiments and the end of the experiments. These photographs would be submitted to the Government at the conclusion of the experiments.
- e) You would keep full account of the transgenic materials and seeds, if any, set in the transgenic plants. All materials after experimentation including the seeds of cotton for the trapper rows would be fully accounted for and the information would be documented and preserved in a bound book that would be available to the Government as when requested for. The harvested crop from the border rows and leftover plant and plant parts from entire experimental plot shall be destroyed by burning after completion of the experiment.
- f) You would collect and provide all the experimental data to the Government for inspection whenever required.
- g) You would ensure that while performing the experiments, the "Recombinant DNA Safety Guidelines-1990" and "Revised Guidelines for Research in transgenic plants & Toxicity and Allergenicity evaluation of transgenic seeds, plants and plant parts" of the Government of India would be strictly adhered to. Accidents, if any, arising out of the experiments may be brought to the notice of the Government immediately.
- h) You would further ensure that only company authorized personnel would be permitted to visit the experimental plot and persons visiting the experimental plot shall enter the name, designation and purpose of visiting the experimental plot in a bound book which should be made available to the Government when requested for.
- i) You would extend full cooperation to the authorized personnel of the RCGM/MEC as well as the State Government Officials or their nominee to inspect the experimental sites and **to have access, for official use only**, the experimental results of the above.

j) You would inform the concerned State Government Officials about the conduct of the experiments in their states.

2. The Limited Trial Permit is valid for **Kharif Season 2005** only from the date of issue and would lapse automatically after the season.

Please acknowledge the receipt of this letter.

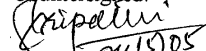
Yours faithfully,



(T.V. Ramanaiah)

Member-Secretary, RCGM &
Scientist-F, DBT

Countersigned:



(K.K. Tripathi)

Scientist-G, DBT

Copy for information and necessary action wherever applicable to:

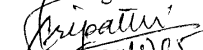
1. The Secretary, Ministry of Environment and Forests, Paryavaran Bhawan, CGO Complex, Lodi Road, New Delhi - 3.
2. The Secretary, Ministry of Agriculture, Krishi Bhawan, New Delhi -110 001.
3. The Chairman, GEAC, MoE&F, Paryavaran Bhawan, C.G.O. Complex, New Delhi-110 003.
4. The Director General, ICAR, Krishi Bhawan, New Delhi -110 001.
5. The Chairman, MEC & Director, IARI, Pusa, New Delhi - 110 012
6. The Chief Secretary, Government of Maharashtra, Mumbai.
7. The Chief Secretary, Government of Tamil Nadu, Chennai.
8. The Chief Secretary, Government of Karnataka, Bangalore.
9. The Chief Secretary, Government of Haryana, Chandigarh.
10. The Chief Secretary, Government of Rajasthan, Jaipur.
11. The Commissioner Agriculture, Maharashtra State, Central Building, Pune (Maharashtra).
12. The Commissioner of Agriculture, Department of Agriculture, Cheupak, Chennai.
13. Agriculture Commissioner, Department of Agriculture, Haryana State, Chandigarh.
14. Director of Agriculture, Rajasthan State, Jaipur.
15. The Commissioner of Agriculture, Directorate of Agriculture, Sheshadri Road, Bangalore.
16. File No. BT/BS/17/02/94-PID



(T.V. Ramanaiah)

Scientist-F, DBT

Countersigned:



(K.K. Tripathi)

Scientist-G, DBT

ANNEXURE - I

Experimental details

- Completely Randomized Block Design (CRBD) with five replications and four treatments in each of the trials.
- In all three trial sets, based on Brinjal fruit type (driven by consumer/ regional preferences) trials would be conducted.

| Trial No. | Treatments: |
|-----------|--|
| Trial-1 | Mahyco hybrid MHB-11 Bt, MHB-11 Non-Bt, Local check, Mohini hybrid. |
| Trial-2 | Mahyco hybrid MHB-39 Bt, MHB-39 Non-Bt, Local check, Pragati hybrid. |
| Trial-3 | Mahyco hybrid MHB-112 Bt, MHB-112 Non-Bt, Local check, Krishna hybrid. |

Location Details:

| Trial No. | Hyb No. | Location | State |
|-----------|---------|---------------------|--------------------------|
| Trial-1 | MHB-11 | Coimbatore Akola | Tamilnadu Maharashtra |
| Trial-2 | MHB-39 | Dindigal Kolar | Tamilnadu Karnataka |
| Trial-3 | MHB-112 | Karnal Jaipur | Haryana Rajasthan |

Description of Field, Plot and Treatments

Each of the trials would be taken up at the locations specified during Kharif-2005. Sprays for sucking pests will be undertaken for all treatments including Bt Brinjal hybrids as per the recommended regional practices. Bt Brinjal hybrids and checks will be given sprays for fruit and shoot borer based on ETL. Record of sprays shall be maintained to calculate economic benefits.

Experimental Details:

| | |
|---------------------|---------|
| Number of trials | : Three |
| Experimental Design | : CRBD |
| Treatments | : Four |
| Replications | : Five |

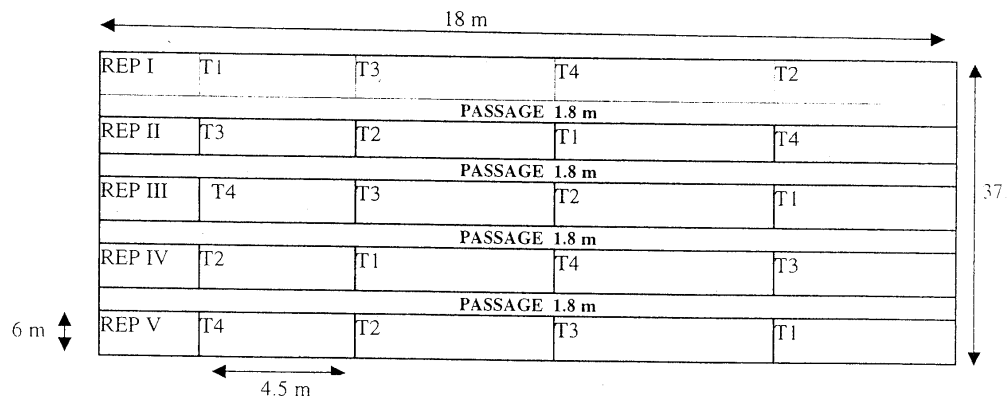
Trial –I T1- MHB-11 Bt, T2 - MHB-11 Non-Bt,
T3- Local check, T4- Mohini Hybrid.

Trial –II T1- MHB-39 Bt, T2 - MHB-39 Non-Bt,
T3- Local check, T4- Pragathi Hybrid.

Trial –III T1- MHB-112 Bt, T2- MHB-112 Non-Bt,
T3- Local check, T4- Krishna Hybrid.

Replications : 5
 Treatments : 4
 Rows per plot : 5
 Row length : 6 m
 Plants per treatment : 50
 Spacing between rows : 90 cm
 Spacing between plants : 60 cm
 Space between replications : 1.8 m
 Gross experimental plot area : 669.60 Sq .m
 Net plot experimental area : 540 Sq.m
 Net plot per treatment : 16.2 Sq.m
 Net area under Bt Brinjal hybrids : 81 Sq.m (16.2 Sq.m x 5)

Field Layout of Replicated Research Trial: (18m x 37.2m)



Farmers and Locations Details

| State | District | Village | Farmers Name |
|-------------|------------|--------------------|--|
| Tamil Nadu | Coimbatore | Nadegoundam pudhur | Mrs Nanjamal W/o Subbusamy Goundar, Mangalathal Koil Thottam |
| Maharashtra | Akola | Bhawrad | Mr Vijay Rambhau Tawre |
| Tamil Nadu | Dindigal | Virupachi | Mr. M. Boopathi S/o Murugesha Goundar |
| Karnataka | Kolar | Alambagiri | Mr. Muniswamy |
| Haryana | Karnal | Kalwhedi | Mr. Bhuvneet Kaliana |
| Rajasthan | Jaipur | Niwaru | Mr. Hazarilal Saini |

Summary Results

Replicated research trials of Bt brinjal hybrids for different market segments were approved by RCGM in 2005. Deployment of Bt brinjal technology is a new strategy for effective control of Brinjal Fruit and Shoot Borer (BFSB). Primary objectives of these trials were:

- To assess the yield potential of the Bt brinjal hybrids, as compared to non-Bt brinjal counterparts and checks.
- To compare lepidopteran pest (BFSB) load and feeding damage between Bt brinjal hybrids and non-Bt hybrids.
- To study possible effects of the Bt gene against secondary lepidopteran pests and non-target insects.

Three Bt brinjal hybrids, their non-Bt counterparts and checks were grown in completely randomized block designs with five replications at six locations. Trials of MHB-11 Bt were conducted at one location each in Maharashtra and Tamil Nadu, trials of MHB-39 Bt were conducted at one location each in Tamil Nadu and Karnataka, trials of MHB-112 Bt were conducted at one location each in Haryana and Rajasthan. Test entries for all the trial sets included Bt-hybrid, the non-Bt hybrid counterpart, local check and a commercial check. Data was collected on Brinjal fruit and shoot borer infestation and related damage to plant parts, fruit count and brinjal yield over pickings. Additional information was collected on secondary lepidopteran pests, sucking pests and beneficial insects.

Results from these experiments indicate that the Bt brinjal hybrids had potential for higher marketable brinjal fruit yield in comparison to the non-Bt counterparts and checks. All Bt hybrids also had significantly higher fruit counts as compared to all three checks. Marketable fruit yield is a measure of quality.

For BFSB related observations, significant differences were detected between hybrids based on presence or absence of Bt gene. For BFSB count, significant differences were detected between Bt hybrids (containing *cryIAc* gene) and all three non-Bt checks. All Bt hybrids were significantly lower in number of BFSB larvae. Differences were also measured between the Bt hybrid and non-Bt check hybrids for

shoot damage to plants from BFSB infestation. Percent damage to shoots were significantly lower for the Bt group as compared to non-Bt hybrids. The degree of such differences in BFSB feeding damage between Bt hybrids and non-Bt hybrids was significant.

Observations were taken till completion of all pickings, for the presence of secondary lepidopteran pests, sucking pests and beneficial insects. No significant differences were noted between Bt hybrids, the non-Bt counterparts and checks in terms of incidence of sucking pests (aphids, jassids, whitefly) and beneficial insects (chrysopa, lady-bird beetle, spiders).

Results of these multi-location replicated research trials indicate that the Bt brinjal hybrids provide adequate level of tolerance to BFSB and show good yield potential, BFSB efficacy and marketable yield. Bt brinjal hybrids showed a significantly lower damage resulting from BFSB feeding in comparison to non-Bt brinjal. Bt brinjal hybrids did not have any effects on non-target insects, including beneficial insects; and therefore can play a positive role within integrated pest management strategies for sustainable brinjal cultivation.