Report of the

Independent Expert Committee on Bt Brinjal

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Centre for Sustainable Agriculture (Hyderabad)
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The Independent Expert Committee

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Retired recently as Professor of Economics and Dean, School of Social Sciences, University of Hyderabad. His specialization includes science policy studies, Political Economy of Development and Labour Economics. He was a member of the Farmers' Welfare Commission set up by Andhra Pradesh government to look into the issue of farmers' suicides and agriculture crisis.

SUMMARY

"An independent expert committee to look into issues related to Bt Brinjal found that DBT guidelines were not always followed by the developers of transgenic brinjal. It found that some tests as laid down by the DBT were not taken up in the safety evaluation or test protocols were not as per the specifications laid down in the guidelines. The Committee acknowledged that recent published evidence on the genes and vectors used in evolving Bt Brinjal show that caution needs to be exercised in using these materials in plants and plant products meant for human use. Results of some toxicity tests show that unless raw data is examined and full report seen, it is not possible to arrive at any meaningful conclusions regarding safety of the product.

On the agronomic trials, Bt Brinjal is not compared with the best agronomic and pest management practices available nor have trials been taken up for at least three years. Committee observed that parameters prescribed in the DBT's permission letter were not always meticulously followed by the company. Committee found that data from the company is insufficient for any conclusion to be drawn about the efficacy of Bt Brinjal. From AICRP data, there is not much hope for Bt Brinjal, the Committee noted.

Committee made several relevant observations which need to be taken cognizance of, on the protocols used for other studies like pollen flow, cooking studies, aggressiveness & weediness, soil impacts study etc.

Committee noted that impact assessment beyond biosafety, including addressing issues like genius loci, of IPR regime on farmers' rights etc. is important. The Committee called for a holistic approach rather than a reductionist approach here.

The Committee noted that the issue of the Indian sub-continent being the Centre of Origin for brinjal is of considerable relevance and that need for Bt Brinjal should be re-examined against this background.

Given that there is uncertainty and variability in the risk assessment process and given that satisfactory methodologies to measure possible long term health effects or unintended/unexpected adverse effects of GM foods are yet to be evolved, the Committee noted that it is prudent to follow a precautionary approach".

Report of the Independent Expert Committee on Bt Brinjal

Terms of Reference for the Independent Expert Committee

I. To evaluate the data presented by Mahyco on Bt Brinjal and its biosafety. On the biosafety tests, specifically compare the protocols used for various tests with the official DBT guidelines evolved as part of the Environment Protection Act.
II. To evaluate the feedback sent to the GEAC from civil society.
III. To look at issues beyond biosafety, including the need for Bt Brinjal.
IV. To suggest a future course of action to GEAC, other regulators, GoI and state governments.

Sessions

The committee met in two sessions on 27th and 28th of September, 2006, and 20th of October, 2006, at Centre for Sustainable Agriculture, Hyderabad and also had discussions over email and a teleconference.

1st session: 27th and 28th September, 2006

Members present: Dr A Narayanan, Acting Chairperson
Dr Ramesh V. Bhat
Dr Ghafoorunnissa, and
Prof. K P Prabhakaran Nair (through teleconference)

2nd session: 20th October, 2006

Members present: Dr. D. Narasimha Reddy, Acting Chairperson
Dr. Ramesh V. Bhat, and
Dr. M. S. Chari.

Preamble

The members decided that the committee will confine mainly to scientific, economic and socio-cultural aspects related to Bt Brinjal so that the findings of the committee may assist the GEAC, developers of the technology, various civil society groups and consumers in general in understanding the facts and in arriving at appropriate conclusions.

I. MAHYCO'S TESTS VIS-À-VIS DBT'S BIOSAFETY GUIDELINES

The committee looked into whether the tests were conducted as per the guidelines proposed by DBT which were in vogue at the time when M/s Mahyco initiated studies on Bt Brinjal[1].

1. The DBT guidelines for sub-chronic oral toxicity study on goats for 90 days specify that Indian ***Barberi*** breed should be used whereas ***Osmanabaadi*** breed was used in the experiments for Bt Brinjal.

2. The skin sensitization test of transgenic material in guinea pigs as laid down in the DBT guidelines has not been taken up.

3. Sub-chronic oral toxicity of leaves of transgenic plants on male rabbits prescribed in DBT guidelines was not tested.

4. The allergenicity of the protein extract from transgenic brinjal was carried out in *Brown Norway rats* and not in *rabbits or guinea pigs* as suggested in DBT guidelines.

5. DBT guidelines prescribe ***in vitro*** and ***in vivo*** immunological assays for the detection of reactogenic antibodies in the test sera. ***In vivo*** assays (PCA and PK tests) were not done.

6. The DBT guidelines state that the characteristics of the donor organisms, of the vectors used, of the transgenic inserts and of the transgenic plants are required to be generated. The Committee notes the following in this regard -

    a. Though *Cry1Ac* gene was earlier considered generally innocuous, recent published evidence indicates that Cry1Ac protein from *Bacillus thurengiensis* is a potent systemic and mucosal adjuvant as potent as the cholera toxin which enhances mostly serum and intestinal IgG antibody responses specifically at the large intestine (Vazquez et al, 1999). Also another study (Vazquez-Padron et al, 2000) demonstrates the possible interaction *invivo* of Cry proteins with animal bowel. According to Moreno-Fierros et al (2000), caution needs to be exercised while using Cry-containing plants and plant products for human use.

    b. Recent reports on *CaMV 35S* (Myhre et al, 2006) note that promoter gene expression in human enterocyte-like cells might have GE food implications.

[1] Revised Guidelines For Research In Transgenic Plants & Guidelines For Toxicity And Allergenicity Evaluation Of Transgenic Seeds, Plants And Plant Parts, 1998 - Department Of Biotechnology, Ministry Of Science And Technology, Govt. Of India

c. Regarding the *aad* gene used in developing Bt Brinjal [streptomycin resistant gene], this Committee notes that according to the EFSA, this is a potentially dangerous marker to animals and human beings and should not be used in the case of GM plants used as food.

d. The *Agrobacterium tumefaciens* medium was used for the transformation process of development of Bt Brinjal. Strains of agrobacterium were earlier implicated in incidence of bronze wilt in cotton in the US (McGraw, 2000). It is not clear whether its potential impacts have been studied carefully in this case.

In summary, the Committee notes the following:

Study details	Prescribed specifications in DBT guidelines	What was carried out	Other Comments
Sub-chronic oral toxicity study on goats for 90 days	Indian ***Barberi*** breed should be used	***Osmanabaadi*** breed was used	
Skin sensitization test of transgenic material	Use guinea pigs	Test not done	
Sub-chronic oral toxicity of leaves of transgenic plants	On male rabbits	Test not done	
Allergenicity of the protein extract from transgenic brinjal	Use rabbits or guinea pigs	Done on *brown Norway rats*	
Detection of reactogenic antibodies in the test sera	To do ***in vitro*** and ***in vivo*** immunological assays	Only *in vitro* tests carried out. ***In vivo*** assays (PCA and PK tests) were not done	
Characteristics of the donor organisms, the vectors used, the transgenic inserts and the transgenic plants are required to be generated			Recent reports indicate: • Cry1Ac is a potent systemic and mucosal adjuvant. Possible interaction of Cry proteins with animal bowel is demonstrated • In the case of

			CaMV 35S, promoter gene expression in human enterocyte-like cells has GE food implications • Aad gene is a potentially dangerous marker, not recommended for use in GM plants used as human food • Agrobacterium used in transformation process implicated in bronze wilt

GENERAL OBSERVATIONS

The committee is not certain whether the testing laboratories follow Good Laboratory Practice (GLP) procedures and whether they are accredited. It is also not clear how testing materials were ascertained and whether they are authentic. The laboratories undertaking the test must ensure their authenticity by independently getting them analyzed.

The committee was constrained to some extent since the details of the experiments [protocol, full data, statistical evaluation etc.] were unavailable for it to critically examine the issues involved. In fact, in many instances only the summary of reported work carried out was available.

ADDITIONAL COMMENTS

Since conclusive experimental evidence is available to indicate that root exudates from GM crops perceptibly alter the soil microflora profile (Vadakattu & Watson, 2004), it is disturbing that no data has been generated on this very crucial aspect, which can substantially alter the inherent fertility status of the soil in which these crops are grown. The long-term effects of such changes can vastly affect the productive capacity of soils in which these crops are grown. Such data must have been provided to eliminate any doubt on the possible adverse effect of root exudates on soil's inherent fertility status.

II. EVALUATION OF MAHYCO'S TESTS AND DATA

A. On Mahyco's Agronomic trials and other tests related to pollen flow, aggressiveness etc.

1. To critically understand Genotype-Environment-Pest interactions, at least three years' data is required (over the same season) and not just two or three crops grown in a year. This is because the pest incidence is seasonal and is highly influenced by the environment.

2. The trials cannot be termed as Multi-locational trials since each hybrid has been tested at the most at two locations.

Further, the trials for each specific hybrid were not continued in the subsequent year in the same location whereas such trials should be with the same hybrid, in the same location for at least three years.

3. Similarly, consistent data over at least three years on insect susceptibility/resistance and efficacy of the method as per ICAR norms is required to arrive at any meaningful conclusions.

4. Data provided by Mahyco on non-target pests is only for one season and one centre. It should be from all the centres, over seasons.

5. Trials are not compared with best agronomic and pest management practices already existing (IPM, NPM, organic). Available data generated by institutions like Indian Institute of Horticulture Research, Tamil Nadu Agriculture University, Anand Agriculture University, Centre for World for Solidarity/Centre for Sustainable Agriculture clearly show that other efficient pest management practices are possible. Therefore, the best available management practices should be the check for testing Bt Brinjal's efficacy.

6. There are some important questions unanswered on the data parameters, listed below:

- % damage of terminal buds and shoot damage is an important parameter that needs to be assessed
- toxicity and efficacy of the technology on the first instar larvae of Fruit & Shoot Borer (FSB) is something that needs to be clearly assessed
- data on little leaf incidence is to be generated and reported
- plant population at the time of harvest is important
- all the trials should also take up entomological evaluation in addition to agronomic evaluation. Wherever the plant population is low or the trials are not reported, the reasons for that should be indicated and assessed.
- in some cases, pest incidence (of FSB) is below ETL even in control plot.
- some data like 80 larvae/plant is exaggerated and unbelievable.
- ETL across locations is highly variable (CD and CV should be mentioned and data needs to be analysed statistically).

7. In Mahyco's trials conducted in 11 locations, data have been presented only from 9 locations. The data from Hissar and Alwar are not presented.

In Kharif 2005, permission had been accorded for 15 trials. However, data for only 3 hybrids from six locations has been presented. How is this accounted for or justified is not clear.

8. No statistical analysis has been done in reports by the company. Without statistical analysis no meaningful conclusions can be drawn.

9. Data on the number of (pesticide) sprays, quantity of fertilizer used and the type of fertilizers used for the different treatments are not available.

10. No information on the plant population at the time of harvest is available. This is extremely crucial for a reliable statistical scrutiny.

11. It is not clear who has taken the count of various pests, insects, diseases etc., from the trial plots and who has supervised data collection from the farmers' fields.

12. Data have not been presented along the checks and the non-Bt counterparts. Selective presentation of data is noticed.

13. The variation in yields across locations is very high and it is not clear whether the same management practices were used across locations for drawing any conclusions.

14. While permitting limited field trials, the DBT asked for the evaluation of field infestation levels of Fruit and Shoot borer (*Leucinodes orbonalis*), Gram caterpillar/fruit borer (*Helicoverpa armigera*) and Stem borer (*Euzophera perticella*). However, details of the evaluation carried out are not available for any meaningful conclusion. This should have been presented for all the treatments for various pests for any such conclusion to be drawn.

15. As per information available, baseline susceptibility data was generated only for one pest [*Leucinodes*] for one year [2004] even though the DBT's permission letter given to the company for conducting multi-locational trials in 2004 as well as 2005 requires the company to generate such data for at least three pests - Fruit and Shoot borer (*Leucinodes orbonalis*), Gram caterpillar/fruit borer (*Helicoverpa armigera*) and Stem borer (*Euzophera perticella*), over a minimum of two years (two seasons).

16. The DBT has asked the company to assess the Cry1Ac protein expression levels once every 15 days in their permission letter. This Committee notes that this was not meticulously followed.

17. The Committee is unable to locate any data on the economics of pesticide use, the cost-benefit ratio, as required to be generated from all trial locations as per the DBT's

permission letter in 2005. In the absence of all such data, no valid and dependable conclusion can be drawn on the efficacy of Bt brinjal.

18. **On the ICAR-supervised field trials**: The agronomic trials data presented by the company from its trials has to be read along with the ICAR-supervised All India Coordinated Research Project – Vegetable Cultivation (AICRP) results on Bt Brinjal, which are part of the regulatory requirements. From the data presented in the AICRP annual report for 2005-06, there is not much promise for Bt Brinjal.

Further, even in the AICRP data, a critical statistical analysis will present a reliable picture of the actual situation. For example, the fruit borer infestation figures between centres like Varanasi and Hyderabad show a great variation and only statistical analysis will help to evolve a better picture.

In the ICAR trials, there is high variation and the results are erratic. No meaningful conclusions can be drawn on the performance as data is very insufficient.

Unfortunately, even with the best of efforts, this Committee was unable to obtain information on the AICRP Bt Brinjal trials in 2004 and these trials are not reflected in the AICRP Annual Report, which is intriguing. The Committee also would like to emphasise that Mahyco's presentation to the GEAC does not have any data from the ICAR-supervised trials. .

19. Mahyco's document entitled "Development of Fruit and Shoot Borer Resistant Brinjal" on page 4 under 'Biology of the Plant System' says that ***"Brinjal plant is usually self-pollinated, but the extent of cross-pollination has been reported as high as 48% and hence it is often classified as cross-pollinated crop".*** However, the pollen escape studies conducted by Mahyco over only one year, 2002, show an outcrossing percentage of 1.46% to 2.7%. This is very intriguing.

The pollen flow studies do not capture the possible [resistant] trait transfer and the extent of such transfer. They also do not capture the outcrossing situations between Bt Brinjal and native brinjal species, wild relatives or with plants of the *Solanaceum* family.

20. Mahyco's report on the study on Aggressiveness and Weediness [on page 6 of Brinjal I document on MoEF website] says, ***"If any plant growth occurred, the same was checked with ELISA to determine if it was transgenic or not"*** It then goes on to add, ***"There was no brinjal plant observed to grow or germinate in this plot for the period of the study".*** This is incomprehensible and the report seems to have been written in a lackadaisical manner.

The protocol for assessing aggressiveness and weediness is unclear.

21. Cooking studies should take into account complex food matrices and not just raw seed or fruit or vegetable in isolation. In the Cooking Studies and Protein Estimation, the

company has tested for the Cry1Ac protein but has not checked for the metabolites. While the ELISA test might show negative for the protein, it does not account for the complex food matrix situations, as the Starlink Corn case has demonstrated (Siruguri *et al*, 2004).

B. On the Biosafety tests of toxicity, allergenicity etc.

1. The Committee notes that the report on Sub-Chronic Oral Toxicity Tests on Sprague Dawley rats for 90 days given in summary by the laboratory and the company raises the following questions:

1. The reports say that "there were isolated instances of necropsy findings" – isolated is how many? Clarification on 'isolated' is missing.
2. The incidence of 'pathological lesions'…'being extremely small' needs further explanation.
3. They conclude that it is not dose-dependent when apparently only one dose of 1000 mg was used. The three doses of 250, 500 and 1000 mg/kilo of body weight were used only in the dose-ranging study of 14 days and not the main study of 90 days as per the report. It is not clear how the main study report [of 90 days] then concluded that it is not dose-dependent.

Unless the raw data is examined and the full report is seen, it is not possible to arrive at meaningful conclusions regarding the safety of the product.

2. The Committee also notes in the report on Sub-Chronic Oral Toxicity Study on Goats for 90 days that statistically significant changes were found in the haematological as well as the clinical parameters. But despite this, they are not considered to be of physiological significance. In the absence of raw data, including the range of control values, it is difficult to further comment on this aspect on the safety or otherwise of the product.

The reported significant difference in the hay consumption of the transgenic Bt Brinjal-fed group is also of concern.

III. EVALUATION OF FEEDBACK SENT TO GEAC BY CIVIL SOCIETY

The Civil Society has carried out an admirable job in bringing out awareness on various aspects of Bt Brinjal to the notice of the GEAC and the general public. It has meticulously collected the literature, scrutinized the data generated and sent feedback to the regulators. Civil society groups have also done some field work. As expected of them, the civil society groups have taken an aggressive stand, voiced strong concerns and questioned the regulatory mechanism and the machinery.

Most of the scientific issues raised and questioning the authenticity of the data generated and their interpretation, appeared to be valid.

The Centre for Sustainable Agriculture (CSA), Hyderabad, has shown that in the case of Bt Brinjal in Andhra Pradesh the supply chain was contaminated with the Bt brinjal from the trial being sold in the local market which clearly highlighted the absence of fool-proof monitoring mechanism. The data from only one farmer in one trial among the 17 field trials on Bt brinjal carried out over two years was investigated. A similar study all over India, if it had been carried out by any civil society/ Government organization, would have provided valuable data.

The civil society recorded the farmer's observation that the colour of the Bt Brinjal would change (fade) as the day progressed and thus would fetch a lower price. Such critical observations have not been documented either by the company or in the ICAR-coordinated field trials. However to stand scientific scrutiny, such new observations have to be consistently documented.

Regarding the field trials, information as to which farmer is conducting the trial, where and when is he conducting it etc., is not in the public domain. Though the GEAC is supposed to be informed and they in turn alert the State Governments about the GM crop trial, it appears that only after the crop season is over, the names of the farmer/location is released by the company. This is a clear violation of precautionary principle as no purpose would be served when the crop season is over. Much of critical data would go unrecorded. The civil society has done a good job in bringing these issues to the notice of GEAC.

The civil society had also brought to the notice of GEAC various aspects of Bt Brinjal that need to be re- examined such as pollen flow studies, agronomic trials, soil impact studies, toxicity and allergenicity tests, food cooking & protein estimation studies, biodiversity issues, socio-economic impact assessments, rights of the farmers and consumers etc, and, finally, whether India needs a Bt brinjal, when other alternatives are available.

IV. ISSUES BEYOND BIOSAFETY

1. Concept of "genius loci" (spirit of the place)

This concept refers to the sensory food quality including smell, taste and appearance, which is specific to a particular location and which lends a particular preference in the minds of users/consumers to the product/location. This is especially true for brinjal in the Indian context. For example, the brinjal grown in Matti village in Udipi distirct, Karnataka has been considered "sacred" and having special taste since 15th century and even today its special taste is recognized and relished by consumers of brinjal in parts of Karnataka.

2. Holistic approach

Introduction of Bt Brinjal or for that matter any GM food crop calls for a holistic approach, rather than a reductionist approach. This is particulary so in view of the fact that India is the home for this vegetable and its cultivation goes back to a few millennia.

In the reductionist approach, the concept of food quality is cleaved. In the holistic approach it can be considered that the overall food quality is composed of three components of measurable quality (presence/absence of nutrients, toxins, microbes etc), consumer quality (consumers subjective assessment of foods in terms of colour, flavour, texture as well as emotional, social and ethical issues) and environmental quality (whether its production contributes to the maintenance of a sustainable and bio diverse system of agriculture).

3. Precautionary approach

One of the frequently quoted definitions of precautionary principle is "when an activity raises threats of harm to the environment or human health, precautionary measures should be taken even if some cause and effect relationships are not established sufficiently". It often guided decisions on food safety. The principle is also part of Cartagena Protocol. Uncertainty and variability in the risk assessment process are well known. Satisfactory methodologies to measure possible long term health effects or unintended/unexpected adverse effects of GM foods are yet to be evolved. Under these circumstances it is prudent to follow a precautionary approach.

4. IPR issues

Impact of pushing the brinjal crop into an IPR regime and the consequences of the same for farmers' rights (in using their own seed will be jeopardized; perpetual dependence on the external agencies will be created) and the resulting consequences on the consumers need to be assessed.

5. Issue of Origin of Brinjal

The issue of Indian subcontinent being the Centre of origin for Brinjal (Paroda and Arora, 1999) is of considerable relevance here. According to these authors, the wild forms of brinjal, *Solanum incanum* and *S. melongena* var. *insanum* are distributed from peninsular region to the foot of the Himalayas. Under these conditions the wisdom of introducing Bt Brinjal is to be considered carefully.

The Cartegena protocol, to which India is a signatory, recommends a cautious approach to impact assessment with regard to transgenic crops in their Centres of Origin. The need for Bt Brinjal should be re-examined against this background.

In Mexico, the centre of origin of maize, two legal requirements i.e. a map of the depicting the centers of origin and genetic diversity of maize, and a program outlining measures to protect native species, have to be met before approval for field trials for GM maize could be granted.

6. Available alternatives

The main argument for advocating Bt Brinjal by the developers of this technology is to reduce pesticide use. In India, considerable progress has been achieved through Integrated Pest Management [IPM], Non-Pesticidal Management [NPM], pest control through use of non-chemical products, inter cropping, crop rotation, trap cropping and Organic farming all of which would address the pest management issues in Brinjal.

V. SUGGESTED FUTURE COURSE OF ACTION

1. Epidemiological study on allergenecity (occupational/accidental/deliberate consumers)
2. Studies on Indian farmers' perception of risk and benefits and cost-benefit ratio to be carried out in a systematic way, preferably by Indians
3. Data on human/animal consumption of brinjal
4. Implementation of mandatory regulations and evolving mechanisms for the purpose
5. Revising guidelines as per latest recommendations of the ***Codex Alimenatrius*** commission under the UN system
6. Ministry of Health, with the help of Indian Council of Medical Research (ICMR), to evolve and implement guidelines on biosafety. Ministry of Agriculture with the help of Indian Council for Agricultural Research (ICAR) to evolve and implement guidelines on field trials and cultivation. Evolving guidelines should not be confined to the organization meant for promoting Biotechnology.
7. Guidelines should be provided to the state Governments on the issues related to GM crops and foods.
8. Labeling issues and providing choice for consumers need to be worked out. This is extremely important in the Indian context where most of the produce is sold in scattered unorganized markets with very little possibility of implanting any effective labeling mechanism.
9. All the data, both scientific and civil society-related, on Bt Brinjal must be made available in the public domain ensuring absolute transparency.

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