

torch cell you throw away when once the zinc casing has mostly dissolved away.

I am not certain of the ergonomics of the 'biobattery' but it would really have been impressive if the authors had considered the possibility of methane being cogenerated with the hydrogen near the anode and managed to demonstrate either that it happens or that it does not. If methane is formed at all under the relatively aerobic conditions of the 'biobattery', it would have been an 'advance' on (not really a mimic of) the as yet poorly understood process of biomethanation which takes place only under the exclusively anaerobic conditions of the rumen (of cows, sane or insane) or biogas digester. Or, there would have been real novelty to the authors' efforts had their aim been to isolate and immobilize an

oxidizing enzyme from a natural source, one that utilizes oxygen from the air to oxidize the hydrogen produced near the anode. Commercially viable or not, replacing manganese dioxide by such an enzyme for overcoming 'polarization' would have been a real breakthrough for developing a 'biobattery'. Maybe, the electric eel has all the answers – it utilizes energy from combining what it ate of the prey that it has stunned with an electric shock with the oxygen dissolved in seawater, having met the challenge of inventing a true 'biobattery' millions of years ago!

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This is with reference to the article 'Biobatteries to utilize bioenergy from fruit and vegetable wastes'. The item is misleading because the source of electrical energy in these batteries is the dissolution of Zn. The standard potential for the Cu/Zn couple is 1.1 V and it is not surprising that when these two electrodes are immersed in a paste of vegetable or fruit waste, a current is observed. The readers of *Current Science* deserve better.

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NEWS

R&D efforts in industry – National awards for 1996 presented

In order to provide recognition to the efforts of industry towards innovative research and technological development, the National Awards for R&D efforts in industry were instituted in 1987 by the Department of Scientific & Industrial Research (DSIR). These awards are in the form of silver shields and are presented along with citations at the inaugural session of the 'National Conference on in-house R&D in Industry', held annually by DSIR.

Professor Yogendra Alagh, Union Minister of State for Science & Technology presented the shields on 10 December 1996 in New Delhi to the

following seven winners of awards for 1996:

- **Armour Polymers Limited, Mumbai**, for the development and commercialization of the amoxidation process for the manufacture of cyano pyridines.
- **Hetero Drugs Ltd., Hyderabad**, for overall bulk drugs and intermediates.
- **Lamco Lightning Arrester Mfg. Company Pvt. Ltd., Hyderabad**, for the development and manufacture of lightning arrester disconnectors.
- **Patwa Kinarivala Electronics Ltd., Vadodara**, for the development of a

range of instruments and electronic control systems for the textile industry.

- **Titan Industries Limited, Hosur**, for the design, development and commercialization of ultra-thin quartz analog watch movements.
- **Ankur Seeds Pvt. Ltd., Nagpur**, for breeding superior hybrids of cotton, okra, chilli, brinjal, cucurbits and sunflower.
- **Jain Irrigation Systems Ltd., Jalgaon**, for fully absorbing technology, imported for micro-irrigation and sprinkler systems, and improving it by their own R&D.

RESEARCH NEWS

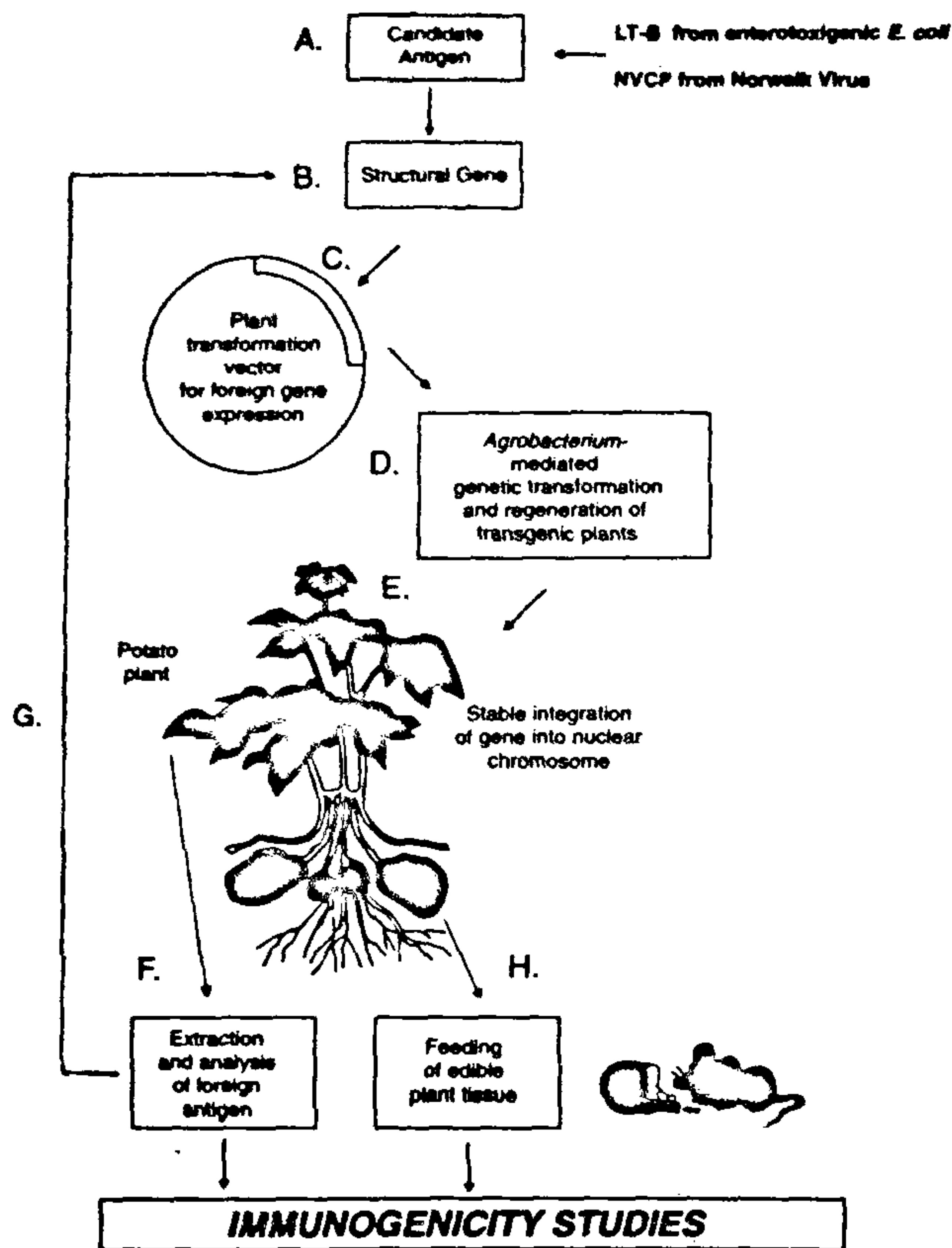
Vaccine production in transgenic plants

P. Suprasanna, T. R. Ganapathi and P. S. Rao

The development of novel techniques of tissue culture and molecular biology to genetically transform plant species has revolutionized plant biology, and there is an enormous interest to design and produce plant species with desirable characteristics such as insect resistance, disease resistance, herbicide tolerance and delayed fruit ripening. The notable achievements

in these areas led to explore the possibilities of employing transgenic plants with modified biosynthetic potential for new products¹. Biologically active peptides and proteins have many potential pharmaceutical applications including use as vaccines, immuno-modulators, growth factors, hormones, blood proteins and enzymes². There has been a recent interest

in this direction in producing functionally active proteins, peptides of medical importance, in transgenic plants. On a global scenario, the developing countries are often the target of several infectious diseases. Preventive medicine (for example, vaccines) has proceeded rapidly in the last decade as biotechnology has been applied in several areas. These medical



- A. Optional factors for choosing a candidate antigen
 - Purified protein is sufficient to promote immunity to the disease.
 - Purified protein is orally immunogenic.
 - The gene for the protein is cloned.
 - The protein maintains its antigenic epitopes and assembles into its native form when expressed in a heterologous system.
- B. Factors to consider in selecting a gene for plant expression systems
 - Modification of the gene with plant specific regulatory elements.
 - Analysis and reconstruction of antigen-coding sequence for optimal plant expression.
- C. Create a plant transformation vector
- D. Transform plant tissue
- E. Generate plants for analysis
- F. Analyse recombinant protein for antigenic epitopes and proper assembly within plant tissue
- G. Optimize production of recombinant protein in plant tissue by redesign of gene or transformation vector
- H. Test oral immunogenicity of the vaccine-containing transgenic food
 - Test for induction of an immune response to the recombinant protein.
 - Challenge immunized animals (if possible) with the disease to determine if the candidate vaccine can induce a protective immune response.

Figure 1. Production of candidate vaccines in plants is shown schematically along with factors to consider for various steps. (From Richter *et al.*⁵; Reproduced with permission from Dr C. J. Arntzen.)

advances are not likely to have significant impact in developing countries because of prohibitive costs of production and delivery of the recombinant proteins which comprise the new vaccines (Arntzen, pers. comm.). The economic conditions and inaccessibility to world markets also limit the use of vaccines for immunization to large population. In this regard, the concept of vaccine production in transgenic plants assumes greater significance for a relatively low-cost, agriculture-based system, rather than the sophisticated and expensive cell culture and fermentation-based vaccine production.

The concept was first introduced by Charles J. Arntzen and his group in a report of vaccine production in tobacco plants genetically engineered with plant-derived recombinant hepatitis B surface antigen (rHBsAg) (ref. 3). In this study, they have demonstrated that the HBsAg was expressed in tobacco and the partially

purified protein when injected into mice, elicited antibody response, similar to that obtained with 'recombivax' (a commercial hepatitis B vaccine). This study clearly demonstrated close-relatedness of the antigens and also the potential of producing immunologically active vaccines in transgenic plants.

Potential recombinant vaccines for viral and non-viral organisms have been produced in transgenic plants. Norwalk virus causes epidemic, acute gastroenteritis. The recombinant capsid protein (NVCP) was expressed in transgenic tobacco leaves and potato tubers⁴. The capsid protein could be extracted from tobacco leaves in the form of 38 nm Norwalk virus-like particles. rNV expression levels up to 0.23% of total soluble proteins in tobacco leaves and up to 0.37% in potato tubers were obtained. The partially purified recombinant NVs from tobacco were given orally, and potato tubers expressing NVCP were fed to mice. In both the cases,

stimulation of antibody (IgG) specific to rNV was noticed. This study also demonstrated that oral consumption of recombinant plant tissues can evoke specific immunoglobulin production, furthering the concept of 'edible vaccines'.

In another study⁵, tobacco and potato were transformed with genes encoding the binding subunit of *E. coli* heat labile enterotoxin (LT-B). Worldwide, enterotoxigenic *E. coli* and *Vibrio cholerae* cause acute watery diarrhoea by colonizing the small intestines. Transgenic plants having the LT-B gene expressed the protein similar to the bacterial protein. Transgenic tubers when directly fed to mice, elicited the development of both serum IgG and mucosal IgA antibodies to LT-B, in contrast to no LT-B specific antibodies in mice fed with non-transformed tubers.

Research on transgenic plant-based production of vaccines has just begun and already studies on optimization of accu-

mulation of recombinant proteins in edible plant tissues and feasibility including economics of such approaches are being made. These investigations generate a hope towards developing low-cost edible vaccines from plants that are commonly available, compared to expensive recombinant vaccines. In order to be applicable to large section of population in a edible and un-cooked form (to ensure, there is no denaturation), production of vaccines in candidate crops like banana can be

invaluable to many tropical countries in the developing world where these are grown, richly available at lower costs and consumed as staple food.

1. Mason, H. S. and Arntzen, C. J., *TIBTECH*, 1995, **13**, 388-392.
2. Lyons, P. C., May, G. D., Mason, H. S. and Arntzen, C. J., *Pharma News*, 1996, **3**, 7-12.
3. Mason, H. S., Lam, D. M. K. and Arntzen,

C. J., *Proc. Natl. Acad. Sci. USA*, 1995, **92**, 3358-3361.

4. Mason, H. S., Ball, J. M., Shi, J-J., Xi, J., Mary, K. E. and Arntzen, C. J., *Proc. Natl. Acad. Sci. USA*, 1996, **93**, 5335-5340.
5. Richter, L., Mason, H. S. and Arntzen, C. J., *J. Travel Med.*, 1996, **3**, 52-56.

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SCIENTIFIC CORRESPONDENCE

Assessment of plant bio-diversity at Kalakad-Mundanthurai

T. Ganesh *et al.* (*Curr. Sci.*, 1996, **71**, 379-392) have generated interesting phytosociological information for the Tiger Reserve in the Agasthyamalai range.

While highlighting the importance of quantitative analysis in an area of 3.82 ha, the pioneering work of Champion¹ and Puri *et al.*² is referred to as based on qualitative criteria such as physiognomy and dominant species, ignoring quantitative data.

Champion's treatise on the vegetation types came out in 1936, later on revised by Champion and Seth in 1968 (misquoted as 1936 in the reference). This study covered the entire sub-continent. The idea was to come out with a classification good for the country as a whole, with spot descriptions. Significantly, Champion chose to call his *magnum opus* 'a preliminary survey'. Had he confined himself to small plots with quantitative statistics, the concept of which barely existed in his time, perhaps, the publication would not have yet seen the light of the day.

Contribution of Puri *et al.* of a later date (1983) relies on the vegetation maps of the French Institute at one millionth scale based on the criteria of dominance-abundance-fidelity of species. The series recognized for the Kalakad-Mundanthurai region is *Cullenia-Mesua-Palaquium*. It is within this series that the quantitative study has established *Cullenia-Alagaia-Palaquium* sub-type considering an area of 3.82 ha.

The question is for how large an area

would this sub-type remain valid, latitudinally and altitudinally? How many sub-types founded quantitatively go into one series, viz. *Cullenia-Mesua-Palaquium* of the southern part of the Western Ghats? How many hundreds (or thousands) can be counted in Peninsular India? What is good for a 4 ha study carried out in considerable detail may not be manageable at the level of the sub-continent. The question of scale should not be lost sight of.

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Reply by T. Ganesh *et al.*:

We recognize the pioneer and monumental contributions of Champion¹ and Puri *et al.*² to the description of forest vegetation in India, but reiterate that plant ecologists in India must move beyond qualitative description of vegetation, particularly in light of the current need for assessment and monitoring of biodiversity. Our paper represents an effort towards quantification of tree diversity in an area that is poorly known, but has a high level of biodiversity. Incidentally, Meher-Homji and his

colleagues, notably Pascal³ from the French Institute, themselves have been pioneers in collecting quantitative data on the distribution of tree species in plots throughout the Western Ghats.

We also wish to point out that the 3.82 ha that Meher-Homji refers to is a sub-sample of an area over 100 ha. Nevertheless, his question for how large of an area would this sub-type remain valid is an appropriate one. Although we recognized the forest type as *Cullenia-Aglaia-Palaquium* type, we did so in the context of types and sub-types established by Champion and others. We now feel that the concept of types and sub-types based on dominance of certain species is of limited utility, and may not be valid in many cases. Plant communities change over time and space. The static description of vegetation does not capture the dynamics of natural communities. Understanding dynamics is key to long-term conservation of biodiversity. In fact, one of the points of our paper is that our results do not conform to the vegetation described for the area because the vegetation is expected to vary over small spatial scales. Therefore, quantitative studies are required to document patterns of diversity and long-term monitoring is essential to understand dynamics. Regrettably, in trying to make our point about changes in community structure over space and time, we fell into the trap in claiming the existence of another sub-type. Thus, we are very grateful to