A report on the VIII International Congress of Plant Tissue and Cell Culture

Organizing an international congress every four years is a significant activity of the International Association of Plant Tissue Culture. Its main purpose is to present advances in the state of the art and stimulate new research and find solutions to difficult problems. The previous two congresses, held in the US (1986) and The Netherlands (1990) were attended by over 2000 scientists. However, only 1300 scientists from 55 countries were registered for the present congress, held in Firenze, Italy, from 12 to 17 June, 1994. The fall in attendance could be attributed to two main factors: (i) many scientists had to opt between this congress and the one on Plant Molecular Biology scheduled to be held one week later in Amsterdam, and (ii) world-wide financial crunch. The organizers were unable to provide any financial support to the participants, not even waiving of the Registration Fee. This could have dissuaded the participation of younger scientists, especially from developing countries, including India. Countries such as Bangladesh, Nepal, Pakistan and Sri Lanka were totally unrepresented. My participation in the congress was made possible by pooling partial travel grants from the University of Delhi, CSIR and DST. The purpose of this article is to highlight the proceedings for the benefit of Indian investigators in this field and to enlighten those who are working on crops crucial to our economy.

The programme comprised 7 plenary lectures, 20 symposia and keynote addresses, 14 workshops and 2 poster sessions. The posters contributed more original information than oral presentations at the symposia. Of the twenty symposia the following four included over 100 reports: (i) *in vitro* regeneration – 189, (ii) genetic transformation – 128, (iii) somatic embryogenesis – 118 and (iv) micropropagation – 105. Considerable emphasis was laid on cereals and brassicas as experimental materials. As three symposia and five workshops were held concurrently, it was not possible to attend all the sessions. Understandably, this report is based on the sessions attended by me.

In the opening plenary lecture, I. K. Vasil presented the success story of genetic transformation of wheat, largely based on the pioneering work done by him, Vimla Vasil, and other members of his group. Several other papers also described heritable transformation achieved in wheat. There was a clear message in the Congress that rice and wheat, the two most important food crops of the world, are now amenable to molecular breeding. This has been possible largely due to the establishment of embryogenic cultures of cereals and the use of the particle gun method of DNA delivery. Indeed, with the available methods of gene delivery, the insertion of desirable foreign genes into plant cells has become a routine laboratory exercise. Some of these methods do not require the rather arduous step of plant regeneration from isolated protoplasts or cells. A good protocol for regeneration from explants, which is now available for a large number of crop plants, is all that is required to obtain transformation. The major limitation, however, is the availability of agronomically useful genes and their molecular characterization. A word of caution raised in one of the workshops during the congress was that one or two years of field testing of transformed plants may not be sufficient to claim stable transformation as the introduced trait may be lost even after 5–6 sexual cycles.

Isolation of male and female gametes of maize, their controlled fusion and regeneration of fertile plants from the naked 'zygote', reported by Kranz and Lörz (Germany) can be considered a major breakthrough in the area of sexual reproduction in higher plants. In the same symposium, Holm (Denmark) reported the spectacular success of his group in regenerating fertile plants of barley and wheat from mechanically isolated fertilized eggs by co-culturing with the microspores of barley, undergoing embryogenesis. In both these studies, unfertilized eggs failed to divide *in vitro*. It may now be possible to use the isolated zygote for genetic transformation by electroporation. *In vitro* fusion of isolated egg and sperm cells (IVF) extends the scope of wide hybridization. This system should enhance our basic knowledge about the intricate mechanism of gametic fusion in higher plants. The ability to culture isolated zygotes and early globular embryos should be expected to add to our understanding of the nutrition of embryos at early developmental stages.

Anther and pollen culture of *Brassica napus* has been highly successful, as pollen embryos are being used for genetic transformation. The major gain in this technique is the rapid production of homozygous transformants. Lörz (Germany) reported the production of homozygous transgenic plants of barley by transforming microspores before they were cultured for androgenesis. Our recent communication (Bhojwani and Agarwal) about enhancing the androgenic response in anther cultures of *Brassica juncea* to 18% (as against the earlier reports of 3%) invoked considerable interest. It is now possible to attempt isolated pollen culture of this crop and use it for genetic transformation. A private seed company in Canada is starting work on conventional and molecular breeding of *B. juncea* to develop cultivars suited to dry areas.

Somatic embryogenesis was reported in several species but the success has been largely with embryonic explants. The treatments used to induce embryogenesis are highly varied, suggesting that we still do not fully understand the mechanism of induction of this important process, regarded as a potential method for large-scale propagation of plants. Pilot-scale propagation of *Coffea* (Sondahl, USA) and *Clematis* (Weber, Germany) by somatic embryogenesis in 1–5 litre capacity bioreactors has been successfully accomplished. Merkle (USA) described somatic embryogenesis in a number of tree species belonging to the genera *Magnolia* and *Liriodendron*. Direct planting of somatic embryos in perlite was not only reported to be more successful but yielded better quality plants, with profusely branched root system, than seedlings raised on agar medium.

An important development was the realization that liquid medium is superior to agar medium for micropropagation, including shoot proliferation. This not only reduces the cost of production but also opens out the possibility of using bioreactors. Teisson (Belgium) described a culture system involving temporary

immersion of the cultures in liquid medium which improves the quality of plants multiplied through somatic embryogenesis as well as shoot proliferation. Immersion of cultures four times a day, for 15 min each time, gave excellent results with a wide range of crop plants (*Citrus*, *Coffea*, *Eleis*, *Hevea* and *Musa*). The use of robots has not made the expected breakthrough in micropropagation (Debergh, Belgium).

Although date palm micropropagation has been commercialized, several laboratories are involved in developing more efficient protocols for somatic embryogenesis in this crop. Scientists from the Philippines reported success with the micropropagation of coconut palm from immature embryos (Orense *et al*) and rachilla explants (Ebert *et al.*). This finding should be important for India as coconut is an important, open-pollinated crop with erratic yields and all attempts for *in vitro* propagation have so far been unsuccessful. High frequency regeneration of shoots (some of which also rooted) from the callus derived from microcuttings of cashewnut, reported by Bessa (Portugal), should be of equal interest to Indian scientists.

Disease-free micropropagated banana plants have been produced in Taiwan at the rate of 2 million per year. So far over 15 million plants have been distributed for field plantation throughout the country, which has stabilized their banana export industry.

It was gratifying that the workshop on 'Technical Problems in Plant Tissue Culture', chaired by me, was most well attended. Problems of systemic infection and use of antibiotics were discussed at length. The consensus was that it is impossible to establish bacteria-free cultures and undue concern should not be directed to eliminate benign bacteria in cultured tissues or regenerating plants. The use of antibiotics to control harmful bacteria should be based on a detailed study of the nature of bacteria and their sensitivity to various antibiotics. Some bacteria may be useful for the growth of plant tissues. Use of bactericidal compounds other than the traditional antibiotics, such as neem products, to contain this problem was also recommended. Nairn (New Zealand) presented experimental evidence to suggest that the hyperhydration effect of gelrite is due to its constituent, sulphated galactans.

The next Congress of Plant Tissue and Cell Culture will be held in 1998, in Jerusalem, Israel.

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SCIENTIFIC CORRESPONDENCE

Reproductive biology: An aid in the classification of bamboos

The importance of bamboos to the national economy as an industrial raw material, besides their more than thousand uses in day to day life, is well known. Bamboos have fascinated scientists and laymen for centuries. To laymen all bamboos look similar but there are more than 1200 species described under nearly 50 genera. Most of the woody bamboos (in which there is no sporadic flowering) flower and seed after an exclusive vegetative growth for a species specific supra-annual interval, ranging between 3 and 120 years[1]. There are two types of flowering: (i) gregarious and (ii) sporadic. In gregarious flowering all members of a cohort (plants from seeds of common origin) enter the reproductive phase approximately at the same time, and after flowering and seeding the parents die *en masse*. This death of the bamboo parents used to be given more importance, probably because of their long intermast periods and arborescent habits. It has to be considered as a character bamboos share as members of the grass family[2]. In sporadic flowering, members of a cohort enter the reproductive phase at different times, or at irregular intervals, and after flowering (and seeding?) the parents do not die but revert to vegetative growth. Due to this peculiar flowering behaviour in bamboos, flowers and seeds are available only at very long intervals. This has resulted in a poor understanding of their inter-relationships, besides making the perennial raising of plantations using seeds and hybridizations difficult. Selection is the only method available at present for bamboo improvement[3]. It is possible now to induce flowering in bamboos by tissue culture methods[4]. Induction of flowering *in vitro* can be used for perennial seed production and hybridizations. To plan a hybridization programme it is necessary that the inter-relationships between the bamboo species and genera are well understood.

Bamboos and herbaceous bambusoid grasses are grouped into the sub-family Bambusoideae or tribe Bambuseae under the family Graminae (Poaceae). Most botanists agree to this sub-family or tribe position. There are also arguments in favour of conferring an independent family status to this group[5]. Soderstrom and Calderon[6] are of the opinion that the bamboos and the bambusoid grasses evolved from a common stock. In herbaceous bambusoid grasses botanists have satisfactorily delimited the genera. In bamboos the generic delimitations still remain incomplete[7]. In 1913 Camus[8] proposed a modification of Bentham's[9] classification for Bambuseae. In 1935 Camus[10] expanded it and suggested seven tribes, *Arundinareae*, *Arthrostylidae*, *Chusqueae*, *Bambuseae verae*, *Hickelieae*, *Synandrae* and *Bacciferae*. He divided the tribe *Bacciferae* into four sub-tribes – *Dendrocalaminae*, *Melocanninae*, *Pseudocolxinae* and *Perrierbambusinae*. In Holttum's[11] opinion both the systems of classifications (Bentham's modification of Munro's and Camus') do not conform with the natural order. He stressed the need for a natural system of classification based on many characters, and proposed a system based on the structure of the ovary[12]. According to this system there are four types of ovaries: *Schyzostachyum*, *Oxytenanthera*, *Bam-*