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"REVOLUTIONARY" NEW GENE TRANSFER METHOD DEVELOPED

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A Canadian research team headed by an expatriate Sri Lankan scientist claims to have developed a simple and fast method to transfer plant genes.

It has been described as a discovery "that could revolutionise genetic engineering" and accelerate the improvement of a broad range of crops.

In 1989, the same research team, working at the Department of Crop Science in the University of Guelph in Ontario, Canada, won international recognition for developing what they called *"artificial seeds"* – dry plant cells that grow into genetically identical plants. These were produced using tissue culture methods.

"Artificial seeds" technique has already stirred excitement in plant breeding circles because it offers a simple and easy way to multiply rare plants.

Artificial seeds could also prove helpful in saving endangered species of plants, and enable horticulturists to exchange plant genetic material without fear of spreading plant disease.

"This (technique) will be particularly beneficial to developing countries, whose plant quarantine regulations are not so strict. The dried seeds, produced in sterile laboratory conditions, will not harbour disease–causative micro organisms," said Dr. Tissa Senaratna, the Sri Lankan plant physiologist heading the Guelph research team.

The team has now demonstrated an apparently simple method to insert a desired single gene into artificial seeds – by simply putting the seeds into water containing that gene !

As the seed soaks up moisture, it would pull the gene inside and adopt it. The gene's characteristics will be expressed in the plant that grows from the treated seed.

Dr. Senaratna calls it *"the surfboard phenomenon"*, drawing an analogy to a surfer riding the crest of a wave.

"It's very simple," he quipped. "I think it surprised everybody."

It certainly did surprise the first audience of scientists to hear his ideas and the results of his experiments at the International Tissue and Cell Culture Congress in Amsterdam last summer.

The process of gene transferring is crucial to gene splicing, one of the most prominent applications in genetic engineering since 1973. Gene transferring has enabled scientists to produce some of the marvels of molecular biology, such as human insulin produced by bacteria, plants that grow their own herbicides, growth hormones that increase milk production in cows etc.

Similarly, with such gene transference, it is possible to develop new characteristics in plants selectively and immediately. This contrasts with the time–consuming, imprecise method of cross–breeding practised for centuries.

Gene transfer systems so far involved one of three basic techniques : bacteria, micro–injection or ballistic.

The bacteria approach involves several steps: loading the desirable gene onto a bacteria; having the bacteria penetrate a cell of the target plant or bacteria; then having them dump that genetic load into the cell and having the cell adapt them into its genetic code.

So far, the bacteria approach has only worked with dicotyledons, such as beans, and not monocotyledons, such as wheat. Canada's agriculture features far more monocotyledon crops than dicotyledons, so any genetic engineering technique that works for monocotyledons will be of "immediate and particular interest to Canada's plant breeders," noted one Canadian newspaper which highlighted the development.

In the micro injection technique of gene transfer, microscopic surgical instruments, (such as a needle) are used to insert a gene into a plant. And in the ballistic approach, it involves loading the genes on tiny pellets that are literally blasted into a cell with a .22 calibre rifle.

Dr. Senaratna says his new approach depends on two factors.

First, the cell membrane has to open. That is *not* normal for ordinary cells, but does happen briefly with artificial seeds, both when they are drying down to become artificial seeds, and again when those dry seeds are moistened to develop into plants.

Second, the gene–carrying water has to move across that open membrane into the seed.

This is why the method would not work with ordinary cells. Ordinary cells have a membrane that acts like a skin to hold its components in place. This membrane does not open up, even briefly, like those in artificial seeds.

Dr. Senaratna, while satisfied with their progress so far, stresses that much more remains to be done.

To begin with, the artificial seed technique has so far been only tried and proven on a few species of plants – alfalfa, geraniums, microspores involved in canola–breeding programmes and embryos from black spruce trees.

Dr. Senaratna is confident that the artificial seeds technique will eventually work for many more species, including wheat. "But we haven't got around to trying it yet."

If the artificial seed technique works on a given plant, then the new genetic engineering system should also work. There too, more work remains to be done to determine whether it will work for a broad range of genes.

The Guelph experiments were done with the GUS gene – glucuronidase, a popular "marker gene" that yields an easy–to–see blue colour.

Dr. Senaratna's team, which included Drs. Bryan McKersie, Ken Kasha and Doug Procunier, had their first success with gene transfers in early 1990.

"We literally jumped up and down with joy, shouted and hollered when we saw it worked," Dr. Senaratna recalled.

The team has been going through exciting times since the artificial seeds breakthrough the previous year. Using soy been and a few other crops, they produced thousands of dry "seeds" from a single plant. Unlike natural seeds – the product of sexual reproduction in plants – "artificial" seeds were produced using tissue culture.

In tissue culture, researchers fiddle with individual cells taken from any part of a plant, inducing them to develop just like fertilised eggs. With the addition of proper nutrients and hormones, these cells ultimately develop like a "seed".

Tissue culture has been tried on many food crops, horticultural and forest tree species. It enables scientists to produce a large number of young plants without having to depend on naturally occurring plant reproduction.

However, no one had previously been able to dry these "seeds" from their test tubes, to make packaging and transport easier. Precisely this is what was achieved with "artificial seeds".

Once dried, "artificial seeds" can be packed into a seed coating using various nutrients. This protects the seeds from mechanical damage. Seeds can also be coated with substances which will imbibe water at a faster rate, giving them extra initial vigour – an edge over ordinary seeds.

When conditions are right, artificial seeds can germinate just like normal seeds. However, says Dr. Senaratna, they take "a few days longer" to grow.

Dr. Senaratna, now perfecting his gene transfer technique, believes that the animal kingdom might find some uses of his research with plants. For example, he notes that sperms can be frozen; he thinks that sperms may be dried, just as they were able to dry plant seeds.

If that proves possible, the new gene–transferring technique might work for animals and birds as well.

That, in turn, leads Dr. Senaratna to ideas already being explored by another researcher at Guelph. Dr. Jim Squires in the Department of Animal and Poultry Science has just begun experiments to test his idea that sperm cells could be a simple, easy and cheap way to accomplish genetic engineering.

But that is another story.

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(An earlier version of this article appeared in *The Island* in December 1990; it has since been edited and parts re–written).