

Using plants as human gene vehicles

By the FAAR Biotechnology Group, for the Canadian Food Inspection Agency

Even though Plant Molecular Farming (PMF) is an emerging biotechnology sector, the concept that genetically engineered plants can be used as vehicles for commercial or large scale production of valuable products is in fact not a new idea. Some of the very first transgenic plants ever produced were those in which the proteins that normally accumulate in seeds were altered to contain foreign amino acid sequences. The goal of these modifications was to provide commercial scale production of seed with increased nutritive content.

Once this concept is appreciated, it is a short jump to using seed-specific promoter elements to drive the expression of any number of different gene products. Any plant gene promoter can serve as a tool for expression of a foreign gene product in any number of different plant cells.

It is not the fact that plants can be used to express non-plant genes, or genes from other species of plants, that has recently made PMF more visible. It is the production of pharmaceutical or therapeutic agents that is often the key issue.

The possibility of using plants to produce human gene products is attracting growing media attention, which in turn is creating public concern.

The production of recombinant proteins for use in human medicine originated with

the discovery that foreign DNA could be incorporated into bacteria which could be used to express foreign genes. The first recombinant pharmaceutical products — insulin and interferons — were produced in transgenic *E. coli*. The interest in using such an approach is obvious because recombinant bacteria can rapidly produce large quantities of recombinant proteins at comparatively reduced costs in fermentation cultures.

But the ability of bacteria to make, assemble and correctly modify different types of mammalian proteins proved inadequate for more complex proteins.

Recombinant production systems in cells of higher organisms, beginning with yeast, were subsequently tested and implemented as they were able to process mammalian genetic messages accurately.

A recent survey suggests that at least 369 different recombinant biotechnology medicines directed to more than 200 different diseases are currently under development.

The US demand alone for biotech pharmaceuticals is expanding at the staggering rate of 13 per cent annually and is expected to reach a market value of \$US28.6 billion by 2004. About 100 of these products under development are vaccines.

Another area of enormous interest and

commercial potential is the production of therapeutic and diagnostic antibodies.

Currently the first six leading antibodies that are directed against bronchial pneumonia, anti-platelet GPIIb, Non-Hodgkins lymphoma, breast cancer, Crohn's disease and kidney transplant rejection factors, represent a \$US1.5 billion market. It is anticipated conservatively that 100 such antibody products will be developed within the next five to seven years. Sales are projected to be \$US4 billion this year.

The outcome of this impressive growth is that the ability to produce these products by traditional fermentation methods will soon become the limiting factor. So the large-scale production of recombinant proteins and other products in plants is poised for rapid expansion.

Certainly plant-based systems will not be suitable for the production of all products but, with the tremendous increase in the demand for production, plant-based systems offer many advantages. Chief among these is cost of production (low capital cost for facilities) and safety (plants rarely harbour or transmit human viruses or other infectious agents).

In addition to the simple production of established therapeutic molecules, it is anticipated that many treatments may be tailored for individuals by specific expression of their own individual genes. This would allow, for example, the mass production of a patient's own antibodies. These could subsequently be harvested from plants and provided to enhance the patient's ability to fight a disease such as cancer.

The human genome project and other sequencing projects are providing new genes at an accelerated rate. The discovery of genes that are related to diseases, especially those where there are no current effective treatments, will result in an unprecedented increase in potential new drugs and therapeutic agents. The demand for recombinant protein production is expected to skyrocket.

So the issue of how molecular farming in plants might be accommodated and regulated is pressing.



Plant-based systems to produce therapeutic and diagnostic antibodies have the two big advantages of comparatively low production costs and clinical safety. (PHOTO: Medicago)

WHAT PMF PRODUCTS?

Although the production of drugs and other health care products is the most talked about and controversial aspect of molecular farming, it is clear that plants can be used as production vehicles for any number of products. It is true that plants may not always be able to make specialised protein modifications exactly as in human or animal counterparts — but there is no specific limitation to the production of any gene product in plants.

Products that are manufactured may have an animal or other origin, but it is anticipated that many of the new biotech medicines will be made from entirely synthetic peptide or protein sequences which cannot be classified as either plant or animal. The number of different commercially valuable products that can be made correctly in plants is not known, but DNA sequences from animal or other genes can often be modified slightly to be more easily ‘read’ by plant cells.

Table 1 is a summary of the types of PMF products that could be made.

Crops under consideration for production purposes

The procedures used to grow, harvest and process the plant material will also influence the degree to which humans, animals and the environment are exposed to the molecular farmed products. The processing of material on site, methods chosen for transportation off site, either processed or unprocessed, and disposal of residual biomass are other considerations.

A further and critically important consideration is the species of plant chosen as host for molecular farming activity. This will determine the potential for a molecular farming product to come into contact with humans, animals or the environment. In addition, the use of food and feed crops for molecular farming will raise the issue of accidental mixing, since most plants being used for production of valuable products will be visually indistinguishable from non-modified crops of the same species.

Another consideration is the technology required to use a specific crop species for manufacture of a product. This includes husbandry of the crop as well as the technical considerations related to introducing the new gene.

New technology means that the range of plant species that can be transformed is now essentially unrestricted, provided that appropriate tissue culture methods are available.

For obvious reasons, more information

TABLE 1: Possible molecular farming products

Primary products
Antibodies, antibody fragments
Enzymes: industrial; therapeutic; diagnostic; cosmetic
Structural proteins: peptides; hormones
Antigens (vaccines)
Anti-disease agents, drugs
Enzyme inhibitors
Derived products
Bio-plastics
Vitamins, co-factors
Nutraceuticals
Secondary metabolites: phenolics; glucosinolates; tannins; starches; sugars; fragrances; flavours; alkaloids
Fibres

is known about transformation of commercially important species.

So one of the main considerations for regulatory oversight and determination of safety issues is the crop species being used for production of the product.

The scope of host plants could include:

- Major food crops such as corn, wheat, soybean, rice, canola and potatoes;

- Lesser food crops include beans, peas, vegetables and so on;
- Animal feed crops such as lucerne and grasses;
- Non-food crops like cotton, hemp and tobacco;
- Ornamental plant species;
- Forestry tree species; and,
- Aquatic plant species.

PROCEED WITH CAUTION

PMF is an emerging industry that promises many benefits and opportunities but presents many challenges.

If consumers and industry are to benefit from PMF, every effort must be made to ensure that the environment and human and livestock health are protected and that no unnecessary risks will ensue from movement of the foreign genes into other plants.

Issues such as the use of food and feed crops, and segregation from commodities, must be carefully examined.

Where movement of transgenes via pollen cannot be wholly prevented, assessment processes must be in place to ensure that any potential effects on human or animal health or the environment will have been anticipated and that commercial and regulatory activities are structured accordingly.

