

Commercialization of Plant-Derived Vaccines in Canada: A Distant Dream?

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Abstract

Sequencing of various pathogen genomes in the past two decades has ushered the discovery of recombinant DNA derived subunit vaccines to fight infectious diseases. Western Canada is an innovation hotbed for platform vaccine research and development as technologies created by NRC-PBI, VIDO, and universities. Since 1995, we have promoted the idea that subunit plant-derived vaccines (PDV) could emerge as third generation agricultural-biotechnology products. This idea represented a revolutionary advantageous technological platform, which should mitigate technical limitations and associated socio-economic challenges of traditionally produced vaccines in various animal or microbial cells.

The move beyond the proof of concept will prepare the groundwork for expeditious licensing of pioneer PDV and catalyze the prerequisite paradigm shift. Implementation of PDV introduction at the global level is urgently needed, especially for developing countries. Preliminary studies conducted in the local biotechnology cluster reveal some factors that retard the commercialization process. Initially, a lack of a fully mature regulatory framework for licensure alone presents a major hurdle. Protecting intellectual property (IP) requires careful management of planning and financial resources. Also, the public acceptance of these “innovations” can be sensitive and public acceptance of the new paradigm unpredictable. Ideologically the two sides could clash, considering proponents of PDV would never wish PDV end up being rejected like GM crops in Europe.

PDV as an alternative, if not a complement, to the conventional vaccines have defined challenges to their introduction to the marketplace. We will present a priority list of a pathway that identifies challenges and will try to offer solutions to deal with these issues globally. The

research will add to the growing body of literature pertaining to regulation, IP protection and social issues of novel biotechnology products moving from lab to the commercial marketplace.

Introduction

Vaccines are considered to be one of the most successful and cost-effective weapons against infectious diseases to this date.¹ Currently vaccines have been developed using mammalian cell lines, yeast and the common enteric bacterium *Escherichia coli*. But mammalian systems (cell and tissue culture) have been traditionally plagued by low-scalability and contamination with animal viruses and prions. Yeast cells are competitive but perform poorly with respect to certain post-translational modifications (e.g. glycosylation) in comparison to higher eukaryotes.

For worldwide success an ideal vaccine should address issues such as availability, accessibility, costs, activity, stability, effectiveness and amenability for combination vaccines. Plants as a production platform when compared to other systems stand at a much better position in light of the above reasoning. Plant systems are scalable to agricultural plots or farms, are not infected by animal viruses or prions and are capable of complex glycosylation. Research in this field for over a decade has solved some of the problems with further work in progress.

Experts working in this field believe plant-derived vaccines (PDV) will not supplant many conventional vaccine technologies, but will probably provide an alternative and economical system for production of vaccines with promising application in underdeveloped countries.² The efforts involved in commercialization of PDV have yielded mixed results with a few products in the early clinical trials and many in the preclinical pipeline. Yet, even after decades

of research and development a PDV candidate for humans has not reached the marketplace. This makes us wonder about the barriers facing commercialization of PDV. Obstacles to commercialization of biotechnology products have been discussed in detail including competitiveness and lessons to be learned.³

We have been investigating barriers to commercialization of plant-made pharmaceuticals (PMP), including PDV. Currently we are engaged in interdisciplinary research with the following objectives:

- Analyzing commercialization attempts of PDV in Canada,
- Anticipating and creating a priority list of challenges facing the commercialization process and where possible, offering feasible solutions, and
- Studying challenges from three perspectives: regulatory framework, intellectual property rights and commerce, and society.

This research should add to the growing body of literature pertaining to regulation, IP protection and social issues of novel biotechnology products moving from lab to the commercial marketplace.

Research and Development Background

Selected milestones

Since the reporting⁴ in 1992 of expression of a vaccine antigen (Hepatitis B surface Antigen/HBsAg) in a transgenic plant many R&D milestones have been reached. In 1994 the first confined field trial of GM canola expressing pharmaceutical in Canada took place.⁵ The Canadian Food Inspection Agency (CFIA) held a series of consultations from 2001 until 2006 with various stakeholders. In April 2003, Canada implemented a directive for confined field trials of Plants with Novel Traits (PNT) intended for Plant Molecular Farming (PMF),⁶ the framework of which will be discussed in detail later.

Being the first to reach an important milestone of getting an approval (January 2006) for a plant-made vaccine (PMV), Dow AgroSciences demonstrated that this new technology fits within the existing US Department of Agriculture (USDA) Center for Veterinary Biologics regulatory approval process.⁷ Dow's poultry vaccine is against Newcastle Disease Virus (NDV) and is expressed in plant cell culture.

Researchers in this particular domain are targeting diseases that infect through the mucosal system such as tuberculosis, pneumonia, influenza, diarrheal diseases, sexually transmitted diseases, HIV, etc. The principal reasoning behind targeting these diseases is that plant-based vaccines stimulate the immune response at the mucosal level and thus would be especially effective against them.⁸ Antigens from a variety of sources including viral (e.g. HBsAg, HIV), bacterial, enteric pathogens (e.g. Enterotoxigenic *E. coli* heat labile toxin B), non-enteric pathogens and self-antigens (e.g. immunocontraceptives) have been expressed in plants.⁹

Research and Development in Canada

There has been work on PDV in Canada for a considerable time. Many groups have been engaged in the production of a human cytomegalovirus (HCMV) subunit vaccine in plant and plant parts. The University of Ottawa, Health Canada, the Canadian Red Cross Society, Agriculture and Agri-Food Canada (AAFC), Prairie Plant Systems Inc. and others have been involved at times in the R&D and testing efforts.¹⁰

Another effort that has brought together AAFC, University of Guelph's Department of Plant Agriculture (formerly Crop Science), Ontario Veterinary College (OVC) and Ontario Research Enhancement Program (OREP) was a project for production of an oral vaccine for Porcine Reproductive and Respiratory Syndrome (PRRS) virus in a non-food crop bioreactor system.¹¹

In Saskatoon, AAFC has also been involved with the Vaccine and Infectious Diseases Organization (VIDO) on a project to produce an oral vaccine for Porcine Parvovirus (PPV) of swine produced in transgenic plants.¹² AAFC was also involved with the Canadian Department of National Defence (DND) in expression and processing of influenza and encephalitis vaccines in plant cells.¹³

There are instances where collaborations between Universities and private companies have taken place in Canada. One such example is execution of a license agreement between the University of Guelph and Dow AgroSciences LLC that provides Dow AgroSciences with exclusive rights to the leukotoxin antigen LKT-50 to develop plant-cell vaccine to protect cattle from shipping fever.¹⁴

Framework

A regulatory oversight is needed to mitigate the several risks involved during production and delivery stages of PDV, with potential impact on the environment and on human

health. Canada is claimed to have one of the safest, most effective regulatory systems for biotechnology products in the world and the Government continues to assure Canadians that the products and processes of biotechnology are subject to the highest standards of scientific testing for health, safety, and environmental impact.¹⁵ The regulatory system is science based, requires firms to deliver raw data and summaries of all tests performed and delivers consistent decisions.¹⁶ This science-based system is in line with principles laid out by global organizations such as the World Health Organization (WHO), the Organisation for Economic Co-operation and Development (OECD), the United Nations Environment Programme, the Food and Agriculture Organization (FAO), the International Plant Protection Convention (IPPC), the Codex Alimentarius Commission, and the Office International des Épizooties.¹⁷

Depending on the stage of production/disposal, regulation of PDV is largely shared between the federal departments of Health, Environment and the CFIA (federal agency). The Health Products and Food Branch of Health Canada is responsible for regulating these products under the *Food and Drug Act* and is usually involved during clinical trials and pre- and post-marketing of biological products. CFIA's Plant Biotechnology Office is responsible for environmental release of plants with novel traits (PNT) intended for plant molecular farming (PMF). CFIA regulates these products for confined and unconfined environmental release under the *Seeds Act* and the Seeds Regulations (Part V) and safety assessment for use of by-products as feed is carried out under the Feeds Act and Regulations. For conducting confined research field trials the CFIA issued interim amendments to its Directive 2007-08 to address the additional potential risks confined research field trials of PNT intended for PMF may pose to the environment. As a result, under these amendments applications for confined research field trials of such PNTs are assessed on a case-by-case basis only.¹⁸ The CFIA advises that avoiding feed/food crops may simplify the path through the regulatory process¹⁹ and recommends establishing a licensing system for "molecular farmers".²⁰ The Government of Canada is investigating policy options for commercial plant molecular farming.²¹ To date, no PNT intended for PMF has been granted authorization for its commercial cultivation in any Canadian jurisdiction.

Analysis

We started our initial investigation focussing on Saskatoon. Saskatoon is home to Canada's largest and fastest growing agricultural biotechnology cluster with about 30% of this sector's national activity.²² This cluster is home to more than

40 companies (2007 statistics) engaged in agricultural biotechnology R&D and builds on the research potential of the University of Saskatchewan (UofS) (including VIDO), AAFC Saskatoon Research Station, National Research Council-Plant Biotechnology Institute (NRC-PBI) and others.

Guardian Biotechnologies

Guardian Biotechnologies Inc. (Guardian) is a privately owned company incorporated in 2002²³ located in the Industrial Partnership Wing of NRC-PBI facility at Saskatoon. It is involved in molecular farming of vaccine antigenic proteins (poultry diseases) in plant and plant parts. Guardian has agreement with VIDO (animal tests), partnership and collaboration with the Universities of Saskatchewan and Guelph, NRC-PBI and others.

A part of Guardian's strategy is their ability to transform plants that are non-traditional North American recombinant crop plants. The underlying reasoning is to be in a position to avoid or reduce regulatory issues that surround the use of North American crop plants in molecular farming. The company was involved in R&D of vaccine candidates against Coccidiosis (protozoan disease) and NDV (viral) and these projects were funded partially by Industrial Research Assistance Program (IRAP) of the NRC. In 2004 Guardian obtained and confirmed proof-of-principle for their candidate antigen proteins.

Our intention was to follow the lead product candidates from beginning in order to identify and investigate barriers to their commercialization. We contacted Guardian about their PDV project in late July 2007 and made an appointment in early August 2007. Our initial contact, Dr. Jim MacPherson provided a tour of the facility and some discussion to indicate that Guardian had aborted the PDV project. According to Dr. MacPherson the principal reason for stopping the PDV project was the fact that Canada doesn't have protocols in place yet to regulate plant made therapeutics. It seemed likely that this situation will remain the same for several years yet. Guardian had consultations with the CFIA (Plant Protection Branch and Veterinary Biological Group), Health Canada and Environment Canada about the regulatory clearance. Also, according to Dr. MacPherson, politicians were unwilling to risk their careers on this issue. This reasoning may be interpreted to be of greater geo-political importance if the association representing the biotechnology industry in Canada has limited access or support within the government as compared to its United States counterpart which is an authoritative voice when speaking on issues affecting the industry¹³.

Conclusion

Our preliminary studies indicate the presence of bottlenecks or barriers to commercialization of PDV in a Canadian agricultural biotechnology cluster. Our hypothesis is that plants are a safe and convenient platform for production of subunit vaccines and have the potential to become a novel solution to conventional vaccine manufacturing. Our proof-of-concept rests with the fact that a number of research groups worldwide have investigated plants as a manufacturing platform. Researchers are affiliated with various disciplines ranging from science and technology to scientific regulations to intellectual property and social science. The main barrier now is the prerequisite work specifically in increasing the scale of production and antigen yields. There are many regulatory issues that need to be addressed in order to reduce potential concerns about risk to human health and environmental impacts of such production systems. Oversight is needed principally because they are based on transgenic plants. This must be balanced with the associated financial burden deterring R&D and commercialization activities. It is difficult to implement an existing IP regime to protect PDV because of their multiple classifications as a plant variety, a pharmaceutical, a biotechnological innovation and a region-specific commodity. Therefore, the move beyond the proof of concept will involve a paradigm shift, preparing the groundwork for expeditious licensing of pioneer PDV, i.e., development of case specific regulatory framework at scientific, IP and commercial levels. It is our primary contention that the successful strategy for PDV has to rely on continued public support as an investment in public good.

We wish to follow up on our study and proceed to different clusters involved in expression of vaccines in plants. We plan to conduct studies in Saskatoon (Saponin Inc.) and the Québec City cluster (Medicago Inc.). With the prospect of PDV widespread deployment in the developing world our results would be timely.

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1. World Health Organization (WHO), "Immunization, Vaccines and Biologicals," online: WHO <<http://www.who.int/immunization/en/index.html>>.

2. Y. Thanavala, Z. Huang & H.S. Mason, "Plant-derived vaccines: a look back at the highlights and a view to the challenges on the road ahead" (2006) 5 Expert Review of Vaccines 249.

3. M. Leopold, "The Commercialization of Biotechnology - the Shifting Frontier" in George T. Tzotzos, ed., *Biotechnology R & D Trends: Science Policy For Development* (New York: New York Academy of Sciences, 1993) 214.

4. H.S. Mason, D.M. Lam & C.J. Arntzen, "Expression of Hepatitis B Surface Antigen in Transgenic Plants" (1992) 89 Proceedings of the National Academy of Sciences of the United States of America 11745.

5. Canadian Food Inspection Agency, "Table 1: Summary of Confined Research Field Trials of Plants with Novel Traits (PNTs)," online: <<http://www.inspection.gc.ca/english/plaveg/bio/mf/sumpnte.shtml>>.

6. Canadian Food Inspection Agency, "Confined Research Field Trials of PNTs Intended for PMF," online: <<http://www.inspection.gc.ca/english/plaveg/bio/mf/confinee.shtml>>.

7. Dow AgroSciences, "Dow AgroSciences Achieves World's First Registration for Plant-Made Vaccines" (31 January 2006), online: <<http://www.dowagro.com/animalhealth/resources/news/20060131b.htm>>.

8. C.J. Arntzen & R.T. Mahoney, "Plant-Derived Vaccines: A New Approach to International Public Health" (2004) 69(1) Journal of Food Science CRH8.

9. Dwayne D. Kirk *et al.*, "Risk analysis for plant-made vaccines" (2005) 14 Transgenic Research 449.

10. Canadian Food Inspection Agency, "Plant Molecular Farming Discussion Document," online: <http://www.inspection.gc.ca/english/plaveg/bio/mf/mf_disde.shtml#G5>. Office of the Auditor General of Canada, *Petition No. 94 - Biotechnology and "Pharming Crops"*. (Ottawa: Office of the Auditor General of Canada 2004) online: <<http://www.oag-bvg.gc.ca/domino/petitions.nsf/viewe1.0/6771E58EEAE924EE8525704C00783A3A>>.

11. Office of the Auditor General of Canada, *Ibid.*

12. *Ibid.*

13. *Ibid.*

14. "Dow AgroSciences, University of Guelph Sign License Agreement to Develop Plant-Cell Vaccine to Protect Cattle from Shipping Fever", online: Dow AgroSciences <<http://www.dowagro.com/animalhealth/resources/news/20070827b.htm>>.

15. Office of the Auditor General of Canada, *supra* note 10.
16. Stuart Smyth, George G. Khachatourians & Peter W.B. Phillips, *Governing Innovative Science: Challenges Facing the Commercialization of Plant-made Pharmaceuticals*, ed. by R.E. Evenson & V. Santaniello, *International Trade and Policies for Genetically Modified Products* (Wallingford: CABI Publishing, 2006). Stuart James Smyth, *A Decade of Regulating Agricultural Biotechnology Liability in Canada: A Case Study From 1994 - 2004*, (MSc Interdisciplinary Studies, University of Saskatchewan, 2005) online <<http://library2.usask.ca/theses/available/etd-02072005-135411/>>.
17. Office of the Auditor General of Canada, *supra* note 10.
18. *Supra* note 6. G. Koivisto, "CFIA's regulatory perspective—challenges and opportunities to ensure science-based policies" (Presented at the Bio-Based Molecular Production Systems Workshop, Canadian Agri-Food Research Council, Ottawa, 2004), online: <<http://www.carc-crac.ca/common/BMPS%20Federal%20Perspectives/CFIA%20english.pdf>>.
19. Koivisto, *Ibid*.
20. J.W. van der Laan *et al.*, "WHO informal consultation on scientific basis for regulatory evaluation of candidate human vaccines from plants, Geneva, Switzerland, 24-25 January 2005" (2006) 24 Vaccine 4271.
21. Canadian Food Inspection Agency, "Plant Molecular Farming", online: <<http://www.inspection.gc.ca/english/sci/biotech/reg/pmfamve.shtml>>.
22. National Research Council of Canada, "Saskatoon Agricultural Biotechnology, Nutraceuticals, and Bio-Products", online: <http://www.nrc-cnrc.gc.ca/clusters/saskatoon_e.html>.
23. "Corporate overview", online: GuardianBiotechnologies <<http://www.guardianbio.com/2004/company/company.htm>>.