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ENVIRONMENTAL SCAN OF COMMON PRACTICES IN GENOME EDITING AND CRISPR IN CANADIAN PUBLIC RESEARCH INSTTUTIONS

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This research is undertaken in collaboration with the Johnson Shoyama Centre for the Study of Science and Innovation Policy.

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Executive Summary

More food of greater nutritional value needs to be produced with an ever-shrinking amount of non-renewable resources. We are all also aware that climate change is making this challenge even more daunting. However, there is now reason for genuine hope because sustained research in biology has yielded a set of genetic engineering techniques collectively known as “genome editing.” Among these techniques, one stands out for its immense versatility and the relative ease with which it can be used, namely Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR). What is so novel about genome editing, is that it offers greater precision than precursor technologies such as transgenic modification (often shortened to GM).

For crop breeders, these techniques diversify the tools used in the research that could eventually lead to the design of crops that can be grown in increasingly hostile climates (due to climate change), and that can use inputs more efficiently (fertilizers, pesticides, etc.). Across Canada there are public breeding institutions with highly capable personnel that have access to cutting-edge technology that helps them produce novel crop varieties for domestic and foreign use. But it is unclear if or how they are using genome editing tools. This report attempts to answer whether Canadian public crop breeders are using genome editing techniques in their research and breeding programs and why or why not.

Initially, this study took an indirect approach by scrutinizing all publicly available information pertaining to public crop breeding programs, across Canada. A recent survey showed evidence that Canadian crop breeders (both private and public) are aware of these techniques but there was little hard evidence of if or how they were using the tools.

This second and more direct approach involves semi-structured, one-on-one discussions with Canadian public crop breeders from six institutions (Annex, Figure 1). Only a few crop breeders that participated in the discussions reported using CRISPR/Cas9 in their research, but none use it in their breeding programs. Many noted that genome editing is not suitable for all crops and that in spite of the rhetoric, the time it takes to develop a new crop variety with these tools is about the same as with conventional tools. If the aim is to enable plant breeders the use of genome editing among Canadian public crop breeders, funding is needed to reduce this tool to practice.

Acronyms

Canadian Food Inspection Agency	CFIA
Canadian Intellectual Property Office	CIPO
Crop Development Centre	CDC
Clustered Regularly Interspaced Short Palindromic Repeats	CRISPR
Deoxyribonucleic acid	DNA
Environment and Climate Change Canada	ECCC
Freedom-to-Operate	FTO
Genetically Modified or Genetic Modification	GM
Genetically Modified Organism	GMO
Intellectual Property	IP
New Breeding Technologies	NBTs
Organization for Economic Co-operation and Development	OECD
Plant Biosafety Office	PBO
Plants with Novel Traits	PNTs
Ribonucleic acid	RNA
Tangible Property	TP
Technology Readiness Level	TRL

1.0 Introduction

Sustainable food production is an issue of world concern. According to the FAO (2021), "to be sustainable, agriculture must meet the needs of present and future generations, while ensuring profitability, environmental health, and social and economic equity." How crop production – the basis for food, feed, fiber and biofuel production – advances is of the utmost importance for sustainability. Production methods themselves are greatly influenced by the crop varieties that are cultivated. Therefore, crop breeding has key a role to play in the development of sustainable agricultural production systems (Fess et al., 2011). Novel crop varieties need to constantly be developed to adapt to an increasingly changing climate, evolving markets and consumption trends.

Crop breeding is the science of changing and improving the heredity of agriculturally important plants (Dixon et al., 2014; Poehlman, 2013). Most crop breeding programs rely on the introgression of existing genetic variation, which requires extensive crossing and access to germplasm resources, and culminates with the selection of the progeny lines with desired traits. The entire process can be slow, expensive, and largely imprecise. The three current main methodologies for crop improvement in modern agriculture are cross breeding, mutation breeding (mutagenesis), and transgenic breeding (Dixon et al., 2014; Oladosu et al., 2016).

Classical breeding methods have provided a sustained period of both increasing crop yields and increasing food production (Dixon et al., 2014). However, the highly uncertain impacts of climate change on future agricultural productivity increases the urgency to develop resilient agricultural production systems (Knox et al., 2012; Müller, 2011; Müller & Robertson, 2014; Sapkota et al., 2019). New breeding technologies are now needed to sustain an increase in yield and food production, but also wealth creation in the context of climate change, water scarcity, energy shortages, and decreasing natural resources (phosphorus, land, etc.).

A suite of techniques collectively known as genome (or gene) editing have been developed and now allow users to make targeted DNA sequence modifications in organisms (Bogdanove et al., 2018; Chen et al., 2019; Hua et al., 2019). By harnessing natural DNA repair mechanisms, genome editing techniques can overcome many of the constraints to crop breeding, especially breeding precision and time needed to make genotypic changes (Qi, 2019). Among these techniques are oligomer directed mutagenesis (ODM), zinc finger nucleases (ZFN), transcription activator–like effector nucleases (TALENs) and clustered regularly interspaced short palindromic repeats (CRISPR)/Cas9 (Schenkel & Leggewie, 2015). All of these techniques produce genome edits at high frequency; however, ZFNs or TALENs are costly and time intensive (Puchta & Fauser, 2014). In contrast, CRISPR/Cas9 systems have proved to be more versatile and robust genome editing tools for crop improvement and have facilitated significant progress in basic plant research (Ricroch et al., 2017). These novel techniques, CRISPR/Cas9 specifically, are now being used for basic research and applied to crop breeding.

A recent wave of surveys of experts from around the world have identified the potential benefits of genome editing techniques for crop breeding (Lassoued et al., 2019a), and show that they seem to pose no greater risk than conventional breeding methodologies (Lassoued et al., 2019b). Experts think that new breeding technologies (NBTs) should not be the subject of extreme regulation, given that when applied, most of them yield products whose DNA is indistinguishable from products obtained through conventional breeding methods (Lassoued et al., 2020). Currently, Canada's Plant with Novel Traits (PNT) system does not differentiate between any of the genome-editing technology applications. Thus far, the CFIA has received, assessed, and approved two varieties of herbicide tolerant canola developed through site-directed mutagenesis (genome editing) (CFIA, 2019).

Canada is a world leader in food production and a leading centre of research into wheat, canola and pulses. In addition to these crops, across six Canadian universities at least 30 other distinct crop types are researched and bred, albeit on a significantly smaller scale than wheat, canola, and pulses. With such a diversity of crop research and breeding programs across Canada, and the amount of resources devoted to and generated from these efforts (Groenewegen et al., 2016), it is surprising how little is actually known about what is done in the labs.

This paper explores what GE techniques and technologies Canadian public breeders are using in their research and breeding programs and why. In effect, we asked: Are genome editing techniques, particularly/including CRISPR/cas9, being used in Canadian public research institutions to breed crops? Why or why not? What do Canadian public crop breeders think about these technologies? Their responses to both of these questions are presented in this report.

The first part of this paper reports on what breeders are doing and why. The second part assesses the validity of the rationales for the choices expressed.

2.0 Current adaptation and adoption of genetic editing in Canadian Public Plant breeding

Canadian universities that have a plant science department that research, and/or breed crop varieties were included in this report (Table 2). An environmental scan of crop types being bred and researched, and the techniques used to breed or research them was undertaken. This was achieved by gathering all the information available on their websites. Professors or lead researchers with a lab that undertake research in genetics, genomics, plant biology, bioinformatics, and crop breeding were documented as well as the main crop/plant (or group of plants and crops) of their research focus (Annex). Their peer-reviewed publications were scrutinized for use of genome editing or CRISPR/Cas9. For each academic institution included, documentation indicating any reservations or restrictions on genome editing or genetic modification were sought. The plant science departments of the universities included in this project were then scrutinized for evidence of genome editing or CRISPR/Cas9 use. Department news or updates, reported projects or funding, researcher produced peer-reviewed literature, and researcher profiles were all scrutinized for this information. This approach failed to produce any evidence of genome editing or CRISPR/Cas9 use by public Canadian crop breeders.

To corroborate collected information from Canadian university plant breeding programs, the Canadian Food Inspection Agency's (CFIA) field trial database of approved plants with novel traits (PNTs) was also consulted (Annex, Figure 2). Any authorizations would be an indirect indicator that genome editing or CRISPR/Cas9 are, or have been, used to develop PNTs.

None, of these initial efforts yielded any evidence of genome editing or CRISPR/Cas9 use in Canadian public crop breeding. Gleim et al. (2020), however had previously surveyed Canadian crop breeders, both public and private, and found that all of them were aware of novel crop breeding technologies being used in other countries. Thus, the authors of this project suspected that publicly available information was not truly representative of the use of genome editing or CRISPR/Cas9 by public Canadian crop breeders. To be as thorough as possible, we undertook one-on-one interviews over the internet with breeders working at Canadian universities. This second step gave a better understanding of the complexity surrounding the use of genome editing and CRISPR/Cas9, in Canadian public crop breeding. Based on the information collected from both approaches, a concise analysis about the state of genome editing use in Canadian public crop breeding programs is provided.

The second approach taken in this project was more direct. That is, we conducted semi-structured one-on-one discussions with Canadian public breeders from all six institutions noted in Table 1 (Annex). Crop breeders were directly asked what use they were making of genome editing or CRISPR/Cas9 technology in their breeding programs and their reasons. Over the span of one month, the discussions took place over the internet. Their unaltered, but anonymous, accounts are summarized in the section below.

Table 1 **Crop types bred and researched at Canadian Universities**

Public Breeding Institution	Department/Division Name	Crop Type Bred/Researched	Number of Faculty and Staff
McGill University	Agricultural and Environmental Sciences -Plant Science	Cereals, Pulses	25
University of Alberta	Department of Agricultural, Food & Nutritional Science	Algae, Cereals, Pulses, Oil Seeds, Forages	83
University of British Columbia	Faculty of Land and Food Systems	Fruit, Grape Vine, Cereals	61
University of Guelph	Plant Agriculture	Cereals, Fruit, Oilseeds, Pulses, Vegetables	34
University of Manitoba	Plant Science	Cereals, Oilseeds, Pulses	53
University of Saskatchewan	Plant Science/Crop Development Centre	Cereals, Pulses, Oilseeds, Forages, Fruit	45

Notes: Not all faculty and staff in these Departments are directly involved with plant breeding. Most do research that is peripheral to plant breeding i.e. study plant physiology, insect-plant interactions, microbe-plant interactions, etc.

A few crop breeders that participated in the discussions reported to using CRISPR/Cas9 in their research, but not in their breeding programs. No other genome editing technique was reported as being used. Breeders reported that CRISPR/Cas9 applications so far have been solely for proof-of-concept purposes only and that the resulting products have only been grown in contained and well secured growth chambers.

These efforts remain relatively low key in the public record for a few reasons. As long as experiments remain contained, they do not need to be disclosed or reported to regulators, which explains why there is no public information indicating that gene editing or CRISPR/Cas9 is being used in Canadian public breeding programs. Moreover, breeding programs compete amongst each other and with private crop breeding firms; not all activities done by a breeding program are made publicly available in order to maintain a competitive advantage. Information about how traits within a crop were developed would not be known publicly until that crop is in regulated confined field trials, which means they are two to three years from the market. At least one breeder also suggested that given that there is no genomic difference between a trait developed through certain genome editing techniques or CRISPR/Cas9 and conventionally derived traits, reporting on gene editing and CRISPR/Cas9 applications might not be strictly necessary in Canada.

It was puzzling that of all Canadian universities with a plant breeding program, all of which have sophisticated laboratories and highly capable professors and researchers, only one example of CRISPR/Cas9 application to modify a crop variety was found (a peer-reviewed publication of a researcher applying CRISPR/Cas9 to a canola variety). After all, a Canadian private firm has already successfully applied genome editing technology (RNA interference) to develop Arctic® apples and moved it through the Canadian regulatory process, thus Canadian breeding institutions are capable of applying advanced biotechnologies (Brooks, 2016; Waltz, 2016). Why then does it seem that Canadian public breeding institutions are not using the latest genome editing technologies in their plant breeding programs?

We used a series of interviews with breeders to explore what factors discouraged them actively testing and using genetic editing in their programs. Not unsurprising, most saw this technology wrapped up in the controversies of transgenic technology. Researchers were also asked about whether they used transgenic (GM) technology use in their breeding programs. There was an almost unanimous consensus that crop breeders at Canadian academic institutions have given up on transgenic research completely. Their reason was simply that after 25 years the technology still carries a negative connotation that it is not able to shake off. From the breeder perspective, it seems unlikely that the negative perception around this technology will ever dissipate enough to merit the use of the technique in their breeding programs. The breeders in the CDC, in particular, collectively decided not to use transgenics based on a cost-benefit analysis that was not in its favour. The CDC policy is that scientists have the freedom to use any tool or breeding technique they would like, so long as they do not harm the industry, the University, or the customer base. The collective judgment was that while there might be some small technical advantages to a crop breeding program, the cost of regulatory compliance and managing market uncertainty outweigh any benefits.

With this context, we identified four main issues that influenced choices about using genetic editing in plant breeding.

First, few could identify any specific barriers. Asked directly about the existence of barriers (internal or external) to the use of gene editing and CRISPR/Cas9 at their host institution, most breeders consulted reported that there were none. It is noteworthy, that a significant number of breeders consulted admitted that they were not knowledgeable about the specific rules at their host institution regarding the use of genome editing or CRISPR/Cas9.

Second, a major concern of most breeders was even if they successfully developed a trait through genome editing or CRISPR/Cas9, they would not be able to navigate Canada's regulatory regime in order to bring the resulting crop to market. Only two Canadian public crop breeders that participated in the discussions reported being knowledgeable enough to successfully navigate Canada's regulatory regime in order to take a genome edited trait to market.[1, 2]

Third, pressed on their lack of use of novel crop breeding technologies, a significant number of breeders noted that consumer perceptions were an important factor in their decision to use or go without new technology in their breeding program. Many asserted that they would not use the genome editing or CRISPR technologies yet because consumer opinions about these remain largely unknown. Particularly for those breeders working on crops destined for foreign markets (crops for export), the technology is not really an option until those markets have a shift in policies and consumers signal they will accept genome edited crops.

Fourth, some of our respondents asserted that while genome editing and CRISPR/Cas9 can be powerful tools, they are not necessarily the appropriate tool for every job. In fact, for some crops (e.g., lentils and sunflower), due to the biological complexity underpinning the most important traits or their recalcitrancy to tissue culture, genome editing are not currently viable tools. Some breeders opined that the genome edits that have been published so far are relatively trivial and for which conventional tools could have been easily applied with the same results. In most cases, breeders look for what is "cheapest" and "fastest." From a Canadian public breeder's

[1] The Economic Strategy Table - has discussed that regulations play an important role in the agri food's development: http://www.ic.gc.ca/eic/site/098.nsf/eng/00015.html

[2] Echoing the Economic Strategy Table, the Industry Strategy Council has also called for a novel approach to innovation regulation https://www.ic.gc.ca/eic/site/062.nsf/eng/00118.html#s-3.3.1.5

perspective, cheapest and fastest takes into account that once the laboratory process is over (be they conventional methods or genome editing), the crop still has to then be grown many times under field conditions (plant breeding), and regulatory approval subsequently obtained. As an example, one breeder stated:

> "I can either use CRISPR/Cas9 to make an edit, without fully understanding gene function, hoping to obtain a disease resistant crop, or I can hire a summer student to apply a disease to a plot of my crop, and whatever survives is deemed resistant to the disease in question. I can then multiply this surviving plant, and I now have a variety in almost the same amount of time as I would have if I had used CRISPR/Cas9, but now I don't have to know about functional genomics or go through a regulatory approval process."

Thus, for many crops, conventional breeding methodologies will continue playing an important role in the development of new varieties for many years to come. Genome editing and CRISPR/Cas9 have simply joined the 'tool-box' now available to Canadian public crop breeders.

Overall, the barriers appear to reflect the uncertainties of the technology – of its regulation, ownership, market acceptance and efficacy – than of any overt barriers. The next section explores the validity of these concerns. We also add one more variable – who owns the technology?

3.0 Assessing the reasons for not using genetic editing

Below we discuss the context for each of the four competing and/or complementary rationales for not using this new technology. Section 3.1 discusses how agriculture is regulated in Canada. Section 3.2 discusses the apprehension of some crop breeders about market acceptance. Section 3.3 briefly discusses the biological complexity of genome editing tools and the technology readiness of the technology in the context of Canadian public crop breeding. Section 3.4 discusses the question of ownership - the legal battle over who owns CRISPR/Cas9 in Canada has yet to begin.

3.1 Agricultural regulation in Canada

The top-of-mind concern for most breeders was if or how the products of genetic editing would be regulated which, as we discuss below, remains somewhat uncertain.

The basis for agricultural regulation in Canada is the Seeds Act, which sets out the broad parameters of Canada's seed regulatory framework (SSCP, 2017). The original version of this Act dealt simply with the higher productivity of agricultural seeds. The goal was to ensure new cultivars offered some gain over the existing seed stock. New varieties under the Seeds Act are assessed by Recommending Committees made up of breeders, pathologists, crop quality experts and agronomists, who compare new varieties against commonly used check varieties across a wide range of phenotypic categories.

With the introduction of GMO varieties in 1995, the system expanded, with a range of new assessments for environmental and health and safety of the new traits. In Canada, products created through the use of biotechnology are regulated by several federal institutions (Table 2). The three main regulators are: The CFIA, Health Canada, and Environment Canada. The CFIA is responsible for regulating both the performance (or efficacy) and the environmental safety of plants with novel traits. The agency also conducts sustained monitoring and inspection to make sure that registered products continue to meet quality and safety standards after their approval. Health Canada is tasked with setting the standards for the safety of food supply, including food products of biotechnology (novel foods). Environment and Climate Change Canada (ECCC) is responsible for assessing all other products of biotechnology, such as animals and microbes.

Table 2 Canadian biotechnology regulatory institutions and legal instruments		
Agency	**Product**	**Act**
CFIA	Plants with novel traits Novel livestock feeds Veterinary biologics	Seeds Act[3] Feeds Act[4] Health of Animals Act[5]
Health Canada	Novel foods	Food and Drug Act[6] Pest Control Products Act[7]
Environment and Climate Change Canada	All animate products of biotechnology for uses not covered under other federal legislation	Canadian Environmental Protection Act (1999)[8]
Source: CFIA (2016) Note: For analysis of Canadian agricultural biotechnology regulation see Smyth (2019).		

The CFIA has developed a series of broad regulatory directives and principles to be followed when evaluating products of agricultural biotechnology. They:

1. Build on existing rather than create new legislation where possible;
2. Regulate products based on the expression of a novel trait, rather than the method used to develop the product; and
3. Conduct evaluations of each product on the basis of its unique characteristics and establishes appropriate safety levels based on the best scientific evidence.

For the CFIA, safety is not the complete lack of risk, rather the level of 'acceptable risk.' If the level of risk is unacceptable, a product will be rejected. For plant varieties deemed to be carrying a novel trait, prior to commercial production, approval, registration, or licensing might be required. This ensures sustained adherence to approval criteria. Moreover, the CFIA treats all plant variety submission information as confidential business information (Smyth, 2019). Information about varieties only becomes available once the CFIA posts it on its online repository after their review.

Since 1995, technologies approved in Canada have usually required additional review in countries seeking to produce the varieties or import them for food or feed. As a result, we have a much more complex regulatory process. GMOs divided the world, with 29 countries as of 2019 approving at least one GM plant and more than 44 countries approving imports of GM crops as

[3] Seeds Act (justice.gc.ca)
[4] Feeds Act (justice.gc.ca)
[5] Health of Animals Act (justice.gc.ca)
[6] Food and Drugs Act (justice.gc.ca)
[7] Pest Control Products Act (justice.gc.ca)
[8] Canadian Environmental Protection Act, 1999 (justice.gc.ca)

food and feed (ISAAA 2021). But a portion of the world went GM-free, banning production or import of some or all GM crops.

Genome editing is similarly dividing the world, with some countries regulating it in the same way as GMs (European Union) while others have decided to not regulate it any differently or significantly different than conventionally bred varieties (Lassoued et al., 2020). Canada does a bit of both. In the specific case of genome editing or CRISPR/Cas9 use, certain applications of do not result in a novel trait (Chen et al., 2019) and thus are not subject to higher regulation; other edited crops may require the same process as required for GMs.

3.2 Market acceptance

Market acceptance was a common consideration, which given the evidence is a valid concern. Breeders correctly interpret that consumer opinions and acceptance of the applications of these technologies remain largely unknown. As far as the authors of this report are aware, Gatica-Arias et al. (2019) are the only ones to have surveyed consumers about their perceptions of genome edited or CRISPR/Cas9 derived foods so far. That survey, albeit of Costa Rican consumers and with methodological issues that can be critiqued, showed that consumers are not against these technologies being used in food production. Almost half of those interviewed reported that they would purchase a kilo of genome edited rice or beans, if the price were the same as a conventional alternative. In an assessment of hypothetical application of novel technologies to food production, Yang (2018) used insights from behavioral economics to focus on three particular factors that motivate disparate assessments of food technologies: logical scientific vs. narrative information to communicate about food biotechnology; cultural worldview; and intermediary food-related values. Yang found that information framing induces different attitudinal changes and food choice behaviors. While logical scientific information is more trustworthy and credible, consumers report that narratives are easier to understand. Moreover, deep-seated human worldviews do have significant influence on how people respond to novel food technologies. People with hierarchical (vs. egalitarian) and communitarian (vs. individualistic) worldviews tend to hold more positive attitudes and be more accepting of agricultural biotechnology. Finally, Yang found that intermediary food-related values and their relative importance to consumers, have significant power in explaining attitudes and choices about foods produced through nanotechnology. Thus, consumers' attitudes and food choices related to innovative food technologies are affected by both 'inside' individual factors, such as underlying human values (i.e., cultural worldviews and food-related values), and 'outside' environmental factors, such as the information framing (i.e., narrative communication). Similar results have been found in other consumer surveys in the U.S. exploring the acceptance biotechnology derived foods and personality traits (Lin et al., 2019), as well as outside environmental factors such as food labels (Kolodinsky & Lusk, 2018).

A confounding problem is that consumer opinions and attitudes towards novel technologies in food production vary widely in different markets, justifying Canadian public crop breeders' apprehension about consumer acceptance of genome edited foods especially outside the Canadian jurisdiction. Gleim et al. (2020) show that for some Canadian public breeders, the decision to stay away from genome editing is largely driven by market perceptions. Most crop production in Canada is destined for international markets, which for the most part, are concerned with the processes used and the embodied product attributes. There is no example of international trade in any genome edited crops yet. Thus, Canadian crop breeders' aversion to a technology not yet widely accepted in international markets is understandable.

The European Union (EU) is the most notable example of a group of countries that have shut down imports of agricultural biotechnology products except for specific products that very closely regulated. The EU, in particular, has taken the same stance on products derived from site-directed mutagenesis (e.g., CRISPR/Cas9), which has discouraged Canadian plant breeders from using genome editing in their plant breeding programs (Eriksson et al., 2019; Gleim et al., 2020).

Canada has a strong memory of the international market reacting negatively to crop innovation. GM flax was developed in Canada and licensed for commercial production in both Canada and the United States in the late 1990s but withdrawn from the market when the EU failed to approve its cultivation or importation. Almost a decade later, on September 8th 2009, Germany issued an EU-wide Rapid Alert notification confirming the presence of GM flax in some samples of flax imports from Canada (Viju et al., 2014). All EU flax imports, the vast majority directed to the industrial oils market and destined for paints or linoleum production, were embargoed until Canadian exporters could comply with EU standards. Canada was the world's largest producer of flax, so the EU had no alternative source of supply. The market shutdown thus imposed significant costs on both the Canadian and EU flax industries (Booker et al., 2017).

No doubt, this episode has made public crop breeders reconsider the use of breeding technologies not yet approved in the destination market(s) for their crops.

3.3 Technology readiness

This is still a relatively young and immature technology.

The objective of genome editing is to precisely edit nucleic acids[9] in living cells. It is accomplished by taking advantage of the natural DNA repair mechanism to create desired DNA sequence modifications. A highly efficient way to achieve precise modifications in plant genomes, is to take advantage of the double-strand-break (DSB) repair mechanisms (Voytas, 2013). The two main mechanisms used to correct DSBs are homologous recombination (HR), and non-homologous end joining (NHEJ) (Waterworth et al., 2011). In HR, DNA templates bearing sequence similarity to the break site, are used to introduce sequence changes to the target site. In NHEJ, broken chromosomes are rejoined, often imprecisely, thereby introducing nucleotide changes at the break site.

All genome editing techniques rely on the single step of engineering an enzyme (i.e., the nuclease), that induces a DSB at a specific site of the DNA that is to be edited. It does not matter which nuclease is used to induce the break (ZFN, TALEN, meganucleases or CRISPR RNS-guided nuclease), the biological outcome is always the same (Bogdanove et al., 2018). CRISPR/Cas9 (and the CRISPR-associated protein) is an adaptive virus immunity system in bacteria (Zhu et al., 2020). CRISPR/Cas9 systems can be programmed with relative ease, to make DSBs at any desired target segment at a minimal cost (Mali et al., 2013).

However, to be able to use genome editing a thorough understanding of the plant's genome is needed. Functional genomics is the field that concerns itself with understanding the relationship between the information contained in an organism's genome, and its physical characteristics (National Academies of Sciences & Medicine, 2020). This field was born 20 years ago when the human genome was first sequenced; it then became clear that the next step in biology was to understand the function of genes (Function, 2000). Twenty years on, moving from phenotype to genotype remains difficult because the biological mechanism that translates between them has yet to be figured out.

For wheat, the most important crop for Canadian agriculture, protocols already exist to use novel genome editing technologies (e.g., Bhalla & Singh, 2017). However, even the pioneers in this field recognize that without knowing what specific genes encode for (genomic information),

[9] Nucleic acids are the main information-carrying molecules of the cell. They determine the inherited characteristics of every living organism.

technologies to edit them are of limited use. Thus, even if policies are specifically tailored to foster the use of genome editing and CRISPR/Cas9 in Canada, the technical constraint must be overcome (understanding gene function) for these technologies to be exploited to their full potential.

From discussions with crop breeders, the technology readiness levels (TRL) of their breeding programs with respect to genome editing and CRISPR/Cas9 were assessed. The TRL, developed by the American National Aeronautics and Space Administration, are a type of measurement system used to assess the maturity level of a particular technology (NASA, 2012). There are 9 technology readiness levels, with 1 being the least ready and 9 being already used in real-life conditions.[10] This measurement system was adapted by Innovation Canada, and is appropriate in this case because breeders are familiar with the system, as it is used by various programs for funding purposes (Innovation Canada, 2018).

In addition to the TRL assessment of their programs, breeders were also asked to self-assess their TRL. Both scores were added and averaged to get a more balanced TRL measure (Table 3). There was considerable variability in TRL scores (between Level 1 and 7) among breeding programs that participated in the discussions. The Canadian average level was 4, or "basic technological components are integrated to establish that they will work together." In this particular case, that means that CRISPR/Cas9 has been used and its products have been grown under controlled conditions. However, using these technologies does not mean that breeders *a priori* know what the modified plant traits will do, or how they will manifest themselves. Thus, the real field-level impacts remain unknown and purely theoretical at this point.

Table 3 TRL level in different technologies of participant breeding programs

Crop	**Genome Sequencing**	**Molecular Markers**	**Genome Editing**
Grains (barely, wheat, oats)	High	High	Low
Pulses (peas, lentils)	High	High	Low
Oilseeds (Canola, Sunflowers, Soybeans)	High	High	Low
Perennial Crops	Medium	Low	Low
Fruits (Apples)	Low	Low	Low

Notes: In this codification a Low TRL level is a score of 1-3, a Medium TRL level is a score of 4-6, and a High TRL level is a score of 7-9.

[10] https://www.ic.gc.ca/eic/site/080.nsf/eng/00002.html

3.4 Who owns CRISPR in Canada?

Ownership and control of CRISPR-Cas9 is in dispute globally. In May 2012, the University of California (UC) filed the first of many patent applications for CRISPR-Cas9 on behalf of a group of researchers at multiple institutions. The group includes Professors Emmanuelle Charpentier, originally from Umeå and now with the Max Planck Institute in Berlin, and Jennifer Doudna of UC Berkeley. Together these researchers were awarded the Nobel Prize in Chemistry for their work in 2020. Then in December 2012, a research group at MIT and Harvard University (the Broad Institute) filed a competing patent claim for CRISPR-Cas9. As is often the case, the patent applications remained secret for 18 months, so that it was only afterwards that the two teams realized they have competing claims. The same cross filings were made in Europe. In 2016 both teams began to prosecute their applications in the United States, seeking exclusive rights to CRISPR-Cas9. So far, the rights have not been conclusively assigned or divided.

In Canada, who owns CRISPR/Cas9 technology has also not yet been established (CIPO, 2021). While over 3,400 patents and pending patent applications refer to CRISPR and Cas9, there are only 3 claims over the foundational technology open in Canada (Table 2). To commercialize technologies that use the CRISPR/Cas9 system in Canada, licenses to the foundational patents and particular applications may be needed (Lipkus, 2018). Thus, if Canadian public crop breeders develop a crop variety through CRISPR/Cas9, they will most likely have to seek a license to commercialize their product. But from whom? No party listed in Table 2 has been assigned patent rights. In fact, in Canada the CRISPR patent dispute has yet to begin, and this lack of clarity may already be stifling innovation (Lipkus, 2018). Currently, even if a novel crop variety was developed through CRISPR/Cas9 it is unclear if it could be commercialized in Canada. While basic research or proof-of-concept studies often proceed without licensing technologies, if they generate new traits of value, they could be subject to retroactive licensing once patents are issued, which could significantly reduce the bargaining power of the inventor. For that reason, many researchers will avoid using the technology.

Table 2 Parties Claiming CRISPR system ownership in Canada

Patent Application	Title	Inventors	Applicant	PCT Filing Date
CA3081937	Type V Crispr/Cas Effector Proteins For Cleaving Ssdnas And Detecting Target Dnas	Doudna, Jennifer A. Chen, Janice S. Harrington, Lucas Benjamin Ma, Enbo	The Regents of the University of California (United States of America)	2018-11-20
CA2930877	CRISPR-CAS System Materials and Methods	Charpentier, Emmanuelle Chylinski, Krzysztof Fonfara, Ines	CRISPR Therapeutics AG	2014-11-17
CA 2932439	CRISPR-CAS Systems and Methods For Altering Expression Of Gene Products, Structural Information And Inducible Modular Cas Enzymes	Zhang, Feng Zetsche, Bernd	The Broad Institute & the Massachusetts Institute of Technology	2014-12-12

Source: CIPO (2021)
Notes: All three parties have filed a patent claim with the Canadian Intellectual Property Office (CIPO) through the Patent Cooperation Treaty (PCT). The PCT is an international treaty providing standardized filing procedures for foreign patents in the countries that have signed the treaty (Canada is a signatory). As per current regulations, 'CRISPR Therapeutics AG' requested their application be 'examined' five years after their initial submission on 2019-11-18. It is now up to CIPO to consider the merit of the claim.

One complication is that the technology is not that difficult to use. At an OECD meeting on genome editing in 2018, Fyodor Urnov, Professor in the Department of Molecular & Cell Biology at UC Berkeley, asserted that "there are no trade secrets at this point in this field; a high-school student should be able to put together a genome editor using the CRISPR/Cas9 system" (Friedrichs et al., 2019, p. 422). In effect, the 'know-how' of making a genome editor was essentially in the public domain. Thus, it is unclear whether even if a patent is assigned it will have retroactive effects, and in practical terms, will the patent holder be able to collect royalties from the invention.

4.0 Conclusion

CRISPR/cas9 technology is currently being used by some Canadian public crop breeders, but only for research purposes. A complex set of decisions are made around a whole host of traits that are selected every time a crop variety is developed. The decision of whether or not to use genome editing or CRISPR/cas9 technology is but one factor in this complex interplay of decisions. We found an interesting framing of this decision process coming from our discussions with a group of XX Canadian public plant breeders.

When asked about their practices, no breeders said they were prohibited from using the technology. Instead, breeders identified a range of concerns in the following order, implicitly ranking them from greatest to least:

1. regulatory hurdles
2. market acceptance
3. technological readiness.

None directly alluded to IPRs and freedom to operate.

But as we got into the discussion, it became clear that the biggest barrier was the competitiveness of the technology itself. Their decision to use the tool or not ultimately comes down to: (1) what is the most appropriate tool for the task at hand (not always a biotechnology tool), (2) whether consumers and the market would accept the product resulting from the application of the tool they choose, followed by (3) concerns about regulatory hurdles. None ever mentioned the legal aspects of using CRISPR/Cas9 or the CRISPR System and the challenges of securing freedom to operate. Conceptually, the ‘weight’ of each factor can be depicted as:

1. fitness of the technology to the crop and trait
2. market acceptance
3. regulatory hurdles
4. freedom to operate

Above all, CRISPR/cas9 or genome editing use comes down to whether they are the “cheapest” tools that will achieve a crop breeder’s objectives the “fastest.”

Understanding the function of genes (functional genomics) is a necessary first step to wider adoption. If the aim is to help crop breeders employ genome editing or CRISPR/Cas9 in their breeding programs, funding can and probably should be directed to basic biology research. This deficit in knowledge is a serious constraint on novel crop breeding techniques in the public sector.[11]

A secondary, but still important set of considerations, are the traditional GE3LS concerns.[12] Canadian public plant breeders signal they would benefit from any insight into Canadian consumer perceptions and willingness to accept products of genome editing. Funding this stream of research could indirectly benefit Canadian public crop breeding programs, as it would help them develop research and breeding objectives with better market information.

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[11] The private crop breeding sector has more knowledge about gene function in general, but this data is not publicly shared.

[12] https://www.schoolofpublicpolicy.sk.ca/csip/documents/2017_GE3LS%20Book.pdf

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Annex: Canadian Institutions Capable of Using Genome Editing

The first approach to exploring the common practices of genome editing and CRISPR use in Canadian public research institutions, was to scrutinize the information each institution makes public (available over the internet). In other words, the first approach taken was indirect.

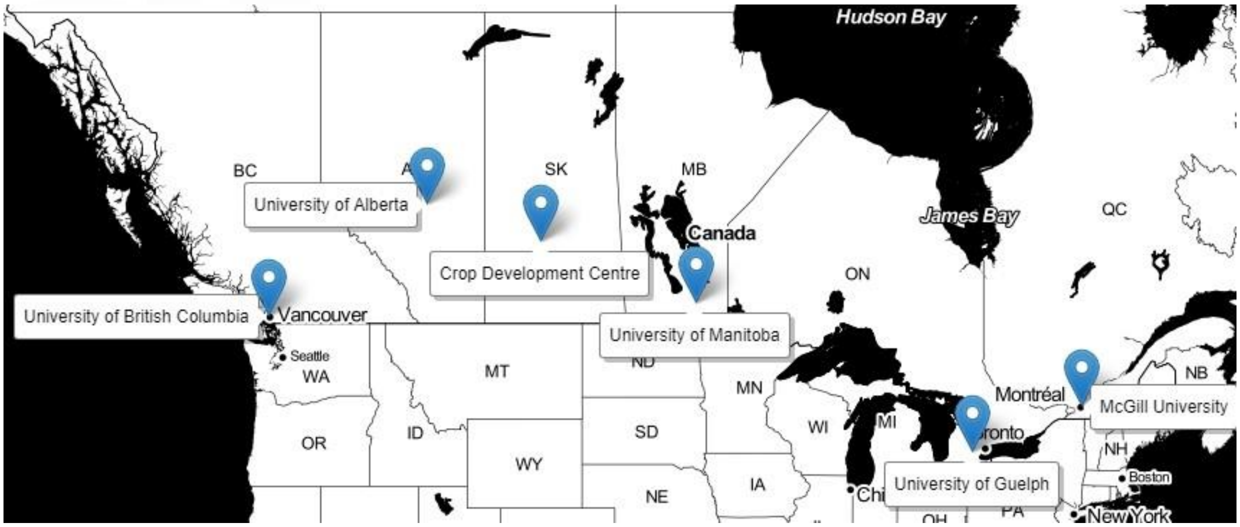

Figure 1 Canadian institutions capable of doing agricultural genome editing research

Source: Authors

Figure 1 depicts all Canadian universities that have well trained scientists, researchers, and appropriately equipped laboratories that are capable of genome editing agriculturally important crops. There are a total of 25 different crop types being researched and bred at six universities across Canada. However, while at first glance this number might give the impression of research diversity, upon closer inspection wheat and canola are the main focus of Canadian public breeding and research (Figure 2). By focus, it is meant that these are the crops to which the most resources, both in financial terms and number of professionals researching them, are devoted.[13] Wheat is researched and bred at six universities across Canada, and while canola has a significant amount of resources devoted to it as well, this crop is only bred and researched at the University of Saskatchewan, the University of Alberta and the University of Manitoba.

[13] In discussions with researchers of minor crops, some of them expressed uncertainty as to whether their breeding and research programs would continue after they retired.

University of Alberta

The Department of Agricultural, Food & Nutritional Sciences at the University of Alberta attracts over $20 million in research funding every year.[14] Within this Department, a group of faculty and research staff focuses on Plant Biosystems research. Of this group, there are three distinct (with overlapping research interests) subgroups that focus on (1) plant breeding and genetics, (2) crop biotechnology and (3) plant physiology.

Plant breeding and genetics

The focus of this subgroup or researchers is the conventional and biotechnology driven breeding of crops for improved quality, yield and organic production.

Table 3 University of Alberta capacity to undertake genomic research

Model crop	Methodology used to breed crop
Canola	Conventional, cell tissue and molecular marker techniques
Wheat	Conventional and Genetic Research (no genetic transformation technologies)
Cereals	Bioinformatics and computational genomics
Canola	Disease management
Higher Plants, Algae and Yeast	Biotechnology (no crop transformations)

Crop Biotechnology

The main focus of this subgroup is to apply molecular biology to identify, characterize and validate novel genes for crop improvement, with particular emphasis on oil seed lipid synthesis and, as well as the biosafety of transgenic crops.

Table 4 University of Alberta capacity to undertake genomic research

Model crop	Methodology used to breed crop
Wheat and Canola	Genetic Study and Crop Management
Canola	Genomic Study
Lentils	Genomic Study

[14] https://www.ualberta.ca/agricultural-food-nutritional-science/index.html

Plant Physiology

The main focus of this group is to understand biochemical and genetic regulation of vegetative and reproductive growth and development in agronomic crops.

Table 5 University of Alberta capacity to undertake genomic research

Model crop	Methodology used to breed crop
Legumes	Genetic Study and Crop Management

University of Manitoba

The Department of Plant Science within the Faculty of Agricultural and Food Sciences at the University of Manitoba,[15] maintains an active research program with the aim of developing superior cultivars and new production systems suited to the changing needs of Manitoba farmers, and the agri-food industry.

Table 6 University of Manitoba professors with capability to undertake genomic research

Model crop	Methodology used to breed crop
Cereal Crops	Biotechnology
Wheat	Genetics and Conventional Breeding
Grains & Oilseeds	Conventional Breeding
Corn, Soybean	Molecular Biology
Canola	Genetics and Conventional Breeding
Canola and Wheat	CRISPR/Cas9 and Molecular Genetics
Plants	Bioinformatics
Canola	Plant genomics and molecular biology

[15] https://umanitoba.ca/afs/plant_science/

Table 6 University of Manitoba professors with capability to undertake genomic research	
Canola	Molecular Biology

McGill University

The Department of Plant Science is within the Faculty of Agricultural and Environmental Sciences at the University of McGill.[16] Members of the Plant Science Department have differing research focus, most of which is peripheral to crop breeding and research.

Table 7 McGill University professors with capability to undertake genomic research

Model crop	Methodology used to breed crop
Plant Cell	Genomics
Pulses	Genomics and Plant Phenomics
Wheat and Barley	Genomics and Genome Editing
Agricultural and forestry crops	High-throughput "omics" methods: bioinformatics, genomics, proteomics and metabolomics-based methods to study resistance to fungal diseases
Barely, Legumes	Molecular and genomic tools to develop enhanced crop plants
Various (potato, eucalyptus,	Genomics and Bioinformatics
Legumes	Genomics
***Brachypodium* and Hemp**	Genomics

[16] https://www.mcgill.ca/plant/

University of British Columbia

The Faculty of Land and Food Systems at the University of British Columbia is the umbrella institution for various research groups. All groups have as their guiding goal the development of sustainable food systems.

Table 8 University of British Columbia professors with capability to undertake genomic research

Model crop	Methodology used to breed crop
Wheat	Genetics, Genomics, Conventional Breeding
Grape for Wine	Molecular biology
Potato, rice, beans, wheat	Plant Genetics

University of Guelph

The University of Guelph houses the Department of Plant Agriculture. It is a research-intensive department dedicated to teaching, research and service related to horticultural crops, turf grass, landscape species and field crops. It is the largest research department at the University of Guelph (over $16M/y in research grants and contracts).[17] The Department is made up by 34 faculty, 40 Staff, 60 Contract Staff, and 110 Graduate Students. The areas of expertise of both faculty and staff are in plant breeding, plant physiology, statistics/bioinformatics, molecular genetics and bioproducts, among others.

The end result of much of the work undertaken by the Department is the development of new crop varieties that are licensed both in Ontario and beyond (rest of Canada and parts of the United States). Royalties collected from seed sales of multiple varieties, and germplasm released by the Department's plant breeding programs, bring in over $800,000 to the University annually. Making it the largest and most consistently successful University of Guelph intellectual property revenue stream.

[17] About Us | Plant Agriculture (uoguelph.ca)

Table 9 University of Guelph professors with capability to undertake genomic research	
Model crop	**Methodology used to breed crop**
Berry Crops	Conventional breeding
Soybean	Conventional and molecular genetic-based methods
Canola	In vitro and conventional
Maize (Corn)	Quantitative developmental genetics
Field bean	Genetic Modification Molecular biology and Genomics
Soybean	Conventional and molecular biology
Tree Fruit	Cell culture and molecular genetics

Crop Development Centre (University of Saskatchewan)

The Crop Development Centre (CDC) is a field crop research organization within the Department of Plant Sciences at the University of Saskatchewan. CDC scientists integrate basic research with genetic improvement of spring wheat, durum, canary seed, barley, oat, flax, field pea, lentil, chickpea, fababean and dry bean.[18] The CDC has released over 450 new varieties of wheat, durum, barley, oats, flax, field peas, lentils, chickpeas, canary seed, and dry beans since its inception (Groenewegen et al., 2016).

Table 10 University of Saskatchewan professors with capability to undertake genomic research	
Model crop	**Methodology used to breed crop**
Barley and oat	Conventional breeding
Pulse crops, dry bean	genomics and conventional breeding
Forage Crops	Conventional breeding
Fruit crops	Conventional breeding
Bread wheat	Conventional breeding and genetics
Wheat	Conventional breeding and genetics
Chickpea and Flax	Conventional breeding and genetics
Lentil, fababean and special crops	Conventional breeding

[18] https://agbio.usask.ca/research/centres-and-facilities/crop-development-centre.php

Field pea & soybean crops	Conventional breeding and genetics

CFIA Field Trial Database

The confined research field trial program at the CFIA provides breeders with the opportunity to grow PNTs for research purposes.[19] Specific terms and conditions of confinement, which are designed to minimize any impact the PNT may have on the environment, vary by crop. CRISPR-Cas9 was first used to edit cells in 2013 (Cong et al., 2013; Mali et al., 2013), thus the CFIA's data base was scrutinized from this year onwards for any field trial of a PNT by a Canadian public institution.

Between 2013 and 2019 there were 1,275 field trial authorizations for PNTs in Canada. Of these, 86 were done by a public institution, or 6.7% of total trials (Figure 2). There has been a marked decline in the number of PNT trials authorized after 2014, in particular to public institutions. There is no ordinance of any kind prohibiting Canadian public breeding institutions the use of GM technology. The decline in PNT authorizations to public institutions might be an indirect indicator, that these institutions are forgoing the use of this technology of their own volition. A possible explanation might be that because Canadian consumers are generally averse to GM crops (Macall et al., 2021). If so, then there may be no point in using this technology. Canada's experiences with wheat and canola show the range of possibilities. The most publicly funded researched crop in Canada is wheat, where producers and markets have both signaled they are unwilling to adopt GM varieties in part because as a whole food the genetic transformant is in the end product. Canola, in contrast, has used GM technologies, in part because the processed oil has no detectable genetic material while the meal is mostly fed to animals. Other crops fall on this spectrum from avoidance to embrace depending on their degree of processing, the specific traits involved and their end use.

[19] See: https://www.inspection.gc.ca/plant-varieties/plants-with-novel-traits/approved-under-review/field-trials/eng/1313872595333/1313873672306

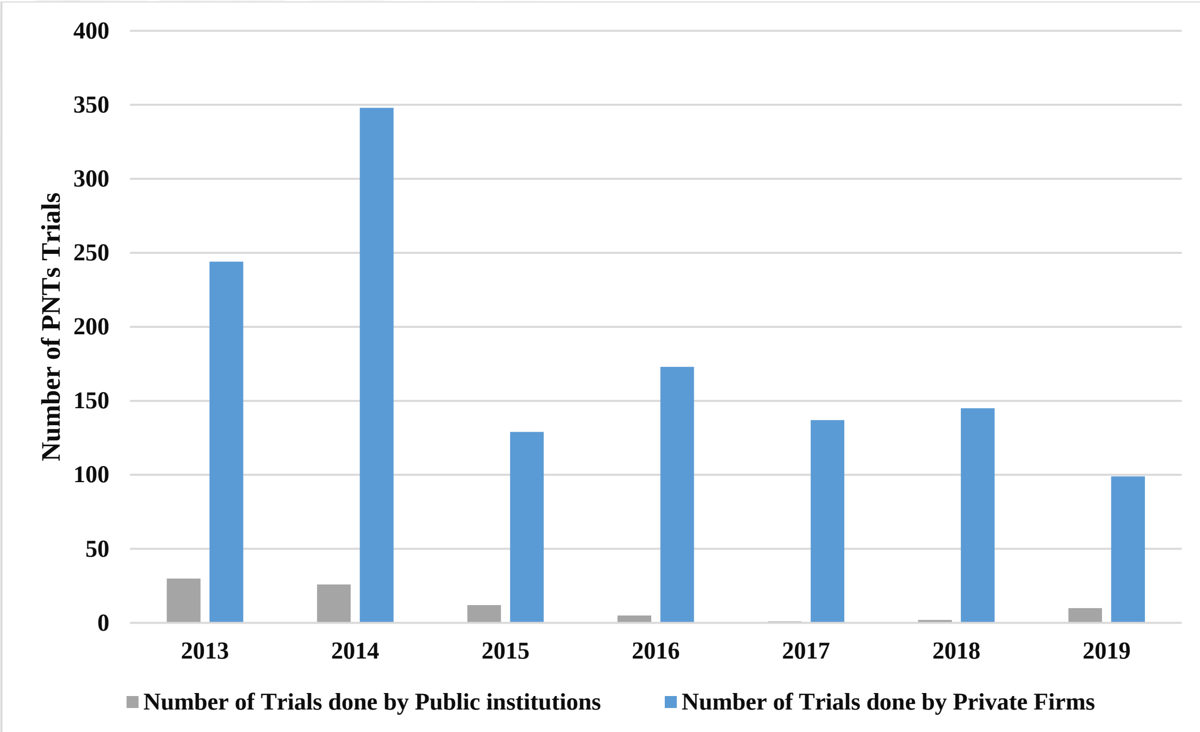

Figure 2 Number of PNTs Field Trials done in Canada 2013-2019

Source: CFIA (2020)