Markers Assisted Selection for the Canadian Swine Industry

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1 Canadian Swine Industry

The Canadian pork industry enjoys a solid reputation world wide for superior quality and health status. The Canadian pork exports have continued to increase during the recent years. Canada ranks number one among the pork exporting countries of the world. Domestically, the pork industry is also recognised as a major industry sector in agriculture, contributing to the trade surplus.

The Canadian Swine Improvement Program has a proven history of genetic improvements over the past several years. Over 90,000 pigs are tested each year on the Canadian Swine Improvement Program. There are 120 participating herds across Canada and over 9000 nucleus sows. The program involves selection and genetic improvement of three major breeds: Duroc, Yorkshire and Landrace. Over 7,000 new records are added every month in the national database and genetic evaluations are computed for growth, feed efficiency, carcass traits and sow productivity.

The swine improvement program is supported and operated by the Canadian Centre for Swine Improvement (CCSI) and member organisations. CCSI is a national organisation created by the swine industry to provide leadership, coordination and services relation to genetic improvement. CCSI operates a research program with industry partners, research institutions and Canadian universities. The current research plan includes integration of molecular genetics in selection programs as one of the key components along with research on selection strategies, genetic evaluation methods and optimum use of genetics by the producers.

2. Current Tools, Selection Methods and Results

Genetic evaluations are computed for age at market weight (100 kg), backfat thickness, lean yield, loin eye area, loin muscle depth, feed conversion and litter size at birth. In the province of Ontario, the evaluations also include age at first farrowing, farrowing interval and weaning weight. These evaluations are based on multiple trait BLUP. Evaluations are currently under development for piglet survival, meat quality and conformation.

In addition, a number of tools are available to the users to help in use of the evaluation and selection decisions. For example a web application, "Computer dating", allows the users to define their own breeding goals, develop and rank pigs on customised selection

indices and calculate expected genetic gains based on tentative matings. A Windows based program, "Breeding for Profit" helps to compare different selection scenarios and make selection decisions at the time of probing based on Estimated Breeding Values (EBV's), customised indices and expected genetic gains in various traits.

Over the past years the program has made significant genetic progress using the polygenic EBVs and quantitative selection methods. The gains during the last five years amount to a reduction of about 7.5 days of age to market weight, 1.4 mm reduction in backfat, 106 gm less feed per kg of growth, 0.67% higher lean yield and 1.2 sq cm larger loin eye area in the market hogs and about one extra pig in the litter. As a result, an average commercial sow is about $130 (US) more productive from 2002 genetics compared to sows from 1997 genetics.

3. Use of Molecular Information

Recent developments in molecular genetics provide the opportunity for increasing the rate of genetic progress further by a combination of molecular information and performance records through marker assisted selection.

A number of genes and markers have been identified in swine with varying degree of success in the limited field experiments. There is a "cautious optimism" in the industry towards the use of markers assisted selection. There is a need to set priorities by establishing which major genes and genetic markers are the most promising for commercial application and to develop selection and economic models to evaluate the benefits and costs associated with each promising gene or genetic marker before substantial funds are invested in their use. Therefore, a project has been recently launched with support from the CCSI's member organisations, breeders and the federal government.

This project is a research initiative to evaluate and incorporate the important discoveries into industry breeding programs. This includes evaluating the current advancements in molecular genetics and the economic benefits from their application to the swine industry, and developing a strategy and methods for integrating molecular genetic advancement into Canadian swine breeding programs.

The project will open up opportunities for further research and innovation in this area by providing a strategic plan for swine molecular genetic research in Canada. This plan will contain stepwise priorities for research and possibilities of collaboration between industry, federal research labs and universities.

3.2 Evaluation of the Current Situation

A large number of genes and QTLs have been identified and mapped through the use of molecular genetic technology over the past years in the main livestock species. In swine, the number of genes mapped is close to 2000**.** Some of these genes contain the genetic

code for a protein and therefore have a functional role in the physiologic affect of the trait. However, a large number of identified genes are non-functional genes often referred to as genetic markers for the specific traits. The genetic markers are expected to be associated with genes that affect the quantitative trait or the so-called quantitative trait loci or QTL. Some of the important candidate genes and mapped QTLs for economic important traits are reviewed.

3.2.1 Industry Use of Candidate Genes

The effects of the Halothane gene have been known to the swine industry since about thirty years ago because it is bound with PSS that is associated with sudden death losses, the incidences of pale soft exudative pork (PSE), dark firm dry meat (DFD), and back muscle necrosis. The gene was also reported to result in lower feed intake, lower daily gain, and smaller litters born and raised. In Canada, the HAL-1843 trademark DNA test developed by Canadian researchers at the University of Toronto was widely applied to screen Canadian swine populations. Halothane gene frequency and its negative effects on Canadian swine population were investigated by many research groups (e.g. Gibson et al. 1996; Murray and Johnson 1998). During recent years, Halothane gene was generally screened among breeding sires. The positive sires were eliminated. In some regional centers of swine breeding, dams have also been tested and culled from nuclear populations. This independent culling on the Halothane gene might have some negative effect on selection for other desirable traits, however, it is difficult to quantify and perhaps the effects is very low in view of the current rates of genetic progress.

RN (the rendement napole) gene is another gene tested in Canada. RN gene was first documented by Leroy et al. (1990). Carriers of this dominant gene show lower pH, higher surface and internal reflectance values, lower water-holding capacity, greater cooking loss and lower Napole yield (yield after curing and cooking). The RN gene has undesirable effects on the processing quality of pork and can lead to estimated industry losses of $10.5 (US) for each hog carrying the gene.

A joint study was carried out by the Canadian Centre for Swine Improvement Inc.(CCSI), the Food Research and Development Centre (FRDC) of Agriculture and Agri-Food Canada, and the National Institute for Agricultural Research in France (INRA)(Houde et al. 2002). The study was done on a sample of 305 boars from artificial insemination centres from all areas of Canada, in order to evaluate the frequency of the gene in the main breeds used in the country. The pigs sampled from these three breeds for the study did not carry the RN gene. Therefore, it is very unlikely the gene is present in the larger population of these breeds. The results of this study were very useful for commercial producers that have been using the Canadian Duroc, Yorkshire and Landrace breeds, and for the abattoirs and processors that purchase their pigs. The absence of genes like RN with a negative effect on meat quality and the extensive use of the Duroc breed known for its superior meat quality, are some of the reasons for the reputation of Canadian pork in domestic and international markets.

These genes (Halothane and RN) are mainly responsible for negative effects on meat quality. Tests for these genes are mainly used for screening of the commercial populations and independent culling in the nucleus populations. The major Canadian breeds are rather free from the RN gene and the frequency of the Halothane gene is also reducing due to extensive testing at the nucleus level. Having reduced these problems, there is still a large variation for meat quality that can be addressed through selection on some other candidate genes that have positive effects. Examples of such genes are: Heart Fatty Acid Binding Protein gene (HFABP) for increasing intra muscular fat without increasing backfat, Melanocortin-4 receptor (MC4R) for backfat, growth and feed intake, Calpastatin (CAST) gene for tenderness, Fructooligosaccharide (FOS) gene for proportion of white fibres in the muscle, etc.

A mutation of the c-Kit receptor gene known as the Dominant white allele is the result of a gene duplication (Marklund et al. 1998), which disrupts normal melanocyte development in the embryo. This gene is therefore useful in detection of unwanted color transmission in white breeds. Some countries consider the color uniformity seriously, as an indicator of breed purity. DNA tests for this gene can be useful for color uniformity of crossbreds. A patented gene test (Andersson et al. 2001) is available that identifies the Dominant white allele by its extra copy of the normal c-Kit receptor sequence. Test matings were made between white boars and Duroc sows. DNA samples from litters exhibiting colour and those that do not were analysed, to assess the potential usefulness of the c-Kit receptor gene test at predicting colour-free matings. Results showed that a variation of the Dominant white allele appears to exist in the commercial white pig population. The inheritance of coat color is not very straight forward. The frequency of pigs with coat color in the Canadian white breeds is also low, which makes it difficult to obtain sufficient samples for analysis.

A large number of other promising candidate genes have been reported by different research groups. There is a need to investigate the usefulness of these genes for the Canadian swine industry and develop specific methods and guidelines for their use in genetic improvement programs. There is a special interest in improving female reproduction and meat quality traits by using molecular information.

3.2.2 Promising Candidate Genes and Markers

A review was conducted regarding possible markers as well as genetic parameters of the markers such as number of alleles, levels of dominance, linkage to specific QTLs, and allelic frequencies in the tested populations etc. The usefulness of different markers was evaluated based on the extra response to selection on the marker compared to direct selection on the quantitative trait, effect of early selection and other practical considerations such as additional costs and benefits. Some of the promising genes and markers have been identified for potential use by the industry.

The heart fatty acid–binding protein (FABP) is one of the promising candidate gene for intramuscular fat content because of its large effects and because HFABP polymorphisms

might be implemented in marker-assisted selection to improve IMF content independently from backfat thickness (Gerbens et al. 2000). North American markets as well as the Japanese market are increasingly demanding a proper IMF content in fresh pork. Improvement of IMF content is not only important for consumers' acceptance of marbling level, but it has also an important effect on eating quality such flavor, tenderness and juiciness. A recent survey has indicated that there is a large proportion of pork loin which is under consumers' acceptance threshold. Trends of low IMF are expected to continue with further selection for lower backfat and higher lean yield, because of the positive correlation between IMF content and backfat thickness. HFABP gene offers the opportunity to increase IMF content without affecting backfat thickness, therefore having fast growing leaner market hogs with desirable level of IMF content.

Insulin-like growth factor 2 (IGF2) gene plays an important role in mammalian growth, influencing foetal cell division and differentiation, and postnatal muscle growth. Based on its physiological function, IGF2 has been considered as a candidate gene for a quantitative trait locus (QTL) in pigs affecting muscularity. The large effect of IGF2 gene on lean meat content and backfat thickness of swine has been detected by several studies (e.g. Jeon et al 1999; Nezer et al. 1999; Lee et al. 2001). The IGF2 QTL is highly polymorphic. Polymorphism exists between breeds (e.g. between wild boars and Large White, between Large White and Pietrain, and between Berkshire and Yorkshire) and within purebred populations. The IGF2 gene is imprinted in pigs (paternally expressed). This imprinting nature of this candidate gene might be very useful for practical animal breeding.

The QTL at IGF2 gene is paternally expressed only. The genes from boars should show the full effect on progeny, regardless of the sows' genotypes. Use of homozygous terminal sires in producing hogs should be able to increase the lean meat content uniformity of hogs because a sire, especially an AI sire, can produce a large number of progeny and the dams' QTL at IGF2 gene will not cause any phenotypic variation in progeny. Using IGF2 gene to increase the uniformity of pork leanness is not just a theoretical potential. It was observed by breeding practices (e.g. Kris et al. 2002) as well.

Sow longevity (or lifetime reproduction) was said to be reducing as a result of the genetic selection for increasing leanness and lowering the fat deposition. Some studies (e.g. Brisbane and Chesnais 1996; Stalder et al. 2001) reported the association between backfat and sow longevity that has been defined as the lifetime number of litters produced by a sow. Body fat deposition is necessary to sustain sow reproduction performance, for example to supply adequate milk production and to limit body weight loss (Ranford et al. 1994). The selection for leaner carcass demanded by the packing industry and consumers may conflict with the sow longevity and lead to increased replacement costs of sows in swine production. The candidate gene IGF2 might provide a possibility to overcome this conflict. The imprinting mechanism of this major gene might be used for producing commercial hogs with required leanness from fatter dams since only the favourable alleles from homogenous sires at IGF2 are expressed in progeny.

Investigations of some other promising genes and markers are still being conducted. The opportunities are being investigated for exploiting these genes in commercial pig populations.

3.3 Methods for Incorporating Molecular Information in Current Evaluation Systems

The areas where DNA information is especially useful are the traits not measurable on living animals, such as meat quality traits; traits of low heritability, such as sow reproduction; sex-limited traits; disease resistance and early stage selection. For most of candidate genes (or SNPs) and QTLs, the best strategy is to evaluate them together with phenotypic information under optimal selection principle.

To incorporate candidate genes and QTLs into a current genetic evaluation system, the statistical model to be evaluated based on the gametic model proposed by Fernando and Grossman (1989) and ignoring epitasis is:

$$y_i = \sum_{j=1}^{M} g_j + \sum_{k=1}^{Q} (v_{ik}^1 + v_{ik}^2) + u_i + ... + e_i$$

Here, g, v and u are for effects of the candidate gene, QTL and polygene, respectively. Candidate gene effects are taken as fixed. The selection criterion will be

$$I_i = \sum_{j=1}^{M} b_j g_j + \sum_{k=1}^{Q} b_k (v_{ik}^1 + v_{ik}^2) + u_i$$

The breeding objective is to maximize the accumulative genetic response for a given period, e.g. t generations to avoid long-term disadvantage of marker-assisted selection (Dekkers et al. 1998). In Canada, the national genetic evaluation system based on animal model has been successfully used for many years. To keep the current framework of genetic evaluation and incorporate molecular information into the current system, the gametic effects at QTLs are included with the polygenic effects,

$$a_i = u_i + \sum_i (v_i^1 + v_i^2)$$

Accordingly, the mixed model equations are

$$\begin{bmatrix} X'X & X'Z & ... \\ Z'X & Z'Z + A_a^{-1}\lambda_a & ... \\ ... & ... & ... \end{bmatrix} \begin{bmatrix} \hat{\beta} \\ \hat{a} \\ ... \end{bmatrix} = \begin{bmatrix} X'y \\ Z'y \\ ... \end{bmatrix}$$

where,

$$A_a = A_u \frac{\sigma_u^2}{\sigma_a^2} + \sum_{loci} A_v \frac{2\sigma_v^2}{\sigma_a^2}$$

The A_v here can be converted from gametic relationship matrix G:

$$
G_{2n \times 2n} =
\begin{bmatrix}
\cdot & \cdot & \cdot & & \cdot & & \cdot & \cdot & \cdot & \cdot \\
\cdot & \cdot & \cdot & & \cdot & & \cdot & \cdot & \cdot & \cdot \\
\cdot & \cdot & g_{ij}^{11} & g_{ij}^{12} & \cdot & \cdot & \cdot & \cdot \\
\cdot & \cdot & g_{ij}^{21} & g_{ij}^{22} & \cdot & \cdot & \cdot & \cdot \\
\cdot & \cdot & \cdot & & \cdot & & \cdot & \cdot & \cdot & \cdot \\
\cdot & \cdot & \cdot & & \cdot & & \cdot & \cdot & \cdot & \cdot \\
\cdot & \cdot & \cdot & & \cdot & & \cdot & \cdot & \cdot & \cdot \\
\cdot & \cdot & \cdot & & \cdot & & \cdot & \cdot & \cdot & \cdot
\end{bmatrix}
\Rightarrow
\begin{bmatrix}
\cdot & \cdot & \cdot & \cdot \\
\cdot & r_{ij} & \cdot & \cdot \\
\cdot & \cdot & \cdot & \cdot \\
\cdot & \cdot & \cdot & \cdot
\end{bmatrix}
= A_{n \times n}
$$

since according to Liu et al. (2002):

$$
\frac{g_{ij}^{11} + g_{ij}^{12} + g_{ij}^{21} + g_{ij}^{22}}{2} = r_{ij}
$$

In this way, the number of equations for the polygenic effect and QTLs is equal to the number of animals to be evaluated. In the current genetic evaluation system, the computer programs remain the same. Only the numerical relationship matrix is adjusted.

4 Strategic Options for the Swine Industry

There are opportunities as well as challenges associated with the use of molecular genetic information in selection programs. Use of molecular information has led a clear improvement of the meat quality of Canadian swine. The considerable progress realized over the past years regarding knowledge of the mapped candidate genes and QTLs influencing traits of economic importance have provided molecular tools that allow potentially more exact genetic evaluation and more rapid genetic improvement. The success of integrating molecular genetics in industry breeding programs depends on the implementation strategies that make effective use the molecular and phenotypic information.

The strategic plan will be based on the results of the project. It will list research priorities and opportunities for the coordination of efforts across various research labs and swine improvement organizations across Canada. The plan will include procedures for collection of DNA samples from the nucleus and commercial herds, establishment of a DNA bank as well as specific recommendations for choosing DNA tests and marker genotyping.

The plan will be developed by the researchers and finalized after discussion by a working group of experts from government, research institutions and the Canadian pork industry.

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