

Use of IGF2 Gene Test for Genetic Improvement of Sow Productivity

2006L005R

Project Supported by
Alberta Livestock Industry Development Fund (ALIDF)



Final Report
February 2009

Acknowledgements

The project team would like to express its gratitude to the breeders, AI and regional centres who participated actively in the project and provided their valuable time, their pigs and their support in various project activities:

- Acadia Breeders Ltd.
- Apple Valley Farm
- Atlantic Swine Centre
- Bloomsbury Farms Ltd.
- Bodmin Ltd.
- Centre de Développement du Porc du Québec inc.
- Cobiporc- Québec inc.
- Centre d'Insémination Porcine du Québec
- Edulia Farms Ltd.
- F&S Hog Farm
- Fast Genetics Inc.
- Fermes de Lourdes inc.
- Fermes des Marées inc.
- Ferme Jacques Ouellet inc.
- Ferme Porasseny inc.
- Ferme Liloporc inc.
- Five Lakes Farms Ltd.
- Gwynne Vista Farms
- Huvenaars Farms Ltd.
- International Genetics PEI
- Jakubec Farms
- Novastar Genetics Inc.
- Ontario Swine Improvement Inc.
- Orford Swine
- Monoway Farms Ltd.
- PEAK Swine Genetics Inc.
- Rose Briar Farm
- Sand Ridge Farm Ltd.

Special thanks to Dr. Nadine Buys from Belgium who participated as a consultant to the project.

The support from the Alberta Livestock Industry Development Fund (ALIDF) is gratefully acknowledged. It would not have been possible to carry out the project without their funding support.

Project team

The project team consisted of members from Western Swine Testing Association (WSTA), the Canadian Centre for Swine Improvement (CCSI) and the University of Guelph.

The project was led by the Western Swine Testing Association (WSTA). WSTA is a not-for-profit organization that provides testing and evaluation services to the swine industry in Western Canada. A variety of commercial industry and seedstock oriented services are offered including: technician or owner tester genetic improvement programs, carcass evaluation services, commercial testing of replacement gilts or ultrasonic evaluation of fat, loin size and lean yield % for lean growth curves and ultrasonic technician training. More information about WSTA is available at www.wsta.ca. WSTA coordinated the research activities with the producers and other project team members.

CCSI is a national non-profit organization created by the industry to provide leadership and services related to swine genetic improvement in Canada. Its members include Canadian Pork Council (CPC), Canadian Meat Council (CMC), Canadian Swine Breeders Association (CSBA) and regional swine improvement centres in Western Canada, Ontario, Quebec and Atlantic Canada. More information about CCSI is available at www.ccsi.ca. CCSI geneticists assisted in project design, data analysis, development of recommendations and guidelines as well as the development of computer-based applications for effective use of the technology.

The Laboratory Services, University of Guelph is a multi-faceted analytical and diagnostic laboratory which serves a broad range of agri-food and veterinary clients within government, commercial and academic sectors on a fee-for-service basis. It is a state-of-the-art ISO 9001:2000 registered facility. The laboratory extracted DNA from tissue samples including blood and completed the genotyping of boars and sows for the IGF2 gene.

Table of contents

1. ABSTRACT.....	5
2. BACKGROUND INFORMATION.....	6
3. PROJECT OBJECTIVES	9
4. PROJECT DESIGN AND METHODOLOGY	10
5. RESULTS	14
6. IMPLICATIONS OF THE PROJECT WITH REGARDS TO THE IMPROVEMENT OF ALBERTA LIVESTOCK INDUSTRY	23
7. LITERATURE CITED.....	25

1. ABSTRACT

The IGF2 gene is located on porcine chromosome 2 and a single nucleotide polymorphism (SNP) in intron 3 of this gene has been reported to have large effects on certain carcass quality traits such as lean meat content. Several studies have also described the use of this paternally expressed gene to produce leaner hogs from fatter sows. Moreover, it has been reported in certain studies that the gene could have an effect on sow productivity traits and longevity. The aim of this study was to investigate the effect of IGF2 gene on several sow productivity traits such as litter size at birth, litter weight at weaning, number of piglets weaned and farrowing interval in Canadian swine populations. As a first step, boars were screened in the main Canadian swine breeds in order to find heterozygous boars (AG) for the IGF2 gene. Daughters of heterozygous (AG) boars that inherited either the A or G allele from their sire were studied in Yorkshire and Landrace breeds. In total, 1234 litters born from 441 sows were included in the analysis. Results show that a difference between Landrace and Yorkshire breeds in the effects of the IGF2 gene on sow productivity traits exists. In Yorkshire breed, sows having inherited the paternal A allele had more piglets born (+1.34) than their sisters having inherited the G allele. Yet, in both breeds, a similar tendency was seen, with sows that inherited the paternal G allele weaning more piglets (+0.17 and +0.24 piglets/litter in Landrace and Yorkshire, respectively) and heavier litters (+2.5 and +2.3 kg/litter in Landrace and Yorkshire, respectively) in comparison to daughters of AG sires that inherited the paternal A allele. Further research is required to investigate other potential candidate genes located in close proximity to the IGF2 gene which may affect litter size. There is also a need to accumulate more genotype information and records in order to have more and larger sire families included. This project has helped getting a better understanding of the IGF2 gene effects on reproduction traits and its optimal use by breeders and producers.

2. BACKGROUND INFORMATION

2.1. Use of IGF2 Gene Test for Genetic Improvement of Sow Productivity

Large effects of Insulin-like Growth Factor 2 (IGF2) gene on certain carcass quality traits such as lean meat were reported by several studies. The imprinting effect allowing to use fat sows to produce leaner hogs has been described in several studies as well. However, very few studies have been conducted on the effect of the IGF2 fat genotype on sow productivity.

Overview of research completed on IGF2 in swine

- Andersson-Eklund *et al.* (1998), in a collaborative study between Swedish and British researchers, found numerous Quantitative Trait Loci (QTL) which had an effect on percentage of lean meat and the proportion of muscle to bone in wild boar and Large White intercross. One of these QTL was mapped to chromosome 2. This QTL had a moderate effect on lean meat content as well as on lean meat plus bone.
- Jeon *et al.* (1999) re-analyzed the data from Andersson-Eklund *et al.* (1998) and detected a maternally imprinted QTL at the distal tip of chromosome 2 that had a significant effect on lean meat content in ham, longissimus dorsi muscle, heart weight and backfat thickness as well as a moderate effect on reflectance value. The QTL linked to IGF2 could explain 15-30% of the phenotypic variation in muscle mass as well as 10-20% of the phenotypic variation in backfat thickness.
- Nezer *et al.* (1999), researchers from Belgium, found that IGF2 is imprinted in tissues of pigs as well as in human and mouse.
- Laere *et al.* (2003) detected that the causative IGF2 mutation was caused by a nucleotide substitution (G→A) in intron 3 at nucleotide position 3072. The assumption that selection for lean growth caused a reduction of the IGF2 G allele in swine operations was suggested.
- Buys (2003) studied the segregation pattern of the IGF2 alleles. She found that the A allele of the IGF2 gene increased meat production between 3-4% and reduced backfat by 2.3 mm on average. She also found that using homozygous AA boars to produce market hogs could bring 1.98% more lean meat and greater carcass uniformity than the other genotypes.
- Jungerius *et al.* (2004) looked at IGF2 genotypes in white European and Meishan crossed swine. They found that the IGF2 polymorphism could explain a substantial part of the variance in backfat thickness and showed that on average animals that

- inherited the G allele from their father, had approximately 1.6 millimeters more fat than animals that inherited the paternal A allele.
- Estellé *et al.* (2005) found that in Large White commercial pigs, Iberian and Landrace F2 intercross, the A allele had a significant interaction with backfat thickness, lean muscle area and ham weight. The authors reported that the A allele had a tendency to decrease backfat thickness while increasing carcass weight, lean muscle area, ham and shoulder weight.
 - Buys *et al.* (2006) examined the effect of IGF2 on sow productivity in Large White and Landrace breeds. The author reported that sows which inherited the paternal G allele from heterozygous sires had significantly higher number of piglets born alive per litter, higher total born piglets per litter, and higher number of piglets weaned. These sows which inherited the G allele also had greater average number of parities than sows that inherited the A allele. In contrary to other studies (Andersson-Eklund *et al.*, 1998; Jeon *et al.*, 1999), reported no IGF2 effect on carcass measurements.
 - Vykoukalová *et al.* (2006) looked at four different Single Nucleotide Polymorphisms (SNPs) in the IGF2 gene. The authors reported that the IGF2 A allele is fixed in Duroc and Hampshire swine populations. They also found a significant difference between IGF2-intron 7-G6162C and backfat. The authors suggested further research to study the effect of the polymorphism of intron 7 on fatness traits and its usefulness for selection.
 - Van den Maagdenberg *et al.* (2007) investigated the effects of IGF2 on proteolytic and lipolytic enzyme activity. They found that hogs that inherited the paternal A allele had relatively higher weight at slaughter than hogs inheriting the paternal G allele. The authors also suggested that the increased muscle mass in animals that inherited the paternal A allele may be associated with decreased protein degradation and increased myoblast fusion or both.
 - Gardan *et al.* (2008) looked at the effect of IGF2 genotype on adipocytes (fat cells) and on subcutaneous tissues. The authors found that IGF2 mRNA was higher in muscles of pigs that inherited the paternal A allele than those that inherited the G allele but not in subcutaneous tissues. They also observed that the number of adipocytes were not different between both groups, just that the animals with paternal A allele had smaller adipocytes. The authors concluded that the IGF2 mutation favours myofiber hypertrophy and reduces fat cell development in subcutaneous tissues.

2.2. Current status of research on IGF2 in Canadian swine populations

As part of a project on meat quality funded by ALIDF in 2005, 255 purebred pigs were tested for IGF2. The results of the project suggested that the IGF2 gene has a large, favourable and significant effect on backfat thickness, lean yield, lean depth and loin eye area. The pigs with

lean genotype (AA) had 7.11 mm less backfat, 4.3% more lean meat, 7.41 mm higher lean depth and 7.22 sq cm larger loin eye area compared to the fat genotype (GG). Results from this study corroborate results from previous studies on IGF2 and is novel in the sense that it offered a preliminary view of IGF2 frequencies and associations to performance traits in Canadian swine populations.

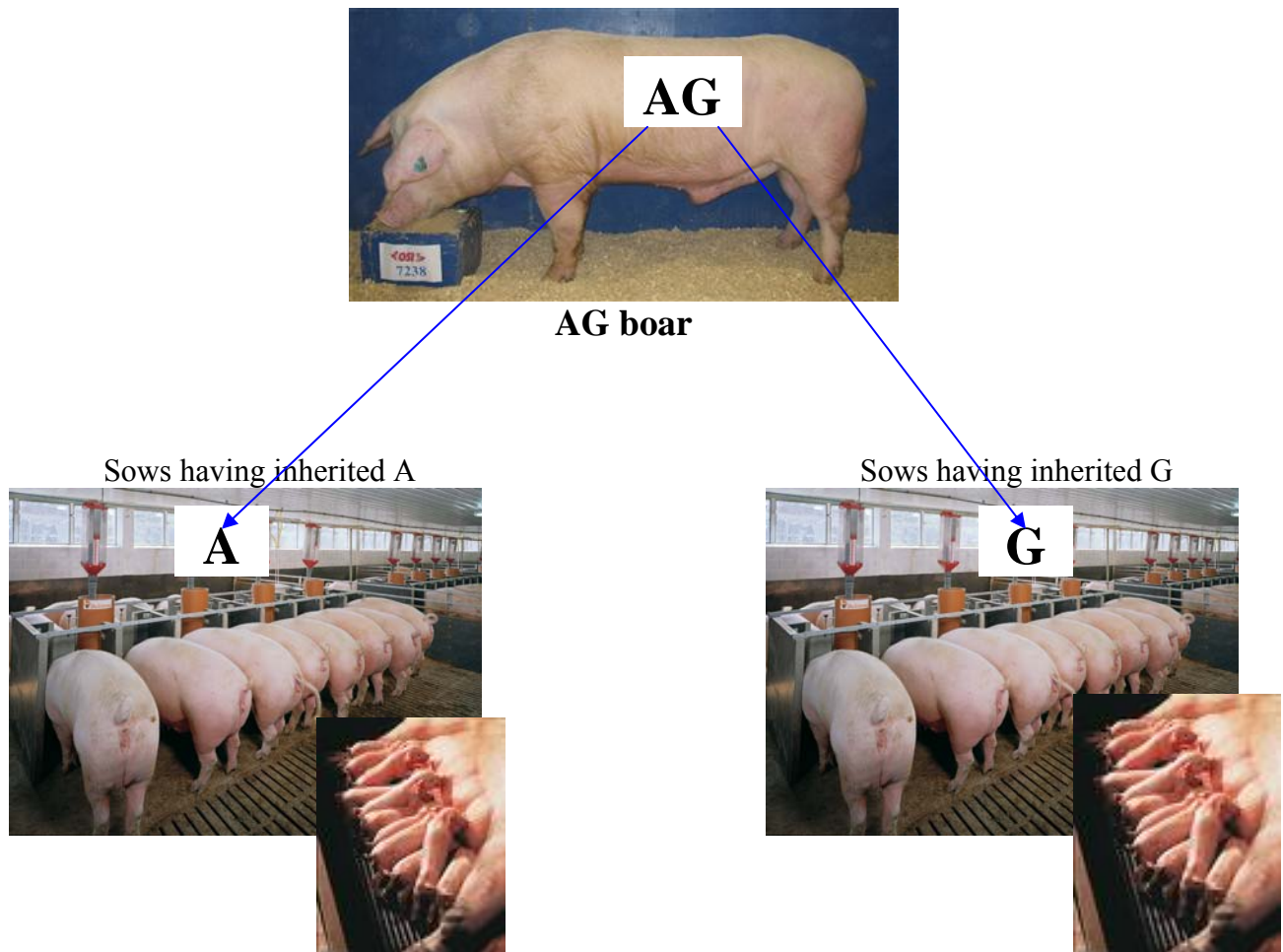
It is possible that the increased selection for lean carcasses has resulted in the increase of the mutant A allele in swine populations. It has also been suggested that IGF2 affects sow productivity and longevity (Buys *et al.*, 2006). So far, few studies have been done on the effects of IGF2 gene on sow productivity traits. Mathur *et al.* (2003) suggested that sows that inherited the paternal G allele would have a higher prolificacy and better body condition. This project aims to document the effect of the paternally inherited IGF2 alleles on various sow productivity traits in Canadian purebred sows.

3. PROJECT OBJECTIVES

The objective of this project was to evaluate the usefulness of the new and innovative approach of using DNA tests to better manage sow productivity traits, in particular the test for the IGF2 gene. Specific traits of interest included total number of pigs born per litter, perinatal piglet survival, number of pigs weaned, litter weight at weaning and farrowing interval.

4. PROJECT DESIGN AND METHODOLOGY

The first phase of genetic screening was to find heterozygous boars (AG) with purebred daughters having litter records known in the CCSI database. The second phase was to test the daughters of AG boars to identify which ones received the A allele and which ones the G allele from their sire. The goal was to compare the productivity of daughters that have inherited the paternal A allele versus those that have received the G from their sire.



4.1. Animals

4.1.1. Screening for heterozygous boars

Blood samples from boars in various Western Canadian AI centres and breeding herds were sampled and sent to the University of Guelph for IGF2 testing. This step served as a screening process in order to identify boars heterozygous for IGF2. Initially the screening was on

available sires in participating herds in Western Canada, mainly in Alberta, and relatively few AG boars were discovered. Additional screenings focused on sires with largest number of active daughters in breeding herds, but again relatively few AG boars were found. As more animals were genotyped, probabilities of genotypes in non-genotyped pigs were estimated using software which was developed at CCSI for defining the parental origin of IGF2 in large pedigrees (Jafarikia *et al.*, 2008). A final screening was focused on sires which had the highest probability of carrying the G allele. This turned out to be very effective at increasing the number of AG sires found. Neither AG, nor GG boars were found in Durocs, where the A allele may be fixed, as found in other populations (Vykoukalová *et al.*, 2006).

4.1.2. Sampling of daughters from heterozygous boars for IGF2 testing

As soon as an AG boar was identified, the CCSI database was searched to find all female progeny with litter records and potentially still available for blood sampling. Breeders were also asked to sample other daughters that did not yet have litter records in the CCSI database, but which had been kept for breeding. The dams of the daughters were sampled as well if available. Since no AG boar was found in Duroc, the sampling of daughters was limited to Landrace, Yorkshire and Lacombe breeds.

Table 1 shows the numbers of heterozygous (AG) sires that were identified, and the numbers of their daughters that were genotyped by breed. Blood sampling of daughters of AG boars to form large sire families for analysis proved to be a large challenge, in part due to the difficulty in finding AG sires and in part due to the quick turn-over in breeding herds resulting in many daughters having been culled before they could be sampled.

Table 1. Number of heterozygous (AG) boars found and number of daughters genotyped

<i>Breed</i>	<i>Number of AG Sires</i>	<i>Daughters of AG sires</i>
<i>Landrace</i>	23	384
<i>Yorkshire</i>	12	136
<i>Lacombe</i>	2	13
<i>Duroc</i>	0	0
<i>Total</i>	37	533

In an earlier study by CCSI (unpublished), the G allele frequency was found to be higher in Landrace in comparison to Yorkshire (Table 2). As could be expected, there were not as many potential sire families found in Yorkshire compared to Landrace in this study. In Lacombe, the G allele frequency was found to be relatively high the previous study. However, because of the small population size of this breed, a limited number of boars was available for testing, and the records on the small number of daughters tested (13) were not included in the analysis. Crossbred daughters tested for IGF2 under the project were also excluded from the analysis due to the low number of crossbreds sampled.

Table 2. Frequency of IGF2 alleles by breed found in an early study (CCSI, unpublished)

<i>Breed</i>	<i>Sires</i>		<i>Females</i>	
	A	G	A	G
<i>Duroc</i>	1.00	0.00	1.00	0.00
<i>Lacombe</i>	0.75	0.25	0.22	0.78
<i>Landrace</i>	0.86	0.14	0.65	0.35
<i>Yorkshire</i>	0.96	0.04	0.85	0.15
<i>Duroc</i>	1.00	0.00	1.00	0.00

4.2. Tissue sampling, DNA extraction and Genotyping of IGF2

Two to five milliliters of blood were sampled from each animal in one EDTA tube per animal. Subsequent to blood collection, the tubes were labeled, kept on ice and shipped to the University of Guelph laboratory for IGF2 testing. In some cases, ear notches or semen were sent to the laboratory for DNA extraction and IGF2 testing. Upon arrival at the laboratory, DNA was extracted from samples and the genetic material was prepared and cleaned for genotyping. Genotyping and sequencing of IGF2-intron 3-3072 was completed following the method outlined by Laere *et al.* (2003).

4.3. IGF2 genotype reporting

Reports containing the IGF2 results were sent to the individual breeders and IGF2 results were also stored in CCSI's database for further joint analysis with sow productivity traits. IGF2 genotypes were also made available to breeders for consultation on CCSI's website using search tools such as PigsOnTheWeb. PigsOnTheWeb is a useful tool for breeders to search the database for specific individual pigs or a group of pigs so that they can access all of their pedigree, performance and genetic information including IGF2 results.

4.4. Data analysis

4.4.1. Computation of IGF2 genotype probabilities

A simple two-step method for estimation of genotype probabilities for animals with unknown genotypes was developed as part of another research project at CCSI (in progress). In the first step, the available genotypes from each animal and its parents were used to estimate the parental allele probabilities. In every round of the second step, Bayes' theorem was used to estimate the genotype probabilities of each parent using the average genotype probabilities of all its progeny and then estimated genotype of each parent was used to re-estimate the genotype probabilities for progeny. The algorithm was successfully applied to this project using the genotype and pedigree information (Jafarikia *et al.*, 2008).

4.4.2. Statistical analysis of sow productivity data

Sow productivity data collected by breeders under the Canadian Swine Improvement Program were used in this project, in order to ascertain if sow productivity differences exist between daughters that inherit the IGF2 paternal A allele versus the daughters that inherit the paternal G allele. The traits analyzed included: total number of piglets born per litter, number of piglets born alive per litter, number of stillborn piglets per litter, number of piglets weaned per litter, litter weight at weaning and farrowing interval.

Litter records from genotyped sows born from AG sires were used in the analysis. Data was analyzed using the GLM procedure from SAS software, including inherited IGF2 paternal allele (A or G) by the sow, contemporary group (combination of herd, year and quarter), sire, parity (1, 2, 3, 4-5, 6 and higher), service sire breed and inbreeding level of both the sow and the litter. These are the same fixed effects as in the model used in national genetic evaluations for these traits plus the effect of the paternal allele for IGF2.

5. RESULTS

5.1. Genotype frequencies of daughters of AG sires

5.1.1. AG sires

The data set presented in Table 3 includes 23 Landrace and 12 Yorkshire sire families. There was a large range in the sire family sizes, with the number of daughters per sire ranging from 1 to 65. There was an average of 12.6 daughters per sire with an average of 2.8 litters per daughter. In each breed, one or two boars had a large number of daughters with litter records.

5.1.2. Genotyped sows

Among the genotyped progeny of AG boars, 60 daughters (54 Landrace and 6 Yorkshire) did not have any litter records known in CCSI's database. In some cases, this could have occurred because the sow was supposed to be kept for breeding but was culled, or because litter records have not yet been sent to CCSI's database. The litters of sows in contemporary groups (combination of herd, year and quarter) with less than 10 records were also excluded from the analysis. This step excluded 17 Landrace and 2 Yorkshire sows. Table 3 shows the numbers of genotyped sows originating from AG sires and used in the analysis, within each breed. The sows under study were present in 16 Landrace herds and 11 Yorkshire herds.

Table 3. Numbers of daughters of AG boars having litter records in the CCSI database

<i>Breed</i>	<i>AG boars</i>	<i>Originating herds of AG boars</i>	<i>Daughters by genotype (number of litters)</i>			
			<i>AA</i>	<i>AG</i>	<i>GG</i>	<i>Total</i>
<i>Landrace</i>	23	10	144 (437)	156 (415)	13 (41)	313 (893)
<i>Yorkshire</i>	12	10	75 (186)	51 (151)	2 (4)	128 (341)

5.1.3. AG daughters

In this project, the IGF2 allele received from the sire (A or G) was of primary interest, since the IGF2 gene is paternally expressed in the progeny. This means that only the allele transmitted by the sire is expressed. Daughters of AG sires which have been genotyped AA have received the A allele from their sire. Similarly, the GG daughters have received the G allele from their sire. In the case of AG daughters, they can be either AG (A received from the sire and G received from the dam) or GA (G received from the sire and A received from the dam). When the dam had a homozygous genotype (AA or GG), it was possible to determine which allele the

daughter inherited from its sire. For instance, an AG daughter born from an AG sire and an AA dam must have received the G allele from the sire since the G allele is not present in the genotype of the dam. In this project, relatively few dams were available to be genotyped (39 Landrace and 6 Yorkshire). In order to determine the probability of inheriting the paternal G or A by the daughter, the genotype probabilities computed for the AG daughters were based on all genotype information available in the population.

Figure 1 shows the distribution of the probability of having inherited G from the sire for the Landrace AG daughters. Figure 2 shows the distribution for Yorkshire AG daughters. The frequency of the G allele in an earlier study conducted at CCSI was 35% in Landrace females and 15% in Yorkshire females (Table 2). Thus, when the dam's genotype is unknown, there is a greater probability that AG daughters will have received the G allele from their AG sire. This is especially true for Yorkshires, where the frequency of the G allele in the population is low. The estimation of the probabilities shown in these figures is a function of the overall population frequencies for the breed and sex, as well as the known genotypes of relatives. The probabilities of AG daughters having received the G from their sire are higher on average for Yorkshire compared to Landrace, as can be seen in figure 2 compared to figure 1.

For the purpose of this study, heterozygous daughters with a probability higher than 50% of having inherited the paternal G allele were classified as GA. It is recognized that this approach will tend to underestimate any true effects of the IGF2 allele, since several daughters which have been assumed to be GA (G allele from the sire) might actually be AG (G allele from the dam). Although beyond the scope of this project, more sophisticated genetic models could be used to provide unbiased estimates of the effects of IGF2. Such an approach is being developed under a separate project which is currently in progress at CCSI. Based on this simplified classification, sows were grouped into three categories for further analyses: those assumed to have received the paternal A allele, those assumed to have received the paternal G allele and those for which the paternal allele could not be determined. Consequently, in Landrace breed, from the 156 AG sows, 135 were classified as GA, 4 as AG and 17 as unknown. The 17 unknown genotypes were offspring of an AG sire and an AG dam. In this situation the progeny has a 50% chance of being either AG or GA. In Yorkshire, the 51 heterozygous sows were classified as 48 GA and 3 as unknown.

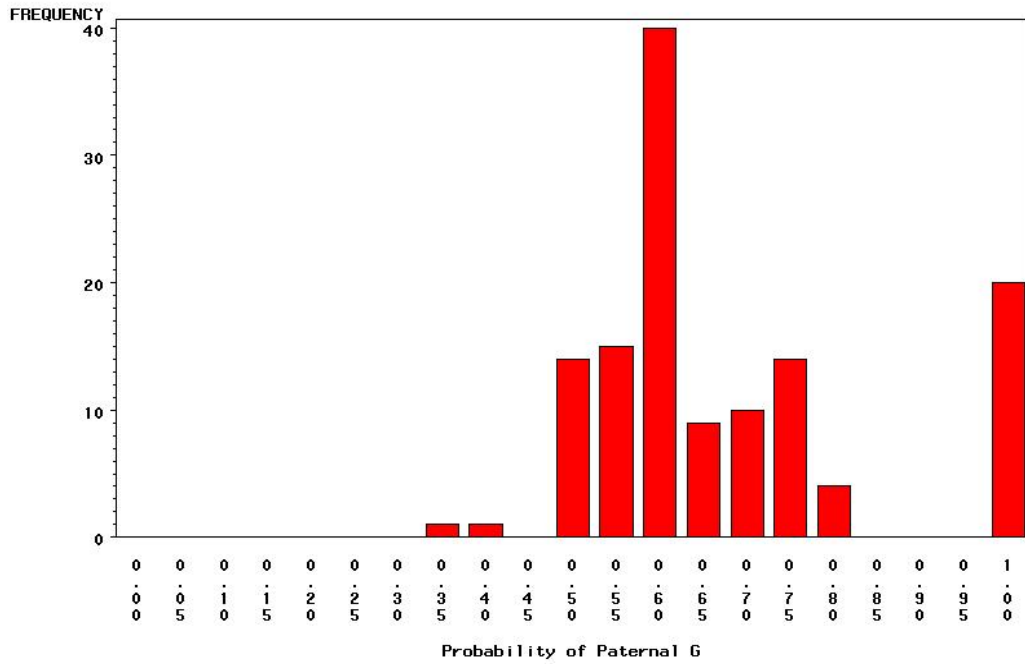


Figure 1. Distribution of the probability of having inherited IGF2 G allele from the sire in heterozygous Landrace sows

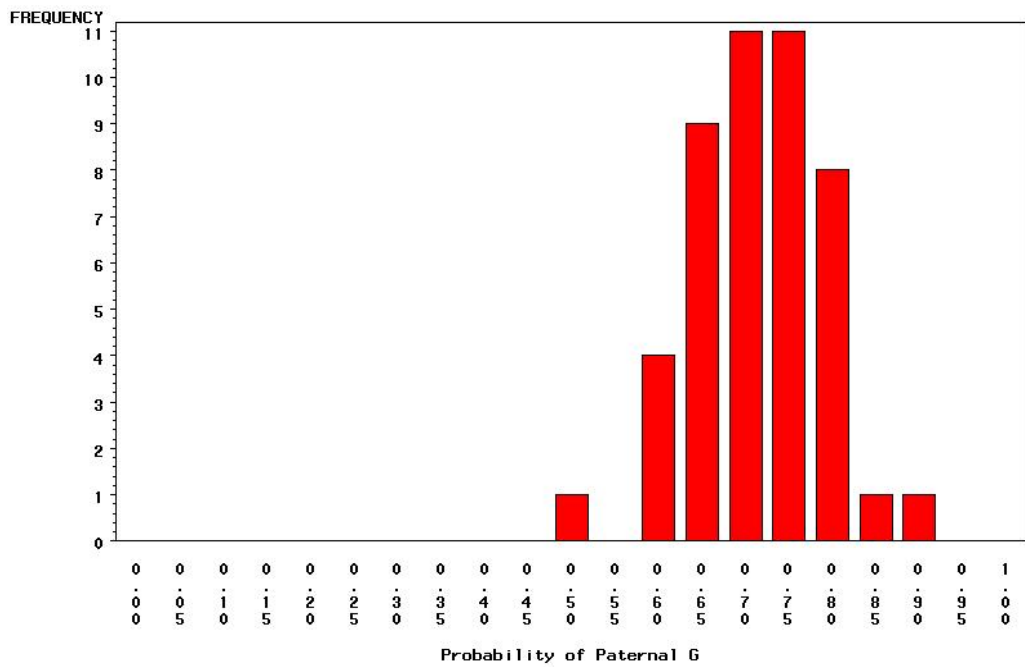


Figure 2. Distribution of the probability of having inherited IGF2 G allele from the sire in heterozygous Yorkshire sows

5.2. Effect of IGF2 paternal allele on sow productivity traits

5.2.1. Descriptive statistics

Sample statistics for sow productivity traits of daughters of AG boars are shown in Table 4, regardless of what IGF2 allele was inherited (paternal A or G). In total 1234 litters from 441 sows were analyzed, among which 78% had litter size at weaning and 42% had litter weight at weaning. Note that average parity was relatively low in this study since only younger sires were available for genotyping in most cases. The exception was for older AI sires that were previously genotyped for IGF2, or where there was tissue or DNA stored from earlier projects.

Table 4. Sample statistics for sow productivity records available on daughters of AG boars

	<i>Landrace (n=313 sows)</i>			<i>Yorkshire (n=128 sows)</i>		
	# records	Average	Std Dev	#records	Average	Std Dev
<i>Average parity</i>	893	2.61	1.62	341	2.58	1.45
<i>Total piglets born/litter</i>	893	11.87	3.12	341	11.84	3.10
<i>Piglets born alive/litter</i>	893	11.02	3.17	340	10.96	2.96
<i>Stillborn piglets/litter</i>	893	0.85	1.34	340	0.88	1.40
<i>Piglets weaned/litter</i>	664	9.80	1.69	304	9.94	1.44
<i>Litter weight at weaning (kg)</i>	428	64.9	15.0	85	63.7	14.0
<i>Farrowing interval (days)</i>	528	145.3	9.8	184	145.2	9.4

5.2.2. Gene effects

Table 5 shows the adjusted means by sire allele class for total born, born alive, stillborn, number of piglets weaned per litter, litter weight at weaning and farrowing interval in Landrace and Yorkshire breeds.

Table 5. Least-square means (\pm standard error) for sow productivity traits

<i>Sow breed</i>	<i>Landrace</i>		<i>Yorkshire</i>	
	A	G	A	G
<i>Paternal allele inherited</i>	A	G	A	G
<i>Number of sows</i>	148	148	75	50
<i>Number of litters</i>	448	373	186	149
<i>Average parity</i>	3.10	2.61	2.39	2.85
<i>Total piglets born/litter</i>	11.92 \pm 0.30	11.66 \pm 0.31	12.95 \pm 0.69**	11.61 \pm 0.69**
<i>Piglets born alive/litter</i>	11.18 \pm 0.30	10.84 \pm 0.31	11.83 \pm 0.65**	10.80 \pm 0.66**
<i>Stillborn piglets/litter</i>	0.75 \pm 0.12	0.83 \pm 0.12	1.15 \pm 0.28*	0.84 \pm 0.29*
<i>Piglets weaned/litter</i>	9.79 \pm 0.17*	9.96 \pm 0.17*	9.34 \pm 0.39	9.58 \pm 0.38
<i>Litter weight at weaning (kg)</i>	64.1 \pm 1.0*	66.6 \pm 1.1*	58.4 \pm 2.3	60.7 \pm 2.7
<i>Farrowing interval (days)</i>	145.8 \pm 0.7	146.1 \pm 0.7	145.9 \pm 1.1*	148.2 \pm 1.2*

**Significantly different at 99% confidence level ($p < 0.01$)

*Significantly different at 90% confidence level ($p < 0.10$)

In Landrace, tendencies were observed for number of weaned piglets and litter weight at weaning ($p < 0.10$). Daughters with the paternal G allele averaged 0.17 more pigs weaned and 2.5 kg heavier litter weight at weaning than daughters with the paternal A allele. This is in spite of the fact that total pigs born was higher in the A daughters compared to G daughters.

In Yorkshire, sows that inherited the A allele from the sire had significantly more piglets born per litter (+1.34) than their sisters that inherited the G allele from the sire ($p < 0.01$). A statistical significance in the number of piglets born alive was also observed with an advantage of +1.03 for sows that have inherited the paternal A allele ($p < 0.01$). Two tendencies showed that sows that inherited the paternal G allele had fewer stillborn piglets per litter ($p < 0.10$) and a longer farrowing interval ($p < 0.10$) than sows that inherited the paternal A allele from their sire. In spite of significantly larger litters at birth, there was no significant difference in number weaned or weaning weight between the Yorkshire A daughters compared to G daughters.

Although significant and showing some tendencies in some cases, the results should be interpreted with caution. The number of daughters per sire family ranged from 1 to 65, which created a relatively unbalanced data structure. This potentially created some estimation issues. In

addition, sows are spread out in many different farms sometimes with small contemporary groups.

More sire families and more daughter productivity records per sire would help to have a clearer conclusion. To some extent this will happen naturally as the existing daughters are given the opportunity to have more litters. Additionally, although beyond the scope of this project, a more sophisticated genetic analysis (i.e. marker assisted BLUP using genotype probabilities for all animals) could also give clearer conclusions. This more sophisticated analysis will be done as part of another research project which is in progress at CCSI. There might also be additional influencing factors such as other major genes segregating in these families, or other genes associated by chance with IGF2 in some sires. As more sows sired by AG boars have litter and performance records sent to CCSI database, and with the more sophisticated genetic analysis, a clearer image should emerge.

5.2.3. Example of two sire families

Due to the variation in sire family structure and the possible interaction between some environmental and genetic effects in the analysis, a look at differences in performances of sows having received the A versus the G allele from their sire, but within a specific large sire family in each breed, was undertaken.

Table 6 shows adjusted means by sire allele class, for total born, born alive, stillborn, number of piglets weaned per litter, litter weight at weaning and farrowing interval within the two largest sire families in the dataset. One Landrace boar had 62 daughters (66 litters) in 7 herds and one Yorkshire boar had 63 daughters (201 litters) in 10 herds.

Table 6. Adjusted means for sow productivity traits within two large sire families

	<i>Landrace Boar</i>		<i>Yorkshire Boar</i>	
	A	G	A	G
<i>Paternal allele inherited</i>				
<i>Number of sows</i>	17	45	34	29
<i>Number of litters</i>	17	49	107	94
<i>Average parity</i>	1.00	1.08	2.80	3.07
<i>Total piglets born/litter</i>	11.14 ± 1.39	10.92 ± 1.23	13.68 ± 0.90*	12.45 ± 0.90*
<i>Piglets born alive/litter</i>	10.19 ± 1.35	9.55 ± 1.20	12.73 ± 0.85	11.98 ± 0.86
<i>Stillborn piglets/litter</i>	0.95 ± 0.59	1.37 ± 0.52	1.01 ± 0.40*	0.54 ± 0.40*
<i>Piglets weaned/litter</i>	-	-	10.09 ± 0.54	10.29 ± 0.54
<i>Litter weight at weaning (kg)</i>	57.92 ± 11.62	62.31 ± 6.42	-	-
<i>Farrowing interval (days)</i>	-	-	152.2 ± 1.6	1.53 ± 1.7

* Significantly different ($p < 0.05$)

From the within-family results shown in table 6, two different patterns are observed. In the Landrace family there were no significant differences found, possibly due to the relatively low number of records. In the Yorkshire family, significantly larger litter size at birth (+1.23 pigs) was found for daughters that inherited the paternal A allele, but these daughters also had significantly more stillborn piglets per litter (+0.47) than daughters that inherited the paternal G allele. Not surprisingly, the results within the family of this Yorkshire boar were similar with what was seen in the overall Yorkshire sample (Table 5), since daughter records from this boar represents more than 50% of the Yorkshire data set.

5.3. Discussion and recommendations

This study found that the paternal A allele in Canadian Yorkshire was associated with significantly larger number of pigs born and born alive, while there was no significant difference for these traits in Canadian Landrace. This result is in contrast to what was found in a study on Large White and Landrace lines in Belgium, Buys *et al.*, (2006), which reported that sows which inherited the paternal G allele from AG sires had significantly higher number of total born and born alive. On the other hand, the results of this study provide an indication that there may be a positive association of the paternal G allele of IGF2 with number weaned and weaning weight. This is consistent with the results of Buys *et al.*, (2006).

The results varied somewhat among the traits and between breeds in this study, and compared to the results of Buys *et al.*, (2006). This was illustrated by examining the two largest sire families. A few large sire families accounted for the majority of the data in this study,

especially for Yorkshire where the largest family represented more than half of the data. One possibility is that the effect of IGF2 varies with individual animals or families. This could be caused by non-additive genetic effects such as interactions with other genes, or by linkages to other genes on the same chromosome that also affect sow productivity traits.

The first phase in this project involved a screening to find heterozygous (AG) boars for IGF2. Unfortunately, screening of sires turned out to be impractical since most of the sires that had mature daughters were no longer available for sampling. This created practical limitations which resulted in relatively few sire families, unbalanced family sizes and generally only young families with relatively few daughters available for testing in the second phase. Further, most of the dams of the daughters of AG sires were also no longer alive and therefore could not be genotyped. This resulted in uncertainty with regards to whether an AG daughter received the G from her sire or her dam, thus reducing the statistical power of the analysis. A simple solution to both these issues for future projects requiring DNA testing would be to routinely store tissue samples from all parents of nucleus animals. If such a resource had been available for this project, more informative results could have been achieved and it could have been done with much less time and effort.

As more data accumulate on the young daughters that were genotyped in this project, further analysis could reveal more clearly the effects of IGF2 on the traits studied. It could also allow investigation of an association with longevity, as in Buys *et al.*, (2006) which showed an indication of an association between the G allele and higher average parity.

Further projects should involve more data and more genotyped animals, especially in such a design where 15 to 20 participating herds contribute pigs and records. While it is a great advantage to have so many herds participating in such a project and providing access to many animals within the large populations of Canadian maternal breeds, it also represents a statistical challenge to properly estimate the various environment effects when families are not large enough.

A very promising avenue is to look at IGF2 gene effects using the genotype probabilities instead of only the genotyped animals. CCSI has developed a program to evaluate IGF2 genotype and phase probabilities for a whole breed, based on the genotype information available on a limited number of genotyped pigs (Jafarikia *et al.*, 2008). For instance, in Canadian Landrace, Yorkshire and Lacombe breeds, the genotype data on a few hundreds pigs have been used to estimate genotype probabilities for several hundreds of thousands of pigs known in the CCSI database. These genotype probabilities will be used in the near future, along with sow productivity data to simultaneously compute estimated breeding values (EBVs) and the effects of IGF2 gene on several traits of interest. Ultimately, the genotype information will be used in genetic evaluations for several economically important traits related to carcass quality and sow productivity.

Further projects could also look at the effect of IGF2 genotype in hybrid sows (Yorkshire×Landrace). From this analysis on purebred sows, it seems that the effect of IGF2 gene might not be exactly the same in Landrace and Yorkshire breeds. However, given the small number of sires with the majority of data in this study, it isn't clear whether or not the effects of

IGF2 vary by sire or family, in contrast to overall breed differences. Some research (Buske *et al*, 2006) has suggested that there are other genes on chromosome 2 with large effect on sow productivity, specifically FSHb which has shown a relationship with total born and born alive, and EPOR which has shown a relationship with uterine capacity. IGF2 could potentially be a marker for these other quantitative trait loci (QTL) and have different linkage phases with the other unknown QTL in different families or between breeds. This could explain varying effects of IGF2 on sow productivity traits in different families or between breeds. Since interactions may exist with the genetic background of the sow, it would be interesting to evaluate the impact of the paternal IGF2 allele on an average F1 sow. The applications coming from such a study would be direct and practical for the commercial swine industry.

6. IMPLICATIONS OF THE PROJECT WITH REGARDS TO THE IMPROVEMENT OF ALBERTA LIVESTOCK INDUSTRY

6.1. For breeders

The breeding sector will be able to select sows that have better and more consistent body condition (Buys, 2003), with possible enhanced fertility and sow productivity (Buys *et al.*, 2006). It might be interesting to breed for homozygous GG boars, in order to be able to produce F1 sows born from a GG boar (thus receiving a paternal G allele). This might be easier to do in the Landrace breed, where the G allele frequency is higher. Only one of the maternal lines, such as Landrace, needs to carry the G allele, since only the paternal allele is expressed in the progeny. In other words, all the F1 daughters from a GG multiplier boar would benefit completely from the effects of IGF2, regardless of the IGF2 genotype of the multiplier sow from the other maternal line such as Yorkshire. Ideally, the terminal boar should be AA, in order to confer leaner carcass composition and uniformity to the progeny. This is most likely already the case for the Duroc population, but boars could also be screened within Yorkshire and Lacombe breeds, as well as other terminal sire lines, to ensure that they have the AA genotype for IGF2.

6.2. For producers

The producers will benefit from the improved genetics and methods for their optimum use. According to an earlier research project completed with support from ALIDF, the paternal A allele of IGF2 gene led to an increase of about 4.3% in lean yield of IGF2 AA market hogs compared to the IGF2 GG hogs (Mathur and Liu, 2006). On a typical grading grid used in Canada, a one percent increase in lean yield is worth \$1.17/market hog. In other words, potential benefit to the producer is approximately \$5 for each market hog that doesn't carry the paternal A allele. Alberta producers can ensure that they are getting the paternal A in their market hogs by testing and using homozygous AA terminal boars. To illustrate the significance, for every 1% of hogs marketed in Alberta (about 25,000 hogs) at \$5 more per hog, this would mean a direct benefit to producers of about \$75,000. What percentage of hogs are currently affected by the paternal G allele in Alberta is unknown, but with the exception of Durocs, all the breeds evaluated in this study have much more than 1% frequency of the G allele (see Table 2). For individual producers that are unlucky enough to have terminal boars carrying the G allele, the percentage of hogs affected could in fact be very large.

This study provides an indication that use of the paternal G allele in breeding replacement gilts for commercial herds will enhance productivity through higher numbers weaned and higher weaning weights. This potential benefit in productivity can come without any compromise on the lean yield in market hogs because of the fact that the effect of the G allele from the sow will not be expressed in her progeny. Thus, the benefits are on top of the higher carcass value mentioned above. More study should be done to confirm these effects in nucleus populations, to see if there is an additional benefit from sow longevity, and also to look at the effects directly in the commercial sows.

Moreover, the use of marker assisted selection using IGF2 and other technological advances in genomics may over time increase the accuracy of selection in Alberta swine herds.

This can be done using a DNA sample without requiring the sacrifice of animals and without the costs of measuring traits on the carcass (Fernando and Grossman, 1989; Meuwiswn and Goddard, 1996). Thus, this approach is not only more effective, it is also more practical and economical. The study on the effects of IGF2 on sow productivity was one step in that direction where breeders and producers can benefit from such studies.

6.3. For packers

The packing plants will get more uniform hogs due to similar genotypes with respect to this gene with large effects (Buys, 2003). By reducing the incidence of the paternal G allele in market hogs, the effect is expected to be mostly on the bottom end of the lean yield distribution. In other words, the leanest hogs most likely already have the paternal A allele so won't be getting leaner. It is the fattest hogs that most likely have the paternal G allele. Hence, packers can expect more uniformity with a desirable level of leanness, as found in Buys, 2003. Possibly in the future, they will be able to verify the quality of the pork from the pigs they purchase and demonstrate the superior quality with factual information to their customers. This will improve their ability to target specific markets of value added products like those in Japan and obtain additional premiums. Because of the imprinting effect of IGF2, this gene allows for commercial sows to be better conditioned without any sacrifice to the lean yield in market hogs, provided terminal sires have the AA genotype for IGF2. There will be an indirect benefit to packers from a more productive Alberta sow herd with more pigs weaned per sow and from the contribution of this development to a more economically efficient and sustainable hog supply.

7. LITERATURE CITED

- Andersson-Eklund, L., L. Marklund, K. Lundstrom, C. S. Haley, K. Andersson, I. Hansson, M. Moller, and L. Andersson. 1998. Mapping quantitative trait loci for carcass and meat quality traits in a wild boar x Large White intercross. *J. Anim. Sci.* 76:694-700.
- Buske, B., I. Sternstein, and G. Brockmann. 2006. QTL and candidate genes for fecundity in sows. *Anim. Reprod. Sci.* 95:167-183.
- Buys, N. 2003. IGF2: The use of a paternally expressed QTL influencing muscle mass in Marker Assisted Selection in commercial pig populations. Proceedings of the 28th Annual National Swine Improvement Federation. December 4-5, Des Moines, Iowa, USA.
- Buys, N., G. Van den Abeele¹, A. Stinckens, J. Deley, and M. Georges. 2006. Effect of the IGF2 intron-3-G3072A mutation on prolificacy in sows. 8th World Congress on Genetics Applied to Livestock Production. August 13-18, Belo Horizonte, MG, Brasil.
- Estellé, J., A. Mercade, J. L. Noguera, M. Perez-Enciso, C. Ovilo, A. Sanchez, and J. M. Folch. 2005. Effect of the porcine IGF2-intron3-G3072A substitution in an outbred Large White population and in an Iberian x Landrace cross. *J Anim Sci* 83:2723-2728.
- Gardan D., F. Gondret, K. Van den Maagdenberg, N. Buys, S. De Smet, and I. Louveau. 2008. Lipid metabolism and cellular features of skeletal muscle and subcutaneous adipose tissue in pigs differing in IGF-II genotype. *Domest. Anim. Endocrinol.* 34 (1):45-53.
- Jafarikia, M, B. Sullivan, L. Maignel and P.K. Mathur. 2008. A deterministic procedure for defining the SNP phase in large pedigrees, Canadian Society of Animal Science, Annual Meeting. August 11 - 14, University of Guelph, Guelph, ON, Canada.
- Jeon, J. T., O. Carlborg, A. Tornsten, E. Giuffra, V. Amarger, P. Chardon, L. Andersson-Eklund, K. Andersson, I. Hansson, K. Lundstrom, and L. Andersson. 1999. A paternally expressed QTL affecting skeletal and cardiac muscle mass in pigs maps to the IGF2 locus. *Nat. Genet.* 21:157-158.
- Jungerius, B. J., A. S. van Laere, M. F. te Pas, B. A. van Oost, L. Andersson, and M. A. Groenen. 2004. The IGF2-intron3-G3072A substitution explains a major imprinted QTL effect on backfat thickness in a Meishan x European white pig intercross. *Genet. Res.* 84:95-101.
- Meuwissen T.H.E., and M.E. 1996. Goddard. The use of marker haplotypes in animal breeding schemes, *Genet. Sel. Evol.* 28: 161-176.

- Luoreng, Z., L. X. Wang, and S. D. Sun. 2007. Genetic polymorphism of FSH b subunit gene and correlation with reproductive traits in Beijing Black Pig. *Yi. Chuan.* 29:1497-1503.
- Mathur, P.K., and Y. Liu. 2003. Markers assisted selection for the Canadian Swine Industry. Proceedings of the 28th Annual National Swine Improvement Federation. December 4-5, Des Moines, Iowa, USA.
- Mathur P.K, Liu Y., 2006. Use of IGF2 gene for carcass yield and pork quality in Canadian pigs. Canadian Meat Science Association Annual Meeting. February 8, Quebec City, Quebec, Canada.
- Nezer, C., L. Moreau, B. Brouwers, W. Coppieters, J. Detilleux, R. Hanset, L. Karim, A. Kvasz, P. Leroy, and M. Georges. 1999. An imprinted QTL with major effect on muscle mass and fat deposition maps to the IGF2 locus in pigs. *Nat. Genet.* 21:155-156.
- van Laere, A. S., M. Nguyen, M. Braunschweig, C. Nezer, C. Collette, L. Moreau, A. L. Archibald, C. S. Haley, N. Buys, M. Tally, G. Andersson, M. Georges, and L. Andersson. 2003. A regulatory mutation in IGF2 causes a major QTL effect on muscle growth in the pig. *Nature.* 425:832-836.
- Van den Maagdenberg K., E. Claeys, A. Stinckens, N. Buys, and S. De Smet. 2007.. Effect of age, muscle type, and insulin-like growth factor-II genotype on muscle proteolytic and lipolytic enzyme activities in boars. *J. Anim. Sci.* 85 (4):952-960.
- Vykoukalová, Z. A. Knoll, J. Dvorak, and S. Cepica. 2006. New SNPs in the IGF2 gene and association between this gene and backfat thickness and lean meat content in Large White pigs. *J Anim Breed. Genet.* 123:204-207.