Sampling for Canadian Swine DNA Banking

Introduction

The importance and immediate/potential benefits of having a Canadian Swine DNA Bank have been realized in last few years. The Bank has strategic importance for swine breeding programs, swine genetic research and animal care. The issue now is perhaps the logistics around sampling. This document intends to provide preliminary information regarding effect of sampling on lab results, sampling need according to purpose and some recommendations for creating the bank.

Effect of sampling on lab testing and results

The purpose of collecting samples is for testing. The samples for DNA banking are expected to be used for many and various types of tests in future. The logistics for DNA bank sampling would first need to ensure sufficient quantity and quality of the samples, facilitation of current and future low-cost testing processes, and correct information associated with them.

Sample quantity, quality and type can directly affect quality of results and testing cost. The degree of the effect varies with the type of test involved. For example, a popular PSS test usually needs little DNA to produce a reliable result while an IGF2 test would need much more DNA for an accurate result. It can be very difficult to interpret a result due to either poor quality or low quantity of DNA. Minor sample cross-contamination from using the same tool without proper cleaning or without using a new tool for each sample can result in un-certain or certain but wrong results. For minimizing the testing cost, sample format, in addition to sufficient quantity and quality, should allow high throughput operation, minimal manipulation and automation. For example, one lab technician can handle two batches of 96 samples during a working day for DNA extraction if the samples are fresh or properly-stored liquid blood (i.e. no clogging or degradation), clearly-labeled, no contamination outside the tubes, and well-organized with hard copy and electronic submission forms. If the same samples are received in good shape/information on paper cards, only half (or less) of the samples can be processed for DNA extraction during a working day. The quantity and quality of the DNA extracted from a blood card sample is usually much less than a liquid blood sample. Additional time is always needed to handle samples that are lack of quality (e.g. blood clogged), and required/readable information.

Sampling need according to the purpose

Sampling details such as sample type, size, storage conditions, and required information need to be determined based on the purpose of sampling, and the benefits to be realized. Swine blood (liquid or card), semen (fresh or frozen) and various tissues are all technically acceptable for testing. For creating a swine DNA bank, the sampling purposes can be for (many) multiple tests, for long-term storage (e.g. 15-25 years) and for potential use in many research projects that are

of high interest and value to Canadian swine industry with the lowest possible overall cost for collection, handling, testing and storage. Liquid blood samples can perhaps serve the above purposes well, and help to realize the full benefits of DNA banking through maximizing collaborative high quality research. However, they require trained technicians to take samples in field and designated freezers for storage. Blood samples collected on FTA cards can be stored at room temperature for many years without significant degradation. However, sample quantity retained on the cards are very limited, and may not be sufficient for many multiple usages down the road; the cards can cost much more for handling, sub-sampling and DNA extraction, and have higher risk of cross-contamination. Tissue samples with abundant amount and high quality of information (such as meat quality data) attached to them can be very valuable samples for banking too although it has storage and handling disadvantages. The choice of an appropriate storage mode (original sample, DNA or both) also depends on costs, conservation properties, mid- and long term purpose, etc. Using the current available high throughput technologies, the amount of original sample that can be processed at once is very limited, and may not be sufficient to meet the banking needs. In some cases, extracted DNA is not absolutely necessary. For example, some tests can be done directly using original liquid blood samples without extensive DNA extraction or purification process. While DNA needs to be stored once extracted, storing portions of original samples is perhaps unavoidable.

Recommendations:

1. **Sample type and size:** (a) 5 mL liquid blood in traditional EDTA tubes – the first choice, (b) 5 mL raw semen for boars in plastic semen sampling tubes – the second choice, and (c) other types of samples, such as meat - for special cases. In all the cases, two sub-samples (1.5 mL liquid sample or 1.0-1.5 gram tissue sample each) are taken in the lab and stored in 2.0 mL micro-centrifuge tubes in two locations, the remaining used for initial DNA extraction. Different sample types and sizes usually involve different handling/extraction methods with different costs (short term and long term).

2. **DNA extraction:** Initial DNA extraction is conducted using the best possible existing technologies to obtain DNA with both high quality and sufficient quantity (e.g. 20 µg). The initial DNA extractions are stored in two sets at two different locations.

3. **Sample information:** The amount and quality of the information attached to the samples, such as sample coding, sample tracking and computerization, and security mechanisms to ensure the confidentiality needs to be standardized. The standards adopted should permit sample information sharing for research purpose with minimum risk.

4. **Type of collection:** The full benefits for the swine industry to have a DNA bank can be realized through maximizing collaborative high quality research. To facilitate research use of the samples in a bank, more than one type of collection may be created. For example, the collections can be **anonymous** (for samples with valuable phenotypic data but with no sample IDs or sources), **identifiable** (for samples that are purposely unidentified for research use, but can be linked to their ID/source), or **identified** (for samples with all the information available to interested parties).

5. **Sub-sampling protocol and sample distribution authorization:** A written protocol, including sample distribution authorization, is needed for accessing the sub-samples and their associated information.

6. **Funding:** Once started, banking needs to be an on-going process; collections need to be maintained and continued, and their use optimized. Funding mechanisms need be available to ensure continuation of the process. One possible source of funding can be the payment from research labs that request samples from the DNA bank. The cost to researchers can be covered in their grant applications, and the applications with this component are expected to be more cost-effective, and therefore more attractive, to granting agencies.

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