Partial genetic characterization of a closed bovine herd produced in Canada, called "Lynch Lineback"

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Summary

The Lynch Lineback is a Canadian landrace breed of cattle, originating in Eastern Canada. Developed by a Canadian family (Lynch), this breed has been a closed herd for almost a century. This study provides a partial characterization of the genetic diversity of this breeds in comparison with 10 beef, dairy, and dual purpose breed produced in Canada. A total of 21 microsatellite markers were used to evaluate the genetic profile of the different samples collected for this study. Basic statistical, cluster, and structure analyses were performed to determine the current genetic diversity and the genetic relation with other bovine breeds. Our results revealed that individuals assigned to Lynch Lineback clustered together and this cluster was isolated from all other breeds included in this study. Basic molecular analyses revealed a lower presence of alleles per locus and a lower heterozygosity for this breed. This partial genetic characterization demonstrated that Lynch Lineback breed is a unique population and it could have an impact in the production of beef or dairy cattle.

Keywords: Lineback, microsatellite, genetic diversity, cattle

Introduction:

Canada is rich in livestock diversity. The majority of these Canadian breeds was imported from European countries. Among them, the Lineback cattle made a significant presence during the 19th Century, beginning of 20th Century, which was introduced to North America by British colonists. Unfortunately, Lineback breeders never got together to create a Canadian association and to record the pedigree of the Lineback cattle born in Canada. Thus, the Lineback is not officially recognized as breed produced in Canada (AAFC, 2017). However, a purebred Lineback herd (referred as Lynch Lineback) has been maintained by Lynch family since the early 1900's.

The Lynch Lineback (LL) is a dual purpose breed, milk and beef. These animals are produced based on the following production standards: compact cow, prominent white strip, black feet and long legs, sustain peak lactation production in the heat of summer, and on a

pasture forage only diet, good reproductive life, low complication rate during delivery, and longevity. Overall, this line of Lineback cattle has been selectively bred to withstand the harsh Canadian climate. Lynch Lineback cows have a low impact of milk production compared to Holstein cows, but have a longer lactation and feed to milk conversion in when compared to other dairy breeds (personal data from Lynch Lineback producers). Unfortunately, the status of LL is considered as "Endangered" (between 26 to 75 purebred animals). The goal of this study was to evaluate the genetic diversity of the Lynch Lineback and to evaluate its relation with other cattle breeds (beef and dairy) produced in Canada to determine its importance to be preserved.

Material and Methods:

A- Sampling and genetic analysis

In total, 644 randomly selected animals from 11 populations were genetically analyzed. These populations consisted of dairy (Holstein, American Dairy Lineback, and Kerry), beef (Charolais, Hereford (polled and horned), Angus), dual purpose (Lynch Lineback, Randall Lineback, and White Park), and unknown breed. Animals designated as unknown breed were animals showing phenotypical traits of Lynch Lineback, but pedigree could not be confirmed. Small sampling was obtained for the Kerry and White Park breeds due to their "critical status" in Canada. American Dairy and Randall Lineback samples were obtained from individual located in United States.

DNA material was obtained from hair or sperm samples. DNA was extracted from hair follicles using the same protocol as described by the group of Prystupa (Prystupa et al., 2012). Microsatellites were chosen based on MoDAD microsatellite marker recommendations (FAO, 2011). In total, 21 microsatellites were selected for this genetic analysis (HEL5, HEL9, INRA23, BM1824, INRA35, MM12, INRA32, INRA37, HAUT24, ILSTS005, TGLA126, HEL1, BM1818, ETH185, BM2113, INRA63, ETH3, CSSM66, ETH152, TGLA227, and HAUT27). These loci were genotyped for the different sampling using a commercially available kit (Qiagen Multiplexing Kit; Qiagen Inc., Burlington, Ontario, Canada). Due to the space limit constraint for this manuscript, the allele frequency tables for the different populations are available upon request.

Samples were denatured for 5 min at 95°C, quenched on ice for 2 min and loaded onto a Genetic Analyzer 3500xL (Applied Biosystems) equipped with a 50-cm array and filled with POP7 polymer. GeneMapper® version 5.0 Applied Biosystem) was used to determine genotypes

B- Statistical method

Average (N_a) and average effective (N_e) number of alleles, and inbreeding coefficient (F_{IS}) were determined by using GenALEx version 6.5 (Peakall and Smouse, 2012). Observed (H_o) and expected (H_e) heterozygosities were evaluated using ARLEQUIN version 3.5 (Excoffier and Lischer, 2010). FSTAT version 2.9.3.2 (Goudet, 2005) was used to estimate allelic richness

(AR). Predetermined population were set to assign individual using the "leave out option" and the maximum likelihood method (Paetkau et al., 1995; Paetkau et al., 2004) as implemented in GenALEx version 6.5; Structure version 2.1 (Pritchard et al., 2000), with the parameter settings: K1 to 15, 50 000 burn-in, 100 000 Markov chain Monte Carlo (reaching equilibrium) with 10 replicates was used to assign individuals to inferred clusters Structure Harvester (Earl, 2009) was used to determine the number of inferred clusters. Clumpp version 1.1.2 (Jakobsson and Rosenberg, 2007) was used to cluster the 10 Structure runs into a unified figure and Distruct version 1.0 (Rosenberg, 2004) was used to visualize the cluster output.

Results:

The average number of alleles (N_a) observed for each population varied between 2.41 to 9.23 (see Table 1); White Park had the lowest and the Charolais (CH) had the highest. For the Lynch Lineback (LL), N_a was 3.13. The same trend was observed for the effective number of alleles (N_e). The allelic richness evaluation revealed that LL had the lowest value (2.38) and CH had the highest value (4.36). Observed heterozygosity demonstrated a similar trend; LL had the lowest (0.47) and CH had the highest (0.71). Similar trend was observed for the expected heterozygosity. Inbreeding coefficient varied between -0.2380 (Kerry breed) and 0.2000 (Randall Lineback). Interestingly, the value was -0.0210 for the LL, demonstrating an acceptable level of diversity for this unique breed.

Cluster analysis revealed that LL is a unique population when it is compared to dairy, beef, and dual purpose breeds (see fig 1). In addition, 5 out of 8 samples assigned to the unknown breed population clustered with the LL population suggesting that these animals could be assigned to LL population.

Using a Bayesian approach to investigate population structure, the most probable K value (inferred clusters) was determined to be 5. In the case of the LL samples, very little gene flow was observed (fig. 2). For the unknown samples, gene flow is significantly present suggesting genetic influence by other breeds.

Discussion:

The purpose of this study was to characterize the genetic diversity of the Lynch Lineback (LL) in comparison of dairy, beef, and dual purpose breeds. To date, this is the first study to report a genetic characterization for this breed. Our results demonstrated that the LL population is genetically distinct when compared to other cattle breeds produced in Canada. Our cluster (fig.1) and structure (fig 2) analyses revealed that samples assigned to the LL breed did not cluster with the other breeds included; nor was there notable admixture from other populations. Interestingly, 5 out 8 samples assigned to the unknown population clustered with the LL population. These samples were obtained from cows showing LL characteristics, but the pedigree could not be confirmed. These individual could be considered as LL breed. However, caution must be taken regarding the purity of these cows. Structural analysis revealed a higher level of gene flow

compared for these individual compared to the ones assigned to LL population. Interestingly, Randall (RL) and American Dairy Lineback did not cluster with LL, confirming that LL breed developed its own genetic profile over the time.

To conserve breed standards, the Lynch's have maintained their herds without outcrossing with other breeds. This explains the low average number alleles, low expected Heterozygosity and allele richness coefficients for this closed breed (table 1). However, the LL breed has maintained low inbreeding level over the time (F_{IS} was -0.0210). Recently, several producers have demonstrated an interest in this breed and they worked together to maintain the genetic diversity of this herd, which could explain the inbreeding coefficient result obtained in this study.

Overall, the LL is a unique source of genetic diversity within Canada and it has great value to be preserved. This breed can be considered as a landrace based on our results and are more distinct in comparison to the other populations in this study. It would be advantageous to study the contribution of the LL genome into other breeds to evaluate the potential of improvement of milk yield, fertility rate, and other desired traits in cattle production.

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