

Simulation of Introgression of the Polled Allele into the Holstein Breed via Conventional Breeding versus Gene Editing

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Summary

Introgression of the polled allele into a dairy cattle population via conventional breeding versus gene editing was simulated for three different polled mating schemes. The change in horned allele frequency, level of inbreeding, and genetic gain were determined. Gene editing of the polled locus rapidly decreased horned allele frequency, lowered rates of inbreeding, and increased genetic gain compared to conventional breeding. The optimal polled mating scenario for rapidly decreasing horned allele frequency, maintaining an acceptable level of inbreeding, and increasing genetic gain used both homozygous polled (PP) and horned (pp) sires in combination with gene editing. While conventional breeding methods can decrease the horned allele frequency, the increased inbreeding and loss of genetic gain in the absence of gene editing would be detrimental to genetic improvement in the dairy industry.

Keywords: polled, gene editing, cattle

Introduction

Physical dehorning of cattle is a standard practice to protect both animals and humans from injury. However, it is not only costly to the producer, but also painful and stressful for the animal. Dehorning is currently facing increased public scrutiny as an animal welfare issue (Stafford and Mellor, 2011). Despite these factors, 94% of U.S. dairy cattle producers report routine dehorning (USDA, 2009). Horns are inherited as an autosomal recessive trait and polled cattle result from an autosomal dominant pattern of inheritance (Long and Gregory, 1978). The American Veterinary Medical Association (AVMA) has proposed using polled genetics as an alternative to dehorning (AVMA, 2014). However, there are few polled dairy sires with high genetic merit.

Cole (2015) proposed adding the economic value of polled to selection indices, but showed that this is not particularly effective at increasing the frequency of polled animals in the population. The frequency of the polled allele is so low (0.0071) that carriers are unlikely to be among the top-ranked bulls (Cole, 2015), and increased use of polled dairy sires has been hindered by their unavailability and lower genetic merit. Daughters of Holstein, homozygous polled (PP) and heterozygous polled (Pp) bulls will earn \$301 and \$171 less, respectively, over their lifetimes than daughters of horned (pp) Holstein bulls (Spurlock et al., 2014). Gene editing has the potential to solve these issues by producing high-genetic merit polled bulls, thereby eliminating the need for dehorning (Carlson et al., 2016).

The objective of this study was to simulate the introgression of the polled allele into a dairy cattle population and compare the impact of using the methods of conventional breeding versus gene editing to increase the frequency of the polled allele. The change in horned allele frequency, inbreeding, and genetic gain were determined for both introgression methods in three different polled mating schemes.

Material and methods

Simulation

Computer simulation was used to compare introgression of the polled allele into a dairy cattle population via conventional breeding versus gene editing in three different polled mating schemes. The software program (`geneedit.py`; Cole, 2017b) simulates gene editing applied to a dairy cattle population as an extension of the Cole (2015) program to manage multiple-recessives and was modified to include additional parameters for selection of polled sires.

The simulation procedures were similar to those described in detail by Cole (2015), with the additional polled and gene editing parameters (Cole, 2017a). The base population of each scenario was 350,000 cows distributed over 200 herds and 350 bulls. The frequency of the horned (*p*) allele in the base population was 0.9929 (Null, 2015). True breeding values (TBV) for lifetime net merit (NM\$) were determined by a random sample from a normal distribution, with a mean of \$0 and standard deviation (SD) of \$200 for cows and a mean of \$300 (+1.5 genetic SD of NM\$) and SD of \$200 for bulls. Additionally, to represent the current population of Holstein artificial insemination (AI) sires, base population polled bulls averaged -0.9 SD (*Pp*) and -1.5 SD (*PP*) genetic merit compared to horned bulls (Spurlock et al., 2014). An animal's horned status was determined by randomly selecting one allele each from the sire and dam. The proportion of polled bulls in the base population was 2.5% (18% *PP* and 82% *Pp*) (NAAB, 2017). The population was allowed to grow to a maximum of 500 bulls and 100,000 cows over 20 generations.

Mate allocation

Polled sires were preferentially selected over horned sires in all scenarios but D (Table 1). To establish a baseline for comparison, TBV was the only sire selection criterion used in scenario D. In scenarios A1 and A2, both *PP* and *Pp* bulls could be selected. Only *PP* bulls could be used for scenarios B1, B2, C1 and C2. In scenarios A1, A2, B1 and B2, bulls were allowed a maximum of 5,000 matings per year. If there were too few polled bulls available to service all cows in a generation, a random selection of horned bulls was added to the list of available bulls. Matings were not limited in scenarios C1 and C2.

All scenarios used the modified Pryce (`pryce_r`) scheme to allocate bulls to cows (Cole, 2015). The `pryce_r` scheme penalizes the parent average (PA) TBV to account for inbreeding (Pryce et al., 2012) and the economic costs of a horned (*pp*) calf (Cole, 2015). In all scenarios, an added penalty for producing carrier (*Pp*) offspring was included, so the PA was further adjusted to account for the long term economic impacts of producing a carrier (*Pp*) calf, which was assumed to be half of the cost of a horned (*pp*) calf. The average expected cost of dehorning is \$22.50 per animal (Thompson et al., 2017). To account for breeders' preferences and premium marketing opportunities, the cost of an affected (horned) calf was assumed to be \$40 in all scenarios (Cole, 2015).

Gene editing

Gene editing was modelled as an added step to the production system proposed by Kasinathan (2015), which combines the use of advanced reproductive technologies and somatic cell nuclear transfer cloning. For the three gene editing scenarios (A2, B2, and C2), 1% of the bulls were gene edited for the polled locus in each generation. First, the live bulls and cows were sorted on TBV in descending order. Within the top 1% of bulls, both Pp and pp animals were cloned and edited to be PP. All edited animals were assumed to be PP and were added to the list of live animals. Gene editing was not used in scenarios A1, B1, C1 and D.

Results and discussion

Scenario D, which did not use polled as a separate sire selection criterion, confirms that including an economic penalty for horned (\$40) in the selection index alone is not an effective method for decreasing the horned allele frequency (Figure 1A). The use of gene editing in all polled mating schemes decreased the horned allele frequency as fast as (C2) or faster than (A2 and B2) conventional breeding systems. Scenario C1 (conventional breeding) and C2 (gene editing) resulted in the same change in horned allele frequency, as only PP sires were selected for mating in both scenarios. Scenarios B1 and B2 resulted in the largest difference in change in horned allele frequency, while using the same sire selection criteria. In generation 20, the horned allele frequency in B1 was 97%, whereas in B2 the frequency was 1%. With only a small number of PP sires available in the base population, it is likely that a majority of horned sires were used in B1 instead, so the horned allele frequency did not decrease. When gene editing was used in B2, the number of PP sires increased quickly. This meant that fewer horned sires were used in each subsequent generation, which resulted in a rapid decrease in the frequency of the horned allele.

After 20 generations, inbreeding was 6% in scenario D, which is an average increase of 0.3% per generation. All polled mating schemes, except for B1, resulted in higher levels of inbreeding at generation 20 (Figure 1B). Gene editing in polled mating schemes A and C resulted in lower inbreeding levels than conventional breeding. Scenarios A1 and A2 were not significantly different from each other, as both scenarios were allowed to use a mix of all sire genotypes in each generation. When herds were forced to only use PP sires in a conventional breeding system, inbreeding increased by an average of 1% per generation in C1, ultimately reaching 17% after 20 generations. Gene editing in C2 resulted in 0.4% less inbreeding on average per generation than in C1.

Selection of naturally polled sires resulted in less genetic gain in each polled mating scheme (A1, B1 and C1) compared to Scenario D (Figure 1C). Gene editing achieved superior outcomes for all polled mating schemes (A2, B2 and C2). Scenarios C1 and C2 resulted in the greatest difference, with genetic gain increasing by \$33 per generation compared to conventional breeding. The lowest TBV after 20 generations was seen in C1, which used only PP sires that have lower TBV than pp sires. Only PP sires were used in C2, but the use of gene editing allowed for the top TBV bulls to be PP in just a few generations. The greatest genetic gain, \$166 per generation, was achieved in B2, when herds preferentially used PP sires, but also used pp sires when too few PP sires were available.

These results show that scenario B2, which consisted of using PP and pp sires in combination with gene editing, is the most optimal for rapidly decreasing the frequency of the horned allele, maintaining a reasonable level of inbreeding, and increasing genetic gain (Figure 1). Polled mating scheme C models a case where consumer perceptions, market

expectations, or social license place additional pressure on the dairy industry to completely eliminate dehorning quickly. In this case, the use of gene editing was necessary to rapidly reduce the frequency of the horned allele, decrease levels of inbreeding, and maintain genetic gain.

Conclusion

Gene editing of the polled locus rapidly decreased the frequency of the horned allele, lowered rates of inbreeding, and increased genetic gain compared to conventional breeding. While conventional breeding methods can be used to decrease the frequency of the horned allele in a population, the increased inbreeding and loss of genetic gain in the absence of gene editing would be detrimental to the dairy industry.

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Table 1. Parameters of each mating scenario.

Scenario	Gene editing (1% of bulls)	Bull selection criteria	Sire genotype(s)	Polled matings per year limit
A1	No	Polled	PP and Pp	Yes
A2	Yes	Polled	PP and Pp	Yes
B1	No	Polled	PP	Yes
B2	Yes	Polled	PP	Yes
C1	No	Polled	PP	No
C2	Yes	Polled	PP	No
D	No	TBV	n/a ¹	Yes

¹In scenario D both PP and Pp sires may have been available, but genotype was not included as a selection criterion.

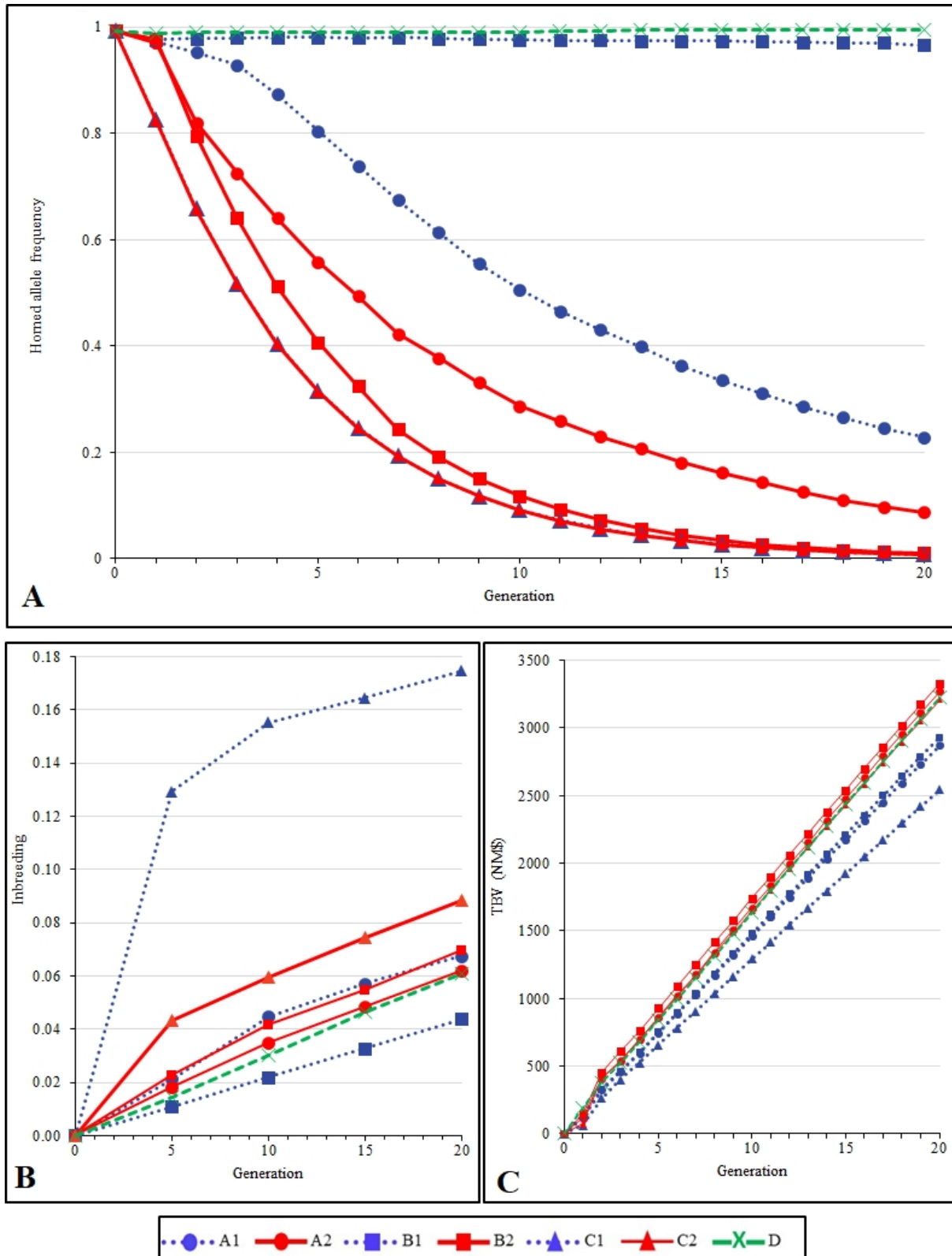


Figure 1. Effect of each mating scenario on A) horned allele frequency; B) inbreeding; and C) TBV. Conventional breeding is a dashed green line, conventional breeding for polled scenarios are dotted blue lines, and gene editing for polled scenarios are solid red lines.