UNIVERSITÀ
DEGLI STUDI
DI PADOVA

HEAD OFFICE: UNIVERSITA' DEGLI STUDI DI PADOVA

DEPARTMENT OF AGRONOMY, FOOD, NATURAL RESOURCES, ANIMALS AND ENVIRONMENT

PhD COURSE IN: ANIMAL AND FOOD SCIENCE

SERIES: XXXVI

FERTILITY IN HOLSTEIN HEIFERS: GENETIC ASPECTS AND OPTIMIZATION OF REPLACEMENT STRATEGIES IN THE DAIRY HERD

Thesis written with the financial contribution of the Italian Holstein, Brown Swiss and Jersey Association

(ANAFIBJ, Cremona, Italy)

Coordinator: Prof. Angela Trocino

Supervisor: Prof. Martino Cassandro

Co-Supervisor: Prof. Mauro Penasa

PhD student: Valentina Ferrari

Declaration

I declare that the present thesis has not been previously submitted as an exercise for a degree at University of Padova (Italy), or any other University. I further declare that this work embodied to be mine.

Contents

Abstract

Heifer management plays a key role in the dairy herd as it can account for more than 20% of total production costs. Fertility is an essential aspect to rear heifers efficiently and maximize lifetime performances, and it can be enhanced through effective management strategies and genetic selection. The recording of pregnancy diagnoses has allowed new traits to be collected early in life such as the interval from first to last insemination. The objectives of the present thesis were to: i) identify genetic and non-genetic factors affecting Italian Holstein heifer fertility, and propose an aggregate index for heifer fertility; ii) investigate genetic and genomic aspects of age at first calving; and iii) describe a practical tool to help farmers optimize replacement management. Chapter 1 investigated environmental and genetic aspects of conception rate, non-return rate at 56 d, interval from first to last insemination, and age at first service in heifers to develop a genetic evaluation. Chapter 2 performed genetic and genomic evaluation of age at first calving. Chapter 3 combined genetic and phenotypic aspects into a tool for the farmers to define the replacement needs and reduce replacement costs. Results of the present work indicated that heifer fertility should be considered as an additional trait in the breeding objectives of Italian Holstein. Genetic selection for age at first calving can reduce the actual phenotypic mean of this trait without negatively affect other reproductive performances. Sexed semen allows farmers to produce more replacements than the actual needs. However, given current market conditions, this is not the most convenient choice for farmers. Thus, providing a tool that helps dairy farmers to optimize replacement strategies based on their herd performances is appropriate. Overall, an optimal replacement management, both from environmental and genetic point of view, is expected to improve animal welfare and contribute to reduce replacement costs and environmental impact.

Key words: heifer, fertility, longevity, profit, genetics

Riassunto

La gestione della rimonta riveste un ruolo chiave in un'azienda da latte in quanto può rappresentare oltre il 20% dei costi di produzione. La fertilità è un aspetto essenziale per allevare le manze in modo efficiente e massimizzare le prestazioni durante la carriera produttiva, e può essere migliorata attraverso strategie di gestione efficaci e la selezione genetica. La registrazione delle diagnosi di gravidanza ha permesso la raccolta anticipata di nuovi caratteri come l'intervallo prima-ultima inseminazione. Gli obiettivi di questa tesi sono stati: i) identificare gli aspetti genetici e non che influenzano la fertilità nella popolazione di manze di razza Frisona Italiana, e proporre un indice aggregato per la fertilità delle manze; ii) studiare gli aspetti genetici e genomici dell'età al primo parto; e iii) descrivere uno strumento pratico per aiutare gli allevatori nelle proprie scelte di gestione della rimonta. Il Capitolo 1 ha indagato aspetti ambientali e genetici del tasso di concepimento, tasso di non ritorno in calore a 56 giorni, intervallo prima-ultima inseminazione ed età alla prima inseminazione per sviluppare una valutazione genetica. Il Capitolo 2 ha sviluppato una valutazione genetica e genomica per l'età al primo parto. Il Capitolo 3 ha combinato aspetti genetici e fenotipici in uno strumento che consente agli allevatori di definire il proprio fabbisogno di rimonta e ridurre il costo di sostituzione. I risultati di questa tesi hanno evidenziato che la fertilità delle manze dovrebbe essere considerata come un carattere aggiuntivo negli obiettivi di selezione della razza Frisona Italiana. La selezione genetica per l'età al primo parto può ridurre l'attuale media fenotipica senza influenzare negativamente altre performance riproduttive. Il seme sessato permette agli allevatori di produrre più rimonta del necessario. Tuttavia, date le attuali condizioni di mercato, questa non è la scelta più conveniente. Pertanto, fornire uno strumento che aiuti gli allevatori italiani ad ottimizzare le strategie per la rimonta in base alle proprie prestazioni aziendali risulta sicuramente appropriato. Nel complesso, una gestione ottimale della rimonta, sia dal punto di vista ambientale che genetico, migliora il benessere animale e contribuisce a ridurre il costo di sostituzione e l'impatto ambientale.

Parole chiave: manze, fertilità, longevità, profitto, genetica

Traditionally, the scientific literature has paid more attention to lactating cows, but more recently the interest towards animals that are the future replacements of the dairy herd has increased. Rearing young stock from birth to first calving affects significantly the costs of a dairy herd and estimates range from $2.33 to $2.38 per head per day (Heinrichs et al., 2013; Overton and Dhuyvetter, 2020). Hence, dairy heifer management has faced several changes to enhance animal lifetime performances and minimize rearing costs. To help dairy farmers in making decisions about heifer management, several strategies and tools have been proposed. Furthermore, it has been demonstrated that careful heifer management leads to animal welfare improvement and environmental impact mitigation (Holden and Butler, 2018). To ensure heifers to calve at desired age and therefore achieve high milk yield and good fertility, specific target body weight should be guaranteed. Indeed, to calve at desired time a dairy heifer should maintain an average growth rate of about 750 g/d (Wathes et al., 2014). Also, to ensure good fertility, heifer conception rate has to be maximized. Heifer fertility depends both on management decisions and genetics, and the major aspects influencing puberty onset are body weight and age (Wathes et al., 2014). Moreover, new available technologies provide farmers with the best breeding strategies for their herds and the combination of such technologies allow farmers to: i) enhance herd genetic progress, ii) optimize breeding decisions to choose the best heifers to keep as replacements and obtain replacement heifers from genetically superior animals, and iii) enhance farm profit from selling crossbred calves. Thus, the identification of management, genetic, and phenotypic aspects involved in the period from birth to first calving is fundamental to optimize and improve animal welfare and farm profit.

Review: Dairy heifer performances as affected by management and genetic factors

V. Ferrari[a,b], J.B.C.H.M. van Kaam[a], M. Penasa[b], M. Marusi[a], R. Finocchiaro[a],

M. Cassandro[a,b]

[a]*Associazione Nazionale Allevatori della Razza Frisona, Bruna e Jersey Italiana, Via Bergamo 292, 26100, Cremona (CR), Italy*

[b]*Department of Agronomy, Food, Natural resources, Animals and Environment, University of Padova, Viale dell'Università 16, 35020, Legnaro (PD), Italy*

Submitted to *Animal*

Abstract

The interval from birth to first calving is an investment period in which dairy farmers should guarantee high management level to maximize the chance that the heifer will become a sustainable cow able to combine high productions, low environmental impact and optimal reproduction performances and longevity. The aim of this review is to provide an overview of management and genetic factors affecting the rearing period, from birth to first calving. More than 50% of total heifer rearing costs are feed costs and different feeding strategies have been proposed to reach desired energy requirements. Given the growing interests towards the rearing period and the increasing availability of new technologies and tools, models that help farmers optimizing their rearing management strategies have been developed. The optimization of replacement management strategies leads to both economic and environmental improvements. An average daily gain of 0.75 kg/d has been suggested for an adequate development and optimal lifetime productive and reproductive performance. The goal is to reach puberty at about 60% of mature weight and 90% at first calving, in order to first calve at 22-24 mo. Furthermore, heifer fertility depends on management and genetic aspects, and to achieve parturition at desired time, heifer conception rate has to be maximized. The combination of genomics and sexed semen (and crossbreeding) offers a variety of possibilities to farmers to 1) reduce generation interval and enhance herd genetic progress; 2) choose only the best heifers to keep as replacements; 3) assure only the necessary female calves from the best heifers thanks to sexed semen use; 4) enhance herd profit from selling crossbred calves yielded with beef semen use on animals of low genetic merit; and 5) increase herd sustainability.

Keywords: breeding, heifer, economic, fertility, longevity

Implications

Dairy heifers represent a relevant cost until the onset of milk production, but at the same time an investment in the genetic progress of the herd. Given current market situation, breeding more replacements than needed is not the best choice neither from economic nor from animal welfare or environmental perspectives (e.g., overcrowding leads to animal welfare reduction). Optimal rearing management is fundamental to both enhance farm profits and meet the increasing consumer's concern towards sustainable breeding methods. Current technologies allow farmers to optimize management strategies within the herd to face economic and environmental issues.

Introduction

The time from birth to first calving is fundamental for dairy farmers because they have to face the complex dilemma of minimizing costs associated with rearing heifers while ensuring or enhancing lifetime economic productivity (Mourits et al., 1997). The cost from birth to first calving for a conventional herd ranges from \$1 700 to \$2 400, considering an overall rearing cost of \$2.33 per head per day at a reference heifer weight of around 318 kg (Overton and Dhuyvetter, 2020; Tranel, 2019). Dairy heifer management has undergone several changes in the last years (Heinrichs et al., 2017), but still the importance of dairy heifer husbandry is probably not sufficiently well recognized by most farmers (Le Cozler et al., 2008). Several studies have been conducted and practical tools have been developed to help farmers. For example, Mohd Nor et al. (2015) proposed a model to determine the optimal number of heifer calves to be reared to minimize the net cost of rearing replacement animals. The literature has focused mainly on dairy heifers in relation to their physiological aspects, available management practices, and lifetime production performances. Advancements in genomics and dairy cow reproductive technologies (e.g., sex-sorted semen) offer an opportunity to apply stronger selection pressure to females (Newton et al., 2018). Since 2008, herd genotyping has become an increasing activity for dairy farmers to identify the best animals in the herd. Moreover, the advent of genomics has led to stronger genetic progress in the dairy populations.

The aim of this paper is to review dairy heifer management and genetic/genomic decisions that can impact future performances of dairy cows. In particular, the first part will review strategies to improve herd management decisions and the second part will focus on genetic and phenotypic aspects that can influence dairy heifer future performances.

Management decisions

Rearing period

Overall, the rearing period is the time from birth to first calving and in dairy cattle corresponds approximately to a 2-yr non-productive period. As such, it has a relevant impact on total milk production costs. Estimating the real replacement cost is not easy as the cost of rearing a heifer is not univocal for all farmers due to difficulties in identifying all costs implicated, and the amount of rearing practices currently used. For example, Heinrichs et al. (2013) assessed a daily rearing cost of \$2.38 per heifer in 44 Pennsylvania herds and Boulton et al. (2017) reported an average of £2.31. Furthermore, Boulton et al. (2017) looked into total rearing costs within 3 development periods: birth to weaning, weaning to conception, and conception to calving. Overall, they observed that the period from birth to weaning accounted for the highest daily cost, about twice that from weaning to conception, and conception to calving (£3.14 vs £1.64, respectively), with feeding accounting for 46% and 32% of the total costs from birth to weaning and from conception to calving, respectively (Boulton et al., 2017).

To attain desired growth rates from birth to weaning, several feeding strategies have been proposed. Akins (2016) compared two feeding programs, namely conventional and intensive liquid feeding. Conventional program consists of administering liquid feed (with 20% protein and 15% to 20% fat) to calves at approximately 8-10% of their body weight, whereas the intensive program consists of feeding liquid feed (26% to 28% protein and fat content similar to previous) at 16% to 20% of body weight. Findings highlighted that, even if costs of feeding an intensive program are higher than a conventional program, calves fed an intensive program have greater growth rates, especially in the first 2 weeks of life. Furthermore, the intensive feeding program has been linked to heifers that are bred and calve 15 to 30 d earlier, and with greater milk yield in first lactation. Accordingly, Davis

Rincker et al. (2011) investigated if the increase of energy and protein intake in milk replacer of calves could affect growth rate and future animal performances; results showed that calves fed diet containing more energy and protein during the preweaning period were 1 month younger, lighter, and tended to reach puberty earlier. Furthermore, Khan et al. (2011) reported that feeding larger amount of milk before weaning could delay physical and metabolic rumen development, leading to lower solid feed consumption around weaning. Consequently, Khan et al. (2011) supported the importance of the transition from liquid (milk or milk replacer) to solid diet (grains or forage) in minimizing weight loss and distress at weaning. Increasing the energy and protein intake in calves has been also shown to increase both the amount of mammary parenchymal DNA and parenchymal tissue at weaning (Brown et al., 2005). This increase in mammary development was not observed once the calves were weaned, indicating the calf is more sensitive to the level of nutrition prior to weaning. Meyer et al. (2006) reported a sensitive increase in mammary fat pad weight in heifers fed at least twice as much milk replacer as the control group before weaning, but the same was not true for parenchymal mass. They indicated that the level of nutrient intake had a limited influence on mammary epithelial cell proliferation, the rate of DNA accumulation in the parenchyma, and the total parenchyma mass. Mammary gland growth is influenced by feeding during the rearing period and diets with an excess of energy have been linked to mammary gland body weight increase due to an increase of the amount of adipose tissue and a consequent reduction of mammary secretory tissue and epithelial cells (Sejrsen et al., 1982). Energy requirements play an important role also in later phases. Hence, diets of post-weaned heifers are important to make sure animals are growing at a rate which guarantees they will be ready for breeding and to use the diets efficiently. It has been reported that heifers fed higher energy during early growth reach breeding size and puberty earlier than heifers fed higher forage to grain ratio (Abeni et al., 2019; Gardner et al., 1988). Conversely, the strategy of limit-feeding has been analysed as a way to improve feed efficiency and reduce nutrient losses while meeting the nutrient needs of growing heifers. Hoffman et al. (2007) evaluated if limit-feeding diets used on heifers would influence future lactation performance.

Limit-feeding diets consist of limiting feeding by 80-90% of the dry matter intake rather than *ad libitum* control diets, while assuring similar daily nutrients intake to heifers. As a result, Hoffman et al. (2007) observed an increase of the feed efficiency in heifers fed restricted diets, and did not report changes of milk yield and content in first lactation cows. Accordingly, Lascano et al. (2009) reported that limiting heifer's feed intake, but with higher energy content, leads to feed efficiency improvement to maintain constant daily gain. Indeed, the most feed efficient heifers ate less, had fewer meals, and spent less time eating than contemporaries (Green et al., 2013). In order to fulfil heifer's feeding behaviour (e.g., time spent feeding and ruminating) and reduce hunger and frustration caused by foraging lack, it has been suggested to add a supplementary low-nutritive long-particle feedstuff such as straw (Greter et al., 2013).

Models to support heifer management decisions

Modelling to improve heifer management has been a more recent purpose of research to make effective management decisions (Heinrichs et al., 2017) and different tools have been investigated to understand the potential impact of different management decisions on herd profit. Knowing the true costs and revenues of raising replacement heifers allows farmers to increase their efficiency. Gabler et al. (2000) elaborated a flexible cost analysis spreadsheet to accommodate various operations to analyse the costs to raise a replacement heifer on an individual operation basis. Operation considered referred to feeding and labour management, and housing system. They estimated, on average, $1 124 from birth until the beginning of breeding ($1.50/day). In their calculations, those authors reported that the two most expensive heifer rearing periods were from day 3 to weaning, and from the beginning of breeding to 3 weeks prior to calving. Furthermore, Boulton et al. (2017) developed a spreadsheet to calculate the total cost of rearing from birth to first calving, also including the opportunity cost of the heifer and the cost of mortality, and the time taken to repay these costs, using data collected in Great Britain. Results showed that the mean cost of rearing a dairy heifer, including fixed and variable costs, was £1 300; if capital and opportunity costs were considered, the mean total cost increased to £1 819 (£2.31/day). In their study, the mean age at first calving (AFC) was 25.8

months; they demonstrated that taking 26 months as the baseline, the mean total cost of rearing decreased by 17.1% for calving at 23 months and increased progressively up to 25.2% for AFC ⩾ 30 months, as AFC had the greatest influence on total rearing costs. Finally, they found that, on average, heifer needs approximately 1.5 lactations to repay the rearing costs.

Mourits et al. (1999, 2000) defined a dynamic programming model to determine optimum rearing decisions of dairy replacements in US. The optimal rearing practices resulted in an average calving age of 20.5 months at an average BW of 563 kg, setting prepubertal average daily gain (ADG) at 0.9 kg/d and maximum post-pubertal ADG at 1.1 kg/d. Sensitivity analysis showed two main influencing factors: growth rate restrictions and variation of reproductive performance. Authors concluded that the optimal rearing patterns came from limiting prepubertal ADG, followed by a post-pubertal compensatory feeding regime.

Reproductive technologies may allow the creation of more dairy heifer calves than needed to replace culled cows. Nevertheless the combination of reproductive technologies and genomic information creates options for various management strategies (Kaniyamattam et al., 2016). Kaniyamattam et al. (2016) developed a “daily stochastic dynamic dairy simulation model” including multitrait genetic models and different reproduction and selection strategies of a dairy herd based on traits included in the 2014 lifetime net merit (NM$) index developed by the Council on Dairy Cattle Breeding in US. Those authors reported that the average true breeding value (TBV) of NM$ of all cows in the herd increased by $263 in a 15-year scenario which combined sexed semen use in heifers and culling of surplus heifers with the lowest estimated breeding values (EBV) of NM$, compared with a scenario that used only conventional semen and where surplus heifers were culled randomly. Certainly, making breeding decisions to maximize the genetic merit of the herd can be complex and it depends also on the uncertainty about which cows will become pregnant with a heifer calf (Johnson et al., 2018). For this reason, studies have compared different mating optimisation approaches for their capacity to maximize expected genetic gain. In particular, the breeding worth (BrW) was used as a tool to measure genetic merit by Johnson et al. (2018), who selected herds from all New Zealand, whereas bulls to mate cows were selected from the active sires list in New Zealand (DairyNZ, 2017) and then, several mating strategies were tested. Individual cow BrW was used to identify the top cows to nominate mating to the top bulls. The most effective strategy to improve BrW was the one in which whole herds were randomly mated to the entire bull team, generated from bulls currently marketed in NZ, excluding low-ranked cows (with the lowest BrW) that were from producing replacement heifers. A limiting factor of these strategies is the need of generating enough replacement heifers, which depends on both herd reproductive performance and the length of AI for seasonal herds. Instead, Bérodier et al. (2021) studied a herd-based mating algorithm to maximize heifer expected genetic gain while limiting the expected progeny inbreeding and the probability to conceive an offspring homozygous for a lethal recessive allele. Moreover, computerized data-driven decision support tools have been developed to assist dairy farmers in herd management. Cabrera (2018), in a recent review, briefly described some tools developed to help dairy farmers with their herd replacement management strategies in order to enhance herd profits (e.g., a software to calculate and compare the economic value of reproductive programs to improve reproductive performance and farm profit; a tool to estimate the net present value difference between a sexed semen program and a conventional program for dairy heifers and the net return associated with the reproductive performance considering the monthly pregnancy rates; a tool regarding the use of beef semen on dairy cows and heifers to calculate the number of eligible heifers and cows to be bred and the production of calves according to semen used, under a defined protocol of using conventional, sexed and beef semen according to breeding services and genetic makeup; a tool to help dairy farmers whether to use genomic testing on their heifer calves).

Environmental impact of heifer management

The environmental impact of intensive farming is undergoing an increasing interest in public opinion. At the same time, dairy production chain aims at performing in a more environmentally responsible way and studies have been conducted to look for strategies to reduce the environmental impact of cattle farming systems. From this perspective, authors focused on studying the impact of different heifer feeding strategies on nutrient flows (Mourits et al., 2000; Hoffman et al., 2001). Indeed, under Dutch traditional rearing scenario, Mourits et al. (2000) observed that a ratio that covers heifers needs' (i.e., ratio consisting of 45.8% grass, 41.3% grass silage, and 12.8% concentrates) resulted in nutrient input of 72.0 kg nitrogen (N) and 21.2 kg phosphorus (P) per heifer per year. Then, balancing nutrient input and output, these resulted in a surplus of 51.4 kg/year/heifer for N and 17.0 kg/year/heifer for P. Furthermore, to meet industry concerns about N excretion, Hoffman et al. (2001) conducted a study to evaluate the effect of dietary protein on growth and N utilization of post-pubertal Holstein replacement heifers (400 kg). In the trial, heifers were fed diets containing 8, 11, 13, or 15% crude protein (CP) and the authors observed that the increase of dietary CP resulted in linear increases of N intake, fecal-N, and urinary-N; they also observed that diets containing 13% CP provided the maximal growth and nutrient utilization of Holstein heifers. Furthermore, Lascano and Heinrichs (2011) reported higher N retention comparing high concentrate with 20% forage (HC) and low concentrate with 80% forage (LC) diets. Moreover, they reported that wet fecal, dry fecal, and fecal water outputs were lower for HC than LC diets; on the other hand, urine output was greater in HC-fed than LC-fed heifers. In addition, emissions of NH_3-N from manure of heifers fed HC were higher per unit of manure, but not on a daily basis. These findings were in line with results of Zanton and Heinrichs (2009), who did not find significant differences in total N excretion and retention between forage level in heifers diets. Also, NH_3 volatilization did not differ between forage levels in diets, but only increased with increasing N intake because of greater levels of urine and total N excretion, as observed by Lascano et al. (2008) who manipulated the forage-to-concentrate ratio in experimental diets.

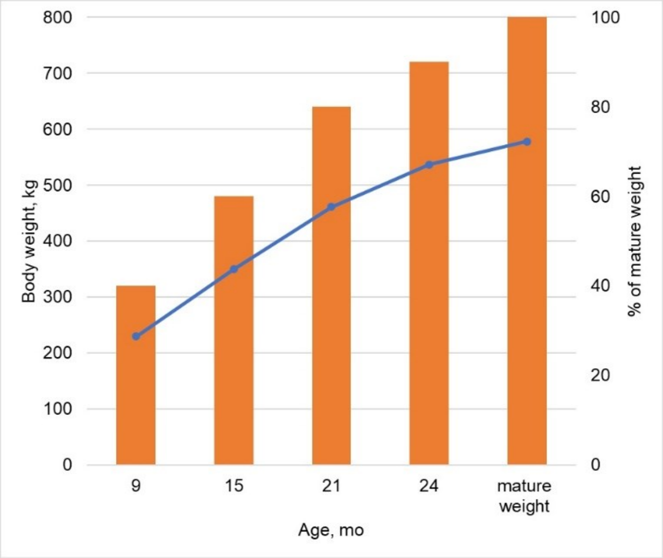
Moreover, Foskolos and Moorby (2018) assessed dairy cattle lifetime N use efficiency incorporating animals' performances, fertility aspects, and diseases both of lactating and replacement animals. They considered the N use efficiency of six categories (heifer growth, heifer removal, pregnancy, cow removal, disease and fertility, and milk production) and according to their findings, a mean replacement N use efficiency (ReplNE) of 23.7% over the total categories was estimated, and the value was most substantially affected by the last stage of heifer growth. Furthermore, results highlighted that replacement rate was negatively correlated with lifetime N excretion and variation in AFC showing larger changes on ReplNE than variation in protein content in diets. A negative correlation of AFC with lifetime and replacement N use efficiency was estimated. In their study, time to first calving affected total feed N requirements for heifer growth and consequently N losses for growth for the period between first service and first calving. They also estimated a mean N requirement for heifer growth of 42.9 ± 5.6 kg of N per heifer (with BW at 1st calving of 544 kg) and feed N utilization efficiency of 25.2%.

Phenotypic and genetic factors

Growth and body weight

Replacement heifers need to reach specific target BW to ensure they calve at desired time, and subsequently achieve high milk yield and good fertility (Bryant et al., 2004). Even though the mature BW is difficult to estimate (Bryant et al., 2004), different BW targets have been recommended for heifers based on their expected mature BW. Furthermore, BW is a key determinant of the time of onset of puberty, because a heifer reaches puberty when she attains 42 to 48% of her mature BW (Bryant et al., 2004). Accordingly, target BW at 6 months, at mating (15 months), and pre-calving (22 months) of 30%, 60%, and 90% of mature BW, respectively, have been suggested as goal standards (Mcnaughton and Lopdell, 2012). Besides that, in order to calve at desired time, a heifer should maintain an average growth rate of about 750 g/d (Wathes et al., 2014). In a New Zealand study, Bryant et al. (2004) presented a model which uses the estimated breeding value for mature BW of individual heifers to calculate target BW at important stages of growth (Fig. 1).

Fig. 1. Target weight of dairy heifers in New Zealand across different rearing phases (modified from Bryant et al., 2004). Blue line represents the average body weight at specific age and orange bars the percentage of the mature weight reached at that specific age.



Furthermore, Hurst et al. (2021) investigated if variables collected during the preweaning phase (daily milk consumption, serum total protein, pneumonia and scours incidences, body size composite, birth weights, and incremental BW) can be used to predict future BW of replacement heifers. They assumed that milk consumption and incidences of respiratory disease occurring in the preweaning phase can influence dairy heifer BW up to 400 d of age, and observed that increased total milk consumption and birth weight resulted in heavier heifers at 400 d of age. Respiratory disease also had a significant effect on predicted BW of heifers up to 400 d. Interestingly, over time, BW of calves treated once, twice, or 3 or more times for respiratory disease did not differ from one another, whereas not-treated-calves were statistically different from the ones treated for respiratory disease. Based on the results, variables collected during the first 60 d of life were able to account for more than 30% of the difference in BW up to 400 d of age. Moreover, Archbold et al. (2012), in an Irish study, assessed the relative influence of BW at mating start date (MSD) in heifers on puberty, lifetime milk production performance, longevity, and profitability. They reported that heifer BW significantly influenced the heifers pubertal rate with higher value at increasing BW at MSD. Heavier heifers at MSD tended to calve earlier in the first lactation and survived longer compared with lighter heifers.

Moreover, heifer BW at MSD was significantly associated with subsequent milk fat and protein yield potential, not only during first lactation but also as a long-term effect. Accordingly, greater BW at first calving leads to lower incidence of dystocia (Hoffman and Funk, 1992). Looking at the relationship between BW of dairy heifers and subsequent milk production, McNaughton and Lopdell (2013) observed a positive relationship between 305-day milk yield in first and second lactation and the percentage of target BW attained by heifers; moreover, a 1% increase in target BW measured between 18 and 21 months was associated with an increase of 23.2 ± 0.2 litres milk in the first lactation and 24 ± 0.9 litres in the second lactation. Handcock et al. (2019) reported that BW from 3 to 21 months of age had significant effect on first lactation milk yield and heifers that were heavier pre-calving produced more energy corrected milk and milk solids (fat and protein yield) than heifers that were lighter. Similar results were obtained observing milk production up to 3rd calving; indeed, heavier heifers produced more energy corrected milk and milk solids compared with lighter heifers. In a subsequent study, Handcock et al. (2020) focused on the relationships between BW and stayability, and BW and calving pattern. Results showed that heavier heifers were more likely to remain in the herd for first, second, and third lactation compared with heifers that were lighter, and they were more likely to have first calving earlier than lighter heifers. Accordingly, Han et al. (2021) observed the relationship between heavy and light heifers at first calving and first lactation milk production and they concluded that there were no significant relationship between BW at first calving and milk yield in the 24 months after first calving, but heifers that reached between 73 and 77% maturity rate at first calving can produce more milk in first lactation without sacrificing long-term milk yield. They also displayed that heifers that were heavier at calving lost more weight during the first month of lactation and subsequently faced higher risk of being culled earlier in life than lighter heifers.

Optimizing AFC

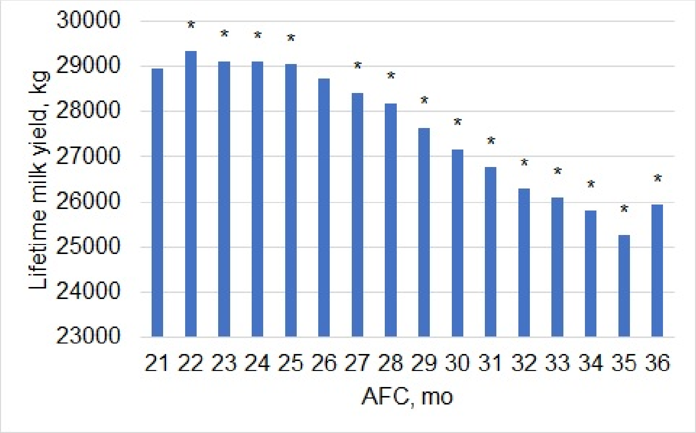
Age at first calving is a key aspect in the dairy herd as it defines the point at which an animal starts to generate profit. Several studies have attempted to define the optimal AFC and its impact on lifetime productive and reproductive performances in cows. Usually, AFC varied between 24 and 36 months and attempts have been made to reduce it (Le Cozler et al., 2008). Results are controversial and defining an optimum AFC is complicated. Some authors have suggested an optimal average AFC between 22 and 24 months (Table 1) without negatively affecting lifetime productive and reproductive performances (Cooke et al., 2013; Ettema and Santos, 2004).

Table 1. Optimal AFC for Holstein dairy cows.

Optimal AFC (mo)	Reference	Country
23-24	Pirlo et al. (2000)	Italy
21-22	Cole et al. (2013)	USA
23-24.5	Ettema and Santos (2004)	USA
23-25	Cooke et al. (2013)	UK
22-26	Moriarty S. (2017)	Irland
22	Beavers and Van Doormaal (2015)	Canada

The relationship between the shortening of AFC and first lactation milk yield and milk components has been demonstrated. Van Eetvelde et al. (2020) reported AFC to be the most influencing variable in first lactation milk yield revealing the lowest 305-d ECM in the youngest heifers (21 months), and the highest reached at 33 months. Additionally, Pirlo et al. (2000) observed that AFC below 23 months has a negative effect on first lactation milk yield and fat percentage. Similarly, Ettema and Santos (2004) reported that heifers with AFC less than 700 d produce less milk and lower fat content (%) than heifers that calve later, and Mohd Nor et al. (2013) observed a significant decrease in 305-d milk yield when AFC moved from 24 to 23 months. Moreover, Sherwin et al. (2016) observed that first lactating cows calving at 23-24 months had the highest probability to calve a second time. Accordingly, Van Pelt et al. (2016) reported that survival rate during first lactation had an optimum between 23 and 24 months AFC and calving at older ages resulted always in a decline in survival rate. In addition, Eastham et al. (2018) reported that heifers calving at 21 months produce less milk than others but, also, heifers that calved earlier than 22-23 months old had the lowest 305-d somatic cell count and calving interval, and the longest lifetime daily milk yield, similar to results observed by Hutchinson et al. (2017) (Fig. 2).

Fig. 2. Least squares means of lifetime milk yield across age at first calving (AFC) of 242 000 heifers in 1 000 Italian herds (ANAFIBJ, unpublished data).



Age at first calving has an impact also on reproductive performances. Indeed, Ettema and Santos (2004) reported lower conception rate (CR) at first service after calving, increased calving difficulties, and higher risk of stillborn calves in heifers that calved at less than 22 months. Furthermore, the relationship between AFC and CR, calving interval, and days open has been investigated by Krpálková et al. (2014) which observed that the group with AFC ≥ 26 months had the highest CR, the highest number of completed lactations, and the lowest percentage of culling cows, whereas AFC between 24.5 and 26 months had lower calving interval and days open and higher milk yield than the group with older AFC. Increasing AFC has also been associated with a reduced probability to complete the first lactation (Heise et al., 2018) and an increased risk of leaving the herd prematurely (Cooke et al., 2013). Finally, Kamal et al. (2014) examined the association between heifers AFC and calf birth weight, and observed that calves born from heifers between 20.3 and 22 months old and 23.5 to 25.5 months old had a birth weight comparable to that of calves born from heifers between 22 and 23.5 months old.

Heifer fertility

In order to achieve expected breeding goals, heifer fertility is an essential aspect and to enhance heifer fertility, maximising heifer CR is desired. In Holstein, it has been suggested that heifers have to conceive at around 15 months of age to reach first calving at desired age (Wathes et al., 2014).

Furthermore, Kuhn et al. (2006) reported that a breeding age from 15 to 16 months maximises CR. As reported by Wathes et al. (2014), the major aspects influencing puberty are BW and age, and heifer fertility depends both on management decisions and genetics. The onset of puberty is the sum of a gradual increase in gonadotropic activity through the luteinizing hormone (LH) and follicle stimulating hormone (FSH), generated by a decline in the negative feedback of estradiol on the hypothalamic secretion of gonadotropin-releasing hormone (Gn-RH), and a subsequent increase of LH secretion (Larson, 2007). Poor reproductive performances are associated with a reduction in circulating concentrations of insulin, IGF-1, leptin, and glucose, and elevated concentrations of BHBA, NEFA, and glucocorticoids (Butler, 2014). Some studies have focused on the effects of different diets on hormones concentration. Mackey et al. (2000) observed that progesterone and estradiol concentrations are unaffected by diet, whereas insulin plasma concentrations increased proportionally as energy diet content increased. Furthermore, growth rate and maximum diameter of the dominant follicle decreased under severe nutritional restrictions. Some authors studied the positive effects of supplemental fatty acids on heifer reproductive performances by increasing energy intake in the diet (Howlett et al. 2003; Larson, 2007). Santos et al. (2008) reported that the most beneficial fatty acids on heifer fertility performances are the polyunsaturated FA of the n-6 and n-3 families. Moreover, Hutchinson et al. (2012) analysed the effect of four different fat supplements (palmitic acid, flax-seed containing α-linolenic acid, conjugated linoleic acids, fish oil) on follicle and corpus luteum development, and steroid hormone in early lactation cows. Results reported that cows fed both with palmitic acid and conjugated linoleic acids had higher plasma progesterone concentration and greater corpus luteum volume during the first 14 d after ovulation, whereas cows fed flax-seed diet had greater plasma IGF-1 concentrations than cows receiving the palmitic acid diet, and cows receiving diet containing palmitic acid had less plasma NEFA concentrations than cows fed other diets. Indeed, Butler (2014) reported that prostaglandin synthesis is more influenced by the ratio n-3 to n-6 fatty acids provided through the diet, rather than absolute levels of a specific polyunsaturated FA. Fertility traits have been collected and stored into national databases since many years but as they are lower heritable than productive traits, only in recent years they have been included in breeding goals worldwide (Van Raden et al., 2004; Miglior et al., 2005). Given that heifer fertility data are available earlier in life and are genetically correlated with lactating cow fertility (Tiezzi et al., 2012), genetic parameters for heifer reproductive performances have been estimated (Table 2, Table 3). All reported studies agreed that age at first service and conception rate at first service revealed the highest heritability between traits evaluated (from 0.23 to 0.05); whereas heritability for other considered traits revealed values between 0.008 to 0.03, depending on trait considered (Table 2). Genetic correlations between fertility traits have been displayed by several studies and similar correlations have been reported in both magnitude and direction (Table 3). Then, the link between estimated breeding value for fertility traits and phenotypic performances has been assessed (Veronese et al., 2019a and 2019b; Meier et al., 2021). Veronese et al. (2019a and 2019b) investigated the association between heifer genomic merit for daughter pregnancy rate (DPR) and calving rate with oestrus characteristics, pregnancies, and steroidal hormones. Those authors reported that DPR was positively associated with PGF2α-synchronized estruses and oestrus manifestation within 7 d of PGF2α treatment, whereas calving rate tended to be positively correlated with pregnancy up to 80 d after enrolment in the reproductive program. Furthermore, in the subsequent study, Veronese et al. (2019b) reported that heifers with high DPR had higher estradiol concentration, larger ovulatory follicle diameter and greater ovulation probability within 96 hours after oestrus expression, whereas heifers with high calving rate had higher concentration of IGF-1 from days 7 to 35 of pregnancy, concluding that selecting animals for DPR and CR could improve animal fitness. Meier et al. (2021) analysed heifers' reproductive performances and their EBV for fertility traits in New Zealand. Based on their results, heifers with a predicted high breeding value for fertility traits reached puberty at an earlier age and lighter BW and showed greater percentage of heifers identified as pregnant compared with heifers with a predicted low breeding value for the same traits.

Table 2. Estimates of heritability for heifer fertility traits.

Trait[1]	Heritability	Authors	Records (n.)	Breed
NR56	0.03 (0.01)[2]	Muir et al. (2004)	476 433	Canadian Holstein
	0.029 (0.006)[3]	Jamrozik et al. (2005)	53 158	Canadian Holstein
	0.012*	Liu et al. (2008)	2 593 985	German Holstein, Red dairy cattle, and Jersey
	0.016 (0.005;0.032)[4]	Tiezzi et al. (2012)	37 546	Brown Swiss
	0.010 (0.001)[2]	de Haer er al. (2013)	315 858	Dutch/Flemish Holstein
	0.01 (0.001)[3]	Oliveira Junior et al. (2021)	434 670	Canadian Holstein
	0.013*	Ferrari et al. (2023)	4 863 802	Italian Holstein
AFS	0.19 (0.02)[2]	Muir et al. (2004)	477 748	Canadian Holstein
	0.134 (0.013)[3]	Jamrozik et al. (2005)	53 158	Canadian Holstein
	0.227 (0.010)[2]	de Haer er al. (2013)	311 408	Dutch/Flemish Holstein
	0.05 (0.001)[3]	Oliveira Junior et al. (2021)	434 676	Canadian Holstein
	0.058	Brzáková et al. (2019)	342 648	Czech Holstein
	0.071*	Ferrari et al. (2023)	4 863 802	Italian Holstein
INS	0.029 (0.006)[3]	Jamrozik et al. (2005)	53 158	Canadian Holstein
	0.026 (0.015;0.041)[4]	Tiezzi et al. (2012)	37 546	Brown Swiss
	0.01 (0.001)[3]	Oliveira Junior et al. (2021)	434 527	Canadian Holstein
IFC	0.030 (0.006)[3]	Jamrozik et al. (2005)	53 158	Canadian Holstein
	0.017 (0.009;0.029)[4]	Tiezzi et al. (2012)	37 546	Brown Swiss
	0.01 (0.001)[3]	Oliveira Junior et al. (2021)	387 677	Canadian Holstein
	0.010	Brzáková et al. (2019)	215 456	Czech Holstein
IFL	0.014*	Liu et al. (2008)	2 593 985	German Holstein, Red dairy cattle, and Jersey
	0.016 (0.002)[2]	de Haer er al. (2013)	315 858	Dutch/Flemish Holstein
	0.012 (0.005)[2]	Muuttoranta et al. (2019)	102 326	Nordic Holstein
	0.015*	Ferrari et al. (2023)	4 863 802	Italian Holstein
CFS	0.20 (0.009;0.037)[4]	Tiezzi et al. (2012)	37 546	Brown Swiss
CR	0.014 (0.002)[2]	de Haer er al. (2013)	315 858	Dutch/Flemish Holstein
	0.008 (0.003)[2]	Muuttoranta et al. (2019)	176 286	Nordic Holstein
	0.012*	Ferrari et al. (2023)	4 863 802	Italian Holstein

[1] NR56 = nonreturn rate at 56 d after first service; AFS = age at first service; INS = number of inseminations to conception; IFC = interval from first service to conception; IFL = interval from first to last insemination; CFS = conception rate at first service; CR = conception rate.

[2] Standard error.

[3] Posterior standard deviation.

[4] Highest posterior density region at 95%.

*Approximate SE were from 0.001 to 0.002 (Liu et al., 2008) and from 0.01 to 0.03 (Ferrari et al., 2023).

Table 3. Genetic correlations for heifer fertility traits.[1]

	NR56	INS	IFC	IFL	CR
AFS	0.08 ± 0.12[2] (Muir et al., 2004) 0.28 ± 0.01[3] (Jamrozik et al., 2005) 0.286* (de Haer et al., 2013) 0.11 ± 0.025[3] (Oliveira Junior et al., 2021) 0.006* (Ferrari et al., 2023)	0.23 ± 0.09[3] (Jamrozik et al., 2005) 0.05 ± 0.026[3] (Oliveira Junior et al., 2021)	-0.14 ± 0.09[3] (Jamrozik et al., 2005) 0.17 ± 0.025[3] (Oliveira Junior et al., 2021) 0.197 ± 0.002[3] (Brzáková et al., 2019)	0.077* (de Haer et al., 2013) -0.089* (Ferrari et al., 2023)	0.082* (de Haer et al., 2013) 0.048* (Ferrari et al., 2023)
NR56		-0.85 ± 0.05[3] (Jamrozik et al., 2005) −0.94 ± 0.026[3] (Oliveira Junior et al., 2021)	-0.66 ± 0.08[3] (Jamrozik et al., 2005) −0.84 ± 0.026[3] (Oliveira Junior et al., 2021)	-0.49* (Liu et al., 2008); -0.588* (de Haer et al., 2013) -0.309* (Ferrari et al., 2023)	0.814* (de Haer et al., 2013) 0.763* (Ferrari et al., 2023).
INS			0.97 ± 0.025[3] (Oliveira Junior et al., 2021) 0.92 ± 0.02[3] (Jamrozik et al., 2005)		
IFL					-0.95 ± 0.35[2] (Muuttoranta et al., 2019) -0.729* (Ferrari et al., 2023).

[1] AFS = age at first service; NR56 = nonreturn rate at 56 d after first service; INS = number of inseminations to conception; IFC = interval from first service to conception; IFL = interval from first to last insemination; CFS = conception rate at first service; CR = conception rate.

[2] Standard error.

[3] Posterior standard deviation.

*Standard error ranged from 0.015 to 0.035 (Liu et al., 2008), from 0.005 to 0.10 (de Haer et al., 2013) and from 0.01 to 0.08 (Ferrari et al., 2023)

Breeding tools and technologies

Breeding technologies and tools are valuable and efficient instruments that help farmers to choose the best breeding strategy for the herd. The combination of them allows farmers to: (1) enhance herd genetic progress through increased selection intensity (Calus et al., 2015); (2) optimize breeding decisions to choose the best heifers in the herd to keep as replacements; (3) control inbreeding and avoid genetic defects (Pryce and Hayes, 2012); (4) enhance farm profit selling crossbred calves (Bittante et al., 2020); (5) obtain replacement heifers from genetically superior animals (Ettema et al., 2017); (6) increase farm efficiency (Holden and Butler, 2018).

Genomic selection

Meuwissen et al. (2001) first published a paper on genomic selection and the possible potential of this tool and since 2009, genomic information started to be included in breeding schemes worldwide as a way to speed up the genetic gain by reducing generation interval (Calus et al., 2015). At herd level, genomic selection can be used in heifers as a valuable rearing management strategy to choose the best animals to become future replacements and to lower herd genetic lag. Actually, heifer's breeding values at birth can be known with a reliability of more than 60% (Pryce and Hayes, 2012) and it has been displayed that heifers with genomic tests will have their genetic merit determined with a greater accuracy than contemporaries without a genomic test (Hjortø et al., 2015). Hence, genomic selection would help farmers in their breeding mating selection, to monitor inbreeding levels and avoid deleterious diseases and genetic defects caused by recessive alleles (Bérodier et al., 2021; Pryce and Hayes, 2012). Weigel et al. (2012) studied how the expected gains in lifetime net merit (LNM$) changed in a US Holstein farm based on routine genomic female testing (using a low-density SNP panel) as a way to enhance the value of future herd replacements. They observed that, when pedigree information was not available, genotyping all heifers was the best strategy that optimised LNM$; whereas, when pedigree information was provided, the genetic gain value depended on the cost of genotyping, then, selecting only a subset of heifers to genotype was more cost effective. Accordingly, Calus et al. (2015) estimated the break-even cost to display the maximum genotyping cost that would be repaid by additional revenues under different scenarios and depending on replacement rate and heifers available. Their results suggested that genotyping all heifers led to positive outcomes in different scenarios (setting genotyping price at approximately 50 €, based on 2015 Dutch situation). Lastly, Hjortø et al. (2015) investigated if the use of genomic tests for breeding decisions would decrease genetic lag within herd. They evaluated 5 strategies of genomic tests use in heifers (from no use to 100% heifers genotyped) and they reported that the effect of genomic tests on the herd genetic level improved when the reproductive performances of the herd was good compared when they were average and when more genomic tests were performed.

Sexed semen

The use of sexed semen has become a commercial strategy in dairy (and beef) sector to enhance the probability to conceive a female by 85-90% (Cerchiaro et. al., 2007); indeed, at farm level it is used to ensure enough replacement heifers. In dairy herds, female calves are required for both replacements and herd expansion but the economics of the use of sexed semen depends on some aspects. The process to separate bull semen into a fraction that contains higher than normal X-bearing sperm concentration takes some time, and the result is a lower sperm count and less motile (Fetrow, 2007). Hence, in commercial dairy herds, its use has been recommended primarily on heifers, given that nulliparous heifer has better pregnancy rates and higher CR than lactating cows (Holden and Butler, 2018). Indeed, DeJarnette et al. (2011) evaluated the effect of conventional versus sex-sorted semen on CR of Holstein heifers and they found that CR of sex-sorted semen were lowered by 16% than that of conventional semen (41 and 57%, respectively). Cabrera (2009) reported that the use of sexed semen in dairy heifers is supported for first sexed semen service if their expected CR is more than 30%, otherwise the economic value of its use is not worth. The use of sexed semen on virgin heifers is also preferred to reduce calving difficulties, because it decreased calving problems incidence (e.g., dystocia) as female calves are lighter than males (Weigel, 2004), and decreased dystocia expected costs (De Vries, 2009).

Crossbreeding

Beef from dairy herds is the meat originated both from cow culling and surplus calves and even if beef is considered as an auxiliary outcome of the dairy herd, it produces a significant revenue in dairy herd cash flow. Indeed, its contribution on total meat production is considerable (Berry, 2021). Nowadays, the interest in beef-on-dairy is increasing worldwide (Fig. 3; Van Doormaal, 2019) and at national level (Fig. 4) thanks to some factors: (1) the use of genomic selection and sexed semen have allowed lower heifer replacements needs (Berry et al., 2014); (2) the exploitation of heterosis; (3) increasing consumer's concern on young dairy (male) calves process (Berry, 2021); (4) higher revenue from crossbred calf sale; (5) improved carcass characteristics (Bittante et al., 2020); (6) breed complementary effect enhancement, in which one breed compensates for the weakness of the other (Berry, 2021).

Fig. 3. Percentage of inseminations with Holstein semen and with beef semen on Canadian Holsteins by year of inseminations (Van Doormaal, 2019).

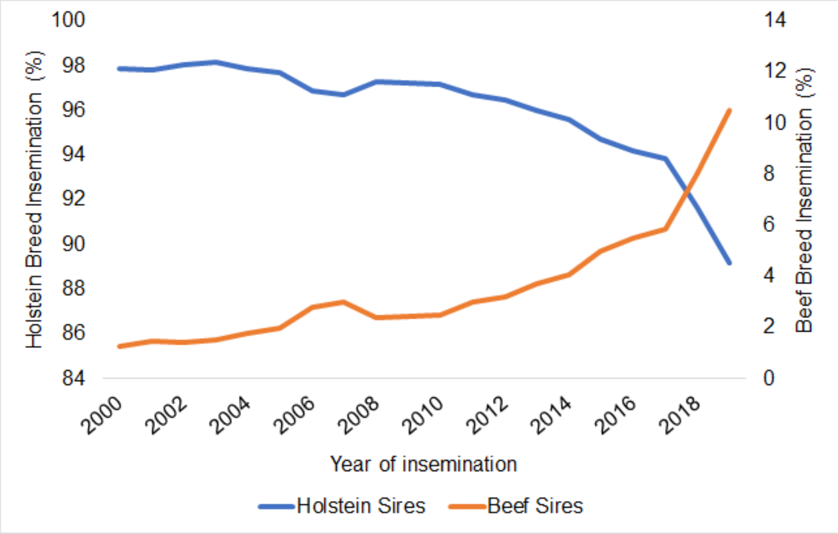
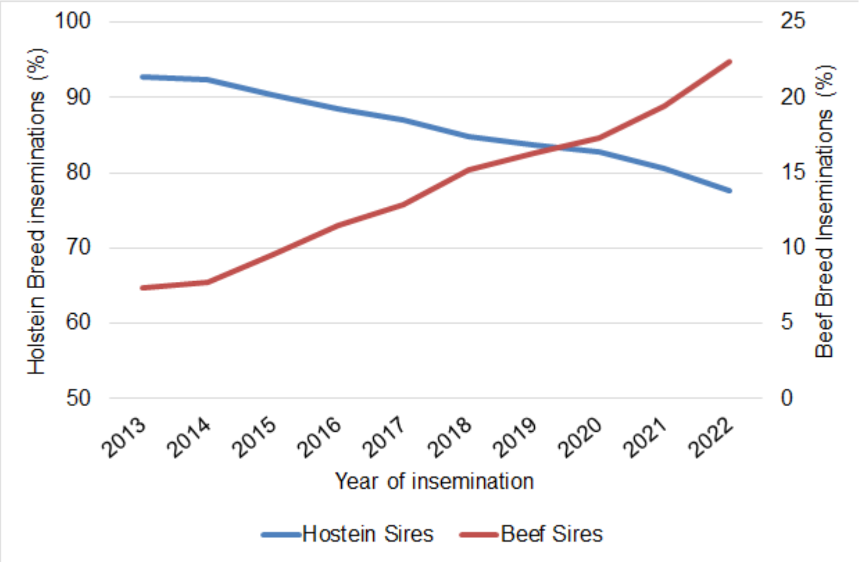


Fig. 4. Holstein breed insemination (%) and beef breed insemination (%) on Italian Holsteins by year of inseminations (ANAFIBJ Database, accessed on 1st December 2022).



Besides that, improving beef from dairy herds, has been shown to decrease carbon footprint of beef production (Holden and Butler, 2018). Crossbreeding is not primarily used on dairy heifers but its use is a direct consequence of an improved replacement's management when combined with other breeding tools to reach better genetic and economic advantages (Clasen et al., 2021); indeed, as a strategic way to increase the herd genetic level, beef semen in dairy herds is often used on inferior genetic merit cows (Berry, 2021; Hjortø et al., 2015). Hence, differences among beef breeds in calving and gestation performances when mated to dairy cows have been discussed (Berry and Ring, 2020a; Fouz et al., 2013). In their study, Fouz et al. (2013) analysed the effect of the use of crossbreeding in dairy herds with respect to calving difficulty and gestation length, and they compared these traits after breeding pure Holsteins or crossbreeding with Belgian Blue (BB), Limousine (LM), and Galician Blonde (GB) bulls. They found that Holstein x BB had the highest calving difficulty, followed by LM and GB; whereas Holstein x LM and GB cows had longer gestation lengths compared with purebred and Holstein x BB cows. Moreover, they observed that both calving difficulty and gestation length were higher in young cows. Accordingly, Scanavez and Mendonça (2018) reported that cows inseminated with Holstein semen have lower gestation length compared to those inseminated with Angus semen. Moreover, Bittante et al. (2020) observed higher CR in dairy crossbreeding, attributed to the effect of heterosis and they argued that the incidence of dystocia depends both on reproduction strategy and the beef breed used. Accordingly, Berry and Ring (2020a) showed the existing variability within beef breeds in terms of calving difficulty, gestation length and calf mortality, and they reported that selecting animals for a dairy-beef index led to an increasing in the overall herd profit. Berry and Ring (2020) evaluated the probability of a female to be mated with a beef or a dairy bull. The factors associated with an increasing probability of being mated with beef semen were age of the cow (higher probability in older cows), poor overall total merit index (in their study they considered the Irish Economic Breeding Index), calving dystocia and veterinary assistance requirement at calving. Lastly, Bittante et al. (2020) displayed the possible use of beef semen on heifers in rotational dairy crossbreeding systems to enhance the NPV of the dairy herd.

Conclusions

This review summarizes the main findings on genetic and management aspects of dairy heifers and it serves as an update on the current state of research. Rearing heifers is challenging given that lifetime performances should be enhanced whilst minimizing rearing costs. To rear heifers efficiently, nutrient requirements must be fulfilled both to reach target BW at specific growing age and to reduce heifers' environmental impact. Besides that, genetic aspects play a relevant role in animals lifetime performances and factors such as growth and age influence both the onset of productive life and future performances. To attain this, good fertility is essential and it can be improved both genetically and phenotypically. Indeed, genetic and genomic aspects play a relevant role to help reaching the best from young stock. In recent years, research has focused on breeding technologies, tools, and the combination of them, to allow farmers optimizing breeding replacement decisions, enhancing farm profits, and reducing herd environmental impact. To support dairy farmers rearing decisions and help them in their breeding definition, models have been developed.

Ethics approval

Not applicable.

Data and model availability statement

None of the data were deposited in an official repository. No new datasets were created.

Declaration of generative AI and AI-assisted technologies in the writing process

The authors did not use any artificial intelligence assisted technologies in the writing process.

Author ORCIDs

V. Ferrari https://orcid.org/0000-0002-7774-4388

J. B. C. H. M. van Kaam https://orcid.org/0000-0002-2592-2461

M. Penasa https://orcid.org/0000-0001-9984-8738

M. Marusi https://orcid.org/0000-0002-2359-3633

R. Finocchiaro https://orcid.org/0000-0002-9058-9992

M. Cassandro https://orcid.org/0000-0002-8709-2870

Author contributions

Ferrari V.: Conceptualisation, Methodology, Investigation, Data curation, Visualisation, Writing - Original Draft; Writing - review and editing.

van Kaam J. B. C. H. M.: Writing - Review & Editing.

Penasa M.: Methodology, Supervision, Writing - review and editing.

Marusi M.: Methodology**,** Conceptualisation, Supervision, Writing - review and editing.

Finocchiaro R.: Methodology**,** Conceptualisation, Methodology, Supervision, Writing - review and editing.

Cassandro M.: Project administration, Supervision, Writing - review and editing; Funding acquisition.

Declaration of interest

The authors declare no conflict of interest.

Acknowledgements

None.

Financial support statement

This study was supported by "Latteco2 project, sottomisura 10.2 of the PSRN-Biodiversity 2020–2023" (MIPAAF, D.M. no. 465907 del 24/09/2021, project unique code J12C21004080005).

References

Abeni, F., Petrera, F., Le Cozler, Y., 2019. Effects of feeding treatment on growth rates, metabolic profiles and age at puberty, and their relationships in dairy heifers. Animals 13, 1020-1029. https://doi.org/10.1017/S1751731118002422

Akins, M. S., 2016. Dairy Heifer Development and Nutrition Management. Veterinary Clinics of North America - Food Animal Practice 32, 303–317. https://doi.org/10.1016/j.cvfa.2016.01.004

Archbold, H., Shalloo, L., Kennedy, E., Pierce, K.M., Buckley, F., 2012. Influence of age, body weight and body condition score before mating start date on the pubertal rate of maiden Holstein-Friesian heifers and implications for subsequent cow performance and profitability. Animal 6, 1143–1151. https://doi.org/10.1017/S1751731111002692

Beavers, L., Van Doormaal, B., 2015. Age at First Calving and Profitability. Canadian Dairy Network.

Bérodier, M., Berg, P., Meuwissen, T., Boichard, D., Brochard, M., Ducrocq, V., 2021. Improved dairy cattle mating plans at herd level using genomic information. Animal 15, 100016. https://doi.org/10.1016/j.animal.2020.100016

Berry, D.P., E. Wall, Pryce J.E., 2014. Genetics and genomics of reproductive performance in dairy and beef cattle. Animal 8:105-121. https://doi.org/10.1017/S1751731114000743

Berry, D.P., Ring, S.C., 2020. Short communication: Animal-level factors associated with whether a dairy female is mated to a dairy or beef bull. Journal of Dairy Science 103, 8343-8349. https://doi.org/10.3168/jds.2020-18179

Berry, D.P., Ring, S.C., 2020a. Observed progeny performance validates the benefit of mating genetically elite beef sires to dairy females. Journal of Dairy Science 103, 2523–2533. http://dx.doi.org/10.3168/jds.2020-19519

Berry, D.P., 2021. Invited review: Beef-on-dairy—The generation of crossbred beef × dairy cattle. Journal of Dairy Science 104, 3789-3819. http://dx.doi.org/10.3168/jds.2020-19519

Bittante, G., Negrini, R., Bergamaschi, M., Cecchinato, A., Toledo-Alvarado, H., 2020. Pure-breeding with sexed semen and crossbreeding with semen of double-muscled sires to improve beef production from dairy herds: Factors affecting heifer and cow fertility and the sex ratio. Journal of Dairy Science 103, 5246-5257. http://dx.doi.org/10.3168/jds.2019-17932

Boulton, A.C., Rushton, J., Wathes, D.C., 2017. An empirical analysis of the cost of rearing dairy heifers from birth to first calving and the time taken to repay these costs. Animal 11, 1372–1380. https://doi.org/10.1017/S1751731117000064

Brown, E.G., VandeHaar, M.J., Daniels, K.M., Liesman, J.S., Chapin, L.T., Forrest, J.W., Akers, R.M., Pearson, R.E., and Weber Nielsen, M.S., 2005. Effect of increasing energy and protein intake on mammary development in heifer calves. Journal of Dairy Science 88, 595–603. https://doi.org/10.3168/jds.S0022-0302(05)72723-5

Bryant, J.R., Holmes, C.W., Mcnaughton, L.R., Brookes, I.M., Verkerk, G.A., Pryce, J.E., 2004. Use of breeding values for live weight to calculate individual live weight targets for dairy heifers. Procc. New Zealand Society of Animal Production, 118–121.

Brzáková, M., Zavadilová, L., Přibyl, J., Pešek, P., Kašná, E., Kranjčevičová, A., 2019. Estimation of genetic parameters for female fertility traits in the Czech Holstein population. Czech Journal of Animal Science 64, 199–206. https://doi.org/10.17221/51/2018-CJAS

Butler, S., 2014. Nutritional management to optimize fertility of dairy cows in pasture-based systems. Animal 8, 15-26. https://doi.org/10.1017/S1751731114000834

Cabrera, V.E., 2018. Invited review: Helping dairy farmers to improve economic performance utilizing data-driving decision support tools. Animal 12, 134-144. http://dx.doi.org/10.1017/S1751731117001665

Cabrera, V.E., 2009. When To Use Sexed Semen on Heifers. Proceedings Dairy Cattle Reproduction Conference, Annu. Conv., Minneapolis, MN, and Boise, ID, DCRC, New Prague, MN.

Calus, M.P.L., Bijma, P., Veerkamp, R.F., 2015. Evaluation of genomic selection for replacement strategies using selection index theory. Journal of Dairy Science 98, 6499-6509. http://dx.doi.org/10.3168/jds.2014-9192

Cerchiaro, I., Cassandro, M., Dal Zotto, R., Carnier, P., Gallo, L., 2007. A Field Study on Fertility and Purity of Sex-Sorted Cattle Sperm. Journal of Dairy Science, 2538-2542. https://www.sciencedirect.com/science/article/pii/S0022030207717514

Clasen, J.B., Kargo, M., Østergaard, S., Fikse, W.F., Rydhmer, L., Strandberg, E., 2021. Genetic consequences of terminal crossbreeding, genomic test, sexed semen, and beef semen in dairy herds. Journal of Dairy Science 104, 8062- 8075. https://doi.org/10.3168/jds.2020-20028

Cole, J.B., Hutchison, J.L., Bickhart, D.M., Null, D.J., 2013. Optimal age at first calving for U.S. dairy cattle. Journal of Dairy Science 96(E-Suppl. 1), 289–290 (abstr. W188)

Cooke, J.S., Cheng, Z., Bourne, N.E., Wathes, D.C., 2013. Association between growth rates, age at first calving and subsequent fertility, milk production and survival in Holstein-Friesian heifers. Open Journal of Animal Sciences 3, 1–12. https://doi.org/10.4236/ojas.2013.31001

Davis Rincker, L.E., VandeHaar, M.J., Wolf, C.A., Liesman, J.S., Chapin, L.T., Weber Nielsen, M.S., 2011. Effect of intensified feeding of heifer calves on growth, pubertal age, calving age, milk yield, and economics. Journal of Dairy Science 94, 3554–3567. https://doi.org/10.3168/jds.2010-3923

de Haer, L.C.M., de Jong, G., Vessies, P.J.A., 2013. Estimation of Genetic Parameters of Fertility Traits, for Virgin Heifers in The Netherlands. Interbull Bulletin 47, 142-146.

DeJarnette, J.M., Leach, M.A., Nebel, R.L., Marshall, C.E., McCleary, C.R., Moreno, J.F., 2011. Effects of sex-sorting and sperm dosage on conception rates of Holstein heifers: Is comparable fertility of sex-sorted and conventional semen plausible? Journal of Dairy Science, 94, 3477-3483. http://dx.doi.org/10.3168/jds.2011-4214

De Vries, A., 2009. The Economics of Sexed Semen in Dairy Heifers and Cows. Edis. Retrieved on 4 May 23 from http://edis.ifas.ufl.edu.

Eastham, N.T., Coates, A., Cripps, P., Richardson, H., Smith, R., Oikonomou, G., 2018., Associations between age at first calving and subsequent lactation performance in UK Holstein and Holstein-Friesian dairy cows. PLoS ONE 13, 1-13. https://doi.org/10.1371/journal.pone.0197764

Ettema, J.F., Santos, J.E.P., 2004. Impact of age at calving on lactation, reproduction, health, and income in first-parity Holsteins on commercial farms. Journal of Dairy Science, 87, 2730–2742. https://doi.org/10.3168/jds.S0022-0302(04)73400-1

Ettema, J.F., Thomasen, J.R., Hjortø, L., Kargo, M., Østergaard, S., Sørensen, A. C., 2017. Economic opportunities for using sexed semen and semen of beef bulls in dairy herds. Journal of Dairy Science 100, 4161-4171. https://doi.org/10.3168/jds.2016-11333

Ferrari, V., Visentin, G., van Kaam, J.B.C.H.M., Penasa, M., Marusi, M., Finocchiaro, R., Cassandro, M., 2023. Genetic and nongenetic variation of heifer fertility in Italian Holstein cattle. JDS Commununications 4, 35-39. https://doi.org/10.3168/jdsc.2022-0270

Fetrow, J., M. Overton, and S. Eicker. 2007. Sexed semen: economics of a new technology. Proceedings Western Dairy Management Conference, March 7-9, Reno, Nev. Retrieved on 24 August 22 from http://www.wdmc.org/2007/fetrow.pdf

Foskolos, A., Moorby, J.M., 2018. Evaluating lifetime nitrogen use efficiency of dairy cattle: A modelling approach. PLoS ONE, 13, 1–22. https://doi.org/10.1371/journal.pone.0201638

Fouz, R., Gandoy, F., Sanjuan, M.L., Yus, E., Dieguez, F.J., 2013. The use of crossbreeding with beef bulls in dairy herds: Effects on calving difficulty and gestation length. Animal 7, 211–215. https://doi.org/10.1017/ S1751731112001656

Gabler, M.T., Tozer, P.R., Heinrichs, A.J., 2000. Development of a cost analysis spreadsheet for calculating the costs to raise a replacement dairy heifer. Journal of Dairy Science 83, 1104–1109. https://doi.org/10.3168/jds.S0022-0302(00)74975-7

Gardner, R.W., Smith, L.W., Park, R.L., 1988. Feeding and Management of Dairy Heifers for Optimal Lifetime Productivity. Journal of Dairy Science 71, 996–999. https://doi.org/10.3168/jds.S0022-0302(88)79646-0

Green, T.C., Jago, J.G., Macdonald, K.A., Waghorn, G.C., 2013. Relationships between residual feed intake, average daily gain, and feeding behavior in growing dairy heifers. Journal of Dairy Science 96, 3098-3107. http://dx.doi.org/ 10.3168/jds.2012-6087

Greter, A.M., Prinsen, M., Duffield, T.F., McBride, B.W., Widowski, T.M., DeVries, T.J., 2013. Growing dairy heifers prefer supplementary long straw when fed a nutrient-dense ration in a limited amount. Journal of Dairy Science 96, 3950–3958. http://dx.doi.org/ 10.3168/jds.2013-6625

Han, L., Heinrichs, A. J., De Vries, A., Dechow, C. D., 2021. Relationship of body weight at first calving with milk yield and herd life. Journal of Dairy Science 104, 397–404. https://doi.org/10.3168/jds.2020-19214

Handcock, R.C., Lopez-Villalobos, N., McNaughton, L.R., Back, P.J., Edwards, G.R., Hickson, R.E., 2019. Positive relationships between body weight of dairy heifers and their first-lactation and accumulated three-parity lactation production. Journal of Dairy Science 102, 4577–4589. https://doi.org/10.3168/jds.2018-15229

Handcock, R.C., Lopez-Villalobos, N., McNaughton, L.R., Back, P.J., Edwards, G.R., Hickson, R.E., 2020. Body weight of dairy heifers is positively associated with reproduction and stayability. Journal of Dairy Science 103, 4466–4474. https://doi.org/10.3168/jds.2019-17545

Heinrichs, A.J., Jones, C.M., Gray, S.M., Heinrichs, P.A., Cornelisse, S.A., Goodling, R.C., 2013. Identifying efficient dairy heifer producers using production costs and data envelopment analysis. Journal of Dairy Science 96, 7355–7362. https://doi.org/10.3168/jds.2012-6488

Heinrichs, A.J., Zanton, G.I., Lascano, G.J., Jones, C.M., 2017. A 100-Year Review: A century of dairy heifer research. Journal of Dairy Science 100, 10173–10188. https://doi.org/10.3168/jds.2017-12998

Heise, J., Stock, K.F., Reinhardt, F., Ha, N.T., Simianer, H., 2018. Phenotypic and genetic relationships between age at first calving, its component traits, and survival of heifers up to second calving. Journal of Dairy Science 101, 425–432. https://doi.org/10.3168/jds.2017-12957

Hjortø, L., Ettema, J.F., Kargo, M., Sørensen, A. C., 2015. Genomic testing interacts with reproductive surplus in reducing genetic lag and increasing economic net return. Journal of Dairy Science 98, 646-658. https://doi.org/10.3168/jds.2014-8401

Hoffman, P.C., Funk, D.A., 1992. Applied Dynamics of Dairy Replacement Growth and Management. Journal of Dairy Science 75, 2504–2516. https://doi.org/10.3168/jds.S0022-0302(92)78012-6

Hoffman, P.C., Esser, N.M., Bauman, L.M., Denzine, S.L., Engstrom, M., Chester-Jones, H., 2001. Short communication: Effect of dietary protein on growth and nitrogen balance of holstein heifers. Journal of Dairy Science 84, 843–847. https://doi.org/10.3168/jds.S0022-0302(01)74542-0

Hoffman, P.C., Simson, C.R., Wattiaux, M., 2007. Limit feeding of gravid holstein heifers: Effect on growth, manure nutrient excretion, and subsequent early lactation performance. Journal of Dairy Science 90, 946-954. https://doi.org/10.3168/jds.S0022-0302(07)71578-3

Holden, S.A., Butler, S.T., 2018. Review: Applications and benefits of sexed semen in dairy and beef herds. Animal 12, s97-s103. https://doi.org/10.1017/S1751731118000721

Howlett, C.M., Vanzant, E.S., Anderson, L.H., Burris, W.R., Fieser, B.G., Bapst, R.F., 2003. Effect of supplemental nutrient source on heifer growth and reproductive performance, and on utilization of corn silage-based diets by beef steers. Journal of animal science 81, 2367-2378. https://doi.org/10.2527/2003.8192367x

Hurst, T.S., Lopez-Villalobos, N., Boerman, J.P., 2021. Predictive equations for early-life indicators of future body weight in Holstein dairy heifers. Journal of Dairy Science 104, 736–749. https://doi.org/10.3168/jds.2020-18560

Hutchison, J. L., VanRaden, P. M., Null, D. J., Cole, J. B., Bickhart, D. M., 2017. Genomic evaluation of age at first calving. Journal of Dairy Science 100, 6853-6861. http://dx.doi.org/10.3168/jds.2016-12060

Hutchinson, I.A., Hennessy, A.A., Waters, S.M., Dewhurst, R.J., Evans, A.C.O., Lonergan, P., Butler, S.T., 2012. Effect of supplementation with different fat sources on the mechanisms involved in reproductive performance in lactating dairy cattle. Theriogenology 78, 12-27. https://doi.org/10.1016/j.theriogenology.2011.12.031

Jamrozik, J., Fatehi, J., Kistemaker, G.J., Schaeffer, L.R., 2005. Estimates of Genetic Parameters for Canadian Holstein Female Reproduction Traits. Journal of Dairy Science 88, 2199-2208. https://doi.org/10.3168/jds.s0022-0302(05)72895-2

Johnson, T., Eketone, K., McNaughton, L., Tiplady, K., Voogt, J., Sherlock, R., Anderson, G., Keehan, M., Davis, S. R., Spelman, R. J., Chin, D., Couldrey, C., 2018. Mating strategies to maximize genetic merit in dairy cattle herds. Journal of Dairy Science 101, 4650–4659. https://doi.org/10.3168/jds.2017-13538

Kamal, M.M., Van Eetvelde, M., Depreester, E., Hostens, M., Vandaele, L., Opsomer, G., 2014. Age at calving in heifers and level of milk production during gestation in cows are associated with the birth size of Holstein calves. Journal of Dairy Science 97, 5448–5458. https://doi.org/10.3168/jds.2014-7898

Kaniyamattam, K., Elzo, M.A., Cole, J. B., De Vries, A., 2016. Stochastic dynamic simulation modeling including multitrait genetics to estimate genetic, technical, and financial consequences of dairy farm reproduction and selection strategies. Journal of Dairy Science 99, 8187–8202. https://doi.org/10.3168/jds.2016-11136

Khan, M.A., Weary, D.M., Von Keyserlingk, M.A.G., 2011. Invited review: Effects of milk ration on solid feed intake, weaning, and performance in dairy heifers. Journal of Dairy Science 94, 1071–1081. https://doi.org/10.3168/jds.2010-3733

Krpálková, L., Cabrera, V.E., Kvapilík, J., Burdych, J., Crump, P., 2014. Associations between age at first calving, rearing average daily weight gain, herd milk yield and dairy herd production, reproduction, and profitability. Journal of Dairy Science 97, 6573–6582. https://doi.org/10.3168/jds.2013-7497

Kuhn, M.T., Hutchison, J.L., Wiggans, G.R., 2006. Characterization of Holstein heifer fertility in the United States. Journal of Dairy Science 89, 4907-4920. http://dx.doi.org/10.3168/jds.S0022-0302(06)72541-3

Larson R.L., 2007. Heifer Development: Reproduction and Nutrition. Veterinary Clinics of North America: Food Animal Practice, 23, 53-68. https://doi.org/10.1016/j.cvfa.2006.11.003

Lascano, G.J., Heinrichs, A.J., 2011. Effects of feeding different levels of dietary fiber through the addition of corn stover on nutrient utilization of dairy heifers precision-fed high and low concentrate diets. Journal of Dairy Science 94, 3025–3036. https://doi.org/10.3168/jds.2010-3831

Lascano, G.J., Zanton, G.I., Suarez-Mena, F.X., Heinrichs, A.J., 2009. Effect of limit feeding high- and low-concentrate diets with Saccharomyces cerevisiae on digestibility and on dairy heifer growth and first-lactation performance. Journal of Dairy Science 92, 5100-5110. https://doi.org/10.3168/jds.2009-2177

Lascano, G.J., Zanton, G.I., Moody, M.L., Topper, P.A., Wheeler, E.F., Heinrichs, A.J., 2008. Short communication: Effect of changing the ratio of forage to concentrate on ammonia emissions by dairy heifers. Journal of Dairy Science 91, 4301–4306. https://doi.org/10.3168/jds.2008-1176

Le Cozler, Y., Lollivier, V., Lacasse, P., Disenhaus, C., 2008. Rearing strategy and optimizing first-calving targets in dairy heifers: A review. Animal 2, 1393–1404. https://doi.org/10.1017/S1751731108002498

Liu, Z., Jaitner, J., Reinhardt, F., Pasman, E., Rensing, S., Reents, R., 2008. Genetic Evaluation of Fertility Traits of Dairy Cattle Using a Multiple-Trait Animal Model. Journal of Dairy Science 91, 4333-4343. https://doi.org/10.3168/jds.2008-1029

Mackey D.R., Wylie A.R.G., Sreenan J.M., Roche J.F., Diskin, M.G., 2000. The effect of acute nutritional change on follicle wave turnover, gonadotropin, and steroid concentration in beef heifers. Journal of Animal Science 78, 429–442. https://doi.org/10.2527/2000.782429x

Mcnaughton, L., Lopdell, T., 2012. Brief communication: Are dairy heifers achieving liveweight targets? Proceedings of the New Zealand Society of Animal Production, July 2012, pp 120–122.

McNaughton, L.R., Lopdell, T., 2013. Effect of heifer live weight on calving pattern and milk production. Proceedings of the New Zealand Society of Animal Production, July 2012, pp 103–107. http://dx.doi.org/10.1016/j.procir.2016.08.002

Meier, S., McNaughton, L.R., Handcock, R., Amer, P.R., Beatson, P.R., Bryant, J.R., Dodds, K.G., Spelman, R., Roche, J.R., Burke, C.R., 2021. Heifers with positive genetic merit for fertility traits reach puberty earlier and have a greater pregnancy rate than heifers with negative genetic merit for fertility traits. Journal of Dairy Science 104, 3707-3721 https://doi.org/10.3168/jds.2020-19155

Meyer, M.J., Capuco, A.V., Ross, D.A., Lintault, L.M., Van Amburgh, M.E., 2006. Developmental and nutritional regulation of the prepubertal heifer mammary gland: I. Parenchyma and fat pad mass and composition. Journal of Dairy Science 89, 4289–4297. https://doi.org/10.3168/jds.S0022-0302(06)72475-4

Meuwissen, T.H.E., Hayes, B.J., and M.E. Goddard, 2001. Prediction of Total Genetic Value Using Genome-Wide Dense Marker Maps, *Genetics* 157, 1819–1829. https://doi.org/10.1093/genetics/157.4.1819

Miglior, F., Muir, B.L., Van Doormaal, B. J., 2005. Selection indices in Holstein cattle of various countries. Journal of Dairy Science 88, 1255-1263. http://dx.doi.org/10.3168/jds.S0022-0302(05)72792-2

Mohd Nor, N., Steeneveld, W., Mourits, M.C.M., Hogeveen, H., 2015. The optimal number of heifer calves to be reared as dairy replacements. Journal of Dairy Science 98, 861–871. http://dx.doi.org/10.3168/jds.2014-8329

Mohd Nor, N., Steeneveld, W., van Werven, T., Mourits, M.C.M., Hogeveen, H., 2013. First-calving age and first-lactation milk production on Dutch dairy farms. Journal of Dairy Science 96, 981–992. https://doi.org/10.3168/jds.2012-5741

Moriarty, S., 2017. Retrieved on 4 May 23 from https://www.icbf.com/wp-content/uploads/2019/11/Optimum-age-at-first-calving.pdf

Mourits, M.C.M., Berentsen, P.B.M., Huirne, R.B.M., Dijkhuizen, A.A., 2000. Environmental impact of heifer management decisions on Dutch dairy farms. Netherlands Journal of Agricultural Science 48, 151–164. https://doi.org/10.1016/S1573-5214(00)80011-1

Mourits, M.C.M., Huirne, R.B.M., Dijkhuizen, A.A., Kristensen, A.R., Galligan D., 1999. Economic Optimization of Dairy Heifer Management Decisions. Agricultural System 61, 17-31.

Mourits, M.C.M., Dijkhuizen, A.A., Huirne, R.B.M., Galligan, D.T., 1997. Technical and Economic Models to Support Heifer Management Decisions: Basic Concepts. Journal of Dairy Science 80, 1406–1415. https://doi.org/10.3168/jds.S0022-0302(97)76070-3

Muir, B.L., Fatehi, J., Schaeffer, L.R., 2004. Genetic Relationship between Persistency and Reproductive Performance in First-Lactation Canadian Holstein. Journal of Dairy Science 87, 3029-3037. https://doi.org/10.3168/jds.S0022-0302(04)73435-9

Muuttoranta, K., Tyrisevä, A., Mäntysaari, E.A., Pösö, J., Aamand, G.P., Lidauer, M.H., 2019. Genetic parameters for female fertility in Nordic Holstein and Red Cattle dairy breeds. Journal of Dairy Science 102, 8184–8196. https://doi.org/10.3168/jds.2018-15858

Newton, J.E., Hayes, B.J., Pryce, J.E., 2018. The cost-benefit of genomic testing of heifers and using sexed semen in pasture-based dairy herds. Journal of Dairy Science 101, 6159–6173. https://doi.org/10.3168/jds.2017-13476

Oliveira Junior, G.A., Schenkel, F.S., Alcantara, L., Houlahan, K., Lynch, C., Baes, C.F., 2021. Estimated genetic parameters for all genetically evaluated traits in Canadian Holsteins. Journal of Dairy Science 104, 9002–9015. https://doi.org/10.3168/jds.2021-20227

Overton, M.W., Dhuyvetter, K.C., 2020. Symposium review: An abundance of replacement heifers: What is the economic impact of raising more than are needed? Journal of Dairy Science 103, 3828–3837. https://doi.org/10.3168/jds.2019-17143

Pirlo, G., Miglior, F., Speroni, M., 2000. Effect of age at first calving on production traits and on difference between milk yield returns and rearing costs in Italian Holsteins. Journal of Dairy Science 83, 603–608. https://doi.org/10.3168/jds.S0022-0302(00)74919-8

Pryce, J., Hayes, B., 2012. A review of how dairy farmers can use and profit from genomic technologies. Animal Production Science 52, 180-184. https://doi.org/10.1071/AN11172

Santos, J.E., Bilby, T.R., Thatcher, W.W., Staples, C.R. Silvestre, F.T., 2008. Long chain fatty acids of diet as factors influencing reproduction in cattle. Reproduction in Domestic Animals 43, 23–30 https://doi.org/10.1111/j.1439-0531.2008.01139.x

Scanavez, A.L., Mendonça, L.G., 2018. Gestation Length and Overall Performance in the Subsequent Lactation of Dairy Cows Conceiving to Holstein, Jersey, or Angus Semen: An Observational Study. Kansas Agricultural Experiment Station Research Reports 4,1-7. https://doi.org/10.4148/2378-5977.7713

Sejrsen, K., Huber, J.T., Tucker, H.A., Akers, R.M., 1982. Influence of Nutrition on Mammary Development in Pre- and Postpubertal Heifers. Journal of Dairy Science 65, 793–800. https://doi.org/10.3168/jds.S0022-0302(82)82268-6

Sherwin, V.E., Hudson, C.D., Henderson, A., Green, M.J., 2016. The association between age at first calving and survival of first lactation heifers within dairy herds. Animal 10, 1877–1882. https://doi.org/10.1017/S1751731116000689

Tiezzi, F., Maltecca, C., Cecchinato, A., Penasa, M., Bittante, G., 2012. Genetic parameters for fertility of dairy heifers and cows at different parities and relationships with production traits in first lactation. Journal of Dairy Science 95, 7355- 7362. http://dx.doi.org/10.3168/jds.2012-5775

Tranel, L., 2019. Heifer Raising Costs in 2019. Retrieved on 10 August 22 from https://www.extension.iastate.edu/dairyteam/files/page/files/whats_it_cost_to_raise_heifers_2019.pdf

Van Doormaal, B., 2019. Use of Beef Sire Semen in the Dairy Industry. Retrieved on 09 September 2022 on https://www.cdn.ca/document.php?id=526

Van Eetvelde, M., de Jong, G., Verdru, K., van Pelt, M.L., Meesters, M., Opsomer, G., 2020. A large-scale study on the effect of age at first calving, dam parity, and birth and calving month on first-lactation milk yield in Holstein Friesian dairy cattle. Journal of Dairy Science 103, 11515–11523. https://doi.org/10.3168/jds.2020-18431

Van Pelt, M.L., De Jong, G., Veerkamp, R.F., 2016. Changes in the genetic level and the effects of age at first calving and milk production on survival during the first lactation over the last 25 years. Animal 10, 2043–2050. https://doi.org/10.1017/S1751731116001282

VanRaden, P.M., Sanders, A.H., Tooker, M.E., Miller, R.H., Norman, H.D., Kuhn, M.T., Wiggans, G.R., 2004. Development of a National Genetic Evaluation for Cow Fertility. Journal of Dairy Science 87, 2285-2292. https://doi.org/10.3168/jds.S0022-0302(04)70049-1

Veronese, A., Marques, O., Moreira, R., Belli, A.L., Bisinotto, R.S., Bilby, T.R., Peñagaricano, F., Chebel, R.C., 2019a. Genomic merit for reproductive traits. I: Estrous characteristics and fertility in Holstein heifers. Journal of Dairy Science 102, 6624-6638. https://doi.org/10.3168/jds.2018-15205

Veronese, A., Marques, O., Peñagaricano, F., Bisinotto, R. S., Pohler, K.G., Bilby, T.R., Chebel, R.C., 2019b. Genomic merit for reproductive traits. II: Physiological responses of Holstein heifers. Journal of Dairy Science 102, 6639-6648. https://doi.org/10.3168/jds.2018-15245

Wathes, D.C., Pollott, G.E., Johnson, K.F., Richardson, H., Cooke, J.S., 2014. Heifer fertility and carry over consequences for life time production in dairy and beef cattle. Animal 8, 91–104. https://doi.org/10.1017/S1751731114000755

Weigel, K.A., Hoffman, P. C., Hoffman, P. C., Lawlor, Jr T.J., 2012. Potential gains in lifetime net merit from genomic testing of cows, heifers, and calves on commercial dairy farms. Journal of Dairy Science 95, 2215-2225. http://dx.doi.org/10.3168/jds.2011-4877

Weigel, K.A., 2004. Exploring the role of sexed semen in dairy production systems. Journal of Dairy Science, 87, 120-130. https://doi.org/10.3168/jds.S0022-0302(04)70067-3

Zanton, G.I., Heinrichs, A.J., 2009. Digestion and nitrogen utilization in dairy heifers limit-fed a low or high forage ration at four levels of nitrogen intake. Journal of Dairy Science 92, 2078–2094. https://doi.org/10.3168/jds.2008-1712

Aims of the thesis

The overall objective of the present thesis was to investigate genetic and management aspects that affect dairy heifer fertility traits. The specific aims were to:

i) identify genetic and non-genetic factors affecting Italian Holstein heifer fertility, and propose an aggregate index for heifer fertility;

ii) investigate genetic and genomic aspects of age at first calving;

iii) describe a practical tool to help farmers optimize replacement management.

Chapter I

Genetic and nongenetic variation of heifer fertility in Italian Holstein cattle

V. Ferrari[1,2,*], G. Visentin[3,*], J.B.C.H.M. van Kaam[1], M. Penasa[2], M. Marusi[1], R. Finocchiaro[1,] M. Cassandro[1,2]

[1]Associazione Nazionale Allevatori della Razza Frisona, Bruna e Jersey Italiana, Via Bergamo 292, 26100, Cremona (CR), Italy

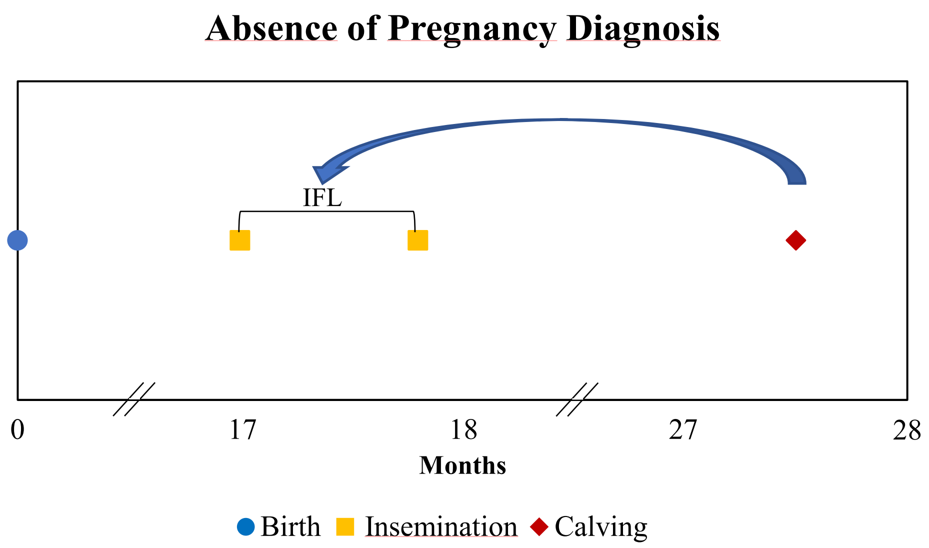
[2]Department of Agronomy, Food, Natural resources, Animals and Environment, University of Padova, Viale dell'Università 16, 35020, Legnaro (PD), Italy

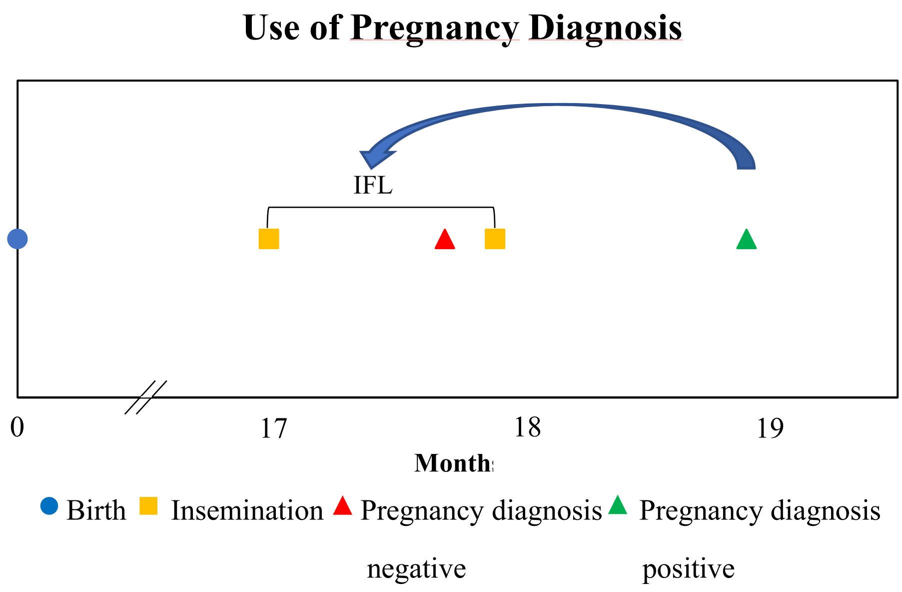
[3]Department of Veterinary Medical Sciences, Alma Mater Studiorum - University of Bologna, Via Tolara di Sopra 50, 40064, Ozzano dell'Emilia (BO), Italy

*These authors equally contributed to the manuscript.

Published in *JDS Communications (2023), 4:35-39*
https://doi.org/10.3168/jdsc.2022-0270

Graphical Abstract





Summary

Pregnancy diagnosis is important to characterize fertility earlier and on a larger proportion of individuals. In the present study, such information was exploited to derive genetic parameters for fertility in nulliparous Italian Holstein heifers. A selection index for heifer fertility was developed using selection index methodology, and phenotypic variation of heifer fertility traits across systematic environmental effects was estimated. The present study demonstrated that although lowly heritable, heifer fertility manifested ample and exploitable genetic variation for selective breeding. The derived

selection index can be included in the national breeding objective as an additional source of information for fertility. Efforts should be made on the collection of pregnancy diagnosis to improve the genetic evaluation of fertility.

Highlights

- Pregnancy diagnosis allows determination of heifer fertility traits earlier.
- Reduction of censored data is achievable by exploiting pregnancy diagnosis.
- Exploitable additive genetic variation exists for heifer fertility.
- Heifer ability to conceive at first service can be improved by selective breeding.

ABSTRACT

Excellent fertility performance is important to maximize farmers' profit and to reduce the number of culled animals. Although female fertility of adult cows has been included in Italian Holstein breeding objectives since 2009, little has been done to quantify genetic variation of heifer fertility characteristics so far. The aim of the present study was to estimate genetic parameters of 4 fertility traits in nulliparous Italian Holstein heifers and to develop an aggregate selection index to improve heifer fertility. Data were retrieved from the national fertility database and included information on insemination, calving, and pregnancy diagnosis dates. The investigated phenotypes (mean ± standard deviation) were age at first insemination (AFI, mo; 17.25 ± 2.89), nonreturn rate at 56 d from the first insemination (NRR56, binary; 0.78 ± 0.41), conception rate at first insemination (CR, binary; 0.61 ± 0.49), and interval from first to last insemination (IFL, d; 26.09 ± 51.85). Genetic parameters were estimated using a 4-trait animal model that included the following fixed effects: herd-year of birth and month of birth for AFI, and herd-year-season of birth and month-year of insemination for IFL, NRR56, and CR; the animal additive genetic effect (fitted to the pedigree-based relationship matrix) was considered as a random term. An aggregate index was developed from the estimated additive genetic (co)variance matrix by considering CR as the breeding goal and AFI, NRR56, and IFL as selection criteria. Heritability estimates from average covariance matrices ranged from 0.012 (CR) to 0.015 (IFL), with the exception of AFI (0.071). Conception rate at first insemination was strongly correlated with both IFL (−0.730) and NRR56 (0.668), and weakly to AFI (−0.065), and the relative emphasis placed on each selection criteria in the aggregate index was 10%, 47%, and 43% for AFI, IFL, and NRR56, respectively. The results of the present study suggest that heifer fertility should be considered as an additional trait in the breeding objectives of Italian Holstein.

Keywords: heifer fertility, Holstein, genetic variation, heritability, genetic correlation.

SHORT COMMUNICATION

Fertility is essential for dairy farmers to sustain and maximize annual farm profit. In young stock, poor fertility increases the non-productive period and increases costs, leading to reduced longevity and lifetime milk production (VanRaden et al., 2004; Wathes et al., 2014). Although lowly heritable, fertility exhibits exploitable additive genetic variation and is negatively genetically correlated to production performances (Wall et al., 2003; VanRaden et al., 2004; Berry et al., 2014). For these reasons, female fertility has become an important driver of breeding decisions in the dairy industry and is nowadays included in merit indices worldwide (Cole and VanRaden, 2018). In general, the main goal is to improve conception and daughter pregnancy rates, and favor shorter calving intervals in lactating cows, and reduce the number of heifers that fail to conceive. The advantages of heifer fertility traits are that they are available early in life (before 18 mo of age) and they are genetically correlated to fertility of lactating cows (Tiezzi et al., 2012). Subsequently, some countries also include heifer fertility traits in their genetic evaluations and most of them consider only conception rate at first insemination (**CR**) or non-return rate at 56 d (**NRR56**; Fleming et al., 2019).

In Italy, heifer fertility data for dairy and dual-purpose cattle breeds, including inseminations and calving dates, have been stored nationally for a long time, and since 2015, pregnancy diagnoses have also been routinely collected and stored. These data are important from different points of view. For example, early information on pregnancy status is helpful to improve reproductive efficiency and pregnancy rate in cattle (Fricke, 2002), and it can reduce the amount of censored data on traits such as days open and interval from first to last insemination (**IFL**; Wiggans and Goodling, 2005). Therefore, information on heifer fertility, combined with pregnancy status data, can be exploited to support farmers in their breeding decision process. The objective of the present study was to quantify genetic and nongenetic variation of fertility in nulliparous Italian Holstein heifers. Data used in the present study were retrieved from the national fertility database managed by the National Breeders Association of Italian Holstein, Brown, and Jersey (ANAFIBJ, Cremona, Italy) for the national genetic and genomic evaluation and thus did not involve animals; prior ethical approval was deemed not to be required. The data set contained information on first inseminations (n = 5,596,046), pregnancy diagnosis (n = 1,561,743), and calving events (n = 4,863,802) recorded since 1994. Furthermore, animal ID, event date, service sire, service code (e.g., AI, natural service), and service sire code (e.g., national or foreign daughter proven or genomic bull) were available for each event. A detailed description of the data quality assessment and editing procedures can be retrieved from Biffani et al. (2003).

Briefly, the aim of the quality control process was to identify the most reliable information to be retained for genetic and genomic evaluations by identifying calving events within an appropriate biological gestation range (i.e., 240 to 300 d), and in which the service sire of the successful insemination is effectively the sire of the newborn animal registered in the herdbook. Only fertility-related events collected on nulliparous heifers were extracted from the national database and used in the present study. Traits considered for subsequent genetic analysis were age at first insemination (**AFI**, mo) restricted to be between 9 to 27 mo, IFL (d), NRR56, and CR. For IFL, a value of 1 was attributed when only one insemination occurred and it was followed by a subsequent calving or confirmed by pregnancy diagnosis. When more than one insemination was necessary to achieve pregnancy, IFL was calculated as the difference between the first and the last insemination if followed by a calving, or if pregnancy was confirmed on the last insemination. When IFL was >230 d and conception was confirmed (or calving occurred), IFL was set to 230 d. Moreover, if the last insemination occurred less than 300 d from the first insemination (and conception was not confirmed nor calving occurred), IFL was set to missing. Otherwise, IFL was calculated as the difference from the first to the last insemination but with a penalty of 64 d (i.e., 3 estrous cycles). All these steps allowed us to reduce the proportion of censored records and therefore to also include phenotypic information on the least fertile animals. Regarding NRR56, 0 was attributed only to heifers for which the second insemination occurred within 56 d from the first insemination and 1 otherwise. Moreover, NRR56 was set to missing if the second insemination was within 14 d from the first service. Finally, CR was set to 1 if the first and only insemination was followed by a calving or a positive pregnancy diagnosis.

Fertility traits were first analyzed through a univariate linear animal model to estimate variance components, which were then used as starting values in a 4-trait animal model to estimate additive genetic and residual covariances among traits. (Co)variance components were estimated using the software AIREMLF90 (Misztal, 2008). To reduce computational time, 10 random subsets of 100 herds each were extracted from the entire fertility data set. This generated 10 data samples on which (co)variance components were estimated. Each herd (and related heifer fertility observations) was represented only once across the 10 generated data samples. Additive genetic (**G**) and residual (**R**) (co)variance matrices estimated in each of the 10 different subsets were then averaged; phenotypic (**P**) (co)variance matrix was calculated as the sum of **G** and **R**. Pedigree of animals in each subset was traced back up to 6 generations, when available. In case of 1 or 2 missing parents, unknown ancestors were assigned to genetic groups based on animal origin and year of birth. The multivariate linear mixed model in matrix notation was as follows:

$$\begin{bmatrix} y_1 \\ y_2 \\ y_3 \\ y_4 \end{bmatrix} = \begin{bmatrix} X_1 & 0 & 0 & 0 \\ 0 & X_2 & 0 & 0 \\ 0 & 0 & X_3 & 0 \\ 0 & 0 & 0 & X_4 \end{bmatrix} \begin{bmatrix} b_1 \\ b_2 \\ b_3 \\ b_4 \end{bmatrix} + \begin{bmatrix} Z_1 & 0 & 0 & 0 \\ 0 & Z_2 & 0 & 0 \\ 0 & 0 & Z_3 & 0 \\ 0 & 0 & 0 & Z_4 \end{bmatrix} \begin{bmatrix} a_1 \\ a_2 \\ a_3 \\ a_4 \end{bmatrix} + \begin{bmatrix} e_1 \\ e_2 \\ e_3 \\ e_4 \end{bmatrix},$$

where y is the vector of phenotypic observations (AFI, IFL, NRR56, and CR), **b** is the vector of fixed effects (herd-year of birth and month of birth for AFI, and herd-year-season of birth and month-year of insemination for IFL, NRR56, and CR), **a** is the vector of random additive genetic effects, **e** is the vector of random residuals; and **X** and **Z** are incidence matrices relating the corresponding fixed or random effects, respectively, to the dependent variable. Variances of the random effects were assumed to be equal to $var(\boldsymbol{a}) = \boldsymbol{A}\sigma_a^2$ and $var(\boldsymbol{e}) = \boldsymbol{I}\sigma_e^2$, where σ_a^2 and σ_e^2 are the additive genetic and residual variances, respectively, **A** is the pedigree-based relationship matrix, and **I** is an identity matrix of appropriate order. Genetic parameters were calculated on the average **G** and **R** matrices of the 10 subsets. Heritability was calculated as σ_a^2 / $(\sigma_a^2 + \sigma_e^2)$, and genetic (r_a) and phenotypic (r_p)

correlations were assessed as $cov_{a(x,y)}/(\sigma_{a(x)}\sigma_{a(y)})$ and $cov_{p(x,y)}/(\sigma_{p(x)}\sigma_{p(y)})$, respectively, where cov denotes the genetic (*a*) or phenotypic (*p*) covariance between trait x and y, and σ denotes the genetic (*a*) or phenotypic (*p*) standard deviation of trait *x* or *y*, respectively. The use of a linear animal model was preferred over the use of threshold animal models for NRR56 and CR. Indeed, linear animal models, applied also to binary traits, produce EBV that are strongly correlated to EBV generated from threshold animal models, and are much easier to be implemented in routine genetic evaluation and less computationally demanding (Malchiodi et al., 2017). Finally, least squares means of the fixed effects were estimated using a univariate linear mixed model by including the aforementioned fixed effects (fitted separately and not as interactions) for AFI, IFL, NRR56, and CR, and by adding herd as a random effect. The analysis was performed using the software Echidna (Gilmour, 2020) on the entire national heifer fertility database.

To generate individual weights for EBV to be combined in the aggregate index (**I**) for heifer fertility, CR was considered as the sole breeding goal and AFI, IFL, and NRR56 were considered as selection criteria. The I can be calculated as $I = b'EBV$, where **b** is the vector of selection criteria individual weights and **EBV** is the vector of selection criteria EBV (Dekkers and Gibson, 1998). According to Schneeberger et al. (1992), **b** can be derived as $b = G_I^{-1}G_{IT}v$, were $\boldsymbol{G_I}$ is the matrix with genetic (co)variances between traits in I (AFI, IFL, NRR56), $\boldsymbol{G_{IT}}$ is the matrix with genetic covariances between traits in I and the breeding goal (CR), and $\boldsymbol{v}$ is the vector of economic values which were assumed to be 1 for all traits. Relative emphasis placed on each individual selection criterium *i* was calculated through the following equation (Berry, 2015):

$$Relative\ emphasis = \frac{|b_i\sigma_i|}{\sum_{j=1}^{n}|b_j\sigma_j|},$$

were σ_i and σ_j are the genetic standard deviations of the trait i and j, respectively. The I was then standardized to mean 100 and standard deviation 5 for proof publication within the national genetic and genomic evaluation.

Descriptive statistics of AFI, IFL, NRR56, and CR, as well as the heritability, and genetic and phenotypic correlations from average **G** and **P** (co)variance matrices are presented in Table 1. Mean ± SD of AFI, IFL, NRR56, and CR were 17.25 ± 2.89 mo, 26.09 ± 51.85 d, 0.78 ± 0.41, and 0.61 ± 0.49, respectively.

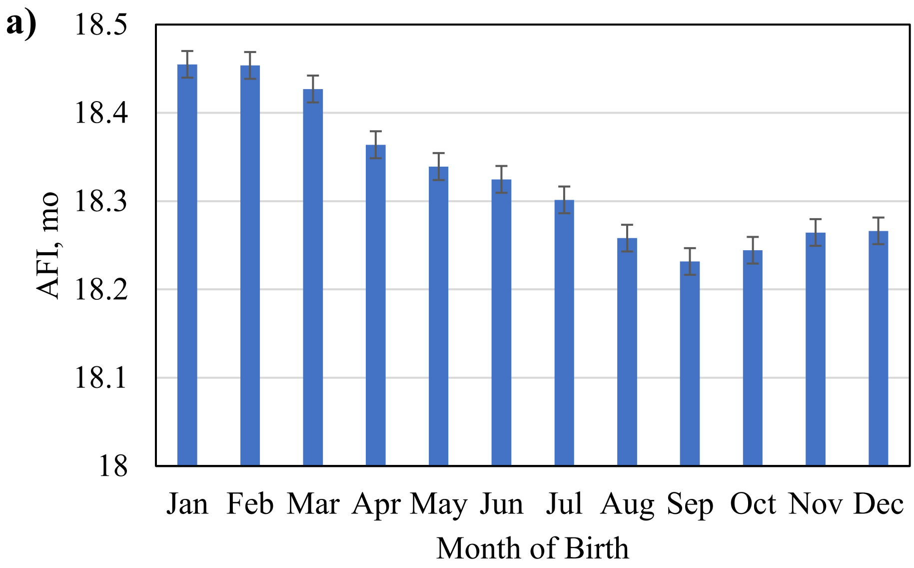
Table 1. Descriptive statistics, heritability (in bold, on the diagonal), genetic correlations (below the diagonal), and phenotypic correlations (above the diagonal) of heifer fertility traits.

	Descriptive statistics				Genetic parameters			
Trait[1]	Mean	SD	Minimum	Maximum	AFI	IFL	NRR56	CR
AFI, mo	17.25	2.89	9	27	**0.071**	-0.089	0.006	0.048
IFL, d	26.09	51.85	1	294	0.050	**0.015**	-0.309	-0.729
NRR56	0.78	0.41	0	1	0.152	-0.256	**0.013**	0.763
CR	0.61	0.49	0	1	-0.065	-0.730	0.668	**0.012**

[1] AFI: age at first insemination; IFL: interval from first to last insemination; NRR56: non-return rate at 56 d; CR: conception rate at first insemination.

Heritability was the greatest for AFI (0.071) and it ranged from 0.012 (CR) to 0.015 (IFL) for the other traits. However, the coefficient of genetic variation (CV_a) was the lowest for AFI (2.75%) and the greatest for IFL (21.22%). The CV_a for NRR56 and CR was 5.88% and 9.99%, respectively. Conception rate was weakly genetically correlated to AFI (-0.065) and strongly correlated to both IFL (-0.730) and NRR56 (0.668); these latter two correlations were very close, in magnitude and direction, to their phenotypic counterparts (Table 1). Age at first insemination was also weakly correlated to IFL ($r_p = -0.089$; $r_a = 0.050$) and NRR56 ($r_p = 0.006$; $r_a = 0.152$). The standard errors of heritability estimates of the four heifer fertility traits at each run ranged from 0.01 to 0.03, and standard errors of genetic and phenotypic correlations from 0.01 to 0.08. The low heritabilities estimated in the current study were expected and consistent with those reported for Canadian Holstein nulliparous heifers (Jamrozik et al., 2005). Results were also in agreement with Liu et al. (2008), who estimated heritability of 0.012 and 0.015 for heifer and cow NRR56, respectively, and 0.014 for heifer IFL in a joint genetic evaluation using Holstein, Red dairy cattle, and Jersey data from Germany, Austria, and Luxemburg. Muir et al. (2004) reported higher heritability estimates for NRR56 (0.030) and AFI (0.190). The present study on heifer fertility is the first one carried out in Italian Holsteins. Indeed, Tiezzi et al. (2012) investigated genetic variation of these traits in Italian Brown Swiss dairy cattle, with heritability estimates that agreed with those of the present study. Genetic correlations estimated in the present study corroborate those reported in previous studies (Jamrozik et al., 2005; Liu et al., 2008) in terms of both direction and magnitude, with AFI being only weakly genetically correlated to other heifer fertility traits (Muir et al., 2004; Jamrozik et al., 2005). The relative emphasis, in absolute value, of the traits included for the calculation of I was 10%, 47%, and 43% for AFI, IFL, and NRR56, respectively, and reflects the genetic correlations between the selection criteria (AFI, IFL, and NRR56) and the breeding goal (CR).

The exclusion of CR from I was because CR is measurable only once calving has occurred, or after a positive pregnancy diagnosis, therefore CR is not measurable in some individuals and could also create a bias in genetic and genomic evaluations, as CR information would not be available for the least fertile daughters. In light of this, encouraging the collection of pregnancy check can be a viable solution to overcome this issue. Although data on pregnancy diagnosis are currently available at the national level, this source of information is not routinely collected by all farmers, and it has been introduced recently in the national fertility database. However, the number of registered pregnancy diagnoses on heifers increased from 180,779 in 2015 to 264,700 in 2021, with Northern Regions contributing the largest proportion (77.90%), followed by Southern (13.60%) and Central Regions (8.50%). This reflects the national distribution of Holstein herds, which are mainly located in the Po valley (Northern Italy), i.e., the area more suited to intensive dairy farming. Results of the present study are of great importance for the national Holstein breeding program not only to provide the farmers with useful information to optimize breeding decisions to improve fertility, but also because genetic proofs of heifer NRR56 can be used to participate to Interbull (2022) Multiple Across Country Evaluation (**MACE**) for the trait "Maiden heifer ability to conceive" (**T1**). Proofs of this trait differ among countries participating in MACE, yet the genetic correlation between Holstein T1 of Italy and T1 of other countries ranges from 0.808 to 0.924, and this correlation is, as expected, stronger with countries that have defined T1 as in the current study (April 2022 evaluation run). Figure 1a depicts the least squares means of AFI across months of birth. Although the effect was statistically significant, differences across months of birth are of little biological impact. Indeed, AFI was the highest for animals born in January and February (18.45 mo; SE = 0.15) and the smallest for animals born between August and October, with the minimum in September (18.23 mo; SE = 0.15). Least squares means across years of birth indicated that AFI progressively shortened from 19.16 mo (SE = 0.15) in 1994 to 17.17 mo (SE = 0.15) in 2018. Raising young stock is a significant cost for farmers (up to 15-20% of total milk production costs; Hutchison et al., 2017), and therefore farmers might prefer to voluntary anticipate animals' productive life. The reduction of AFI across calendar years is consistent with previous studies in US (Hare et al., 2006) and Canadian heifers (Duplessis et al., 2015), and could therefore be related to management (e.g., better calf-raising practises) but also to genetic factors. For example, intense selection for higher milk production may have increased the need for replacement heifers due to unfavorable genetic correlations to other functional traits (Brito et al., 2021). Figure 1b depicts the least squares means of IFL, NRR56, and CR across months of insemination. All traits had an erratic and similar trend and denoted poorer fertility performances (i.e., high IFL, and low NRR56 and CR) in September compared to the remaining calendar months of the year. The similarity of the trend among these traits is likely related to the strong genetic and phenotypic correlations between IFL, NRR56, and CR. Although differences among least squares means of months of insemination within traits were significant, their biological impact was negligible. An indefinite pattern of the month of insemination effect has been observed also by Kuhn et al. (2006) who reported higher CR in April and July and lower in September, October, and November.



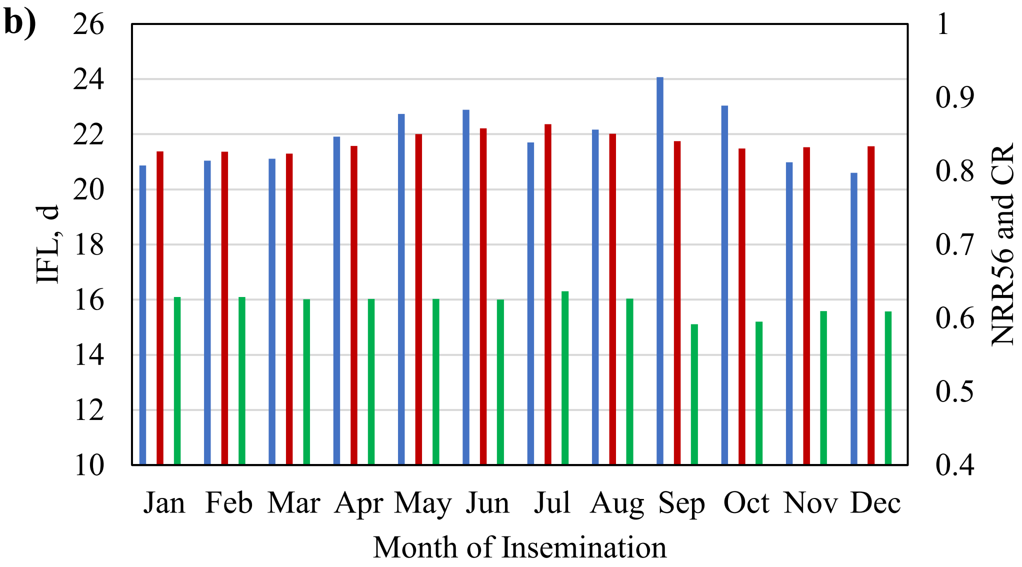


Figure 1. Least squares means of (a) age at first insemination (AFI) across months of birth (SE = 0.15) and (b) interval from first to last insemination (IFL; blue bar, SE = 0.10), non-return rate at 56 d (NRR56; red bar, SE = 0.001), and conception rate at first insemination (CR; green bar, SE = 0.001).

Regarding the year of insemination, results from the linear mixed models indicated that IFL, NRR56, and CR deteriorated. In particular, IFL changed from 21.23 d (SE = 0.19) in 1994 to 24.28 d (SE = 0.15) in 2020. Similarly, in the same time span, NRR56 decreased from 0.88 (SE = 0.001) to 0.78 (SE = 0.001), and CR from 0.68 (SE = 0.001) to 0.63 (SE = 0.001), suggesting that, although EBV for cow fertility, as aggregate index, is included in the national Holstein selection index since 2009, little benefit has been achieved for heifer fertility. Although within trait genetic correlations between heifer and adult cows fertility attributes have never been estimated in Italian Holstein so far, Tiezzi et al. (2012) demonstrated that the genetic correlations between fertility traits measured in Brown Swiss lactating cows and maiden heifers was moderate to weak (0.348 for CR and 0.349 for NRR56 between heifers and first-parity cows, and 0.637 and 0.636 for CR and NRR56 between heifers and second-parity cows). Therefore, the correlated response to selection may prove insufficient. This evidence justifies the inclusion and subsequently the direct selection also for heifer fertility traits in the national breeding program, as currently done in other countries (Miglior et al., 2017). Indeed, heifer fertility traits have gained more relevance into selection indices worldwide (Miglior et al., 2017) and, for example, since 2014 heifer CR has been included in the US selection indices (Cole and VanRaden, 2018).

The present study supports that, as heifer fertility traits are available early in life, their inclusion into the genetic evaluation would be beneficial to improve the overall fertility of the dairy herd. Moreover, new available information on pregnancy diagnoses allows new traits to be collected early in life, such as IFL. The aggregate heifer fertility index calculated in the present study can be included in the aggregate index of cow fertility, which is already published within the national genetic and genomic evaluation and has been included in the Italian Holstein breeding objective since 2009. Moreover, results of the present study can be useful to provide farmers with more detailed information on foreign AI bulls without daughters in Italy but available for the national market thanks to the participation in Interbull MACE for the trait dairy heifers' ability to conceive (T1 trait) as well as due to genomic evaluation.

REFERENCES

Berry, D.P. 2015. Breeding the cow of the future: what do we need? Anim. Prod. Sci. 55:823-837. https://doi.org/10.1071/AN14835

Berry, D.P., E. Wall, and J.E. Pryce. 2014. Genetics and genomics of reproductive performance in dairy and beef cattle. Animal 8:105-121. https://doi.org/10.1017/S1751731114000743

Biffani, S., A.B. Samoré, F. Canavesi, and M. Marusi. 2003. Data quality assessment and preliminary investigations on fertility in the Italian Holstein Friesian. Interbull Bullettin 30:89-95.

Brito, L.F., N. Bedere, F. Douhard, H.R. Oliveira, M. Arnal, F. Peñagaricano, A.P. Schincklel, C.F. Baes, F. Miglior. 2021. Review: Genetic selection of high-yielding dairy cattle toward sustainable farming systems in a rapidly changing world. Animal 15:100292. https://doi.org/10.1016/j.animal.2021.100292

Cole, J.B., and P.M. VanRaden. 2018. *Symposium review*: Possibilities in an age of genomics: The future of selection indices. J. Dairy Sci. 101:3686-3701. https://doi.org/10.3168/jds.2017-13335

Dekkers, J.C.M., and J.P. Gibson. 1998. Applying Breeding Objectives to Dairy Cattle Improvement. J. Dairy Sci. 81(suppl. 2):19-35. https://doi.org/10.3168/jds.S0022-0302(98)70151-1

Duplessis, M., R.I. Cue, D.E. Santschi, D.M. Lefebvre, and R. Lacroix. 2015. Weight, height, and relative-reliability indicators as a management tool for reducing age at first breeding and calving of dairy heifers. J. Dairy Sci. 98:2063-2073. http://dx.doi.org/10.3168/jds.2014-8279

Fleming, A., C.F. Baes, A.A.A. Martin, T.C.S. Chud, F. Malchiodi, L.F. Brito, and F. Miglior. 2019. *Symposium review*: The choice and collection of new relevant phenotypes for fertility selection. J. Dairy Sci. 102:3722-3734. https://doi.org/10.3168/jds.2018-15470

Fricke, P. M. 2002. Scanning the future - Ultrasonography as a reproductive management tool for dairy cattle. J. Dairy Sci. 85:1918-1926. https://doi.org/10.3168/jds.S0022-0302(02)74268-9

Gilmour, A. R. 2020. Echidna Mixed Model Software. Accessed Oct. 25, 2022 https://www.echidnamms.org/wp-content/uploads/2020/10/Echidna-Mixed-Model-Software.pdf

Hare, E., H.D. Norman, and J.R. Wright. 2006. Trends in calving ages and calving intervals for dairy cattle breeds in the United States. J. Dairy Sci. 89:365-370. https://doi.org/10.3168/jds.S0022-0302(06)72102-6

Hutchison, J.L., P.M. VanRaden, D.J. Null, J.B. Cole, and D.M. Bickhart. 2017. Genomic evaluation of age at first calving. J. Dairy Sci. 100:6853-6861. https://doi.org/10.3168/jds.2016-12060

Interbull, 2022. National genetic evaluation forms provided by countries. Accessed on May 5th, 2022. https://interbull.org/ib/geforms

Jamrozik, J., J. Fatehi, G.J. Kistemaker, and L.R. Schaeffer. 2005. Estimates of Genetic Parameters for Canadian Holstein Female Reproduction Traits. J. Dairy Sci. 88:2199-2208. https://doi.org/10.3168/jds.s0022-0302(05)72895-2

Liu, Z., J. Jaitner, F. Reinhardt, E. Pasman, S. Rensing, and R. Reents. 2008. Genetic Evaluation of Fertility Traits of Dairy Cattle Using a Multiple-Trait Animal Model. J. Dairy Sci. 91:4333-4343. https://doi.org/10.3168/jds.2008-1029

Malchiodi, F., A. Koeck, S. Mason, A.M. Christen, D.F. Kelton, F.S. Schenkel, and F. Miglior. 2017. Genetic parameters for hoof health traits estimated with linear and threshold models using alternative cohorts. J. Dairy Sci. 100:2828-2836. https://doi.org/10.3168/jds.2016-11558

Miglior, F., A. Fleming, F. Malchiodi, L.F. Brito, P. Martin, P., and C. F. Baes. 2017. A 100-Year Review: Identification and genetic selection of economically important traits in dairy cattle. J. Dairy Sci. 100:10251-10271. http://dx.doi.org/10.3168/jds.2017-12968

Misztal, I. 2008. Reliable computing in estimation of variance components. J. Anim. Breed. Genet. 125:363-370. https://doi: 10.1111/j.1439-0388.2008.00774.x

Muir, B.L., J. Fatehi, and L.R. Schaeffer. 2004. Genetic Relationship between Persistency and Reproductive Performance in First-Lactation Canadian Holstein. J. Dairy Sci. 87:3029-3037. https://doi.org/10.3168/jds.S0022-0302(04)73435-9

Schneeberger, M., Barwick, S.A., Crow, G.H., and K. Hammond. 1992. Economic indices using breeding values predicted by BLUP. J. Anim. Breed. Genet. 109:180-187. https://doi.org/10.1111/j.1439-0388.1992.tb00395.x

Tiezzi, F., C. Maltecca, A. Cecchinato, M. Penasa, and G. Bittante. 2012. Genetic parameters for fertility of dairy heifers and cows at different parities and relationships with production traits in first lactation. J. Dairy Sci. 95:7355-7362. https://doi.org/10.3168/jds.2012-5775

VanRaden, P.M., A.H. Sanders, M.E. Tooker, R.H. Miller, H.D. Norman, M.T. Kuhn, and G.R. Wiggans. 2004. Development of a National Genetic Evaluation for Cow Fertility. J. Dairy Sci. 87:2285-2292. https://doi.org/10.3168/jds.S0022-0302(04)70049-1

Wathes, D. C., G.E. Pollott, K.F. Johnson, H. Richardson, and J.S. Cooke. 2014. Heifer fertility and carry over consequences for life time production in dairy and beef cattle. Animal 8(suppl. 1):91-104. https://doi.org/10.1017/S1751731114000755

Wall, E., S. Brotherstone, J.A. Woolliams, G. Banos, and M.P. Coffey. 2003. Genetic evaluation of fertility using direct and correlated traits. J. Dairy Sci. 86:4093-4102. https://doi: 10.3168/jds.S0022-0302(03)74023-5

Wiggans, G. R., and R.C. Goodling. 2005. Accounting for pregnancy diagnosis in predicting days open. J. Dairy Sci. 88:1873-1877. https://doi.org/10.3168/jds.S0022-0302(05)72862-9

NOTES

V. Ferrari https://orcid.org/0000-0002-7774-4388

G. Visentin https://orcid.org/0000-0003-0869-5516

J. B. C. H. M. van Kaam https://orcid.org/0000-0002-2592-2461

M. Penasa https://orcid.org/0000-0001-9984-8738

M. Marusi https://orcid.org/0000-0002-2359-3633

R. Finocchiaro https://orcid.org/0000-0002-9058-9992

M. Cassandro https://orcid.org/0000-0002-8709-2870

This study was supported by “Latteco2 project, sottomisura 10.2 of the National Rural Development Program (PSRN) - Biodiversity 2020–2023” [Ministero delle politiche agricole, alimentari e forestali (MIPAAF). D.M. no. 465907 del 24/09/2021, project unique code J12C21004080005], National Breeders Association of Italian Holstein, Brown, and Jersey (ANAFIBJ, Cremona, Italy).

Because no live human or animal subjects were used for this study, no IRB or IACUC approval was required.

The authors have not stated any conflicts of interest.

Chapter II

Genetic and genomic evaluation of age at first calving in Italian Holsteins

V. Ferrari,[1,2,*] F. Galluzzo,[1,3*] J.B.C.H.M. van Kaam,[1] M. Penasa,[2] M. Marusi,[1] R. Finocchiaro,[1] G. Visentin,[3] and M. Cassandro[1,2]

[1]Associazione Nazionale Allevatori della Razza Frisona, Bruna e Jersey Italiana, Via Bergamo 292, 26100 Cremona (CR), Italy

[2]Department of Agronomy, Food, Natural resources, Animals and Environment, University of Padova, Viale dell'Università 16, 35020 Legnaro (PD), Italy

[3]Department of Veterinary Medical Sciences, Alma Mater Studiorum - University of Bologna, Via Tolara di Sopra 50, 40064 Ozzano dell'Emilia (BO), Italy

*These authors contributed equally to this work.

Accepted for publication in *Journal of Dairy Science*

INTERPRETIVE SUMMARY

Genetic and genomic evaluation of age at first calving in Italian Holsteins. Raising replacements heifers is a significant cost for the dairy herd and farmers are interested in shortening the non-productive period. Breeding strategies could be adopted to optimize age at first calving (AFC). In this study genetic and genomic analysis of AFC in the Italian Holstein population was performed and the effects of AFC on productive and reproductive traits were evaluated. Although AFC is lowly heritable, it showed exploitable additive genetic variation for selective breeding. Results supports the inclusion of AFC in an aggregate selection index to help dairy farmers to make proper breeding decisions.

ABSTRACT

Age at first calving (AFC) represents the non-productive period of around 2 yr in Holstein cows and thus it has a relevant impact on the cost of rearing replacements in the dairy herd. In the present study we aimed at evaluating genetic and genomic aspects of AFC in the Italian Holstein population. Data of 4,206,218 heifers with first calving between 1996 and 2020 were used. Age at first calving averaged 26.09 ± 3.07 mo and decreased across years. Heritability was estimated using a linear animal model which included the fixed effects of herd-year-season of birth and classes of gestation length, and the random animal additive genetic effect fitted to a pedigree-based relationship matrix. Estimated breeding values (EBV) and genomically enhanced breeding values (GEBV) were obtained, and they were standardized to mean 100 and standard deviation 5, where animals above the mean are those contributing to reduce AFC. Heritability estimates of AFC ranged from 0.031 to 0.045. The trend of sires' GEBV was favorable and indicated a reduced AFC across years. Approximate genetic correlations between GEBV of AFC and GEBV of other economically important traits were calculated on a subset of genotyped females born after 2015. Moderate favorable associations of AFC with production traits (0.39 to 0.51), udder depth (0.40), interval from first to last insemination in heifer (-0.43), and longevity (0.34) were assessed. Overall, the greatest lifetime productive performances and most favorable days open in first lactation were observed when heifers calved at 22-23 mo. On the other hand, progeny of sires with GEBV of AFC above the mean yielded more milk, fat, and protein in first lactation, and had shorter days open than progeny of sires with GEBV of AFC below the mean. Results suggested that breeding strategies to improve AFC should be pursued, also considering genetic correlations between AFC and traits which are already part of the Italian Holstein breeding objective. The inclusion of AFC in an aggregate index is expected to contribute to enhance farm income.

Keywords: dairy cattle, heifer, heritability, longevity, profit

INTRODUCTION

Age at first calving (**AFC**) is the period from birth to the onset of the productive career of the dairy cow. The AFC is a relevant contributor to the costs of rearing replacement heifers on the farm and, thus, to total milk production costs. Interest exists to shorten AFC and therefore breed heifers with proper growth rate at an earlier age. In Italy, the average daily rearing cost of a heifer has been estimated to increase from €3.20 to €3.30 moving from 24 to 30 mo of AFC, respectively (Paganini, 2023). In UK an average daily cost of £2.31 for rearing a heifer to calving has been reported (Boulton et al., 2017), and in US this cost has been estimated to vary from \$2.46 to \$3.55 (Tranel, 2019; Cabrera, 2023). Differences in costs, especially those related to more recent estimates, are likely due to the strong increase of feed costs. Hence, the direct economic advantage of reducing AFC is immediately derived. The effects of AFC reduction on lifetime performances have been extensively evaluated. Eastham et al. (2018) observed that heifers with AFC to 22-23 mo had, on average, the greatest lifetime milk yield, the lowest SCC, and the shortest calving interval. Accordingly, an average AFC between 22 and 24 mo concurrently with an average daily gain between 0.7 and 0.8 kg have been recommended to achieve a first-calving size adequate to maximise milk production and reduce rearing costs (Abeni et al., 2000; Tozer and Heinrichs, 2001). Furthermore, AFC to 22-23 mo has resulted in shorter days open and calving interval, and greater conception rate in first-lactation cows (Ettema and Santos, 2004; Krpálková et al., 2014). Nevertheless, AFC < 22 mo has been associated to increased calving difficulty, stillborn, perinatal mortality, and risk of dystocia (Ettema and Santos, 2004; Berry and Cromie, 2009).

The AFC depends on a plethora of factors, including management, genetics (Mourits et al., 1999), nutrition, and growth rate (Wathes et al., 2008). Given that AFC depends on both body weight and size, AFC is the result of a proper growth rate within an optimal time interval (Le Cozler et al., 2008). Fertility is characterized by lower heritability compared to production or type traits, however exploitable additive genetic variation has been reported for AFC, along with its genetic correlations with other non-production traits such as calving interval, calving ease, and conception rate (Berry et al., 2014; Heise et al., 2018; Brzáková et al., 2020). Although body weight is more representative of heifer growth rate, its monitoring on a routine cost-effective basis is hardly feasible, whereas information on AFC can be quickly retrieved from herdbook information or easily recorded on-farm. In Italian Holsteins, AFC has been stored in national database since the early 1990s, but information on genetic variation of this trait and its potential use for breeding purposes has not been explored so far. Genetic and genomic proof for AFC could be exploited to support farmers to maximize the profit, which is achievable by reducing heifer rearing costs and maximizing yield output. Hence, the aim of the present study was to investigate genetic and genomic aspects of AFC in Italian Holstein population.

MATERIALS AND METHODS

Data

This study did not involve animals and thus prior ethical approval was deemed not to be required. Data stored in the national database of the Italian Holstein, Brown Swiss and Jersey Association (ANAFIBJ, Cremona, Italy), including AFC of 5,650,513 Holsteins born since 1993 and with first calving date between 1996 and 2020, were used. Animals with unknown parents, AFC outside the range 18 to 36 mo, and gestation length outside the range 240 to 305 d were discarded from the dataset. Contemporary groups were defined as cows born in the same herd-year-season (**HYS**), and HYS with less than 10 individuals were removed. Two seasons of birth were defined, the first from October to April and the second from May to September. After all edits, 4,206,218 Holsteins distributed across 11,528 herds remained for subsequent statistical analyses.

Statistical Analyses

Fixed effects. Numerous fixed effects were tested (herd-year-season of birth and calving; herd-year of birth and calving; year-month of first insemination; month of first insemination; gestation length) and those to be included in the analysis of AFC were identified through a forward stepwise approach based on Akaike Information Criterion, root mean square error, and coefficient of determination in R software version 4.0.3 (R Core Team, 2020). The final model was:

$$y_{ijk} = \mu + HYS_i + GL_j + e_{ijk},$$

where y_{ijk} is AFC; μ is the overall intercept of the model; HYS_i is the fixed effect of the ith herd-year-season of birth (i = 168,135 classes); GL_j is the fixed effect of the jth gestation length class (j = 4 classes: 240 to 260, 261 to 280, 281 to 290, and 291 to 305 d); and e_{ijk} is the random error.

Variance components estimation. The pedigree of cows with phenotypic information was traced back to at least 4 generations. Unknown ancestors were assigned to phantom parent groups based on animal origin and year of birth (296 genetic groups). Variance components were estimated with the Gibbs sampler THRGIBBS1F90 (Misztal et al., 2002; Misztal, 2008) with 160,000 iterations, a burn-in of 10,000, and a thinning rate of 10. The post-Gibbs analysis was carried out with the software POSTGIBBSF90 (Misztal et al., 2002) using the last 150,000 samples. The animal model used to estimate variance components accounted for the fixed effects previously identified; the animal additive genetic effect, fitted to the pedigree-based relationship matrix (**A**), was included as a random term. To reduce computational time, variance components of AFC were assessed in 3 different subsets each including data of animals from 300 herds randomly selected from the whole edited data. Estimated variance components of the 3 subsets were then averaged and the heritability (h^2) was calculated as:

$$h^2 = \frac{\sigma_a^2}{\sigma_a^2 + \sigma_e^2},$$

where σ_a^2 is the additive genetic variance and σ_e^2 is the residual variance.

Breeding value estimation. Estimated breeding values (**EBV**) were obtained with MiX99 software (Lidauer et al., 2019) using average variance components estimated in the previous step and the same aforementioned animal model, and were standardized to mean 100 and standard deviation 5 as undertaken for EBV of other functional traits published by ANAFIBJ. An EBV > 100 identifies animals whose progeny are characterized by shorter AFC.

Genomic prediction and validation. Genomic validation was performed according to Finocchiaro et al. (2012). Estimated deregressed proofs (**EDP**) were obtained for 2 datasets: the full data set with all recorded phenotypes in routine evaluation run (December 2021) and the reduced data set (cut-off set to the year 2017). SNP genotypes were collected using different DNA chip and missing SNP genotypes were imputed with PedImpute software (Nicolazzi et al., 2013) to a standard SNP panel of 69,084 markers. The EDP of the reduced dataset were the response variables to perform genomic evaluation and simultaneously estimate the effects of all SNP (n = 69,084) using the SNPblup model. The SNP effects were used to compute the direct genomic values (**DGV**) of 6,731 genotyped sires with daughters in the full dataset but without daughters in the reduced dataset (4,811 sires, validation set). Finally, current EDP were linearly regressed on DGV of validation sires and the coefficient of determination of this linear regression was the reliability of the DGV of validation sires (Galluzzo et al., 2022). Then, genomically enhanced breeding values (**GEBV**) were estimated by blending DGV and EBV of the full run according to the following formula:

$$GEBV = \frac{EDC_C * EBV + EDC_G * DGV}{EDC_C + EDC_G},$$

where EDC_C is the conventional effective daughter contribution (Fikse and Banos, 2001) and EDC_G is the genomic effective daughter contribution (Patry and Ducrocq, 2011).

Approximate genetic correlations. Approximate genetic correlations of AFC with other production and functional traits evaluated by ANAFIBJ were estimated using Pearson's correlations on a subset of GEBV of genotyped females born after 2015:

$$r_{AFC,y} = \frac{cov_{AFC,y}}{\sqrt{var_{AFC} var_y}},$$

where $cov_{AFC,y}$ is the covariance between GEBV of AFC and trait y, and var_{AFC} and var_y are the variances of GEBV of AFC and trait y, respectively. Proof for the traits investigated are expressed on a different scale based on conventional ANAFIBJ genetic and genomic evaluation. Prior to any standardization, proofs are subtracted to the average of base population, represented by individuals born in the previous 6 to 8 yr, inclusive. Production traits (i.e., milk, fat, and protein yield) are expressed in kg/lactation and higher values are desired. Linear traits proofs, including those considered in the present study (i.e., locomotion and udder depth), are standardized to mean 0 and SD 1, with higher proofs desired. Similar to production traits, fertility proofs are expressed on their original scale. In particular, proofs for heifer and cow interval from first to last insemination are expressed in days, and proofs for heifer age at first insemination are expressed in months; in all instances, smaller values are desired. The GEBV of heifer and cow non-return rate at 56 d from the first insemination (binary traits) are unitless and higher values are desired. Finally, GEBV of the other functional traits, including dystocia, body condition score, somatic cell score, mastitis resistance, longevity (defined, in the present paper, as productive longevity, i.e., from first calving to culling), and direct and maternal gestation length are standardized to mean 100 and SD 5; in all instances, proofs greater than 100 indicate individuals whose daughters will perform better than the average, except for direct and maternal gestation length for which intermediate proofs are the optimum.

Effect of cow AFC and sire EBV for AFC on production and functional traits. The effect of AFC on first-lactation milk, fat, and protein yield, lifetime milk, fat, and protein yield, days open in first lactation, and longevity was quantified through analysis of variance based on the following linear mixed model:

$$y_{ijk} = \mu + \mathrm{AFC}_i + \mathrm{HYS}_j + e_{ijk},$$

where y_{ijk} is the studied trait; μ is the overall intercept of the model; AFC_i is the fixed effect of the ith AFC class of the cow (i = 16 classes, with the first being a class from 18 to 21 mo, followed by 15 monthly classes); HYS_j is the random effect of the jth herd-year-season of birth group (j = 17,989 classes); and e_{ijk} is the random residual term. The best AFC class was the one that maximized first-lactation milk, fat, and protein yield, lifetime milk, fat, and protein yield, and longevity, and that minimized days open.

Subsequently, a second analysis of variance was performed to investigate the effect of sire EBV for AFC on daughters' performance, namely first-lactation milk, fat, and protein yield, lifetime milk, fat, and protein yield, days open in first lactation, and longevity. For this purpose, only sires with an officially releasable EBV for the national genetic and genomic evaluation run were considered (i.e., sires with at least 30 daughters in at least 30 herds, and with EBV reliability ≥ 50%). Sire EBV for AFC were grouped in 7 classes: **-3**: $83 \leq x \leq 87$; **-2**: $88 \leq x \leq 92$; **-1**: $93 \leq x \leq 97$; **0**: $98 \leq x \leq 102$; **1**: $103 \leq x \leq 107$; **2**: $108 \leq x \leq 112$; **3**: $113 \leq x \leq 117$, where x is the standardized sire EBV for AFC. Values above and below 0 identify sires that transmit lower and higher AFC to their progeny, respectively. The linear mixed model was as follows:

$$y_{ijk} = \mu + EBV\_AFC_i + HYS_j + e_{ijk},$$

where y_{ijk} is the studied trait; μ is the overall intercept of the model; EBV_AFC_i is the fixed effect of the ith class of sire EBV for AFC ($i = 7$ classes); HYS_j is the random effect of the jth herd-year-season of birth group ($j = 17{,}989$ classes); and e_{ijk} is the random residual term. The analyses were performed using the software Echidna (Gilmour, 2018).

RESULTS AND DISCUSSION

Age at first calving decreased by 2 mo from 1996 to 2020, from 28.1 ± 2.9 to 26.1 ± 3.1 mo. The frequency (%) of heifers across monthly classes of AFC in 1996 and 2020 are presented in Table 1. When considering heifers calving in 1996, the highest frequency was observed for AFC between 26 and 27 mo, whereas in 2020 the highest frequency was observed between 24 and 25 mo. This trend followed the reduction of age at first insemination reported by Ferrari et al. (2023). Also, a similar trend has been reported by Hutchinson et al. (2017), suggesting that both higher selection intensity for productive and reproductive traits, and improved management practices have indirectly enhanced AFC, allowing farmers to breed animals earlier.

Heritability and Approximate Genetic Correlations

Posterior mean of heritability of AFC, its lower and upper bounds of the 95% highest posterior density (**HPD95**), and the coefficient of additive genetic variation in the three subsets are reported in Table 2. Posterior mean of heritability ranged from 0.031 (subset 3) to 0.045 (subset 2), with the lowest estimate included in the HPD95 of subset 3 (0.020) and the greatest in the HPD95 of subset 2 (0.056). Coefficient of additive genetic variation ranged from 1.30% (subset 3) to 1.56% (subset 2), suggesting a genetic potential to reduce AFC by 10 to 12 d relative to the population mean. Heritability estimates from the current study are in agreement with those of Heise et al. (2018) and Brzáková et al. (2019) in Holstein populations (0.10 and 0.031, respectively). In beef breeds, heritability of AFC has been estimated to be 0.18 and 0.23 in Aberdeen Angus and Charolais, respectively (Brzáková et al., 2020), i.e., higher than that reported for dairy breeds.

Table 1. Frequency (%) of heifers across age at first calving (AFC) in 1996 and 2020

AFC, mo	1996	2020
18-20	0.09	1.08
21	0.30	3.02
22	1.04	8.05
23	3.49	14.30
24	7.53	15.19
25	12.24	14.39
26	13.86	11.19
27	14.35	9.07
28	12.12	6.61
29	9.87	5.02
30	7.35	3.57
31	5.74	2.80
32	4.16	2.00
33-36	7.86	3.71

Table 2. Posterior mean of heritability (h^2) of age at first calving, lower and upper bounds of the 95% highest posterior density (HPD95), and coefficient of additive genetic variation (CV) estimated in the 3 randomly selected subsets

Item	Herds, n	Animals, n	h^2	HPD95	CV (%)
Subset 1	300	101,110	0.037	0.028; 0.046	1.409
Subset 2	300	101,659	0.045	0.034; 0.056	1.563
Subset 3	300	103,000	0.031	0.020; 0.042	1.298

Approximate genetic correlations between cow GEBV of AFC and cow GEBV of production and functional traits are presented in Table 3. The AFC was positively and moderately correlated with production traits (from 0.39 with milk yield to 0.51 with protein yield), which is favorable considering that high GEBV of AFC are desirable. Similarly, Hutchinson et al. (2017) assessed significant positive and favorable genetic correlations between AFC and milk, fat, and protein yield. Weak and moderate approximate genetic correlations were estimated between AFC and locomotion (0.13), and AFC and udder depth (0.40; Table 3), respectively, suggesting that, on average, AFC has an almost negligible impact on locomotion and a more pronounced impact on udder morphology. The approximate genetic correlations between GEBV of AFC and GEBV of fertility traits were favorable, being -0.17 between AFC and cow interval from first to last insemination, -0.43 between AFC and heifer interval from first to last insemination, and -0.73 between AFC and age at first insemination. These associations agree, in both direction and magnitude, with those reported by Brzáková et al. (2019) who assessed the genetic correlations between AFC and cow and heifer interval from first to last insemination in Czech Holsteins. The correlations of AFC with non-return rate at 56 d in heifers (0.23; Table 3) and age at first service in heifers corroborate with Muir et al. (2004). Because interest exists for farmers to shorten to some extent AFC, the aforementioned correlations indicate that genetic selection for AFC can actually improve interval fertility traits, thus resulting in favorable indirect response to selection. Therefore, proper emphasis should be placed to AFC and fertility in the national total merit selection index in order to achieve desired gains for all these traits, which could be achievable given the non-unity genetic correlations among them. Approximate genetic correlations between GEBV of AFC and GEBV of functional traits other than fertility were favorable and moderate to weak, with values from 0.14 (AFC and mastitis resistance) to 0.34 (AFC and longevity), except for the correlation between AFC and direct gestation length and BCS which was unfavorable, despite weak (-0.26 and -0.07, respectively). These correlations agree with those retrieved from the literature (Berry and Cromie, 2009; Zavadilová and Štípková, 2013; Eastham et al., 2018), which reported a favorable association between genetic selection to reduce AFC and animal functionality. These findings support the common choice of farmers to anticipate AFC, as this can lead to lower probability of subclinical mastitis after calving and greater survival to higher parities (Eastham et al., 2018). Shortening AFC by genetic selection could slowly worsen animal body condition, resulting in a non-complete maturation at first calving. To handle this point, at farm level, the time at first insemination should be carefully evaluated along with growth rate and maturation state.

Maximising Phenotypic Performances with AFC

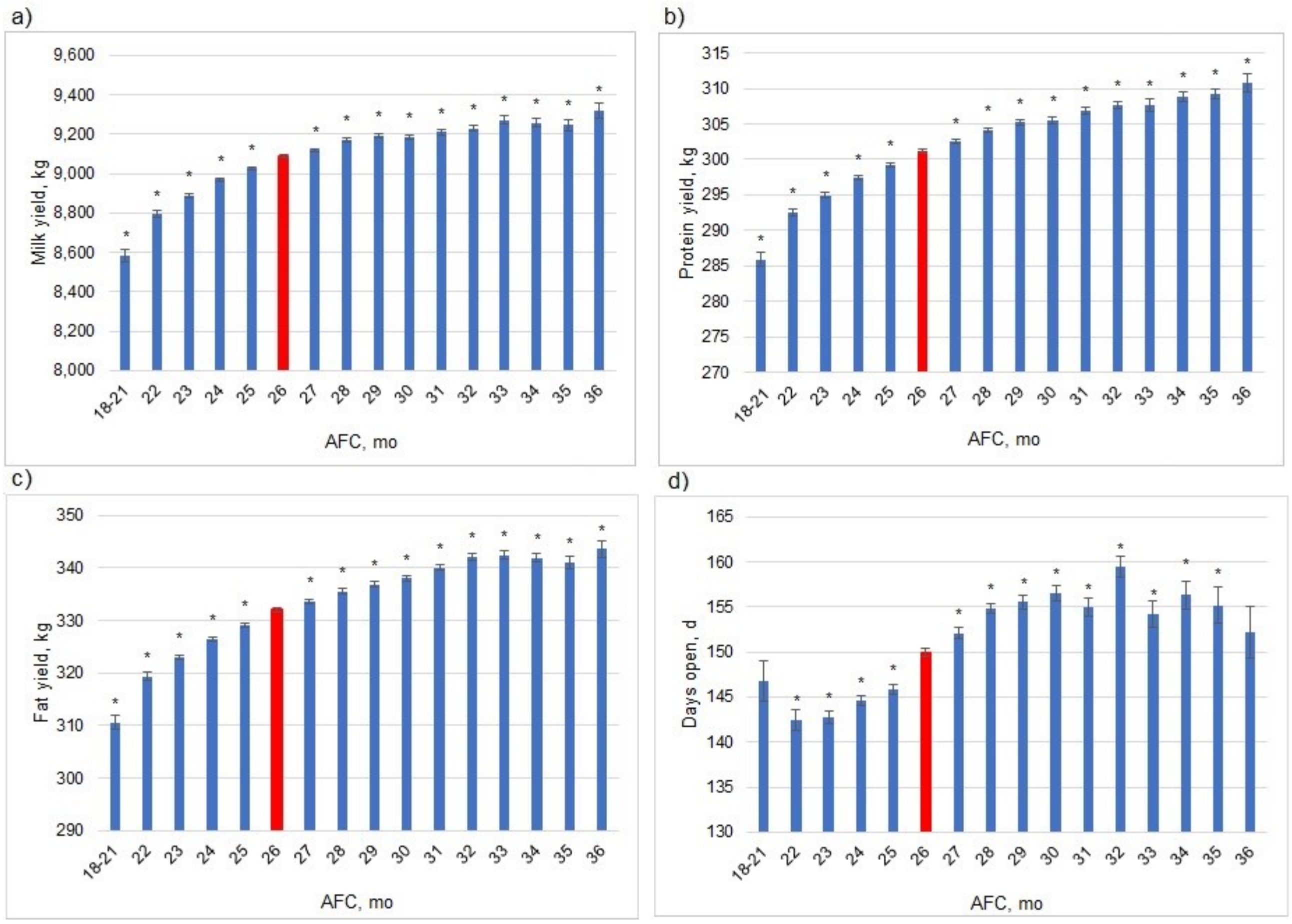
Figures 1 and 2 depict the least squares means of first-lactation and lifetime milk, fat, and protein yield, days open in first lactation, and longevity across monthly classes of AFC. Cows with lower AFC produced less milk, fat, and protein in first lactation compared to animals calving at older age, in agreement with previous studies (Berry and Cromie, 2009; Mohd Nor et al., 2013; Eastham et al., 2018). Cows calving at 36 mo yielded 735 kg more milk, in first lactation, compared to cows whose AFC was from 18 to 21 mo. The same trend was observed for first-lactation protein and fat yield, which increased by approximately 25 kg and 33 kg, respectively, moving from 18-21 to 36 mo of AFC (Figure 1). This trend is likely due to the fact that heifers calving at an earlier age might not have reached a proper body size and maturity, and thus an important amount of the ingested energy during first lactation is partitioned into growth rather than into milk production (Sejersen et al., 2000; Hutchinson et al., 2017). Days open in first lactation increased by approximately 13 d moving from

22 to 32-35 mo of AFC (Figure 1), in accordance with the trends reported by Ettema and Santos (2004) and Zavadilová and Štípková (2013). Moreover, Eastham et al. (2018) reported that the shortest calving interval between first and second lactation was observed when AFC occurred at 23 mo. Days open reduction reflects pregnancy rate improvement.

Table 3. Approximate genetic correlations of cow genomically enhanced breeding values for age at first calving with those for production and functional traits

Trait	Age at first calving	Interpretation
Milk yield	0.39	Favorable
Fat yield	0.40	Favorable
Protein yield	0.51	Favorable
Locomotion	0.13	Favorable
Udder depth	0.40	Favorable
Non-return rate at 56 d (cow)	0.18	Favorable
Non-return rate at 56 d (heifer)	0.23	Favorable
Interval from first to last insemination (cow)	-0.17	Favorable
Interval from first to last insemination (heifer)	-0.43	Favorable
Age at first service (heifer)	-0.73	Favorable
Dystocia maternal	0.25	Favorable
Body condition score	-0.07	Unfavorable
Somatic cell score	0.20	Favorable
Mastitis resistance	0.14	Favorable
Longevity	0.34	Favorable
Direct gestation length	-0.26	Unfavorable
Maternal gestation length	0.30	Unfavorable

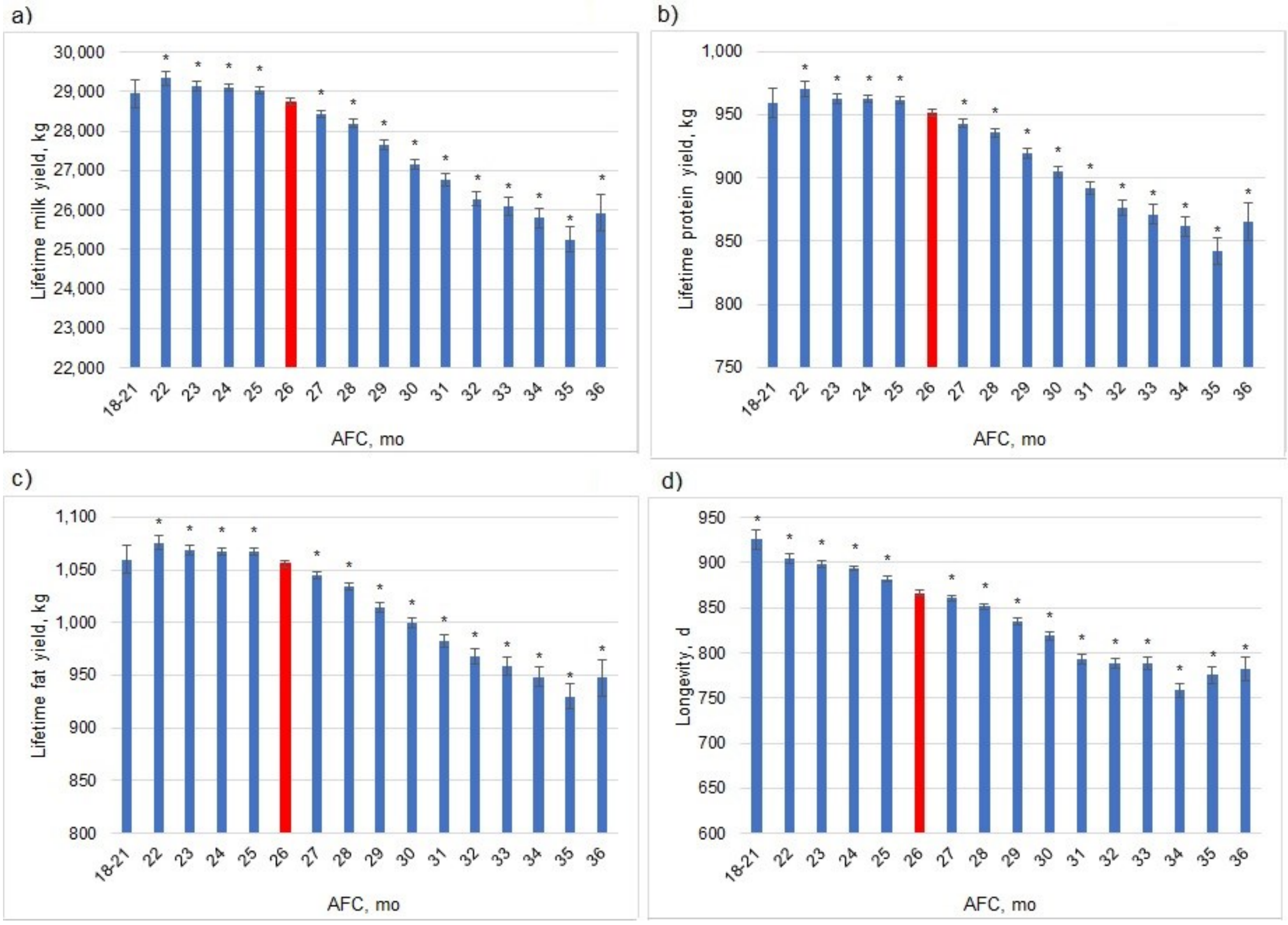
Figure 1. Least squares means and standard error of first-lactation a) milk yield, b) protein yield, c) fat yield, and d) days open for each class of age at first calving (AFC). Red bars represent the actual population mean class. Asterisks indicate the significance of the class with respect to the mean class.



However, when considering lifetime performances, milk, fat, and protein yields were greater in heifers that first calved at lower age (22 to 24 mo) than heifers that calved later (35 to 36 mo). In particular, lifetime milk yield, protein yield, and fat yield decreased by 3,400 kg, 105 kg, and 128 kg, respectively, when AFC moved from 22 to 36 mo (Figure 2). Accordingly, Froidmont et al. (2013) reported the greatest lifetime milk, fat, and protein yield for cows with AFC from 22 to 26 mo than cows with later AFC. This favorable association can be mainly attributed to the longer productive life of cows with earlier AFC, as suggested by longevity in Figure 2d. Indeed, longevity decreased by approximately 140 d moving from 18-21 to 36 mo of AFC, and this trend agrees with findings of

Hutchinson et al. (2017). Similarly, Berry and Cromie (2009) observed that animals that first calved at 24 mo have greater survival probability to higher lactations than animals calving at 36 mo.

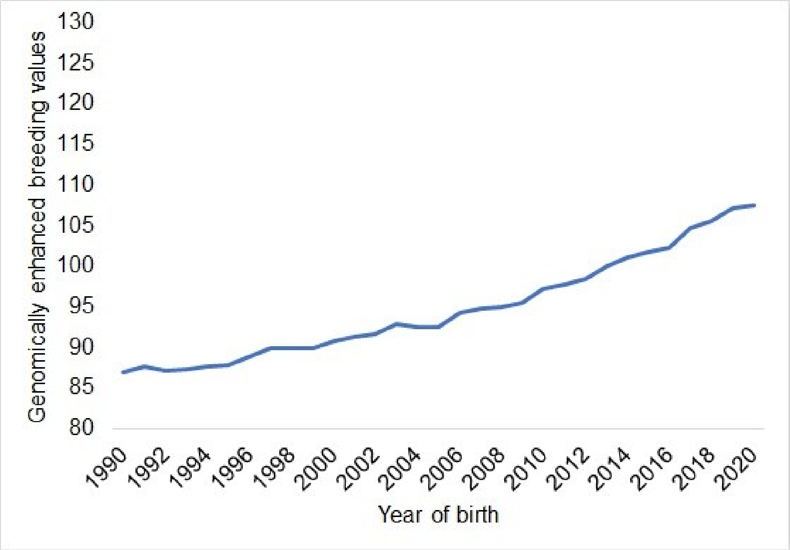
Figure 2. Least squares means and standard error of lifetime a) milk yield, b) protein yield, c) fat yield, and d) longevity for each class of age at first calving (AFC). Red bars represent the actual population mean class. Asterisks indicate the significance of the class with respect to the mean class.



Maximising Sires' Genetic Potential with AFC

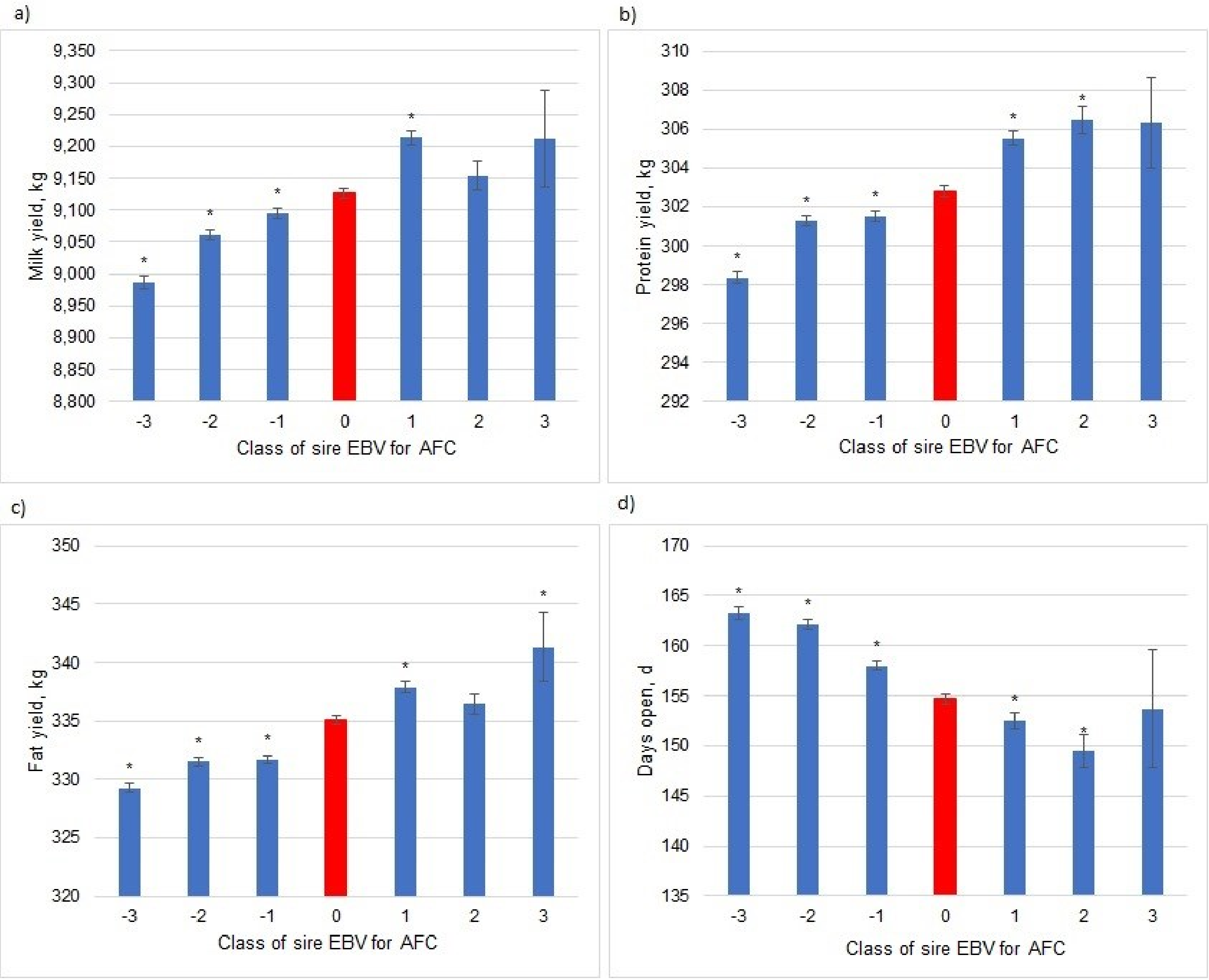
Figure 3 depicts the trend of sires' GEBV for AFC by year of birth. Overall, AFC improved over years, suggesting that indirect selection for this trait occurred, in accordance with results of Amimo et al. (2006) and Hutchinson et al. (2017).

Figure 3. Trend of average sires' genomically enhanced breeding values for age at first calving by year of birth.



Figures 4 and 5 show the trend of first-lactation and lifetime milk yield, fat yield, protein yield, longevity, and days open in first lactation across classes of sire EBV for AFC, where sires above 100 transmit lower AFC. Considering first-lactation milk, fat, and protein yield (Figure 4), sires in class -3 had the lowest production, whereas sires in classes 1 or higher had the highest milk, fat, and protein yield, except for class 2 for milk and fat yield, and class 3 for milk and protein yield which did not differ significantly from the mean. This corroborates with the favorable genetic correlation between production traits and EBV for AFC (Table 3). The trend of days open across classes of sire EBV for AFC depicted in Figure 4 supports the moderate genetic association between AFC and other fertility traits, as indicated by Berry et al. (2014). Indeed, daughters' fertility performances, in terms of days open in first lactation, deteriorates as bulls EBV for AFC decrease, with the exception of daughters of sires in class 3. Overall, it is worth noting that least squares means of production traits, days open in first lactation, and longevity in class 3 (Figures 4 and 5) should be considered with caution due to the low number of sires in this class, which resulted in larger standard errors compared to the other sires EBV classes.

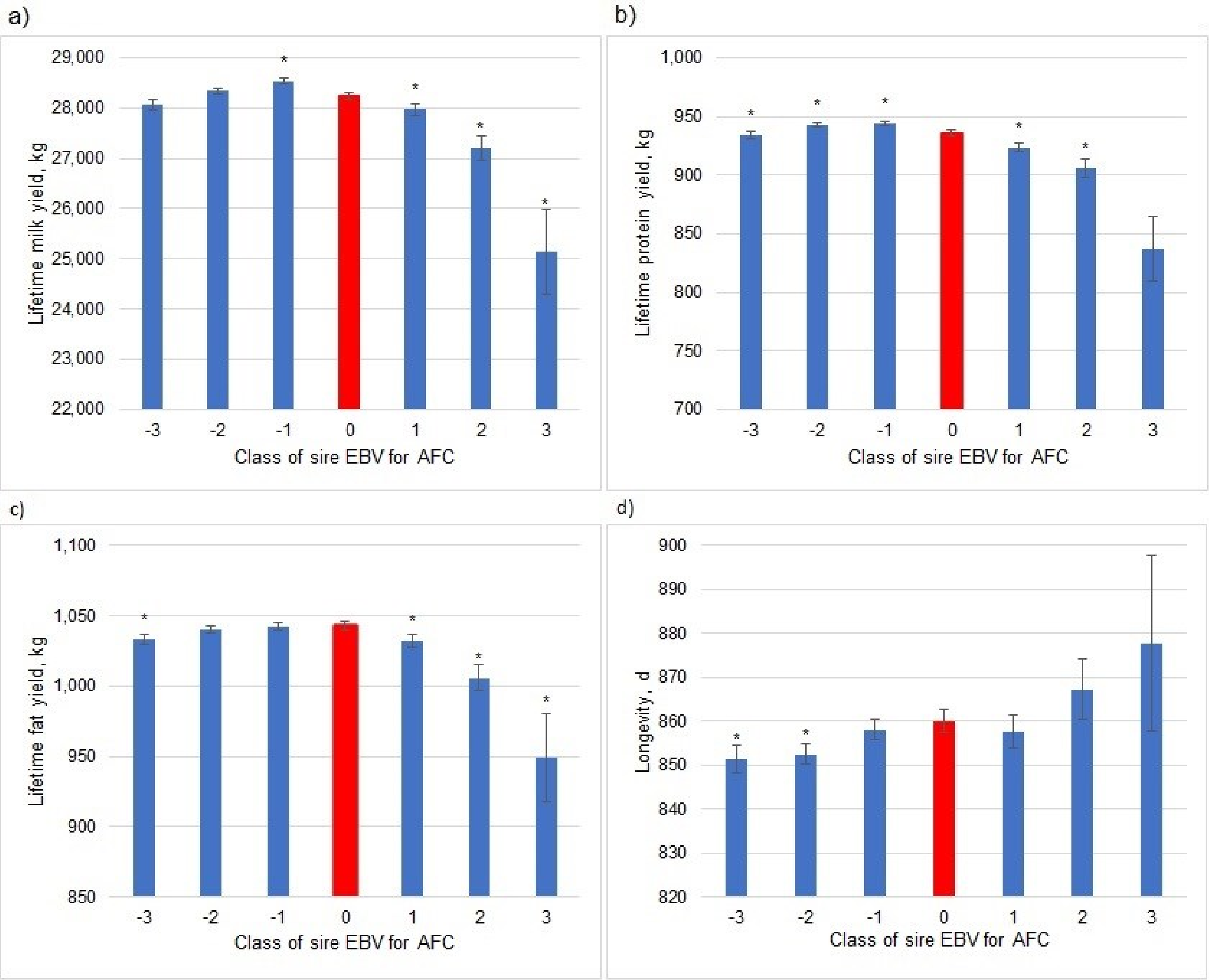
Figure 4. Least squares means and standard error of first-lactation a) milk yield, b) protein yield, c) fat yield, and d) days open across classes of sire EBV for age at first calving (AFC) standardised to mean 100 and SD 5. Classes are: **-3**: 83 ≤ x ≤ 87; **-2**: 88 ≤ x ≤ 92; **-1**: 93 ≤ x ≤ 97; **0**: 98 ≤ x ≤ 102; **1**: 103 ≤ x ≤ 107; **2**: 108 ≤ x ≤ 112; **3**: 113 ≤ x ≤ 117. Red bars represent the standardized population mean class. Asterisks indicate the significance of the class with respect to the mean class.



The pattern of lifetime milk, protein, and fat yield across sires EBV classes for AFC appeared opposite to the trend observed in first lactation (Figure 5). Nevertheless, the biological impact of the differences among least squares means of production traits across classes of sires EBV for AFC was negligible, again with the exception of the last EBV class, which however has to be considered with caution due to the large standard error. For example, by excluding such class, the differences between the greatest and the lowest least squares means for lifetime protein and fat yield was less than 40 kg.

Also, in terms of longevity (Figure 5d) the differences among least squares were small (less than 2 wk) and in many cases did not differ significantly from least squares means of the reference class of sires EBV for AFC.

Figure 5. Least squares means and standard error of lifetime a) milk yield, b) fat yield, c) protein yield, and d) longevity across classes of sire EBV for age at first calving (AFC) standardised to mean 0 and SD 1. Classes are: **-3**: 83 ≤ x ≤ 87; **-2**: 88 ≤ x ≤ 92; **-1**: 93 ≤ x ≤ 97; **0**: 98 ≤ x ≤ 102; **1**: 103 ≤ x ≤ 107; **2**: 108 ≤ x ≤ 112; **3**: 113 ≤ x ≤ 117. Red bars represent the standardized population mean class. Asterisks indicate the significance of the class with respect to the mean class.



CONCLUSIONS

Our results suggested that there is room to lower actual phenotypic mean of AFC in Italian Holstein population to maximise production performances, without negatively affecting fertility. Although relatively small, additive genetic variation exists for AFC, allowing the potential identification of elite sires for this trait. Any possible breeding strategies to improve AFC should be pursued, also considering the genetic correlations between AFC and the other traits included in the national breeding objective. Indeed, approximate genetic correlations of AFC with milk, fat, and protein yield, and fertility traits were favorable. Finally, sires transmitting shorter AFC transmit also improved daughters' production performances in first lactation, yet the impact on lifetime productive lifetime length is small.

ACKNOWLEDGMENTS

This study was supported by "Latteco2 project, sottomisura 10.2 of the PSRN-Biodiversity 2020–2023" (MIPAAF. D.M. no. 465907 del 24/09/2021, project unique code J12C21004080005). Moreover, this study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them. The authors have not stated any conflicts of interest.

REFERENCES

Abeni, F., L. Calamari, L. Stefanini, and G. Pirlo. 2000. Effects of daily gain in pre- and postpubertal replacement dairy heifers on body condition score, body size, metabolic profile, and future milk production. J. Dairy Sci. 83:1468–1478. https://doi.org/10.3168/jds.S0022-0302(00)75019-3

Amimo, J. O., R. O. Mosi, J. W. Wakhungu, T. K. Muasya, and B. O. Inyangala. 2006. Phenotypic and genetic parameters of reproductive traits for Ayrshire cattle on large-scale farms in Kenya. Livest. Res. Rural Dev. 18:147-151.

Berry, D. P., and A. R. Cromie. 2009 Associations between age at first calving and subsequent performance in Irish spring calving Holstein-Friesian dairy cows. Livest. Sci. 123:44-54. http://dx.doi.org/10.1016/j.livsci.2008.10.005

Berry, D. P., E. Wall, and J. E. Pryce. 2014. Genetics and genomics of reproductive performance in dairy and beef cattle. Animal 8:105–121. https://doi.org/10.1017/S1751731114000743

Boulton, A. C., J. Rushton, and D. C. Wathes. 2017. An empirical analysis of the cost of rearing dairy heifers from birth to first calving and the time taken to repay these costs. Animal 11:1372–1380. https://doi.org/10.1017/S1751731117000064

Brzáková, M., L. Zavadilová, J. Pribyl, P. Pešek, E. Kašná, and A. Kranjčevičová. 2019. Estimation of genetic parameters for female fertility traits in the Czech Holstein population. Czech J. Anim. Sci. 64:199-206. https://doi.org/10.17221/51/2018-CJAS

Brzáková, M., J. Čítek, A. Svitáková, Z. Veselá, and L. Vostrý. 2020. Genetic parameters for age at first calving and first calving interval of beef cattle. Animals 10:2122. https://doi.org/10.3390/ani10112122

Cabrera, V. E. 2023. Heifer Breakeven Tool. https://dairymgt.info/tools/heifer_breakeven/ Accessed on Jan. 9, 2023.

Eastham, N. T., A. Coates, P. Cripps, H. Richardson, R. Smith, and G. Oikonomou. 2018. Associations between age at first calving and subsequent lactation performance in UK Holstein and Holstein-Friesian dairy cows. PLoS ONE, 13:e0197764. https://doi.org/10.1371/journal.pone.0197764

Ettema, J. F., and J. E.P. Santos. 2004. Impact of age at calving on lactation, reproduction, health, and income in first-parity Holsteins on commercial farms. J. Dairy Sci. 87:2730-2742. https://doi.org/10.3168/jds.S0022-0302(04)73400-1

Ferrari, V., G. Visentin, J. B. C. H. M. van Kaam, M. Penasa, M. Marusi, R. Finocchiaro, M. Cassandro, 2023. Genetic and nongenetic variation of heifer fertility in Italian Holstein cattle. JDS Commun. 4:35-39. https://doi.org/10.3168/jdsc.2022-0270

Fikse, W. F., and G. Banos. 2001. Weighting factors of sire daughter information in international genetic evaluations. J. Dairy Sci. 84:1759-1767. https://doi.org/10.3168/jds.S0022-0302(01)74611-5

Finocchiaro, R., J. B. C. H. M. van Kaam, and S. Biffani. 2012. The genomic selection system in Italian Holstein. Page 130 in Book of Abstracts of the 63rd EAAP, Bratislava, Slovakia, 27-31 August.

Froidmont, E., P. Mayeres, P. Picron, A. Turlot, V. Planchon, and D. Stilmant. 2013. Association between age at first calving, year and season of first calving and milk production in Holstein cows. Animal 7:665-672. http://dx.doi.org/10.1017/S1751731112001577

Galluzzo, F., J. B. C. H. M. van Kaam, R. Finocchiaro, M. Marusi, S. Tsuruta, and M. Cassandro. 2022. Estimation of milkability breeding values and variance components for Italian Holstein. JDS Commun. 3:180–184. https://doi.org/10.3168/jdsc.2021-0167

Gilmour, A. R. 2018. Echidna mixed models software. Proceedings of the 11th World Congress on Genetics Applied to Livestock Production, Auckland (NZ), 7-11 February.

Heise, J., K. F. Stock, F. Reinhardt, N. T. Ha, and H. Simianer. 2018. Phenotypic and genetic relationships between age at first calving, its component traits, and survival of heifers up to second calving. J. Dairy Sci. 101:425–432. https://doi.org/10.3168/jds.2017-12957

Hutchison, J. L., P. M. VanRaden, D. J. Null, J. B. Cole, and D. M. Bickhart. 2017. Genomic evaluation of age at first calving. J. Dairy Sci. 100:6853–6861. https://doi.org/10.3168/jds.2016-12060

Krpálková, L., V. E. Cabrera, J. Kvapilík, J. Burdych, and P. Crump. 2014. Associations between age at first calving, rearing average daily weight gain, herd milk yield and dairy herd production, reproduction, and profitability. J. Dairy Sci. 97:6573–6582. https://doi.org/10.3168/jds.2013-7497

Le Cozler, Y., V. Lollivier, P. Lacasse, and C. Disenhaus. 2008. Rearing strategy and optimizing first-calving targets in dairy heifers: A review. Animal 2:1393-1404. https://doi.org/10.1017/s1751731108002498

Lidauer, M., K. Matilainen, E. Mäntysaari, T. Pitkänen, M. Taskinen, and I. Strandén. 2019. MiX99: Technical reference guide for MiX99 pre-processor. Release XI/2019. Natural Resources Institute Finland (Luke), FI-31600 Jokioinen, Finland

Misztal, I., S. Tsuruta, T. Strabel, B. Auvray, T. Druet, and D. H. Lee. 2002. BLUPF90 and related programs (BGF90). Commun. No. 28–07 in 7th World Congr. Genet. Appl. Livest. Prod., Montpellier (FR), 19-23 August.

Misztal, I. 2008. Reliable computing in estimation of variance components. J. Anim. Breed. Genet. 125:363-370. https://doi: 10.1111/j.1439-0388.2008.00774.x

Mohd Nor, N., W. Steeneveld, T. van Werven, M. C. M. Mourits, and H. Hogeveen. 2013. First-calving age and first-lactation milk production on Dutch dairy farms. J. Dairy Sci. 96:981–992. https://doi.org/10.3168/jds.2012-5741

Mourits, M. C. M., R. B. M. Huirne, A. A. Dijkhuizen, A. R. Kristensen, and D. T. Galligan. 1999. Economic optimization of dairy heifer management decisions. Agric. Syst. 61:17–31. https://doi.org/10.1016/S0308-521X(99)00029-3

Muir, B. L., J. Fatehi, L. R. Schaeffer. 2004. Genetic relationships between persistency and reproductive performance in first-lactation Canadian Holsteins. J. Dairy Sci. 87:3029-3037. https://doi.org/10.3168/jds.S0022-0302(04)73435-9

Nicolazzi, E. L., S. Biffani, and G. Jansen. 2013. *Short communication:* Imputing genotypes using PedImpute fast algorithm combining pedigree and population information. J. Dairy Sci. 96:2649–2653. https://doi.org/10.3168/jds.2012-6062

Paganini, M. 2023. Quanto costa la rimonta? Accessed Jan. 9, 2023. https://www.pointvet.it/web/index.php?com=contents&option=index&id=141

Patry, C., and V. Ducrocq. 2011. Accounting for genomic pre-selection in national BLUP evaluations in dairy cattle. Genet. Sel. Evol. 43:30. https://doi.org/10.1186/1297-9686-43-30

Tranel, L. 2019. Heifer Raising Costs in 2019. Accessed Jan. 9, 2023 https://www.extension.iastate.edu/dairyteam/content/iowa-dairy-budgets

Tozer, P. R., and A. J. Heinrichs. 2001. What affects the costs of raising replacement dairy heifers: A multiple-component analysis. J. Dairy Sci. 84:1836–1844. https://doi.org/10.3168/jds.S0022-0302(01)74623-1

VanRaden, P. M., and E. J. H. Klaaskate. 1993. Genetic evaluation of length of productive life including predicted longevity of live cows. J. Dairy Sci. 76:2758–2764. https://doi.org/10.3168/jds.S0022-0302(93)77613-4

Wathes, D. C., J. S. Brickell, N. E. Bourne, A. Swali, and Z. Cheng. 2008. Factors influencing heifer survival and fertility on commercial dairy farms. Animal 2:1135-1143. https://doi.org/10.1017/s1751731108002322

Yanga, D.S. and Jaja, I.F. 2022. Culling and mortality of dairy cows: why it happens and how it can be mitigated. F1000Res, 10:1014. https://doi.org/10.12688/f1000research.55519.2

Zavadilová, L., and M. Štípková. 2013. Effect of age at first calving on longevity and fertility traits for Holstein cattle. Czech J. Anim. Sci. 58:47-57. https://doi.org/10.17221/6614-cjas

ORCIDS

V. Ferrari https://orcid.org/0000-0002-7774-4388

F. Galluzzo https://orcid.org/0000-0003-2301-2656

J. B. C. H. M. van Kaam https://orcid.org/0000-0002-2592-2461

M. Penasa https://orcid.org/0000-0001-9984-8738

M. Marusi https://orcid.org/0000-0002-2359-3633

R. Finocchiaro https://orcid.org/0000-0002-9058-9992

G. Visentin https://orcid.org/0000-0003-0869-5516

M. Cassandro https://orcid.org/0000-0002-8709-2870

Chapter III

A tool to optimize dairy herd replacements combining conventional, sexed, and beef semen

V. Ferrari[1,2], M. Marusi[1], M. Penasa[2], J.B.C.H.M. van Kaam[1], R. Finocchiaro[1],

M. Cassandro[1,2]

[1]Associazione Nazionale Allevatori della Razza Frisona, Bruna e Jersey Italiana, Via Bergamo 292, 26100 Cremona (CR), Italy

[2]Department of Agronomy, Food, Natural resources, Animals and Environment, University of Padova, Viale dell'Università 16, 35020 Legnaro (PD), Italy

Submitted to *Italian Journal of Animal Science*

ABSTRACT

A tool to help Italian dairy farmers choosing the most suitable replacement strategy has been developed. The approach aimed to identify yearly female replacement needs based on herd performance level and combination of different semen type (conventional, sex-sorted, and beef semen), with the ultimate goal of enhancing farm profit. A case study based on a 350-cow Holstein herd was used and 3 levels of herd fertility (high, medium, and low) were simulated to define the yearly number of dairy female replacements needed and the number of females yielded under different semen utilization scenario. The number of annual dairy replacements was obtained as the number of cows multiplied by the replacement rate and adjusted by the age at first calving. Number of animals yielded was used to evaluate the replacement cost per 100 L of milk. Then, four strategies of sexed semen utilization were combined with five strategies of beef semen use. Animals that were not inseminated with sexed or beef semen were bred with conventional semen. Regardless of fertility level, the number of dairy female replacement heifers that the farm needs are 110. Increasing beef semen use allows farmer to yield less replacement heifers. Furthermore, as beef semen use increases and the number of replacement heifers decreases, replacement cost per 100 L of milk reduces. Therefore, our results highlighted that replacement costs increase with increasing number of yielded heifers. Hence, combining beef and sexed semen to reach heifer balance close to zero, decreased the replacement cost.

Keywords: heifer, management, tool, crossbreeding, sexed semen

Highlights

- Yielding more heifers than needed is not the most profitable strategy for farmers.
- Combining sexed-sorted semen with beef semen allow farmers to breed less heifers.
- The developed tool will be implemented into a mating program.

Introduction

Farmers have to consider a plethora of factors when defining their mating strategies such as semen type, semen destination use (e.g., dairy or beef), and semen price (De Vries et al. 2023). Furthermore, combining sex-sorted semen with genomic tools can further accelerate the genetic progress of the traits and make more young females available as future replacements in the herd as a result (Borchersen and Peacock 2009; Sørensen et al. 2011; Hjortø et al. 2015). As a consequence, several farmers have an abundance of replacement heifers and it has been shown that culling cows to leave space to replacement heifers is not necessarily the most profitable strategy for a herd (De Vries 2020). Moreover, given the current Italian market conditions, it is not profitable to breed more-than-needed heifers and sell them to other farmers (ISMEA 2023). On the other hand, the higher market value of crossbred dairy calves is an attractive strategy for dairy farmers (Cabrera 2022). The strategy to breed highest genetic merit animals with sexed semen in order to meet herd replacement needs and the remaining cows with beef semen provides an opportunity to farmers to improve both the genetic level of their herds and their profit. Several studies have evaluated different breeding strategies to maximise herd performances (Ettema et al. 2017; Holden and Butler 2018; Clasen et al. 2021). The advantage of sex-sorted semen is maximum when used on high genetic and fertile animals to obtain the number of female calves needed. As conception rate with sex-sorted semen is lower than with conventional semen (Holden and Butler 2018), it is preferably used on virgin heifers and first-lactation cows, given that fertility performances are better for those animals. Furthermore, it has been shown that sexed-sorted semen lowers the incidence of dystocia and stillbirth, as female calves are usually smaller and easy to calve (Holden and Butler 2018; Pahmeyer and Britz 2020). The determination of the number of animals to keep as replacements heifers and the choice of the best strategy are key aspects in herd management (De Vries 2020). Currently, a specific tool to help Italian dairy farmers choosing the most suitable replacement strategy, based on their productive and reproductive data, is not available. Therefore, the objective of the present study was to define a tool for the identification of the annual female replacement needs based on herd performance level and combination of type of semen (conventional, sex-sorted, and beef semen) to optimise economic outcome of the dairy herd.

Materials and methods

The method is based on the approach proposed by Genex Cooperative (Ontario, CA) and adjusted to the Italian herd and market conditions. Italian dairy farmers are rearing more heifers than needed and based on current market conditions the sale of surplus heifers is not cost-effective. Moreover, surplus heifers lead to increasing i) voluntary cow culling to allow heifers to enter the milking herd, ii) rearing costs, and iii) GHG emissions (Holden and Butler 2018; Pahmeyer and Britz 2020). A tool has been developed and reported into an Excel spreadsheet to let users adapting it to their situations.

A case study was simulated assuming a 350-cow Holstein herd (250 cows and 100 heifers entering per year) located in the Po Valley (Northern Italy), targeting 40% replacement rate, 7% stillbirth rate, 5% calves and heifers rearing loss, and 8% pregnancy loss, which represents averages extrapolated by the Italian Holstein, Brown Swiss and Jersey Association (Cremona, Italy). To account for unexpected issues or to allow for more "voluntary" culling, an additional 10% of heifers has been considered. To explore different scenarios, 3 levels of herd fertility were simulated: high (HFL), medium (MFL), and low fertility (LFL). Age at first calving was set at 24 mo (regardless of the fertility level of the herd), conception rate (CR) at 50%, 43%, and 32% for HFL, MFL, and LFL, respectively, and calving interval at 13, 14, and 14.5 mo for HFL, MFL, and LFL, respectively. Values of CR were retrieved from the literature (DeJarnette et al. 2009; Mur-Novales and Cabrera 2017; Li and Cabrera 2019) and then adapted to the Italian average situation. Fertility rate for inseminations with conventional beef and dairy semen was assumed to be the same, whereas for inseminations with sexed dairy semen it was assumed to be 80% that of conventional semen. Percentage of female calves from conventional and beef semen was set at 47%, and from sexed semen at 90%. The method has been developed to enable farmers to adjust all input data described above based on the specific herd conditions and goals. Input variables required by the tool to define the number of dairy female replacements needed in a year and the number of females yielded under different semen utilization are reported in Table 1. All these inputs can be adapted by farmers or technicians to the specific situation of a given herd and current market conditions.

Table 1. Input variables of the heifer management tool. All input data can be changed by the farmer or technician according to specific herd situation.

Variable	Input value
Cows (lactating and dry) (n)	250
Breeding heifers entering the herd (n/yr)	100
Annual replacement rate (%)	40
Annual herd growth rate target (%)	0
Heifers' safety percentage (%)	10
Sex ratio (females/males) by semen type (%)	47/53 (conventional and beef), 90/10 (sexed)
Calving interval according to the fertility level[1] (mo)	13 (high), 14 (medium), 14.5 (low)
Animals not inseminated (%)	2
Pregnancy loss (%)	8
Stillbirth rate (%)	7
Mortality from weaning to first calving (%)	5
Age at first calving (mo)	24
Average heifer rearing cost (€/d)	4.29
Average heifer market value (€)	1800
Average cost for disposal of dead-on-farm cow (€)	300
Average cull cow market value (€)	800
Average purebred male dairy calf market value (€)	51.60
Average crossbred calf market value (€)	245
Milk production (L/d)	31
Total milk sold per year (L)	2,828,750

[1] high = high herd fertility level (50% conception rate and 13 mo calving interval); medium = medium herd fertility level (43% conception rate and 14 mo calving interval); low = low herd fertility level (32% conception rate and 14.5 mo calving interval).

First, the tool was run assuming that all inseminations were performed with conventional semen under a defined fertility level as previously described (HFL, MFL, LFL) and considering that the herd size remained stable to determine the number of dairy replacement calves needed on a yearly basis. The number of annual dairy replacements was obtained as the number of cows multiplied by the replacement rate and adjusted by the age at first calving, in order to account only for heifers that are going to calve during the considered year. Then, four strategies of sexed semen utilization were combined with five strategies of beef semen use. The sexed semen scenarios were: 1) no use of sexed semen (**NO_{SS}**), 2) 100% of heifers inseminated with sexed semen (**H_{100}**), 3) 100% of heifers and 20% of top cows inseminated with sexed semen (**$\text{H}_{100}\text{C}_{20}$**), and 4) 80% of heifers and 20% of top cows inseminated with sexed semen (**$\text{H}_{80}\text{C}_{20}$**). Beef semen utilization was allocated to cows that were not inseminated with sexed semen, according to farm management decisions, at the following percentages: 1) 0%, 2) 25%, 3) 50%, 4) 75%, and 5) 100%. Top cows were identified by farmers based on genetic and/or genomic breeding values, and/or phenotypic performances. Selection criteria of top cows differ among farms given that they depend on specific herd objectives and conditions. All remaining eligible animals that were not inseminated with sexed or beef semen were bred with conventional semen. Under the three fertility levels, from each sexed semen scenario combined with beef semen use, the total number of heifers yielded per year was derived. Then, heifer balance was calculated as the difference between the number of heifers yielded and the annual dairy replacement needs. The number of animals yielded was used to evaluate the replacement cost per 100 L of milk by alternative semen utilization protocols. Feed costs were retrieved from CLAL (2023), and market value of dairy and crossbred calves from ISMEA (2023). Cows were assumed to produce 31 L/d of milk. Replacement cost (**RC**) is the cost to maintain a herd at the same size per 100 L of milk sold and is generally used to compare different breeding strategies. It depends on some economic information, easily collected by farmers: annual replacement rate, heifer rearing cost, and revenue from selling milk (Bethard and Nunes 2011). The RC was calculated as:

$$RC = \frac{cost\ of\ rearing\ replacements - (cull\ cow\ income + income\ from\ male\ calves\ sold)}{income\ from\ 100\ L\ of\ milk\ sold}$$

where cost of rearing replacements included all costs incurred from birth to first calving calculated for all females yielded; cull cow income included the revenue from selling cull cows and heifers; and income from male calves included the revenue from selling dairy male calves and calves from beef when beef semen is used. The 100 L of milk sold has been identified as an appropriate production unit to compare different herd conditions (e.g., size, location, milk production).

Results and discussion

The method presented in the paper is a valuable instrument to help farmers identify the correct number of dairy heifers to be inseminated to maintain constant the herd size (or to set an annual growth rate) and to minimize rearing costs. The annual number of heifers and cows eligible to be mated, the number of services per conception needed to maintain a constant adult herd size, the conception rate under the 3 fertility levels (HFL, MFL, and LFL), and the number of the annual dairy female replacement cows are reported in Table 2. The number of dairy female replacement heifers that the farm needs is 110, for HFL, MFL, and LFL. This number does not reflect differences in reproductive and fertility performances given that the result derived from the number of animals in the herd and the annual turnover rate, adjusted for age at first calving. Moreover, Overton and Dhuyvetter (2020) demonstrated that yielding more heifers than needed is not economically worth for the farm and hence yielding the right number of heifers would enhance farm profit. As mentioned above, we accounted for additional 10% heifers beyond those needed to satisfy replacement needs of the herd to give farmers the opportunity for voluntary culling or unexpected issues. The percentage can be increased or decreased by farmers based on their own herd objectives.

Table 2. Number of heifers and cows to breed, number of dairy replacements needed per year, number of services per conception, and average conception rate (%) needed to maintain a constant herd size under 3 fertility levels[1], assuming 100% use of conventional semen.

Animals	Eligible animals, n	Services/conception, n			Conception rate, %		
		high	medium	low	high	medium	low
Heifers	100	1.8	2.0	2.5	55	50	40
Cows	250	2.2	2.9	4.3	45	35	23
Annual replacements needed	110						

[1] high = high herd fertility level (50% conception rate and 13 mo calving interval); medium = medium herd fertility level (43% conception rate and 14 mo calving interval); low = low herd fertility level (32% conception rate and 14.5 mo calving interval).

Table 3 summarizes the possible pairwise solutions of the tool (replacement costs per 100 L of milk, and heifer balance) that result from the different strategies of beef and sexed semen use under the 3 different herd fertility levels. Larger use of beef semen allows farmers to yield less heifers, on a yearly basis; indeed, when heifer balance is negative, farmers are breeding less heifers than needed, whereas positive values means that farmers are breeding more-than-needed heifers. Therefore, herds aiming at maintaining constant their size have to pursue heifer balance of zero or close to zero. Accordingly, as beef semen use increases and reared heifers reduces, replacement cost per 100 L of milk decreases regardless of reproductive performance. When heifer balance is below zero, replacement cost is reported, but it should be noted that this is not a replacement strategy that should be pursued by farmers, as it means that, if followed, herd size will decrease, or farmers have to buy heifers to maintain their herd size. Furthermore, increasing the use of dairy sexed-sorted semen within the four dairy sexed semen utilization strategies (NO_{SS}, H_{100}, $H_{100}C_{20}$, $H_{80}C_{20}$) leads to an increase of replacement cost (and higher number of reared heifers), regardless of beef semen use. Herd fertility level affects greatly replacement costs and heifer balance (Figure S1). Better fertility level leads to higher number of heifers reared, at the same level of beef and sexed semen use. Within dairy sexed semen strategies, high fertility level showed both greater number of heifers yielded and replacement costs, in accordance with Cabrera (2022), who observed a positive relationship between reproductive performance and replacement balance. Looking at these results, it is clear the positive relationship between replacement cost and heifer balance; indeed, greater replacement costs were obtained with higher number of heifers yielded, which also corresponds to lower use of beef semen. Indeed, the use of beef semen in dairy herds has been observed to positively increase herd net present value (Barrientos-Blanco et al. 2018). Also, the combination of beef semen (on inferior genetic merit cows) with sexed semen speeds up the genetic progress of the herd (Ettema et al. 2017). The highest replacement cost has been obtained with 0% beef semen and $H_{100}C_{20}$ (rearing from 20 to 31 heifers more-than-needed, for MFL and HFL, respectively), whereas the lowest with 100% use of beef semen and NO_{SS} (but rearing from -84 to -83 heifers than needed, for LFL and MFL, respectively, to maintain constant the herd size). The possible choice of pursuing the lowest replacement cost and then buying extra replacements in the market can be followed but it is often not feasible and accepted by most farmers. Thus, dairy farmers prefer to produce their own replacements, while using beef semen (Cabrera 2022). Instead, rearing more heifers than needed and then sell them is not economically convenient, as replacement cost per 100 L of milk increases and actual average heifer market value does not cover rearing costs (ISMEA 2023). Thus, ideal situations can be reached adjusting beef and sexed semen, to reach heifer balance close to zero (Table 3). Indeed, the combination of beef semen and sexed semen, within strategies and reproductive performances, decreased the replacement cost. Within their reproductive performance, farmers should choose the strategy that allow them to reach their annual heifer replacement needs; once obtained, they should select the scheme that decreases the replacement cost. The following step will be to select which heifers and cows have to be inseminated with sexed, conventional, and beef semen. Beef semen in dairy herds is usually used on low genetic merit cows or cows with fertility problems (Ettema et al. 2017), and the combined use of beef semen and sexed semen on heifers produces the highest economic return (Clasen et al. 2021). It is worth noting that this study did not include the effect of gestation length and calving ease that have been linked with the use of beef semen on dairy cattle (Fouz et al. 2013). Besides that, the cost of semen has not been considered given that replacement cost as evaluated by our method only considered the cost of rearing replacements without discriminating different types of semen. This tool will be implemented into ANAFIBJ online mating program to provide farmers an approach to identify the best replacement strategy to follow prior to select which heifers or cows to mate with a given bull in order to enhance herd genetic potential and decrease inbreeding.

Table 3. Replacement costs per 100 L of milk (€) and heifer balance[1] (in parentheses) for different strategies of beef and sexed semen use under different herd fertility levels. Missing values refer to breeding strategies that cannot be pursued.

Beef semen use, %	Dairy sexed semen use[3]			
	NO_{SS}	H_{100}	$H_{100}C_{20}$	$H_{80}C_{20}$
Low fertility level[2]				
0	9.02 (-20)	9.73 (4)	10.00 (16)	9.87 (11)
25	8.52 (-36)	9.18 (-12)	9.50 (0)	9.37 (-5)
50	8.03 (-52)	8.68 (-28)	9.00 (-16)	8.87 (-21)
75	7.53 (-68)	8.18 (-44)	8.51 (-32)	8.37 (-37)
100	7.03 (-84)	7.69 (-60)	- (-)	- (-)
Medium fertility level[2]				
0	9.11 (-16)	9.79 (8)	10.12 (20)	9.98 (16)
25	8.59 (-33)	9.27 (-8)	9.61 (4)	9.47 (-1)
50	8.08 (-50)	8.76 (-25)	9.09 (-13)	8.95 (-18)
75	7.56 (-66)	8.24 (-41)	8.57 (-29)	8.44 (-34)
100	7.05 (-83)	7.73 (-58)	- (-)	- (-)
High fertility level[2]				
0	9.22 (-12)	10.01 (17)	10.4 (31)	10.24 (25)
25	8.70 (-29)	9.50 (0)	9.88 (14)	9.73 (8)
50	8.19 (-45)	8.98 (-16)	9.37 (-2)	9.21 (-8)
75	7.67 (-62)	8.46 (-33)	8.85 (-19)	8.69 (-25)
100	7.15 (-78)	7.94 (-49)	- (-)	- (-)

[1] Heifer balance was calculated as annual dairy replacements needed minus annual dairy heifers yielded.

[2] high = high herd fertility level (50% conception rate and 13 mo calving interval); medium = medium herd fertility level (43% conception rate and 14 mo calving interval); low = low herd fertility level (32% conception rate and 14.5 mo calving interval).

[3] NO_{SS} = no use of sexed semen; H_{100} = 100% of heifers inseminated with sexed semen; $H_{100}C_{20}$ = 100% of heifers and 20% of top cows inseminated with sexed semen; $H_{80}C_{20}$ = 80% of heifers and 20% of top cows inseminated with sexed semen. All remaining eligible animals that were not inseminated with sexed or beef semen were bred with conventional semen.

Disclosure statement

The authors declare that there is no conflict of interest associated with the paper. The authors alone are responsible for the content and writing of this article.

Funding

This study was supported by “Latteco2 project, sottomisura 10.2 of the National Rural Development Program (PSRN) - Biodiversity 2020–2023” [Ministero dell'agricoltura, della sovranità alimentare e delle foreste (MASAF), D.M. no. 465907 del 24/09/2021, project unique code J12C21004080005].

Data availability statement

None of the data were deposited in an official repository. The data that support the findings presented in this study are available from the corresponding author upon reasonable request.

Ethical approval

This study did not involve animals and thus prior ethical approval was deemed not required.

ORCID

V. Ferrari https://orcid.org/0000-0002-7774-4388

M. Marusi https://orcid.org/0000-0002-2359-3633

M. Penasa https://orcid.org/0000-0001-9984-8738

J. B. C. H. M. van Kaam https://orcid.org/0000-0002-2592-2461

R. Finocchiaro https://orcid.org/0000-0002-9058-9992

M. Cassandro https://orcid.org/0000-0002-8709-2870

References

Barrientos-Blanco JA, Thompson NM, Olynk Widmar NJ, Wolf CA, Snyder, LU. 2018. Expected value of crossbred dairy cattle artificial insemination breeding strategies in virgin heifers and lactating cows. Livest. Sci. 211:66-74. https://doi.org/10.1016/j.livsci.2018.03.005

Bethard G, Nunes AL. 2011. Are you efficiently replacing your herd? Proceeding of the Western Dairy Management Conference, Reno (NV), 9–11 March.

Borchersen S, Peacock M. 2009. Danish A.I. field data with sexed semen. Theriogenology. 71:59-63. https://doi.org/10.1016/j.theriogenology.2008.09.026

Cabrera VE. 2022. Economics of using beef semen on dairy herds. JDS Commun. 3:147-151. https://doi.org/10.3168/jdsc.2021-0155

CLAL, 2023; [accessed 2023 March 17] https://www.clal.it/?section=razioni-lombardia-demo

Clasen JB, Kargo M, Østergaard S, Fikse WF, Rydhmer L, Strandberg E. 2021. Genetic consequences of terminal crossbreeding, genomic test, sexed semen, and beef semen in dairy herds. J. Dairy Sci. 104:8062–8075. https://doi.org/10.3168/jds.2020-20028

DeJarnette JM, Nebel RL, Marshall CE. 2009. Evaluating the success of sex-sorted semen in US dairy herds from on farm records. Theriogenology 71:49-58. https://doi.org/10.1016/j.theriogenology.2008.09.042

De Vries A. 2020. Symposium review: Why revisit dairy cattle productive lifespan? J. Dairy Sci. 103:3838–3845. https://doi.org/10.3168/jds.2019-17361

De Vries A., Bliznyuk N, Sharma1 P, Han Y, Pinedo P. 2022. Insemination values to support mating decisions under dairy heifer calf herd size constraints. Proceeding of the 12th World Congress on Genetics Applied to Livestock Production. Rotterdam (NL), 3-8 July. https://doi.org/10.3920/978-90-8686-940-4

Ettema J F, Thomasen J.R, Hjortø L, Kargo M, Østergaard S, Sørensen AC. 2017. Economic opportunities for using sexed semen and semen of beef bulls in dairy herds. J. Dairy Sci. 100:4161–4171. https://doi.org/10.3168/jds.2016-11333

Fouz R, Gandoy F, Sanjuán ML, Yus E, Diéguez FJ. 2013. The use of crossbreeding with beef bulls in dairy herds: effects on calving difficulty and gestation length. Animal 7:211-215. https://doi.org/10.1017/S1751731112001656

Hjortø L, Ettema JF, Kargo M, Sørensen AC. 2015. Genomic testing interacts with reproductive surplus in reducing genetic lag and increasing economic net return. J. Dairy Sci. 98:646-658. https://doi.org/10.3168/jds.2014-8401

Holden SA, Butler ST. 2018. Review: Applications and benefits of sexed semen in dairy and beef herds. Animal. 12:s97–s103. https://doi.org/10.1017/S1751731118000721

ISMEA, 2023; [accessed 2023 March 17 March] https://www.ismeamercati.it/flex/cm/pages/ServeBLOB.php/L/IT/IDPagina/723

Li W, Cabrera VE. 2019. Economics of using beef semen. Proceeding of the Western Dairy Management Conference. Reno (NV), 25-28 February.

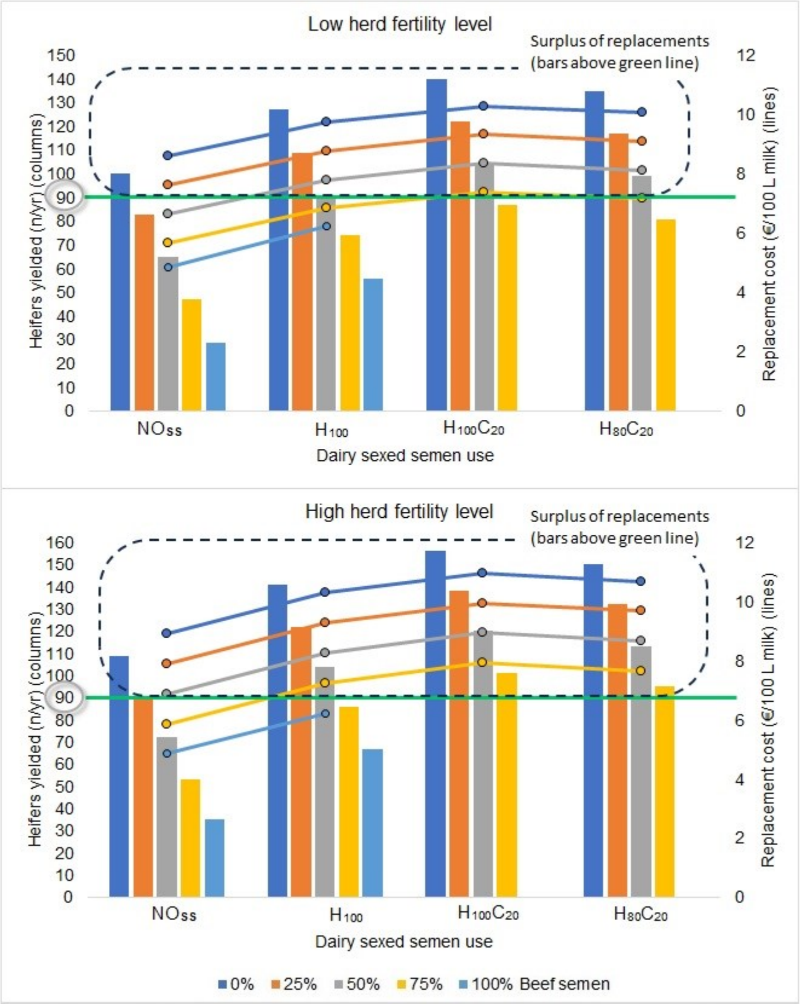
Mur-Novales RM, Cabrera VE. 2017. What type of semen should I use? Proceedings of the Dairy Cattle Reproduction Council Annual Convention, Reno (NV), 7-9 November.

Overton MW, Dhuyvetter KC. 2020. Symposium review: An abundance of replacement heifers: What is the economic impact of raising more than are needed? J. Dairy Sci. 103:3828–3837. https://doi.org/10.3168/jds.2019-17143

Pahmeyer C, Britz W. 2020. Economic opportunities of using crossbreeding and sexing in Holstein dairy herds. J. Dairy Sci. 103:8218–8230. https://doi.org/10.3168/jds.2019-17354

Sørensen MK, Voergaard J, Pedersen LD, Berg P, Sørensen AC. 2011. Genetic gain in dairy cattle populations is increased using sexed semen in commercial herds. J. Anim. Breed. Genet. 128:267–275. https://doi.org/10.1111/j.1439-0388.2011.00924.x

SUPPLEMENTARY MATERIAL



NO$_{SS}$: no use of sexed semen; **H$_{100}$**: 100% of heifers inseminated with sexed semen; **H$_{100}$C$_{20}$**: 100% of heifers and 20% of top cows inseminated with sexed semen; **H$_{80}$C$_{20}$**: 80% of heifers and 20% of top cows inseminated with sexed semen.

Figure S1. Annual number of dairy heifers yielded (columns) and replacement costs (€) per 100 L milk (circles above columns) according to different combinations of sexed and beef semen utilization for low and high herd fertility level. The green line represents the number of annual dairy female replacements needed.

General conclusions

Dairy heifers are fundamental in dairy herds given that they are the future herd genetic progress. The interest towards young animals has increased recently given the need to enhance animal lifetime performance and minimize rearing costs. The present thesis reported how it is possible to enhance heifer fertility traits using genetics, in order to improve future lifetime performances and overall fertility of the dairy herd. Moreover, to enhance farm profit, a strategy is to lower the age at first calving; indeed, results of the present work suggested that it is possible to reduce current phenotypic mean of age at first calving in Italian Holstein population to maximise lifetime production performance, without negatively affect reproductive performance. In addition, to face the tendency of breeding more-than-needed heifers, the tool that has been developed and presented here aims to define the number of herd female replacement needs based on herd performances. This would be beneficial both from an economic and an environmental point of view. In conclusion, understanding the complex interaction between management, genetics, and phenotypic factors during the period from birth to first calving is fundamental to optimize animal welfare and farm profitability. By embracing innovative approaches and using available tools, dairy farmers can secure a more sustainable future for their herds while meeting the evolving demands of the industry.

List of abbreviations

ADG: Average daily gain

AFC: Age at first calving

AFI: Age at first insemination

AFS: Age at first service

AI: Artificial insemination

BB: Belgian blue breed

BHBA: β-Hydroxybutyric acid

BW: Body weight

BrW: Breeding worth

CFS: Conception rate at first service

CR: Conception rate

CP: Crude protein

CVa: Coefficient of genetic variation

DGV: Direct genomic value

DPR: Daughter pregnancy rate

EBV: Estimated breeding value

ECM: Energy corrected milk

EDP: Estimated deregressed proof

FA: Fatty acids

FSH: Follicle stimulating hormone

GB: Galician Blond breed

GEBV: Genomically enhanced breeding values

GHG: Greenhouse gas

GL: Gestation length

Gn-RH: Gonadotropin-releasing hormone

H100: 100% of heifers inseminated with sexed semen

H100C20: 100% of heifers and 20% of top cows inseminated with sexed semen

H80C20: 80% of heifers and 20% of top cows inseminated with sexed semen

HC: High concentrate diet

HFL: High fertility level

HYS: Herd-year-season

HPD95: 95% highest posterior density

IFC: Interval from first service to conception

IFL: Interval from first to last insemination

IGF-1: Insulin-like growth factor 1

INS: Number of inseminations to conception

ISMEA: Institute for Agricultural and Food Market Service

L: Litre

LC: Low concentrate diet

LFL: Low fertility level

LH: Luteinizing hormone

LM: Limousine breed

MACE: Multiple Across Country Evaluation

MFL: Medium fertility level

MSD: Mating start date

Mo: Month

N: Nitrogen

NEFA: Non esterified fatty acids

NH3: Ammonia

NM$: Lifetime net merit

NOss: No use of sexed semen

NPV: Net present value

NR56 (NRR56): Nonreturn rate at 56 d after first service

P: Phosphorus

PGF2α: Prostaglandin F2alpha

RC: Replacement cost

ReplNE: Replacement N use efficiency

SE: Standard error

SNP: Single nucleotide polymorphism

TBV: True breeding value

List of publications

Scientific Publications in International Journals, Conference Abstracts, Conference Proceedings

2021

Finocchiaro R., van Kaam J.B.C.H.M., & **Ferrari V.** 2021. Genetic modelling of heat stress in Italian Holstein cows. In: *Book of Abstracts of the 24th Congress of the Animal Science and Production Association (ASPA),* September 21-24, 2021, Padova, Italy, *Italian Journal of Animal Science*, vol 20 (suppl. 1), p. 236. https://doi.org/10.1080/1828051X.2021.1968170

Ferrari V., Marusi M., & Cassandro M. 2021. A tool to efficiently replace dairy heifers using genomic information. In: *Book of Abstracts of the 24th Congress of the Animal Science and Production Association (ASPA),* September 21-24, 2021, Padova, Italy, *Italian Journal of Animal Science*, vol 20 (suppl. 1), p. 236. https://doi.org/10.1080/1828051X.2021.1968170

Ferrari V., Galluzzo F., Finocchiaro R., van Kaam J.B.C.H.M., Marusi M., Penasa M., & Cassandro M. 2021. Age at fist calving in Italian Holstein Friesian Population: a preliminary study. In: *Book of Abstracts of the 29th Animal Science Days International Symposium (ASD),* September 13-17, 2021, Gödöllő, Hungary.

2022

Ferrari V., Galluzzo F., Finocchiaro R., Van Kaam J.B.C.H.M., Penasa M., Marusi M., & Cassandro, M. 2022. Genetic aspects of age at first calving in Italian Holstein dairy cows. In: *Proceedings of the 12th World Congress on Genetics Applied to Livestock Production (WCGALP),* July 3-8, 2022, Rotterdam, The Netherlands. *Wageningen Academic Publishers*, p. 2895–2899. https://doi.org/10.3920/978-90-8686-940-4_702

Galluzzo F., Van Kaam J.B.C.H.M., Finocchiaro R., **Ferrari V.**, Marusi M., Cassandro M. 2022. Genetic evaluation of gestation length in Italian Holstein breed. In: *Proceedings of the 12th World Congress on Genetics Applied to Livestock Production (WCGALP),* July 3-8,

Rotterdam, The Netherlands. *Wageningen Academic Publishers*, p. 500–503. https://doi.org/10.3920/978-90-8686-940-4_113

Gislon G., **Ferrari V.**, Zucali M., Bava L., Tamburini A., Sandrucci A. 2022. Fertility and management parameters in a tool for the simplified evaluation of global warming potential related to the milk production. In: *Book of Abstract of the 3rd AISSA under 40*, July 14-15, Bolzano, Italy. Available at https://www.aissaunder40.com/_files/ugd/250472_9e281fcbef1f4311b526f54735091355.pdf

2023

Ferrari V., Visentin G., van Kaam J. B. C. H. M., Penasa M., Marusi M., Finocchiaro R., & Cassandro M. 2023. Genetic and nongenetic variation of heifer fertility in Italian Holstein cattle. *JDS Communications*. 4: 35-39 https://doi.org/10.3168/jdsc.2022-0270

Ferrari V., Visentin G., van Kaam J. B. C. H. M., Penasa M., Marusi M., Finocchiaro R., & Cassandro M. 2023. Genetic aspects of heifer fertility in Italian Holstein population. In: *Book of Abstracts of the 25th Congress of the Animal Science and Production Association (ASPA),* June 13-16, 2023, Monopoli (BA), Italy, *Italian Journal of Animal Science*, vol 22 (suppl. 1), p. 34. https://doi.org/10.1080/1828051X.2023.2210877

Gislon G., Zucali M., Bava L., Marusi M., **Ferrari V.**, Finocchiaro R., Cassandro M., & Sandrucci A., 2023. Modelling GWP of milk production using technical efficiency indicators. In: *Book of Abstracts of the 25th Congress of the Animal Science and Production Association (ASPA),* June 13-16, 2023, Monopoli (BA), Italy, *Italian Journal of Animal Science*, vol 22 (suppl. 1), p. 156. https://doi.org/10.1080/1828051X.2023.2210877

Publications in Technical Journals

2020

Ferrari V. 2020. Gestire la rimonta: costo od opportunità? Bianconero, 5(settembre/ottobre), 9-10.

2021

Ferrari V. 2021. Indagine allevatori: gestione della rimonta. Bianconero, 5(settembre/ottobre), 30-31.

van Kaam J.T., Finocchiaro R., Marusi M., **Ferrari V.** 2021. La genomica nella selezione delle bovine da latte. Management della stalla da latte 4.0: governare i cambiamenti. Percorso di formazione a distanza (FAD) abbinato a Summa animali da reddito, 1(gennaio/febbraio), 3-10.

Ferrari V. 2021. Gestione della rimonta: e gli altri Paesi a che punto sono? Bianconero, 2(marzo/aprile), 52-53.

Ferrari V. 2021. La quota di rimonta aziendale: costo od opportunità? Professione Allevatore, 6(1/15 aprile), 63-64.

Ferrari V. Finocchiaro R., Marusi M. 2021. Impatto ambientale dell'allevamento bovino. Bianconero, 3(maggio/giugno), 6-8.

2022

Ferrari V. 2022. Workshop ANAFIBJ: la rimonta e il giovane bestiame. Bianconero, 6(novembre/dicembre), 61-63.

Ferrari V., Fabris A. 2022. Selezionare per il benessere conviene. Allevatori TOP, 9(settembre), 20-23.

Ferrari V. 2022. Strategie selettive per la gestione della rimonta: un'opportunità per il progresso genetico e per il bilancio aziendale. Bianconero, 3(maggio/giugno), 32-33.

2023

Zucali M., Gislon G., Bava L., Sandrucci A., Tamburini A., **Ferrari V.**, Marusi M., Finocchiaro R. 2023. Produzione di latte e impatto ambientale: è possibile stimarlo in modo semplice? Bianconero, 2(marzo/aprile), 13-15.

Ferrari V. 2023. Workshop rimonta: I primi 6 mesi di vita (0-180 giorni) il periodo cruciale per una mandria produttiva e longeva. Bianconero, 2(marzo/aprile), 65-67.

Acknowledgments

During these last three years I met and engaged with many people…I would like to thank them all!

A special thank goes to my supervisor, Prof. Martino Cassandro, and to my co-supervisor, Prof. Mauro Penasa, for their support, patience, and help during this process.

Thanks to Maurizio, tutor at ANAFIBJ and colleague, as he has been the reference point from which to learn every single day of my PhD. I couldn't hope for a better tutor than him.

A special thanks to Ferdinando, Raffaella and all other colleagues from ANAFIBJ for their support and help with my projects…each of them have contributed to a little piece of this thesis.

A special thought to Prof. Giulio Visentin, for his help first as a colleague at ANAFIBJ and then as professor at University of Bologna. Thanks for his precious support in revising my papers.

Thanks also to the University of Padova and the Department of Agronomy, Food, Natural Resources, Animals and Environment to gave me the opportunity to enroll as a PhD student at the Italian Holstein-Friesian breeders association. I would express my gratitude for their remotely support with the bureaucratic fulfillment!

A very big thank goes to my boyfriend Manuel for his patience, support, and daily encouragement…but above all for everyday being simply him!

Thanks also to my parents who have always supported me, to my family and to all my friends. All of them have contributed to this thesis.

Last but not least, thanks also to all reviewers that revised my manuscripts and thesis. I can say that they gave me precious hints to improve the quality of my works.

Author at a glance

Valentina Ferrari was born in Verona (Italy) in 1993. After high school degree, she enrolled at University of Padova (Italy) in 2013, driven by the passion for animal production sector. After a Bachelor in 2016 (105/110) and a Master degree in 2018 (110/110 cum laude) in Animal Science, she entered the world of work with a position at the Italian Holstein, Brown Swiss and Jersey Association (ANAFIBJ, Cremona, Italy). Here, she started her industrial PhD in Animal and Food Science (Department of Agronomy, Food, Natural resources, Animals and Environment - University of Padova) in October 2020 under the supervision of Prof. Martino Cassandro. The PhD project was focused on genetic and management aspects of fertility in Holstein heifers. At ANAFIBJ she works at the I.A. Services office and collaborate with the R&D office. Her research activity is documented by scientific and technical papers published in international peer reviewed and technical journals and presentations at national and international meetings and congresses. Among personal interests, travelling, reading, meeting new people, and discover places emerged.