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TESI

**Stayability approach to longevity evaluation in Italian Mediterranean Buffalo:
genetic parameters and associated traits**

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ABSTRACT

The longevity of dairy animals, particularly Mediterranean Italian buffaloes, is a complex trait influenced by multiple factors, with genetics playing a challenging role due to the significant impact of environmental conditions and farm management strategies. This thesis aimed to estimate the longevity of these buffaloes using the Stayability approach in Italian Mediterranean Buffalo (IMB) herds, analysing its genetic parameters for its potential inclusion in breeding programs and to study the relationship with IMB type traits. Data from 276,415 buffaloes born between 1980 and 2020 were provided by the National Farmer Association of Buffalo Species (ANASB), covering 1,025 farms in central and southern Italy. Stayability was assessed across different parities, defined as the probability of an animal remaining in the herd after each calving.

Genetic parameters, including heritability and estimated breeding values (EBVs), were estimated using a two-model approach implemented in the BLUPF90 suite. Model 1 accounted for fixed herd effects (Herd), herd-year-season effects (HYS), and an additive genetic effect, while Model 2 incorporated milk production EBVs as a covariate to separate genetic effects from production-related influences. Variance components were estimated using Gibbs sampling via GIBBS1F90+, and post-processing was conducted with POSTGIBBSF90.

Results indicate a decreasing stayability trend across parities, with the most significant drop occurring between the first and second parity (0.81 to 0.41), reaching 0.03 by the tenth parity. The culling rate was highest in early lactations and gradually declined in later ones. Heritability estimates ranged from 0.07 to 0.12, with higher values observed in early parities, suggesting that production traits strongly influence culling decisions in younger animals. Genetic correlations between stayability and milk yield EBVs were positive in early parities but weakened in later ones, while negative correlations were observed with fat and protein percentages, potentially due to metabolic constraints on longevity. These findings highlight stayability as a relevant genetic trait for selection, with implications for optimizing breeding strategies to enhance longevity and productivity in IMB herds.

A second study investigates the relationships between morphology and stayability in Italian Mediterranean Buffaloes (IMB). Two analytical approaches were employed to explore these relationships. The first method utilized a mixed-effects logistic regression model to analyse binary stayability outcomes, aiming to identify the morphological traits most associated with prolonged productive life. The second approach estimated approximate genetic correlations between stayability estimated breeding values (EBVs) of artificial insemination (AI) bulls and their official EBVs for

linear type traits, facilitating the assessment of genetic associations between conformation and longevity.

The same dataset from of previous study was used, provided by ANASB, included stayability records from 211,266 buffaloes, spanning parities from stayability 2 to stayability 10. Morphological trait evaluations were available for 51,702 animals from 430 farms. The dataset was merged with test-day milk yield data to control for the confounding effect of milk production. A subset of animals born between 2005 and 2020 and recorded for type traits from 2010 onward was selected. The primary statistical framework applied was a mixed-effects logistic regression model, which incorporated fixed effects for herd-year-season (HYS), age at first calving, and contemporary group, while random effects included the additive genetic effect of the animal. The variance components for stayability were estimated using Gibbs sampling implemented in the BLUPF90 suite, ensuring robust estimation of genetic parameters.

Results from the logistic regression model highlighted that body-related traits, including chest width, body depth, and angularity, were significantly associated with higher survival probabilities. A broader thorax and deeper chest were particularly advantageous, supporting higher milk production without compromising health. Conversely, stature exhibited minimal influence on survival. Among feet and leg traits, heel height positively correlated with longevity, while a wider hock angle negatively affected stayability in later parities. Regarding udder conformation, front udder length and rear udder width were positively associated with survival, whereas deeper udders and excessive rear udder height exhibited adverse effects due to increased susceptibility to mastitis.

The genetic correlation analysis confirmed the phenotypic findings, showing positive correlations between chest depth and stayability, particularly in early parities. Notably, when milk EBVs were accounted for in the analysis (Model 2), genetic correlations between body size traits and stayability became more negative, suggesting that larger animals are retained longer primarily due to their superior production capacity rather than inherent structural advantages. Similarly, udder traits that were favourable for production showed weaker correlations with longevity once adjusted for milk EBVs, emphasizing the importance of disentangling production effects from conformation-related influences on survival.

This study underscores the necessity of incorporating morphological traits in buffalo breeding programs to enhance longevity while maintaining production efficiency. The findings suggest that selecting for animals with optimal conformation, independent of milk production, could improve sustainability by reducing involuntary culling and enhancing overall herd resilience. Future research

should focus on refining selection indices that integrate both productivity and structural soundness to optimize buffalo breeding strategies.

Results from these studies showed that the genetic analysis of stayability had low to moderate heritability values, with an increasing trend for older parities. Productive buffaloes selected during early lactations stay on farms mainly due to their resilience to environmental conditions and good health. The analysis of genetic correlations between longevity and estimated breeding values (EBV) for milk production suggested a possible positive association, although this was less evident in older parities, likely influenced by voluntary culling based on production.

Longevity in buffaloes is heavily influenced by environmental factors, making genetic selection more challenging. Genomic selection could be a solution to better exploit genetic potential and reduce generation intervals, thus accelerating genetic progress.

The association between longevity and morphology showed that some morphological traits traditionally thought to favour longevity, like larger trunk diameters and udder volumes, were more closely linked to higher production than long-term longevity. In contrast, more compact bodies were associated with greater longevity when milk production was controlled for.

The findings suggest that current selection criteria for buffalo morphology should be reconsidered. Advanced genetic models for estimating correlations between longevity, morphology, and productivity can help identify animals with a higher potential for a long, productive career.

Achieving high longevity in buffaloes is crucial for sustainability in dairy production, particularly in markets like Mozzarella di Bufala Campana DOP. Accurate selection for stayability could reduce culling rates, the need for replacements, and environmental impact, supporting more sustainable and profitable buffalo farming practices. Integrated genetic selection programs combined with innovative management strategies could play a key role in the future of the buffalo industry.

1. INTRODUCTION

1.1 LONGEVITY

Longevity, understood as the productive lifespan of a dairy animal, has always been a trait to be carefully monitored, as it significantly impacts farm management and profitability (Van Arendonk, 1985; Allaire & Gibson, 1992). It is certainly closely related to the overall lifespan of the animal but is not strictly correlated with the species' survival capacity. The replacement rate, which refers to the percentage of animals culled each year by the farmer, consists of two different categories: those culled due to death or functional impairments (e.g., mastitis leading to partial or total loss of mammary secretion ability, reproductive failure, etc.), which constitute the involuntary culling rate, and those eliminated by the farmer's choice due to inadequate productive capacity according to farm standards, known as the voluntary culling rate. The increase in the voluntary culling rate in dairy farms has become an essential tool for accelerating the genetic improvement of the herd, allowing for higher per-capita production by introducing younger and more productive animals. Achieving greater longevity in dairy cattle breeds has been the focus of extensive research, especially in recent years (Miglior et al., 2017; Hu et al., 2021; De Vries, 2020). In addition to economic concerns, other crucial factors have emerged, such as environmental sustainability, biosecurity particularly regarding the need to limit antibiotic use in farming and, not least, animal welfare. These factors are increasingly influencing consumer perception of animal-derived products and are driving the search for more acceptable livestock management practices for the future (Barkema et al., 2015; Miglior et al., 2017; Nathan et al., 2020; De Vries, 2020; Brito et al., 2021). Reducing the involuntary culling rate in dairy farming requires selecting more resilient animals that are well-adapted to farm conditions and capable of reaching higher parity orders. These are the stages at which a healthy animal achieves its highest production levels, benefiting overall per-animal productivity. This also allows for optimal voluntary culling rates without increasing the number of animals raised for replacement. Estimating longevity is a complex process that has been addressed using various statistical approaches and indirect estimators. Additionally, definitive data can only be obtained once an animal leaves the farm. This necessitates careful selection of the data used for estimation and reliance on certain assumptions before applying the data. Several approaches have been used for genetic evaluation of longevity in cattle. The table below (from Schuster et al., 2020) outlines the different longevity estimation methods applied so far (with corresponding references listed in the bibliography). The various longevity measurement methods listed in the table are often used differently by various authors and sometimes confused with one another. Frequently, "productive lifespan" or "Herd Life" (HL) is commonly

understood as the duration of life from the first calving (Beaudeau et al., 1995; Hultgren & Svensson, 2009; Brickell & Wathes, 2011), whereas in some cases, HL refers to the total lifespan from birth (Vollema & Groen, 1996).

Term	Definition	Source
Length of productive life	Days between first calving and culling or death	Compton et al. (2017)
	Number of months in milk between first calving and 84 mo of age	Caraviello et al. (2004)
	Days from one calving to the next calving, death, or culling	Sewalem et al. (2005a)
Herd life	Time between birth and last test day	Vollema and Groen (1996)
	Days from first calving to culling	Brickell and Wathes (2011)
	Days from first to last calving	Gill and Allaire (1976)
Productive herd life	Length of time that individual cows remain in herds after their first calving	Hare et al. (2006)
	Days from date of first calving to date of culling or last dry date	Jairath et al. (1995)
Stayability	Stayability until 36, 48, 60, or 72 mo of age and stayability until 12, 24, 36, or 48 mo after first calving	Vollema and Groen (1996)
	Proportion of cows still alive at 48 mo of age	Oltenuacu and Broom (2010)
	Capability of an animal to remain in its herd over time	Essl (1998)
	Measurement of the fraction of animals still alive at a particular age within an opportunity group	Essl (1998)
True stayability	The ability to delay culling	Ducrocq (1987)
Functional stayability	The ability to delay involuntary culling	Ducrocq (1987)
Functional longevity	The ability to delay involuntary culling	Beaudeau et al. (1995)
	Days from first calving to culling, death, or censoring; adjusted for the effect of milk yield	Sewalem et al. (2008)
	Longevity corrected for milk yield relative to the herd mean	Heise et al. (2016)
True longevity	Longevity as actually observed	Beaudeau et al. (1995)

Figure 1. from Schuster et al., 2020

Survival analysis is characterized by data in which the variable used is the time that elapses from an initial reference point to the subject's removal from the herd. Survival analysis employs a proportional hazard model and represents the most widely used methodological approach for assessing longevity in dairy cattle (Vukasinovic et al., 2001; Caraviello et al., 2004; Rostellato et al., 2021). Another way to evaluate an animal's survival in production is by calculating Stayability, or survival rate. Among the first authors to define longevity in terms of stayability, Hudson and Van Vleck (1981) described it as the probability that a cow remains in the herd up to a specific age when given the opportunity to reach it. It is therefore assessed as a binary trait, making it suitable for analysis using either a linear model or a threshold model. Several studies have estimated stayability in dairy cattle using different criteria, most commonly defining it as the probability of remaining in the herd in subsequent years after the first calving (Hudson & Van Vleck, 1981; Vollema & Groen, 1996; Garcia-Peniche et al.,

2006; Hu et al., 2021). Alternatively, as in the case of Hardie et al. (2021), stayability has been evaluated by considering a specific parity order as a reference rather than the age at culling, still measuring the likelihood of remaining in multiple lactations beyond the first.

1.2. SELECTION FOR PRODUCTION IN THE ITALIAN MEDITERRANEAN BUFFALO (BMI)

The Italian Mediterranean Buffalo (BMI) belongs to the River type and is the only officially recognized breed in Italy for milk production, primarily used for the production of Mozzarella di Bufala Campana DOP. The Protected Designation of Origin (PDO) status was granted to this product in 1981, and since then, the Consortium for the Protection of Mozzarella di Bufala Campana DOP has been operating to support and promote this cheese in Italy and worldwide. It is the only organization recognized by the Italian Ministry of Agriculture, Food, and Forestry Policies for the protection, supervision, enhancement, and promotion of this cheese, which is produced in three regions of southern-central Italy: Campania, Lazio, and Puglia. However, the breeding of BMI, initially almost exclusively concentrated in these three regions, has since expanded across the entire national territory, albeit with limited population sizes. As of June 2024, data from the National Livestock Database (BDN) report a total BMI population of 440,023 head, with their regional distribution shown in the table below. Over the past 30 years, the Italian Mediterranean Buffalo population has tripled, increasing from 112,000 to 440,000 head (FAO Database, 2023). Mozzarella has always been the primary product derived from buffalo milk in Italy, and Mozzarella di Bufala Campana DOP is now the third highest-grossing cheese in Italy. The national average milk yield for BMI, recorded by the Italian Breeders Association (AIA), which has registered 74,299 animals across 324 farms, is $2,361 \pm 690$ kg per lactation (AIA, 2023), with average farm production peaks exceeding 3,000 kg per lactation and high-yielding buffaloes producing approximately 5,000 kg per lactation. These high production levels result from a successful combination of advanced management practices and genetic selection. Implementing a selection program is essential for the future development of the breed, as it allows for the improvement of specific traits according to the farming system used. The current selection criteria for BMI take into account various productive and functional aspects that must be considered simultaneously to increase productivity levels while maintaining milk quality. At the same time, selection aims to adapt breed characteristics to evolving ecological and environmental farming conditions. Milk production improvement can be achieved through various means: by increasing milk yield without much concern for other factors or by controlling fat and protein content. The goal of selection for the Italian Mediterranean Buffalo (BMI) has always been to maintain fat and protein levels in milk as stable as possible while striving to improve production volumes. Since 1977, when milk production controls were introduced for animals registered in the Herd Book, and

later in 1986, with the establishment of the first progeny testing programs for selecting genetically superior bulls, the focus has always been on developing a composite index that integrates both milk quantity and quality. Initially, the PKM (Production in kilograms of Mozzarella) index was used, expressing the kilograms of mozzarella obtained from the milk produced during a standard 270-day lactation. This index was based on the following algorithm developed by Altiero et al. (1986), which considered protein and fat percentages in milk, assigning different weights depending on their importance in the coagulation process, as determined through experimental evaluations:

$$\text{PKM (kg)} = (\text{Milk, kg}) * \frac{[3,5*(\text{Protein,\%}) + 1,23*(\text{Fat,\%}) - 0,88]}{100}$$

where:

- PKM = kg of mozzarella cheese yield;
- Milk = kg of 270 d milk yield (standard lactation);
- Protein % = mean percentage of milk protein in 270 d lactation;
- Fat% = mean percentage of milk fat in 270 d lactation.

The estimated weight of mozzarella, derived from the observation of other traits, was used as a selection criterion to improve overall economic returns. However, for all the analyzed traits, including mozzarella production, Rosati and Van Vleck (2002) found relatively low genetic variance estimates, resulting in low heritability estimates. This was largely attributed to inaccuracies in identifying true paternity, due to the almost exclusive use of natural mating in the species at that time. For this reason, efforts have been made to increase the use of artificial insemination to enhance genetic exchange between herds and accelerate the genetic improvement rate. In 2018, a new selection index was introduced by ANASB, replacing and improving upon the previous genetic index. This new tool, called IBMI (Index for the Italian Mediterranean Buffalo), is a composite index. The traits included in the selection index are milk production, fat and protein percentages, the morphological index for legs and feet, and the udder system index. The effectiveness of a selection program is primarily determined by the ability of selection tools to correctly identify genetically superior animals based on predefined selection objectives. The goal of the IBMI is to further emphasize the dairy potential of the Italian Mediterranean Buffalo, while simultaneously enhancing both quantitative and qualitative milk production traits for improved mozzarella yield, as well as functional morphology (favoring productive animals with morphological traits that facilitate housing and management in

stables). The updated weightings, following the introduction of Genetic Groups (Gomez et al., 2021), for the individual genetic indices used to construct IBMI are:

$$\text{IBMI} = 3.2 * (1.55 * \text{EBV Feet\&legs} + 1.45 * \text{EBV udder} + 0.01 * \text{EBV milk} + 4.88 * \text{EBV fat\%} + 18.45 * \text{EBV protein\%}) + 100$$

where:

- EBV Feet&leg = EBV of partial score for Feet&leg type trait evaluation;
- EBV udder = EBV of partial score for udder type trait evaluation;
- EBV milk = EBV of 270 d milk yield;
- EVB fat% = EBV of milk fat percentage in 270 d milk yield;
- EBV protein% = EBV of milk protein percentage in 270 d milk yield.

Pedigree completeness is a fundamental requirement for any genetic evaluation. Thanks to the introduction of genetic groups, which have allowed for more complete pedigrees, the estimation of animal indices has significantly improved in terms of accuracy. This has led to a better assessment of genetic value, ultimately enhancing the genetic selection process for the Italian Mediterranean Buffalo. At this stage, 84 bulls have been included in the progeny test, 27 of which have yet to be evaluated, while 25 have contributed to improvements in both milk production and quality. Since 2024, ANASB has introduced the First-Generation Genomic Index within the IBMI. This new calculation required adjustments to the coefficients used to compute the IGBMI formula, due to the different variances of the genomic indices.

Thus, the new formula is as follows (Source: ANASB online: <https://www.anasb.it/attivita-anasb/indice-di-selezione-genomico-igbmi/>).

$$\text{IGBMI} = 100 + 3.7 * (0.98 * \text{Feet\&Legs} + 1.52 * \text{Udder} + 0.01 * \text{Milk kg} + 5.08 * \text{Fat\%} + 19.34 * \text{Protein\%})$$

where;

- gEBV Feet&leg = gEBV of partial score for Feet&leg type trait evaluation;
- gEBV udder = gEBV of partial score for udder type trait evaluation;
- gEBV milk = gEBV of 270 d milk yield;

- gEVB fat% = gEBV of milk fat percentage in 270 d milk yield;
- gEBV protein% = gEBV of milk protein percentage in 270 d milk yield.

Selection for higher milk production in the Italian Mediterranean Buffalo (BMI) has led to a slow but continuous transformation in the morphology and physiology of the breed. The evolving farming techniques, increasingly focused on intensification, following the model applied to dairy cattle, have significantly boosted farm revenue. However, this has not always translated proportionally into profitability, and despite varying margins, the buffalo sector has been experiencing fluctuating profitability trends for some time.

Over time, higher productive performance in buffaloes has been accompanied by a deterioration in key zootechnical indices, particularly fertility, calving interval, and management-related health issues, such as udder health problems, a pattern already observed in dairy cattle (Rauw et al., 1998; Brito et al., 2021). This has led to a significant increase in culling rates, resulting in a sharp decline in the average parity at the farm level. According to AIA data (2023), the current average number of lactations per buffalo is 3.6, with a culling rate slightly lower than that of dairy cows, but significantly higher than in the past (Zicarelli L., 2004).

1.3. SELECTION FOR MORPHOLOGY IN THE ITALIAN MEDITERRANEAN BUFFALO

The introduction of morphological selection with linear criteria in the selection process of the Italian Mediterranean Buffalo (IMB) was the first approach, dating back to 2005, aimed at obtaining animals that are more resilient to changing farm conditions and better suited to the higher production levels achieved. The importance of morphology in determining the length of a subject's productive life in farming is widely demonstrated by the large number of functional indices used for this purpose in the genetic selection of major dairy cattle breeds. A significant portion of their selection value depends on numerous morphological traits, combined and weighted differently (Cole and Van Raden, 2018). The morphological traits evaluated in the IMB have led to improvements in general conformation, body size, limb defects, and udder characteristics. Approximately 36% of the linear scheme traits in the IMB assess the size and conformation of the udder to enhance anatomical features that influence milkability. The final score, which expresses the overall merit of a buffalo's traits, is obtained as a weighted average of four partial scores:

- Structure, with a weighting of 10% on the final score
- Legs and feet, with a weighting of 30%
- Udder, with a weighting of 40%
- Production potential, with a weighting of 20%

To date, genetic indices have been calculated for the overall score, the four partial scores, and for specific udder traits, chest depth, and leg and foot traits. Since 2018, with the introduction of the new IBMI genetic index, morphology has become an integral part of the overall selection index for the Italian Mediterranean Buffalo. Additionally, it is included in the current genomic index used by ANASB since 2024. Current selection schemes aim to produce animals with good structural conformation, harmonious and proportionate, with a strong topline and a broad, deep chest with good angularity. Legs and feet must be well-formed, with strong pasterns and high heels. The udder should be firmly attached to the abdomen, of good volume in the posterior diameters, and positioned above the hocks. The importance of morphology in influencing the productive lifespan of a subject in farming is widely demonstrated by the numerous functional indices used in the genetic selection of major dairy cattle breeds, where much of their value depends on various morphological traits, differently combined and weighted (Cole and Van Raden, 2018).

1.4. LONGEVITY IN THE ITALIAN MEDITERRANEAN BUFFALO

In the past, the Italian Mediterranean Buffalo (IMB) was characterized by very favorable overall replacement rates, around 10% (Zicarelli L., 2004). These rates applied to herds where genetic selection had little influence, with lower production levels compared to current standards, and a remarkable natural longevity. Over the past 25 years, genetic improvement programs for the IMB aimed at increasing productivity have had the desired effect, resulting in consistently increasing milk production over the years. This has been achieved partly thanks to the use of advanced technologies in intensive farming systems. However, this productivity improvement has negatively impacted the longevity and hardiness of the breed, also contributing to an average increase in feeding and management costs. Currently, the average calving order for the IMB, according to data from herds under functional control (AIA 2023), is 3.57, with an average age at first calving of 3 years. Assuming an average calving interval of 400 days (Zicarelli L., 2010), it is estimated that the most long-lived buffalo are over 15 years old, and the average replacement rate is below 26%. These data reflect a reality found in many farms, and farmers are highly interested in maintaining high longevity in terms of productive life. It is also not uncommon to find farms with peak replacement rates reaching up to 35%. Reproductive issues in the IMB, related to its negative photoperiod-based reproductive seasonality and the need to force the calving schedule to concentrate births during the less favorable period for the species' natural tendency (Zicarelli L., 2004), significantly impact the voluntary replacement rate. This makes it even more important to try to minimize the forced replacement rate. A recent study directly evaluated longevity in the IMB (Gomez et al., 2025), while for the Murrah buffalo breed, two studies from Brazil and one from Bulgaria have evaluated longevity and its genetic components using both stayability and survival analysis approaches (Peeva T & Ilieva Y 2007; Galeazzi, et al., 2010a; Galeazzi, et al., 2010b).

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PART 1:

**GENETIC PARAMETERS OF A STAYABILITY APPROACH TO ITALIAN
MEDITERRANEAN BUFFALO LONGEVITY**

2. INTRODUCTION

The buffalo population in Italy consists of 437,517 animals distributed across 2,271 farms (BDN, 2024), representing 86% of the total buffalo herd in Europe (FAO, 2023). The majority of these buffalo are concentrated in central and southern Italy. According to the Italian Breeders' Association, data from 71,105 lactations recorded in 2023 across 335 farms indicate an average milk yield of 2,350 \pm 697 kg at first calving, with a fat content of 7.75% and a protein content of 4.65%. Although buffalo milk production is lower than that of dairy cattle, profit margins remain high due to the strong global demand for mozzarella cheese, the primary product derived from buffalo milk in Italy. Additionally, buffalo milk from three Italian regions is used to produce *Mozzarella di Bufala Campana*, a Protected Designation of Origin (PDO) cheese that ranks among the highest-exported cheeses in terms of commercial value (ISMEA, 2024). The Italian Mediterranean Buffalo (IMB) is traditionally characterized by its robustness and remarkable longevity (Zicarelli, 2010).

With an average calving interval of approximately 400 days, the longest-lived buffalo have been reported to exceed 15 years of age (Zicarelli, 1997). Although longevity is often associated with lower productivity, it remains a crucial trait for the economic sustainability of dairy species (Grandl et al., 2016; Schuster et al., 2020; De Vries, 2017). Genetic improvement programs for the IMB have consistently focused on enhancing productivity, leading to a steady increase in milk yield; however, this progress has been accompanied by rising breeding costs. In dairy cattle, increased milk production has been shown to negatively impact various fitness-related traits, particularly reproductive performance and health, thereby contributing to higher rates of premature culling and reduced longevity (Knaus, 2009; Oltenacu and Broom, 2010; Hansen, 2000; McConnel et al., 2008; Ingvarlsen et al., 2003).

Furthermore, a higher culling rate compromises selection efficiency by necessitating the replacement of a greater number of heifers, thereby reducing the selection differential for new individuals entering the herd (De Vries, 2017). Strategies aimed at increasing the average herd parity while simultaneously enhancing production could yield significant benefits. Extending the productive lifespan of dairy animals has both direct and indirect advantages, many of which have been extensively documented in dairy cattle (De Vries and Marcondes, 2020; Adamie et al., 2023). This approach may help offset the fixed costs associated with the non-productive period before first calving by optimizing milk production across an extended productive lifespan. Moreover, early culling prevents animals from

reaching their peak lactation performance, which typically occurs between the third and fourth lactation (Najafabadi et al., 2016). Studies have also suggested a positive correlation between greater longevity and farm sustainability, as longer productive lifespans are associated with reduced methane emissions per kilogram of milk produced in bovine species (Grandl et al., 2016, 2019). Additionally, prolonging the productive life of healthy cows could improve public perception of dairy farming by enhancing animal welfare standards and promoting the social acceptability of milk production (Barkema et al., 2015; Röcklinsberg et al., 2016; Schuster et al., 2020).

Various methodologies have been employed to study longevity in dairy cattle (Hu et al., 2021), with one of the most commonly used approaches being the evaluation of stayability. This trait, first defined by Hudson and Van Vleck (1981), represents the probability of an animal remaining in the herd up to a specified age, provided it has had the opportunity to reach that age. Several studies have estimated stayability in dairy cattle using different criteria, with the most common definition referring to the likelihood of an animal remaining in the herd for a set number of years following first calving (Hudson and Van Vleck, 1981; Vollema and Groen, 1996). More recent studies (Garcia-Peniche et al., 2006; Honghong et al., 2021) have introduced an alternative approach, which considers the probability of an animal remaining in the herd for multiple lactations beyond the first, rather than focusing solely on age at culling. This methodology appears particularly promising as it integrates longevity with productivity and fertility.

2.1 AIM OF THE RESEARCH

A more in-depth investigation into the sustainability of buffalo herds would be highly beneficial. In buffalo, longevity has been assessed as the number of days from first calving to six years post-calving using survival analysis in Murrah buffalo (Galeazzi et al., 2010a), and more recently in IMB buffalo as the number of days from first calving to death or culling (Gomez et al., 2024). The stayability approach has only been applied to Murrah buffalo (Galeazzi et al., 2010b), where it was defined as the ability to remain in the herd for consecutive years following first calving. Therefore, the objective of this study was to analyze stayability at different parities, as defined by Hardie et al. (2021), in a large IMB herd population over the past 30 years. Additionally, genetic parameters were estimated to evaluate the potential inclusion of stayability as a selection criterion in breeding programs.

2.2 MATERIAL AND METHODS

For this study, records on the Italian Mediterranean Buffalo were provided by the National Farmer Association of Buffalo Species (ANASB, Caserta, Italy), regarding 276,415 buffaloes born between 1980 and 2020. ANASB operates at the national level as the managing authority of the herdbook, overseeing genetic improvement programs for the species across 1,025 farms, with more than 131,000 animals under official control. The farms included in the dataset are predominantly located in central and southern Italy, within the designated production area of *Mozzarella di Bufala Campana*, a Protected Designation of Origin (PDO) product. The ANASB dataset, which includes each buffalo's date of birth and identification number, was cross-referenced with the milk recording control dataset from the Italian Breeders Association (AIA). The latter provides detailed information on calving and culling dates. To ensure data accuracy and eliminate inconsistencies, the following data validation procedures were applied:

- Date of birth >1989;
- Age at first calving \leq 60 months;
- First calving date
- Date of elimination

The first filter applied to the dataset, date of birth > 1989, ensured the retention of data with more accurate records of 270-day milk yield. The second filter, age at first calving \leq 60 months, was applied to minimize potential recording errors in age at first parity while maintaining inclusivity, in accordance with the mean age at first parity reported by Zicarelli L. (2008) Only animals with a recorded first calving date were retained for analysis. The edited dataset included 211,266 buffaloes from 724 farms. Only animals with a recorded first calving date were retained for analysis. This criterion allowed for the definition of the first stayability value (Stay2) as the probability of remaining in production until the second calving. Subsequent stayability values were defined analogously, up to Stay10, which represents the likelihood of remaining in production until the tenth calving. Evaluating stayability up to the tenth calving was deemed appropriate, as buffaloes frequently surpass 15 years of age in farm conditions, underscoring the species' intrinsic longevity. The variables Stay2 to Stay10 were treated as binary traits, with the phenotype classified as follows:

- **1 = DELETED:** Assigned to all subjects with a culling date, and this value was applied to all parities occurring after the culling date.
- **2 = PRESENT:** Assigned when a valid calving date was recorded for the corresponding stay value, or when, although no valid culling date was available for the current stay value, a valid culling date was reported for a later parity. In such cases, the missing culling date was considered to be “missing by error,” rather than due to culling.

A value of 0 was assigned to missing data, representing buffaloes for which no culling date was available, as well as those without recorded subsequent calving dates. Additionally, a value of 0 was applied to "censored" data, referring to animals for which no culling date was recorded, likely indicating that they were still alive, possibly remaining on the farm and not yet culled. In such cases, the absence of further calving records was attributed to age-related factors preventing additional reproductive cycles. Stayability values were calculated by dividing the number of buffaloes remaining in the herd at each parity (numerator) by the total number of buffaloes present at first calving, excluding individuals categorized as missing (i.e., those assigned a value of 0) from both that parity and all preceding ones (denominator). The average herd culling rate and parity were computed as weighted averages based on stayability values across parities. Data processing and descriptive statistical analyses were performed using *R* (version 4.3.0) in conjunction with the RStudio IDE (version 2023.12.0.369).

2.2.1 HERITABILITY AND VARIANCE COMPONENTS

The pedigree data for the buffaloes included in this study were obtained from the ANASB dataset, which comprised records on 683,766 individuals, including seven genetic groups, as reported by Gomez et al. (2021). Estimates of heritability, variance components, and the calculation of estimated breeding values (EBVs) for Stay2 to Stay10 were performed using the following programs from the BLUPF90 suite, executed in sequence: RENUMF90, GIBBS1F90+, and POSTGIBBSF90 (Miszta et al., 2024). All processing was performed using the IBiSco (Infrastructure for BIg data and Scientific Computing) cluster resource of the University of Naples Federico II.

The statistical model used for estimation was a Univariate Threshold Animal Model (Model 1):

$$y_{ijkl} = H_i + HYSB_j + a_k + e_{ijkl}$$

where:

- Y is the Stayability value at a given parity (Stay);
- H is the herd fixed effect;
- $HYSB_j$ is the random effect Herd-Year-Season of birth;
- a_k is the random effect of the animal;
- e_{ijkl} is the residual error.

Since milk production is supposed to be one of the main factors influencing farmers' culling decisions, a second model was implemented, incorporating the estimated breeding value (EBV) for milk production as a covariate. Consequently, Model 1 was updated as follows, resulting in Model 2:

$$y_{ijkl} = H_i + HYSB_j + a_k + EBVmilk_{ijkl} + e_{ijkl}$$

where:

- Y is the Stayability value at a given parity (StayL);
- $EBVmilk_{ijkl}$, the Estimated Breeding Value for Milk has been added to the previous model as a covariate.

2.2.2 APPROXIMATE GENETIC CORRELATIONS

Genomic EBVs for milk yields (kg, % and cheese yield) as well as the aggregate selection index (gIBMI) were extracted for proven AI bull from the official genetic evaluations published in October 2023 by ANASB.

Then, approximate genetic correlations were calculated between stayability values obtained within Model1 and all previously related EBVs.

Moreover, approximate genetic correlations were calculated among stayability values obtained within Model1 and Model2 and between the two Model within each parity.

The accuracy of genetic predictions for survival was calculated as (Khansefid et al 2021):

$$accuracy = \sqrt{1 - \frac{PEV_{Sire\ i}}{\sigma_u^2}}$$

Where:

- $PEV_{Sire\ i}$ is the prediction error variance for Sire i ;
- σ_u^2 is the estimated sire genetic variance in the model.

Only sires with stayability EBV accuracies ≥ 20 (n=120) were included in the calculation of genetic correlations. Genetic correlations were computed using the *correlate* function from the *corr* statistical package in R.

2.3 RESULTS AND DISCUSSION

Table 1 presents the number of buffaloes recorded at each parity (Total), the number of buffaloes classified as missing and thus excluded from the stayability calculation (Missing), the number of buffaloes still present in the herd at the end of each lactation (Alive), the number of buffaloes removed prior to reaching that parity (Departed), and the corresponding stayability value (Stay).

Table 1 Total number of buffaloes at the different parity, culled at the end of lactation prior to that parity (Culled), number of missing subjects (Missing), number of buffalo still alive at the end of that lactation (Alive), and the corresponding stayability value (Stay).

PARITY	Total (n.)	Missing (n.)	Culled (n.)	Alive (n.)	Stay	Culling rate
2	211,266	17,567	36,117	157,582	0.813	0.187
3	157,582	12,879	30,678	114,025	0.630	0.183
4	114,025	8,432	24,505	81,088	0.470	0.160
5	81,088	5,371	18,394	57,323	0.343	0.127
6	57,323	3,848	14,151	39,324	0.241	0.102
7	39,324	3,118	10,341	25,865	0.161	0.080
8	25,865	2,235	7,477	16,153	0.102	0.059
9	16,153	1,403	5,210	9,540	0.059	0.043
10	9,540	901	3,373	5,266	0.033	0.026

Stayability shows a decreasing trend across successive parities, with a remarkable starting decline from 0.81 between the first and second parity, halving by Stay4, and reaching 0.03 at Stay10. This pattern suggests that, although rare, some individuals remain in the herd beyond the tenth parity. As illustrated in Figure 1, the trend of *Stay* values follows a logarithmic decreasing function as parity increases:

$$y = -0.376\ln(x) + 0.852 (R^2 = 0.991)$$

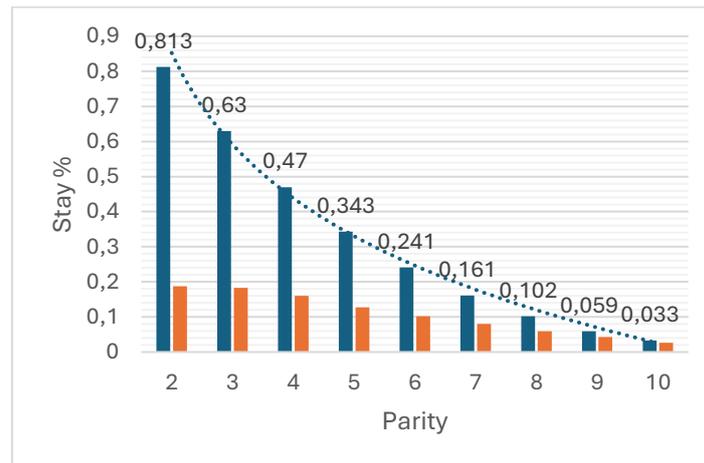
The culling rate is highest between the first and second parity and between the second and third parity (18%), decreases slightly between the third and fourth parity (16%), and then progressively declines by approximately 0.2–0.3 percentage points per parity up to the tenth parity. The mean annual culling rate was 0.15, while the average parity within the dataset was 3.8.

The phenotype stayability was estimated using the full dataset available from ANASB, providing a reliable indicator of stayability for intensive dairy buffalo farming systems. The decision to evaluate stayability up to the tenth parity was based on evidence supporting the greater longevity of buffaloes compared to dairy cattle. It is well established that buffaloes exhibit a significantly higher herd retention rate than dairy cows, with lower overall production levels despite the superior chemical composition of their milk (Zicarelli, 2021; Di Palo, 1997). From an economic perspective, the buffalo species has traditionally compensated for lower milk yields through reduced fixed costs, particularly in terms of replacement management. The overall mean culling rate observed in this study was 15%, slightly higher than that previously reported by Zicarelli (1997).

The data analyzed in this study were derived from buffaloes born since 2005, covering a period of approximately 20 years. During this time, the culling rate increased from approximately 8–10% up until 2009 to around 30% in recent years. Additionally, the average age at calving on farms gradually decreased from approximately 7 years in 2005, stabilizing at around 6 years in 2023 (AIA, 2024). This data has been confirmed by Gomez et al., (2025) that reported an average length of productive life of 7.61 years in IMBs born from 1982 to 2019. In recent years, the need to shorten the generational interval to accelerate genetic progress and achieve higher production levels primarily through the use of artificial insemination with genetically proven sires has led to an increase in the replacement rate, primarily driven by a rise in voluntary culling. Nevertheless, these replacement

rates remain lower than those reported for intensively selected dairy cattle herds (Owusu-Sekyere et al., 2022; Hu, 2021).

Figure 1. Trend of the Stay % value over parity (blue columns) and relative drop % over the parity (orange columns).



Low production levels is probably the primary reason for culling in buffalo herds, as reported by Gomez et al. (2025) and as also has been reported in dairy cow (Vukasinovic et al., 2001, Sewalem et al., 2005, Morek-Kopeć and Zarnecki, 2017, Rostellato et al., 2021). Poor production performance in primiparous buffaloes could be the leading cause of higher culling after the first or second calving even if primiparous buffaloes that compensate for low milk production through efficient reproductive performance demonstrating an optimal calving interval are often retained for an additional lactation. This pattern explains the high replacement rate observed among primiparous and second-parity buffaloes. Beyond third parity, culling decisions are increasingly influenced by factors other than genetic merit. These include reproductive inefficiencies, health disorders, and metabolic conditions, which, as highlighted by Dekkers et al. (1994), are also critical determinants of stayability and culling rates in dairy cattle.

Another significant factor contributing to voluntary culling in buffalo herds is reproductive inefficiency, particularly when it extends the calving interval. This issue is particularly critical in buffalo farming due to the necessity of out-of-season breeding to prevent excessive milk production

during the winter months. Farmers aim to align production with market demand, which peaks in summer or, at most, remains evenly distributed throughout the year (Zicarelli, 2021). The reproductive seasonality of buffaloes, regulated by negative photoperiod sensitivity, needs restricting reproductive activity during the autumn-winter period to prevent milk surpluses during periods of lower market demand. This reproductive constraint may explain the higher average herd age observed in buffaloes compared to intensively selected dairy cattle breeds. Farmers tend to cull low-producing buffaloes at an early age while retaining animals that exhibit superior reproductive performance, even at higher parities. The stayability values observed in dairy buffaloes appear to be consistent with previous studies on stayability in dairy cattle. In Holstein cows, Hardie et al. (2021) reported stayability values that, when adjusted to align with the calculation methods used in this study, exhibit a similar pattern, as their initial stayability value was set at birth. In Jersey cows, slightly lower stayability values were recorded, with 77.9% retention at first parity and 58.02% at second parity, followed by a sharper decline to 0.1% by the tenth parity (Fabbris, 2025). Currently, no directly comparable findings on stayability values in buffaloes exist. However, Galeazzi et al. (2010), who assessed stayability as the probability of remaining in the herd in subsequent years after first calving, reported retention rates of 85% one year after first calving and 63% three years post-first calving.

2.3.1 HERITABILITY AND MODEL COMPARISON

Table 2 presents the results from the two models (Model 1 and Model 2) used to estimate the herd effects (Herd), herd-year-season effects (HYS), and heritability (h^2) values for *Stay* and *StayL* across different parities. The heritability estimates for stayability (h^2) ranged from 0.09 to 0.11 in Model 1 and from 0.07 to 0.12 in Model 2, where milk EBV was included as a covariate. The influence of HYS decreased as parity order increased in both models, with a more pronounced reduction observed in the model incorporating milk production. While the percentage contribution of HYS remained relatively stable across both models, its influence declined in higher parities. Figure 2 shows the estimated h^2 values for Model 1 and Model 2. In general, an increase in h^2 can be observed as parity order rises, though the values do not overlap completely.

Table 2. Variance components, h² value and HYS percentage of total variance in Model 1 and Model 2

MODEL 1					
STAY	Herd	HYS	h²	Confidence Interval (95%)	% HYS
2	0.142	0.436	0.090	0.067 - 0.112	0.276
3	0.129	0.452	0.081	0.063 - 0.099	0.286
4	0.131	0.419	0.084	0.067 - 0.101	0.270
5	0.127	0.387	0.084	0.068 - 0.099	0.256
6	0.136	0.359	0.091	0.073 - 0.107	0.240
7	0.097	0.316	0.069	0.054 - 0.082	0.223
8	0.103	0.292	0.074	0.046 - 0.100	0.209
9	0.119	0.250	0.087	0.051 - 0.121	0.183
10	0.161	0.243	0.114	0.070 - 0.157	0.173

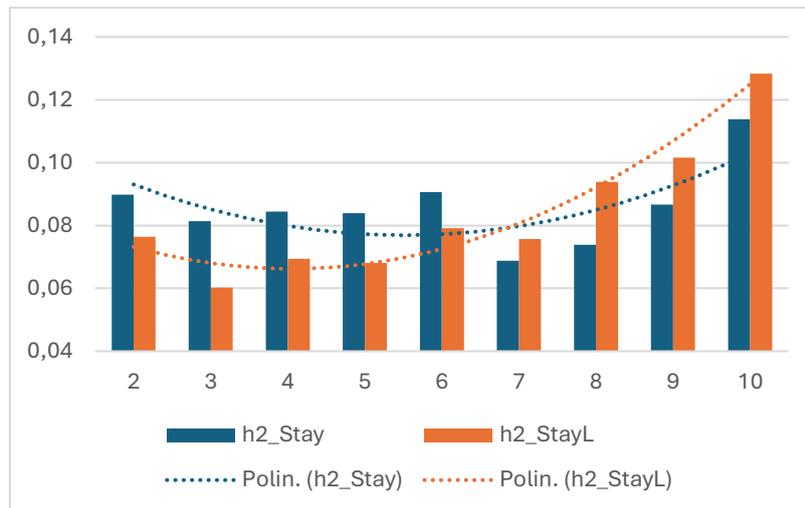
MODEL 2					
STAYL	Herd	HYS	h²	Confidence Interval (95%)	% HYS
2	0.119	0.428	0.076	0.049 - 0.103	0.277
3	0.093	0.445	0.060	0.043 - 0.076	0.289
4	0.106	0.421	0.069	0.054 - 0.084	0.276
5	0.102	0.392	0.068	0.052 - 0.083	0.263
6	0.118	0.368	0.079	0.063 - 0.095	0.248
7	0.109	0.332	0.076	0.059 - 0.092	0.230
8	0.136	0.310	0.094	0.071 - 0.116	0.214
9	0.143	0.261	0.102	0.073 - 0.129	0.186
10	0.185	0.254	0.128	0.089 - 0.167	0.176

Specifically, in Model 1, the heritability trend remained nearly constant from the 2nd to the 6th parity, then decreased at the 7th parity, followed by an upward trend through the 10th parity.

In Model 2, the estimated h² values were lower than those in Model 1 from the 2nd to the 6th parity but became higher from the 7th parity onward, while maintaining a similar overall trend. In Model 1, the HYS value progressively decreased from 0.436 at Parity 2 to 0.243 at Parity 10, with its percentage contribution declining from 27.6% at Parity 2 to 17.3% at Parity 10, indicating a reduced influence of HYS as parity order increased. Similarly, in Model 2, the HYS value followed the same pattern, decreasing from 0.428 at Parity 2 to 0.254 at Parity 10, with the percentage contribution dropping from 27.7% at Parity 2 to 17.6% at Parity 10.

As expected, the estimated heritability values were relatively low, agreeing with previous findings for other dairy species, such as Holstein cows (Hardie et al., 2021; Kern et al., 2014). In other buffalo breeds, such as Murrah, slightly higher heritability estimates have been reported (0.11- 0.23; Galeazzi et al., 2010). However, in those studies, the definition of the stayability was different. Highest h^2 values observed in model1 at early parities confirm and partially explain the previously discussed role of production levels in farmers' culling decisions. Specifically, the higher h^2 values found in this model suggest that stayability is inflated by the effect of production level, that in the model2 was adjusted for.

Fig. 2 Model1 and Model 2 h^2 values of Stay and StayL



From the 7th parity onward, the trend reversed, as animals remaining in the herd having been extensively selected for their production levels, exhibited longevity-related traits, particularly reproductive efficiency and genetic quality that justified farmer decision to maximize the benefits of such animals' offspring. The higher HYS component indicates that a large part of the variation in the observed data is due to environmental factors rather than genetic factors. While it decreased progressively for higher parities, the relative importance of HYS remained relatively stable in both models.

2.3.2 APPROXIMATE GENETIC CORRELATIONS

The approximated genetic correlations among stayability and EBVs for milk production (gMilk) were positive across all stayability classes, with the highest r values observed between Stay2 and Stay5, with an initial decrease followed by stabilisation and a subsequent decline up to Stay10. The observed trend is mainly affected by voluntary culling decisions made by farmers. Indeed, they use to remove less productive animals, particularly in the early parities. In later parities, other factors probably come into play and become predominant in culling decisions, such as reproductive efficiency or health issues, loosening the association between production and longevity and eventually reversing its direction. The observed findings align with previous studies (Fabbris et al., 2025), which reported that high milk production in the early stages does not always translate into long-term longevity due to the increasing metabolic and health demands over time.

Table 3. Approximate genetic correlation among stayability EBVs, buffalo aggregate index (gIBMI), and EBVs for milk related traits.

	Stayability								
	2	3	4	5	6	7	8	9	10
gIBMI ¹	0.05	-0.01	-0.03	0.01	-0.17	-0.05	-0.11	-0.25	-0.25
gMilk ²	0.38	0.19	0.19	0.24	0.13	0.13	0.06	-0.15	-0.10
gFat% ³	-0.35	-0.20	-0.24	-0.22	-0.22	-0.16	-0.13	-0.06	-0.13
gProtein% ⁴	-0.30	-0.17	-0.19	-0.21	-0.24	-0.12	-0.19	-0.11	-0.09
gFatKg ⁵	0.31	0.15	0.12	0.21	0.07	0.10	0.02	-0.20	-0.14
gProteinKg ⁶	0.34	0.16	0.15	0.22	0.08	0.11	0.02	-0.20	-0.12
gCheseeYield ⁷	-0.35	-0.19	-0.23	-0.22	-0.25	-0.16	-0.18	-0.10	-0.09

¹ ANASB Aggregate Index; ²EBV kg milk yield; ³EBV % milk fat; ⁴EBV % milk protein; ⁵ EBV kg milk fat; ⁶ EBV kg milk protein; ⁷ EBV g/kg cheese yield

The approximated genetic correlations between stayability and fat and protein percentage EBVs (gFat% and gProtein%) were remarkably negative, i.e. unfavorable, for both traits, with the highest values at Stay2 and a progressive decline up to Stay10. The decreasing trend across stayability classes

was more marked for gProtein% and slightly less steep for gFat%. The inverse relationship between milk yield and its fat and protein composition partially explains the negative correlation between milk composition and stayability, as most farmers prefer to retain more productive animals, prioritizing milk quantity over composition. The less marked reduction in the association between gProtein% and stayability compared to gFat% could be linked to greater metabolic challenges in buffaloes with high milk fat levels than in buffaloes with high protein levels, eventually affecting their productive longevity. These findings agree with previous studies (Fabbris, 2025; Vitali et al., 2017), suggesting that excessive fat and protein concentrations in milk may be associated with an increased risk of metabolic disorders, negatively impacting overall health and productive lifespan.

The positive correlations between both fat and protein yield (gFatKg and gProteinKg) and stayability in the early stages of productive life (Stay2, Stay3, Stay4), ranging from $r = 0.34$ to $r = 0.21$ for gFatKg and from $r = 0.34$ to $r = 0.15$ for gProteinKg, follow what observed between stayability and gMilk. The negative correlations observed at Stay9 and Stay10 further confirm that longer-lived animals remain in production due to factors beyond their production levels alone. As Fabbris et al. (2025) also found, the balance between productivity and health is indeed crucial in determining buffalo longevity.

The genetic index for cheese yield (gCheeseYield%), which measured the genetic potential to convert milk into Mozzarella, showed constant and negative correlations with stayability ($r = -0.35$ to $r = -0.09$), specular to the trends observed for gProtein% and gFat%. Notably, the correlation values between gCheeseYield% and stayability appear to synthesize the trends of the two qualitative indices, offering a potential insight into its relevance in selection strategies.

Approximate genetic correlations among the stayability values within Model 1 (below the diagonal) and Model 2 (above the diagonal), as well as between the two models (on the diagonal), are presented in Table 4.

Table 4. Approximate genetic correlations among the stayability EBVs from Model 1 (below the diagonal) and Model 2 (above the diagonal), as well as between the two models (on the diagonal)

		Stayability (Model2)								
		2	3	4	5	6	7	8	9	10
Stayability (Model 1)	2	0.940	0.644	0.488	0.420	0.322	0.355	0.309	0.286	0.284
	3	0.756	0.864	0.848	0.782	0.704	0.680	0.633	0.630	0.633
	4	0.660	0.849	0.773	0.931	0.888	0.831	0.761	0.766	0.737
	5	0.586	0.766	0.921	0.725	0.946	0.911	0.858	0.842	0.802
	6	0.510	0.689	0.864	0.929	0.721	0.937	0.888	0.881	0.830
	7	0.504	0.671	0.772	0.862	0.900	0.705	0.964	0.941	0.896
	8	0.367	0.521	0.655	0.783	0.822	0.931	0.768	0.941	0.881
	9	0.243	0.474	0.558	0.686	0.739	0.850	0.900	0.898	0.928
	10	0.285	0.482	0.549	0.637	0.680	0.818	0.850	0.901	0.890

Intra-stay correlations were generally high between consecutive parities but tended to decrease as the interval between parities increased. Higher correlation values were observed for Stay2 in Model 2 than in Model 1, whereas for all other stayability values, correlations were higher in Model 1 than in Model 2. The lowest approximate genetic correlations were found for Stay2 in both models, whereas higher correlations were observed for the remaining stayability values. Similar genetic correlation values between stayability traits were reported by Nascimento et al. (2022) in Jersey cows, suggesting that stayability could serve as an appropriate selection criterion for longevity.

Significant and high values of approximate genetic correlations between stayability traits could be utilized to predict longevity at an earlier stage in buffaloes. However, a more accurate prediction of survival can be achieved from the third parity onward, as the genetic correlation with the tenth parity ranges between 0.6 and 0.92. The genetic correlations along the diagonal between Model 1 and Model 2 are notably high, particularly for Stay2 and Stay3 (94% and 89%, respectively), as well as for the later parities, Stay9 and Stay10 (both 89%). The factors influencing culling decisions at different parities are clearly distinct, and this is reflected in the relationships between their respective heritability values. This distinction is particularly evident in Model 2, where the removal of the milk yield effect allows for a greater emphasis on genetic factors associated with an animal's ability to survive and remain productive in good health.

Survival, however, is influenced by managerial decisions voluntarily made by farmers, which are, in turn, linked to farm-specific contingencies that may vary across years and between farms. These factors contribute to increased variability, thereby lowering the correlation coefficients between stayability traits. Notably, the higher correlation values obtained with Model 2 are strongly influenced by the production level of the animal, which plays a crucial role in determining its likelihood of being retained within the herd. This underscores the impact of milk yield on culling decisions and the genetic relationships associated with stayability.

2.4 CONCLUSION

This study provides valuable insights into the dynamics of buffalo stayability, with significant implications for breeding and management strategies in intensive dairy buffalo farming. It highlights the need for a balanced approach that includes both production traits and health/reproductive efficiency to ensure the longevity of animals in the herd.

Buffaloes generally have lower stayability values as they progress through the parities, with culling decisions primarily driven by production levels in early parities and health/reproductive issues in later ones. This highlights the importance of managing production alongside health and reproductive efficiency to maximize herd longevity.

The heritability for stayability in buffaloes is relatively low, suggesting that environmental factors play a major role in herd retention decisions. A positive correlation was found between EBVs for early stage stayability (Stay2, Stay3) and milk production; this is an interesting point that needs to be verified in further studies, as these results could be biased by the farmer's decision to remove less productive buffaloes, erroneously suggesting that selective breeding for higher productivity can improve stayability in the short term. However, for long-term stayability, it would be essential to also consider traits related to reproductive efficiency and overall health.

The slightly declining influence of herd-year-season (HYS) effects as parity increases emphasizes the growing importance of individual buffaloes' characteristics, such as their genetic potential for survival and reproduction, as they age. This suggests that long-term herd management should focus on identifying animals with good reproductive efficiency and health traits, rather than solely prioritizing milk production.

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PART 2:

THE ROLE OF LINEAR TYPE TRAITS IN PREDICTING GENETIC MERIT FOR LONGEVITY IN ITALIAN MEDITERRANEAN BUFFALOES

3. INTRODUCTION

The National Association of Buffalo Breeders (ANASB) has managed the studbook of the Italian Mediterranean Buffalo since 2000 and oversees selection programs for the genetic improvement of the species across 1,025 farms, encompassing over 131,000 animals. The selection scheme includes also the assessment of type traits to all primiparous buffaloes, being conducted by trained personnel using linear type trait scoring.

The introduction of type trait selection based on linear criteria into the genetic evaluation of the Italian Mediterranean Buffalo (IMB) in 2005 represented the first systematic approach aimed at breeding animals that are more resilient to evolving farming conditions and better suited to higher production levels. The implementation of linear type trait selection in the IMB has led to improvements in overall conformation, trunk size, limb structure, and udder characteristics. Approximately 36% of the traits included in the IMB linear evaluation system pertain to udder size and conformation, with the objective of enhancing anatomical features that influence milk production efficiency.

The final morphological score, which expresses the overall merit of a dairy buffalo's conformation, is calculated as a weighted average of four partial scores: structure (10%), legs and feet (30%), udder (40%), and production potential (20%). To date, estimated breeding values have been computed for the overall score, the four partial scores, and specific traits related to udder morphology, chest depth, and limb structure.

The morphological impact on an animal's productive lifespan is well-documented, particularly in dairy cattle, where numerous functional estimated breeding values are employed in genetic selection to optimize longevity. Many of these indices derive their predictive value from morphological traits, which are combined and weighted according to their relevance (Cole & VanRaden, 2018). Since 2018, certain aspects of the IMB type trait evaluation have been integrated into the selection program (Biffani et al., 2021).

More recently, these traits have also been incorporated into the genomic index used by ANASB since 2024. This index aims to enhance milk yield and quality while ensuring desirable structural conformation, including harmonious and proportionate body structure, a strong topline, a wide and deep chest, and optimal angularity. In addition, ANASB type traits for well conformed IMB feet and legs are strong pasterns and well developed heels, while the udder should be firmly attached to the abdomen with sufficient volume in the rear dimensions and placement above the hocks. (Source: ANASB online: <https://www.anasb.it/attivita-anasb/valutazioni-morfologiche/>). The selection of type

traits aims to develop animals that are better adapted to production, making them more resilient to varying productivity levels and diverse farming conditions. Evidence from dairy cattle breeding suggests that increased milk production may lead to a reduction in the average productive lifespan of females due to established relationships between morphological or genetic traits and various health disorders (Kern et al., 2015). Indeed, traits related to udder morphology and limb structure have been found to correlate with health issues affecting udder integrity and locomotion, respectively, although with varying degrees of significance (Caraviello et al., 2003).

Numerous studies in dairy cattle have highlighted weak to moderate genetic correlations between morphological traits and longevity over the years (Dekkers & Jairath, 1994; Essl, 1998; Vollema, 1998; Vollema & Groen, 1997; Weigel et al., 1998; Buenger et al., 2001; Larroque & Ducrocq, 2001; Williams et al., 2022). However, studies focusing on the buffalo species that investigate the relationships between morphology, production, and longevity are scarce limited. Zhang et al. (2022; 2023) examined the correlations between morphological traits, milk production, and udder health providing theoretical support for early selection with desirable milk performance or less susceptible to mastitis, while further research on the relationship between morphology and longevity is warranted.

3.1 AIM OF THE RESEARCH

The objective of this study was to analyze the relationships between morphology and longevity, the latter being defined as stayability at different parities. Two different approaches were used. The first was based on a mixed effects logistic regression which model binary outcome variables like stayability, i.e. the success or failure to remain in the herd until a given time point. The goal was to identify which morphological traits are most relevant for ensuring prolonged productive life. The second approach was based on the use of approximate genetic correlations among stayability AI bulls'EBVs estimated in part 1 and their official EBVs for linear traits currently available at ANASB. Both approaches, in spite of their simplicity, could further contribute to understanding how selection strategies should be directed to enhance longevity.

3.2 MATERIALS AND METHODS

The dataset used in this study was obtained from ANASB and included stayability information, from stayability 2 to stayability 10, for 211,266 buffaloes. Stayability was defined as the success (1) or failure (0) to remain in the herd until a given time point (e.g. second calving). This dataset was then merged with type trait evaluations and test-day milk yield data. Animals were filtered based on birth year, and only buffaloes with type trait evaluations recorded from 2010 onward, and born between 2005 and 2020, were included in the analysis. When multiple type trait evaluations were available for the same animal, only the most recent record was considered.

The mean age at the time of type trait evaluation was 51.3 months (± 20.9). Production data were 270 days standard lactation records. If production data were unavailable for a given lactation, the average lactation yield of contemporary animals for that specific parity was used. The final dataset included 51,702 buffaloes from 430 farms.

The number of records and herds by each classifier are in Table 1

Table 1. Number of animals and farms per classifier

ID Classifier	Buffalos recorded	Herd visited
1	3356	124
2	15381	263
3	9749	188
4	2425	89
5	1111	63
6	3341	122
7	611	15
8	7548	206
9	3782	138
10	4398	140

Contemporary group was also defined as classifier-herd-date of farm visit (HYI). Contemporary groups with fewer than 10 buffaloes were excluded from the analysis.

After data editing, stayability records with corresponding type trait evaluations and 270-day milk yield records were available for 169,664 lactations (from first to tenth parity) across 51,702 buffaloes. The number of animals per stayability are in Table 2.

Table 2. Number of buffaloes from the first to tenth parity

Parity	Buffaloes
1	51,702
2	49667
3	39686
4	28949
5	20286
6	13836
7	8601
8	4865
9	2591
10	1183

3.2.1 MIXED LOGISTIC REGRESSION

A generalized linear mixed model was used to fit a logistic regression model with random effects.

The R statistical package lme4 (version 1.1-35.1) was used.

The fitted model was:

$$\left[\log \left(\frac{P(Y_i = 1)}{1 - P(Y_i = 1)} \right) \right] = \beta_0 + \sum_{j=1}^p \beta_j X_{ij} + u_k [u_k \sim \mathcal{N}(0, \sigma^2)] + e$$

where:

- $\log \left(\frac{P(Y_i=1)}{1-P(Y_i=1)} \right)$ is the log-odds of the probability that the outcome Y_i (Stay2 to Stay10) is 1.
- X_{ij} are a set of fixed effects.
- β_0 is the fixed intercept.
- β_j are the coefficients of the fixed effects.
- $u_k \sim \mathcal{N}(0, \sigma^2)$ is the random effect for the group HYI contemporaries
- e is the error

Fixed effects included in the model were age at first parity (in months) and type traits. A first model included 13 body related type traits: stature, chest width, chest depth, trunk line, trunk length, rump length, rump angle, ilei width, ischi width, body angularity, hock angle, fetlock strength, heel height. A second model included 8 udder related type traits: udder front attachment length, udder rear attachment width, udder depth, udder rear attachment height, udder support, teat direction, teat length, and teat placement.

All traits were recorded on a scale from 1 to 50. Table 3 presents the mean (\pm standard deviation) of the linear scores, along with descriptions of the extremes of character variability. Contemporary group, as previously defined, was included as random effect.

To check for collinearity among the predictor variables in the generalized mixed model glmer, the Variance Inflation Factor (VIF) was calculated using the car package on an equivalent generalized regression model (glm) without random effects. The average VIF was consistently below 5, with no variable exceeding this critical threshold, indicating an acceptable level of collinearity (Fox and Monette, 1992).

Statistical analyses, including data processing and descriptive statistics, were performed using the R statistical software (version 4.3.0) via the RStudio integrated development environment (IDE) (version 2023.12.0.369).

Table 3. List of all linear type traits with explanation of measurement scale and the corresponding mean \pm sd.

Linear type trait	Score		Mean	\pm sd
	1	50		
stature	low	high	28.61	6.11
chest width	narrow	wide	25.80	5.85
chest depth	shallow	deep	28.78	6.11
dorsal line	curve	straight	24.09	3.83
trunk length	short	long	29.92	5.82
rump length	short	long	28.25	5.22
rump angle	high	wide	24.26	5.87
ilei width	narrow	wide	28.70	6.10
ischi width	narrow	wide	25.27	7.16
body angularity	low	high	28.72	5.52
hock angle	shallow	steep	29.01	4.34
fetlock strenght	weak	strong	21.68	6.00
heel height	low	high	21.56	7.35
udder front attachment length	short	long	21.84	7.39
udder rear attachment width	narrow	wide	23.41	6.21
udder depth	low	high	28.86	6.63
udder rear attachment height	low	high	24.38	6.67
udder support	broken	strong	18.05	8.13
teat direction	divergent	convergent	21.14	4.85
teat lenght	short	long	25.15	6.73
teat placement	close	apart	18.32	6.26

3.2.2 APPROXIMATE GENETIC CORRELATIONS

Approximate genetic correlations were calculated among stayability EBVs estimated in part 1 (with and without adjustment for milk EBV) and ANASB official EBVs for linear type traits. Only EBVs from proven AI bulls with a minimum accuracy of 20 (n=120) for Stayability EBVs were used.

3.3 RESULTS

3.3.1 MIXED LOGISTIC REGRESSION

Results from the mixed logistic regression are summarized for all observed stayabilities in Table 4, which illustrates odds-ratio and significance level

Table 4. Estimated Odds Ratio of survival in the tenth stay for the body type trait score

Body type trait	OR								
	STAY2	STAY3	STAY4	STAY5	STAY6	STAY7	STAY8	STAY9	STAY10
age at first parity	0.80***	0.82***	0.83***	0.82***	0.79***	0.76***	0.75***	0.71***	0.71***
stature	1.01	1.01	1.00	1.00	1.01	1.04	1.02	1.03	1.05
chest width	1.1***	1.07***	1.07***	1.07***	1.10***	1.11***	1.11**	1.07	1.06
chest depth	1.13***	1.18***	1.15***	1.16***	1.14***	1.12***	1.08*	1.12*	1.04*
dorsal line	0.97.	0.99	1.00	0.99	0.99	0.99	0.98	0.98	0.99
trunk length	1.16***	1.13***	1.1***	1.08***	1.05*	1.02	1.06	1.09	1.11
ilei width	1.16***	1.13***	1.14***	1.12***	1.07**	1.05.	1.01	1.05	1.08
ischium width	1.16***	1.17***	1.13***	1.10***	1.11***	1.14***	1.15***	1.13**	1.17**
rump length	1.02	0.99	0.99	1.00	1.00	1.00	1.02	0.97	1.00
rump angle	0.91***	0.90***	0.90***	0.91***	0.89***	0.88***	0.86***	0.85***	0.83***
body angularity	1.07**	1.09***	1.10***	1.11***	1.11***	1.10***	1.12***	1.10*	1.06*
heel height	1.05*	1.03	1.04*	1.06*	1.05*	1.06*	1.08*	1.05	0.98
fetlock strength	0.92***	0.93***	0.95**	0.95**	0.98	0.98	0.97	1.00	1.04
hock angle	0.94**	0.97*	0.97	0.97	0.97	1.00	0.97	0.98	0.98
HYI	3.28	3.6	3.79	4.06	4.4	4.72	5.34	6.85	8.9
Udder type trait	OR								
	STAY2	STAY3	STAY4	STAY5	STAY6	STAY7	STAY8	STAY9	STAY10
age at first parity	0.87***	0.88***	0.88***	0.86***	0.83***	0.79***	0.77***	0.73***	0.73***
front udder length	1.21***	1.15***	1.15***	1.14***	1.13***	1.14***	1.14***	1.15***	1.22**
rear udder width	1.37***	1.36***	1.31***	1.28***	1.28***	1.28***	1.22***	1.14**	1.09
rear udder height	0.96*	0.96**	0.98	0.98	0.99	0.98	0.98	0.98	0.94
udder depth	0.84***	0.85***	0.84***	0.87***	0.88***	0.90***	0.89***	0.91*	0.92
teat length	1.11***	1.12***	1.11***	1.11***	1.08***	1.08***	1.09**	1.12**	1.07
teat direction	1.01	1.01	1.03*	1.02	1.03.	1.03	0.98	1.04	1.11.
teat placement	1.07**	1.06***	1.05***	1.07***	1.06***	1.05*	1.07*	1.04	0.98
udder support	0.95*	0.94***	0.93***	0.93***	0.93***	0.93***	0.98	0.96	0.94
HYI	3.28	3,57	3.80	4,08	4.39	4.71	5.31	6.78	8.82

Significance level : ≤ 0.001 ***; ≤ 0.01 ** ; ≤ 0.05 *

Age at First Parity

Age at first parity was consistently and significantly inversely associated with survival across all parities. The risk for culling increased linearly with increasing parities. These results agree with previous studies in dairy cattle (Rogers et al. 1991; Hultgren & Svensson, 2009; Sherwin et al. 2016; Eastham et al. 2018; Owusu-Sekyere et al., 2022), where an increased age at first calving negatively affects longevity. A possible explanation can be related to a higher risk of calving complications in older heifers, who are prone to accumulate excessive body fat.

Body-Related Linear Type Traits

A higher odds ratio for survival was observed in buffaloes with greater chest depth and width, as well as increased body angularity. These findings are consistent with the results of Gómez-Carpio et al. (2025), who investigated similar relationships between type traits and survival using a Weibull hazard analysis. A broad thorax, particularly in the fore-chest, combined with a well-developed trunk that facilitates higher production without compromising health status, contributes to increased longevity, as previously observed in dairy cows (Veerkamp, 1998; Hiendleder et al., 2003). Moreover, Zhang et al. (2022) reported that chest depth was positively correlated with milk production in buffaloes, while heart girth exhibited a significant positive correlation with milk protein yield a relationship also observed in sheep (Adewumi, 2012).

Culling decisions in buffalo farming, aside from mortality-related causes, are primarily driven by low milk production, reproductive issues, and poor health status factors similar to those reported in dairy cattle (Fetrow et al., 2006; Ahlman et al., 2011) and Murrah buffaloes (Galeazzi et al., 2010). In agreement with previous studies on dairy cows (Caraviello et al., 2003; Sewalem et al., 2004) and buffaloes (Gómez-Carpio et al., 2025), stature was found to be the least influential trait in determining survival.

The odds ratio for rump conformation indicated a higher probability of survival in buffaloes with broad ilei, sufficient ischial width, and, most notably, a less sloped rump, with survival probability increasing with parity. Similar findings were reported by Gómez-Carpio et al. (2025) for buffaloes and by Nascimiento et al. (2022) in Jersey cows. Furthermore, Zhang et al. (2022) found a positive correlation between rump traits and milk production. A well-conformed rump broad in both the fore

and hindquarters, with minimal angulation supports longevity by facilitating ease of calving and enhancing reproductive efficiency in dairy cattle (Sewalem et al., 2004).

Linear Type Traits Related to Feet and Legs

Among the traits within this category, heel height exhibited a significant and positive association with survival up to at least the eighth calving. In contrast, hock angle showed a weaker inverse relationship with survival from the second to the third parity, suggesting that a wider hock structure contributes to improved limb conformation. The observed negative relationship between fetlock strength and survival suggests that reduced rigidity in the fetlock joint is essential for enhancing longevity, particularly up to the eighth parity. Moreover, a wider foot angle, in conjunction with short fetlocks, compromises proper limb support, resulting in a rigid gait and impaired locomotion. Similar findings have been reported by Gomez-Carpio et al. (2025) in buffaloes.

Udder-Related Linear Type Traits

Among the eight udder traits, seven were statistically significant. Positive correlations were observed for front udder length, rear udder width, teat length, and teat placement, although the associations for teat length and teat placement were comparatively weaker. Notably, suboptimal teat placement was associated with diminished efficiency in machine milking and a higher incidence of subclinical mastitis (Singh et al., 2017), a factor that has also been linked to survival in Jersey cattle (Nascimientto et al., 2022). In contrast, udder depth exhibited a strongly significant negative correlation with stayability, while rear udder height and udder support were characterized by weaker negative correlations.

3.3.2 APPROXIMATE GENETIC CORRELATIONS

Body-Related Linear Type Traits

The estimated genetic correlations among stayabilities (from 2 to 10) and the body-related linear type traits are in figure 1. Orange and Blue lines are from stayability EBVs adjusted or not for Milk EBVs, respectively. Focusing on stature, chest width, and trunk length (see Figure 1) we can observe distinct trends between the two models. Indeed, in Model 1 the correlations were relatively low, ranging from

0.1 to -0.1, whereas in Model 2 they were constantly negative, with values spanning from -0.1 to -0.3. Although Model 1 showed a low positive correlation with survival, these values were not statistically significant.

Consequently, a higher EBV for stayability was observed in individuals characterized by a shorter stature, a narrower chest, and a shorter trunk. Similar estimates of genetic correlations were obtained for both models with respect to the dorsal line, chest depth, body angularity, rump slope, the width of the ilei and ischium, and rump length. A less pronounced dorsal line was associated with survival especially during early parities, with the correlation decreasing beyond the third parity. Similarly, although the correlation was weak, a deeper chest appeared to be linked to enhanced survival after the third parity. Notably, increased survival was also correlated with reduced angularity, particularly in Model 2 (Figure 1), a finding that is consistent with the results reported by Williams et al. (2022).

Regarding rump characteristics, reduced anterior and posterior widths were associated with improved survival, especially in Model 2 (Figure 2). In both models, rump length has an unfavorable correlation with survival, with a more pronounced effect in Model 2. Interestingly, the estimated genetic correlations for rump width deviated from the outcomes observed in the phenotypic analysis. Although a wider rump is generally associated with easy calving in dairy cows, it also reflects overall trunk conformation, and larger animals are prone to reduced longevity. In buffaloes, a rump characterized by a proportional diameter and a more compact structure at the ischium has been shown to decrease the likelihood of uterine prolapse (Zicarelli et al., 2010), a condition that remains prevalent in buffalo herds and constitutes a primary factor in premature culling.

The slope of the rump was found to be inversely correlated with stayability at least up to the fourth parity, suggesting that increased longevity is associated with a less pronounced rump slope, particularly in lower parities. However, results from Model 2 are slightly different from Model 1, especially at higher parities, where results are reversed. Generally, the slope of the rump is related to reproductive efficiency by facilitating proper uterine drainage and easing the calving process. However, its effect can change over time due to age-related variations that inevitably alter the expression of conformation traits, as suggested by Williams et al. (2022) in the context of various type traits in dairy cows.

Feet and Leg-Related Linear Type Traits

When moving to the relationship with feet and legs, the hock angle showed an increasingly negative estimated genetic correlation with parity. This pattern suggests that, as parity increases, there is a genetic predisposition toward a wider hock angle. On the contrary, heel height exhibited a weaker yet positive genetic correlation with survival. This result suggest, suggesting that a higher stayability EBV in later parities is associated with increased heel height. The estimated genetic correlation between fetlock strength and survival was weak across all parities, as can be observed in Figure 3. Overall, the results were largely consistent across both models, apart hock angle, which showed decreasing correlations in Model 2 between the sixth and ninth parities.

Udder-Related Linear Type Traits

In Model 1, a positive approximate genetic correlation was observed between survival and front udder length, whereas in Model 2 this correlation was negligible (see Figure 4). Additionally, both rear udder width and udder support were positively correlated with survival in Model 1; however, in Model 2 these traits exhibited an inverse relationship with survival beginning at the third parity. Rear udder height strongly suggested a negative correlation with survival across both models, with the magnitude of this effect increasing in later parities. A rear udder height positioned in close proximity to the vulva has been hypothesized to adversely affect udder health in Jersey cows by increasing the risk of mastitis due to enhanced exposure to fecal pathogens (Williams et al., 2022).

Furthermore, udder depth was significantly and negatively correlated with survival in both models, although the strength of this correlation was reduced in Model 2 (see Figure 4). This finding suggests that buffaloes with deeper udders tend to experience longer lifespans. It is noteworthy that deeper udders are often associated with higher milk production, as evidenced by phenotypic correlations, and that more productive buffaloes are less likely to be culled. Indeed, Table 1 shows that the average linear score for udder depth in buffaloes corresponds to an udder position exceeding 10 cm above the hock a threshold that is considered critical for mastitis risk. This observation may partly explain why buffaloes with deeper udders are not culled.

Regarding teat-related traits, teat direction was consistently and favorably associated with survival across all parities, with the strength of the correlation increasing with age (Figure 5). In contrast, the correlations with the other two teat-related traits changed with advancing parity. Specifically, longer

teats were associated with improved survival in early parities; however, after the fifth parity, longer teats became either neutrally associated with survival (Model 1) or negatively correlated (Model 2). The optimal teat position for survival was found to be central, as evidenced by a mean score lower than the median (Table 3), although the significance of this association diminished in older animals. The declining significance of these correlations over time, analogous to the trend observed for rump slope, is likely attributable to age-related alterations in udder morphology (Williams et al., 2022).

As teat length tends to increase with age, buffaloes with initially shorter teats may be more desirable in later parities. Longer teats, being closer to the ground, are more susceptible to environmental contamination by pathogens responsible for mastitis, thereby increasing the risk of culling (Bharti et al., 2015; Bobbo et al., 2019). While teat position itself is generally stable over time, age-related modifications in udder conformation may affect the interaction between teats and milking equipment, subsequently reducing the strength of the observed correlations.

Notably, the most favorable teat direction was characterized by teats converging toward the center of the udder, with this association becoming increasingly pronounced in later parities, particularly in Model 2 (Figure 5).

3.3.3 DIFFERENCES BETWEEN THE TWO MODELS IN THE ANALYSIS OF THE GENETIC CORRELATIONS

The comparison between the two models, with and without the adjustment of Milk EBV, showed divergent outcomes for specific linear type traits. Differences were observed for three body-related traits: stature, chest width, and trunk length, as well as for four udder-related traits: front udder length, rear udder width, udder support, and teat position. The body-related traits are positively affected by milk production.

Considering that reduced milk yield is the primary reason for voluntary culling in buffaloes, it follows that animals exhibiting larger body structures, and hence greater productivity, are more likely to be retained for a higher number of parities. In contrast, the outcomes of Model 2, which adjusts for the effect of milk production by using milk EBV as a proxy for milk production, underscores the intrinsic influence of these traits on stayability.

These results may thus offer more precise indicators for effective selection aimed at enhancing longevity (Haile-Mariam and Pryce, 2015). Furthermore, selecting buffaloes that maintain

comparable milk production levels while exhibiting a smaller body size may contribute to greater production sustainability. This can be related to the lower environmental impact of smaller animals, due to their reduced feed intake capacity coupled with higher feed efficiency. Similar considerations are true for udder-related traits.

Traits such as front udder length, rear udder width, and udder support contribute to an increased linear score, which is related to larger and more productive udder. Notably, the linear score for udder support tends to be higher in animals with larger udders, reflecting the specific udder conformation characteristic of this species. The observed genetic correlations between type traits and survival may stem from both involuntary culling (e.g., as influenced by rump traits, udder depth, and rear udder height) and voluntary culling (e.g., based on overall body type). Voluntary culling may be partially due to the correlation between type traits and milk yield.

To assess this further, the genetic correlations of survival with type traits were re-estimated after adjusting for milk yield. In instances where no significant association between milk yield and certain type traits was observed, the genetic correlation with survival remained relatively constant, suggesting a mitigation of milk yield's effect on survival. Conversely, for some traits the genetic correlation remained substantially high or even changed in sign despite the adjustment, implying that factors beyond milk yield contribute to the unfavorable correlations between these type traits and survival.

3.4 CONCLUSION

The present study offers significant insights into the relationship between linear type traits and longevity in buffaloes, underscoring the multifactorial nature of survival and stayability. Both phenotypic and genetic correlations reveal that specific body- and udder-related traits substantially influence the productive lifespan of buffaloes, either directly or indirectly through their association with milk yield. The observed positive correlation between broader, deeper body structures and enhanced survival suggests that buffaloes with larger frames are retained for a greater number of parities, likely attributable to their superior productivity.

However, results derived from Model 2 which mitigates the confounding effects of milk production indicate that certain body traits exert an intrinsic influence on stayability, independent of production levels. This finding implies that breeding programs aiming to improve longevity should incorporate

also structural traits to optimize both sustainability and efficiency. Similarly, the analysis of udder-related traits revealed a complex association with survival. While traits such as front udder length, rear udder width, and udder support exhibited positive associations with stayability, deeper udders were negatively correlated with survival, possibly due to an elevated risk of health issues such as mastitis.

Furthermore, the variation in correlation patterns across different parities suggests that the impact of these traits on longevity evolves with age, thereby highlighting the necessity for dynamic selection strategies that account for age-related changes in conformation. Comparing results from the two model helped in disentangling the effects of milk production and the inherent influence of type traits on longevity. Indeed, Model 2, by adjusting for milk yield, provides a clearer delineation of the contribution of conformation traits to stayability.

This distinction is pivotal for refining selection criteria, suggesting that animals with more compact body structures while maintaining equivalent milk production may offer advantages in terms of sustainability, such as reduced environmental impact and improved feed efficiency. Overall, the findings advocate for an integrated selection approach that balances production efficiency with structural integrity to enhance longevity in buffalo populations. Future research should further elucidate the genetic underpinnings of these relationships, thereby optimizing breeding strategies aimed at simultaneously improving productivity and sustainability in buffalo farming.

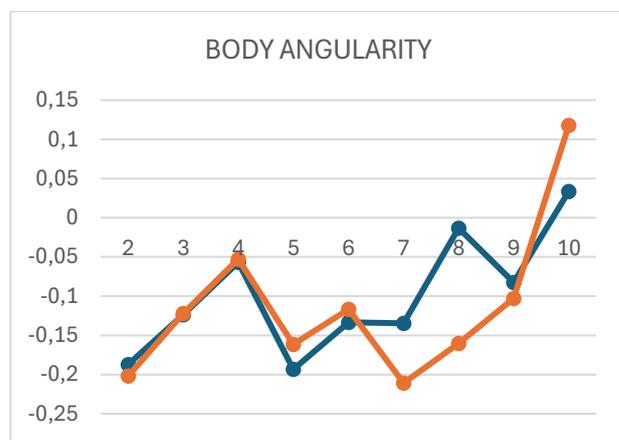
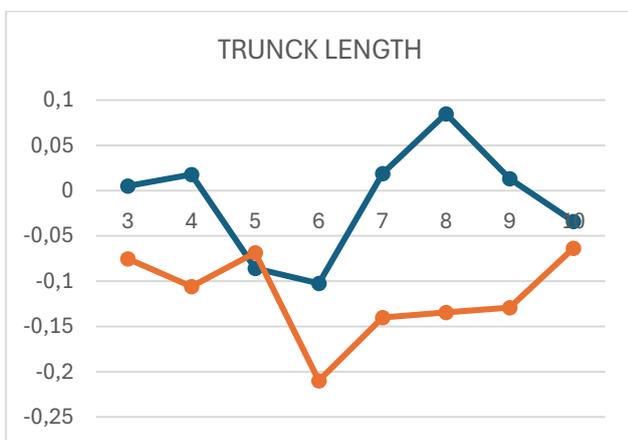
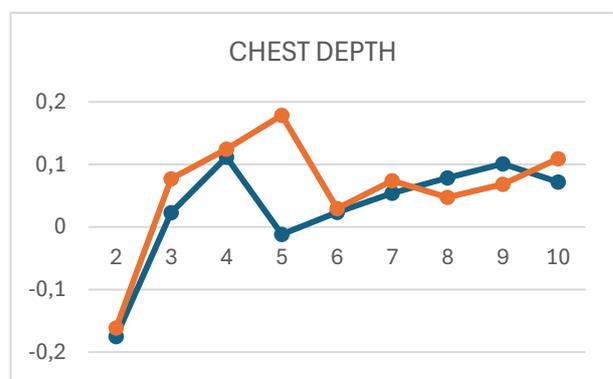
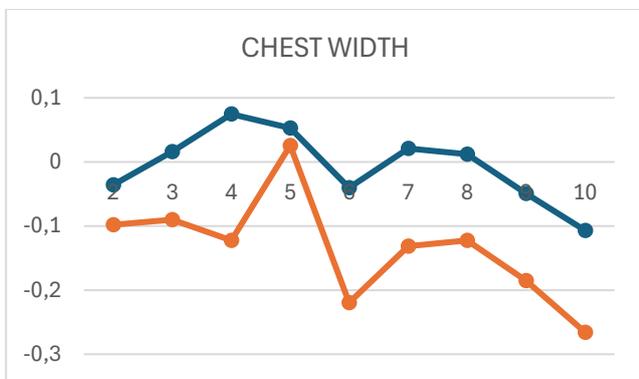
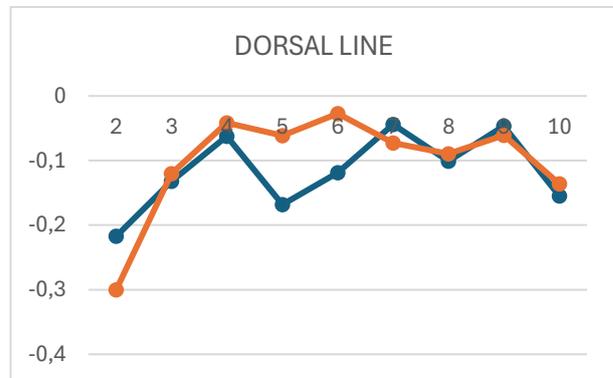
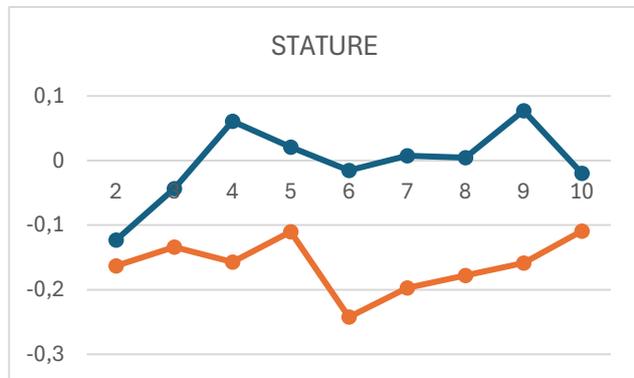


Figure 1. Genetic correlations between survival in each parity and type traits of trunk shape (Model= blue line; Model2= orange line).



Figure 2. Genetic correlations between survival in each parity and type traits of rump shape (Model= blue line; Model2= orange line).

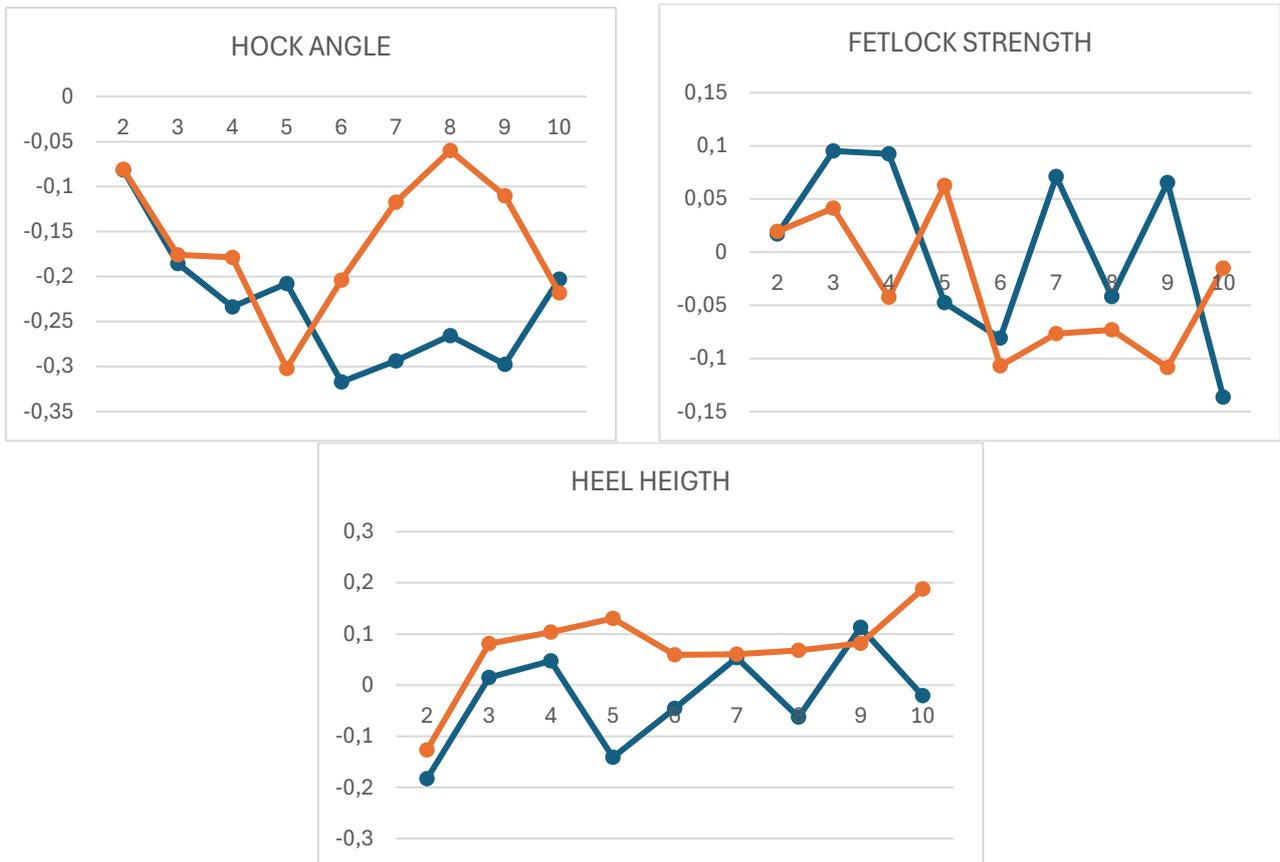


Figure 3. Genetic correlations between survival in each parity and type traits of feet and leg shape. (Model= blue line; Model2= orange line).



Figure 4. Genetic correlations between survival in each parity and type traits of udder shape (Model= blue line; Model2= orange line).

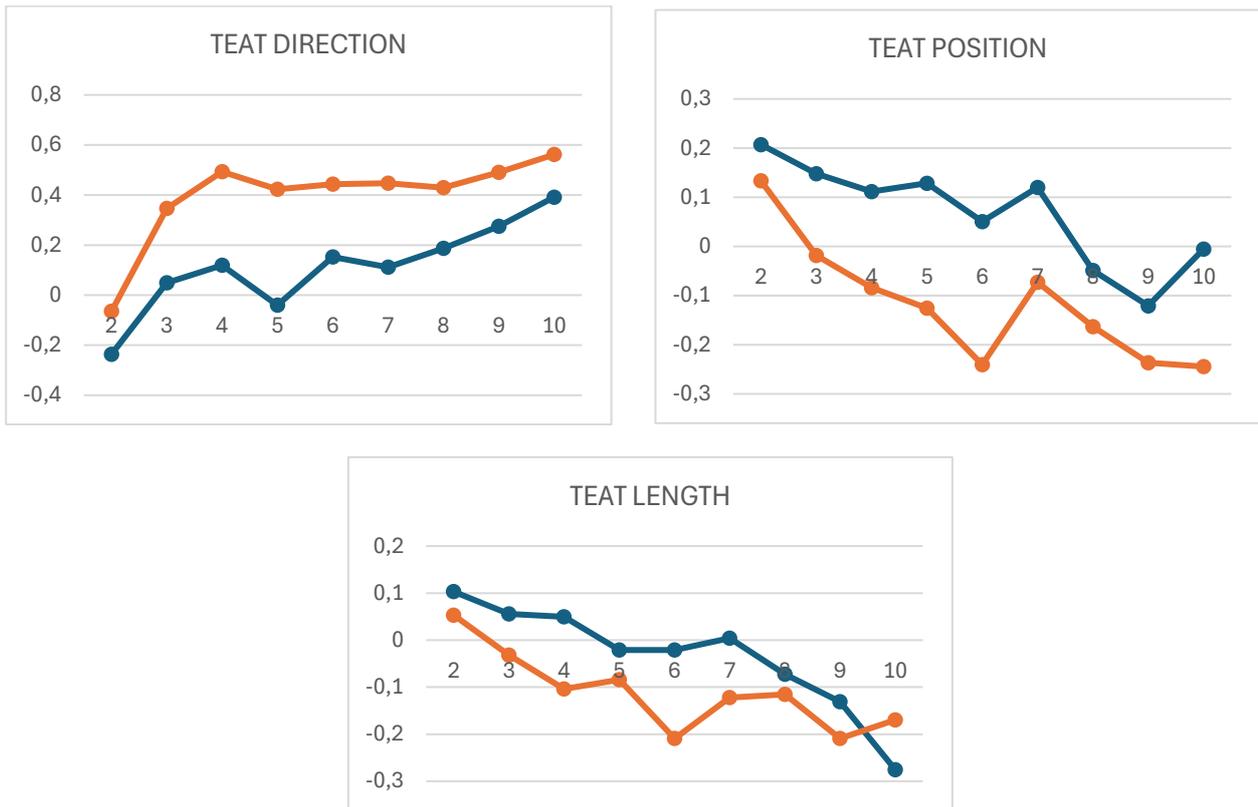


Figure 5. Genetic correlations between survival in each parity and type traits of teat shape (Model= blue line; Model2= orange line).

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4. FINAL CONSIDERATIONS

Longevity in dairy animals is a highly complex trait, resulting from the interaction of multiple factors, among which the genetic component proves to be difficult to extrapolate. This is because environmental on farms conditions and, particularly, management strategies adopted by the farmer often play a dominant role.

The aim of this thesis was to estimate the longevity of the Mediterranean Italian Buffalo using the Stayability approach, which is based on the number of parities rather than the age at culling. This method indirectly provides additional information on the productive efficiency associated with longevity.

The estimation of genetic components for stayability showed low to moderate heritability values, with an increasing trend for elder parities. Once the most productive buffaloes are selected during their first lactations, they remain on the farm primarily due to their resilience to environmental conditions and their ability to maintain good health. The analysis of approximate genetic correlations between longevity and the estimated breeding values (EBV) for milk production revealed a possible positive association. This association deserves further investigation, as, if true, it would suggest that selecting for higher milk production could also increase longevity. However, this relationship appears to be less evident when examining correlations at elder parities. This phenomenon seems to be significantly influenced by voluntary culling based on production, a practice regularly applied by farmers and difficult to separate from the mandatory culling driven by longevity-related issues.

Stayability in buffalo is heavily influenced by environmental factors, complicating genetic selection. Genomic selection offers a potential solution to better exploit genetic potential and reduce the generation interval, accelerating genetic progress. This approach is particularly relevant given the challenges of using survival analysis for longevity studies in buffalo, where complex data and strong environmental interactions limit its effectiveness. Genomics could thus provide a more precise method for identifying animals with higher longevity potential, overcoming these limitations.

Another focus of this thesis was the evaluation of the association between longevity and morphology, using traits recorded in the type trait evaluation of the Italian Mediterranean Buffalo. Two approaches were used: on one hand, identifying the external conformation associated with greater longevity by analyzing the structure of females that remain on the farm up to their 10th parity, and on the other,

assessing the genetic component of morphological traits transmitted by bulls that would correlate with greater longevity in their daughters.

Considering the significant confounding effect of milk production which limits the survival of less productive animals the analysis also employed a model that attempted to minimize the influence of EBV for milk production on the EBV data for morphology.

Both phenotypic and genetic correlations indicate that specific body and udder traits significantly influence the productive lifespan of buffaloes, either directly or indirectly through their association with milk yield. The observed positive correlation between broader, deeper body structures and enhanced survival suggests that buffaloes with larger frames tend to be retained for a greater number of parities, likely due to their superior productivity.

Some morphological traits show significant correlations with the likelihood of farm retention, suggesting their determining role in selecting animals with greater adaptability and resilience over time. It was particularly interesting to find that certain traits traditionally considered to favor greater longevity, such as larger trunk diameters and udder volumes, are more closely linked to higher production than long-term longevity. In contrast, animals with more compact bodies and smaller diameters manage to achieve a greater number of parities when evaluated independently of their EBV for milk production. Another intriguing observation is that some traits measured in young animals may lose significance over time as they undergo changes associated with aging.

Both of these findings suggest the need to reconsider the current criteria used for the morphological selection of buffaloes. The adoption of advanced genetic models for estimating correlations between longevity, morphology, and productive performance represents a valuable tool for identifying animals with a greater propensity for a long and profitable career.

The importance of achieving high longevity in buffaloes is greater now than ever, particularly in relation to environmental sustainability issues that are impacting the entire livestock sector, not just dairy production. Managerial decisions, especially in the context of the Mozzarella di Bufala Campana DOP market, are often influenced by market dynamics, like for instance seasonality. Indeed, the out of season breeding system adopted in many farms and the voluntary culling of less productive animals significantly affect the productive career length of buffaloes.

Looking ahead, accurate selection for stayability could not only improve the efficiency and profitability of buffalo farms but also contribute significantly to the sustainability of the entire

production sector. Increased longevity in buffaloes leads to reduced culling rates, less need for replacement animals, and a smaller environmental impact, aligning with modern sustainable animal husbandry practices. The implementation of integrated genetic selection programs, combined with innovative management strategies, could therefore play a key role in the future of the buffalo farming industry.