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Suckler cow-calf production system in the Mediterranean: feeding behaviour and environmental implications

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Dedicated to my family

For their endless love, and encouragement

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Table of Contents

List of Tables 3
List of Figures 5
General Abstract 6
Introduction 8
1. Cattle production system and economic situation in Sardinia 8
2. Autochthonous breeds 9
3. Suckler cow-calf systems 10
4. Objectives of the research 12
5. References 16
CHAPTER 1 21
1. Introduction 23
2. Materials and methods 25
3. Monitoring Techniques 26
1. Direct Observations 26
2. Automatic Data Recording Methods 28
4. Suckling Parameters 30
5. Factors Impacting Suckling Behaviour 33
1. Calf Age and Weight 36
2. Calf Sex and Genetic Type 37
3. Cow breed and milk production 38
4. Circadian Variation 40
6. Effect of Suckling Behaviour on Reproduction and Production 41
1. Postpartum anoestrous period 41
2. Effect of suckling behaviour on calves' production 44
7. Conclusion 45
8. References 46
CHAPTER 2 60
1. Introduction 63
2. Material and Methods 66
1. Experimental Site and Animals 66
2. Behavioural monitoring data 68
3. Statistical analysis 68
3. Results 70
4. Discussion 73

5. Conclusion 76
6. References 77
CHAPTER 3 86
1. Introduction 88
2. Material and Methods 92
1. Experimental Site and Animals 92
2. Herbage sampling 92
3. Intake estimation 93
4. Measurements 94
5. Statistical analysis 95
3. Results 95
4. Discussion 98
5. Conclusion 101
6. References 102
CHAPTER 4 112
1. Introduction 115
2. Materials And Methods 119
1. Study sites and animals 119
2. Data collection 119
3. Spatial and temporal boundaries of the system 120
4. Estimating the Carbon Footprint 120
5. Estimation of methane emissions 120
6. Estimation of methane emissions from manure 122
7. Estimation of nitrous oxide emissions from wastewater 123
8. Estimation of emissions from energy production and use 125
9. Estimation of emissions from livestock on-farm production and purchased feed. 125
10. Global warming potential and functional unit 127
11. Carbon sequestration 128
3. Results 129
4. Discussion 133
5. Conclusions 135
6. References 137
7. Annexe 1. 145
General Conclusion 148

List of Tables

Introduction

Table 1. The major beef cattle breeds reared in Sardinia (BDN, 2023).....8

CHAPTER 1

Table 1. Literature available on monitoring beef cattle suckling behaviour under pastoral conditions.....33

CHAPTER 2

Table 1. The behavioural categories used to monitor the animals were adapted and modified from Pereira et al. (2020).....68

Table 2. Time devoted to grazing activity by two groups of cows with different calving season for each 30-min subperiods representing the different observation sessions during the day.....70

Table 3. Time devoted to different activities by two groups of cows with different calving season.....71

Table 4. Comparison of body weight and growth rate between the two groups.....72

Table 5. Comparison of calves' growth rates between the two groups…….73

CHAPTER 3

Table 1. Chemical composition of herbage samples.....96

Table 2. HDMIBW, HDMD and FO in cows characterised by distinct calving seasons (winter-calving cows, group W and spring-calving cows, group S) estimated using C31/C32 and C33/C32 alkane ratios.....96

CHAPTER 4

Table 1. Emission factors for purchased feed (Serra, 2014). 126

Table 2. Carbon removal potential, expressed in tons CO2/ha/yr, for different land types. 128

Table 3. Distribution of farm area and herd stock characteristics (data expressed as mean ± standard deviation).....129

Table 4. Carbon balance, total and net carbon footprints, and main average emission sources for the studied farms. 130

List of Figures

Introduction

Figure 1. Cattle farm production systems in Sardinia (BDN 2023)..... 10

Figure 2. Sarda cow and calf on pasture. 12

CHAPTER 1

Figure 1. A halter with tri-axial accelerometers fixed on the right side of the monitored calf (Kour et al., 2018). 29

CHAPTER 2

Figure 1. Comparison of cow body weight changes between groups W and S (S.E. are indicated by vertical lines). 72

CHAPTER 3

Figure 1. Violin Plots for Herbage Dry Matter Intake (expressed as percentage of body weight and estimated by C31/C32 and C33/C32 pairs) of the experimental groups..... 98

CHAPTER 4

Figure 1. The four stages of LCA were conducted according to the ISO 14040 guidelines. 119

Figure 2. Average contribution (in%) of different emission sources to overall impact. 132

Figure 3. Emissions from Enteric Fermentation. 145

Figure 4. Emissions from Manure management. 145

Figure 5. Emissions from Energy. 146

Figure 6. Emissions from Purchased feed. 146

Figure 7. Emissions from On-Farm Feed Production. 147

General Abstract

This thesis studies the suckler cow-calf production system in the Mediterranean region, focusing on feeding behaviour and environmental implications. The dissertation is divided into six sections, including Introduction, four chapters, and conclusion. The introduction section presents a general introduction to beef production in Sardinia and highlights suckler cow-calf operation systems. Chapter 1 provides a comprehensive review of the existing research related to suckling behaviour in beef cattle under pastoral conditions. This review focuses on the factors affecting suckling behaviour and the methodologies employed to monitor this behaviour. In addition, this chapter highlights the challenges of assessing suckling behaviour and discusses the potential of remote monitoring devices. Chapter 2 presents behavioural monitoring data, such as grazing, ruminating, resting, and walking of grazing suckler cows. The results reveal the difference between the two groups of indigenous Sarda cows differing in the calving season in terms of time spent engaging in different behaviours. Chapter 3 focuses on the herbage intake, digestibility, and faecal output of the two groups of cows presented in Chapter 2. This chapter discusses the differences between the two groups in terms of herbage intake and digestibility, with a specific emphasis on the differences related to postpartum recovery which necessitates high energy consumption from the dam. Chapter 4 investigates the carbon footprint and effect of carbon sequestration in mitigating greenhouse gas emissions associated with the suckler cow-calf production system. This study estimates emissions from different sources, such as enteric methane, manure management, energy consumption, and feed production and purchasing. The results also demonstrate the capacity of carbon sequestration to mitigate emissions. Additionally, it highlights the importance of considering the environmental

implications of production systems. This is in addition to the conclusion section, which is based on the findings of different studies. Overall, this thesis provides valuable insights into the suckler cow-calf production system in the Mediterranean region in terms of the different behaviours of beef cattle in extensive systems and the environmental implications of these systems. It also highlights the need for further research to optimise the production system and reduce its environmental impact.

Introduction

Cattle production system and economic situation in Sardinia

To date (BDN, 2023), the cattle production system in Sardinia consists of approximately 282.800 heads distributed over 9000 farms and represents the seventh most important cattle breeding region in Italy. Beef production constitutes 72% (204.600) of the total heads reared in the region, while 15% (43.000) are dairy cows, mainly localised in the western plains of the island, and approximately 13% (35.200) are dual-purpose breeds (beef-dairy).

Table 1. The major beef cattle breeds reared in Sardinia (BDN, 2023).

Breed	**Heads (n°)**	**Percentage (%)**
Crossbred	105,206	51.41
SARDO BRUNA	44,851	21.92
LIMOUSINE	22,528	11.01
SARDA	16,468	8.05
CHAROLAIS	4,467	2.18
SARDO-MODICANA	4,072	1.99
BRUNA SARDA	2,915	1.42
BRUNA	1,357	0.66
Other breeds	2,768	1.35
Total	**204,632**	

Table 1 presents a list of the major beef cattle breeds reared in the region. Notably, only 12% belonged to specialised beef breeds, including Limousine and Charolais. However,

most animals are indigenous and crossbred, such as Sardo Bruna, Sarda, and Sardo Modicana (Table 1).

In 2022, 31,462 cattle were slaughtered in Sardinia, constituting approximately 1.14% of the national value. Approximately 44% of the slaughtered animals were calves and males aged between 10 and 30 months (BDN, 2023). The meat produced from these animals is valued at nearly 102 million euros, which represents approximately 3.64% of the national value (Agriconsulting and Ismea, 2020). Furthermore, in 2019, 76,274 quintals were produced in Sardinia, accounting for approximately 8% of southern Italy's production and 1% of the national production (Agriconsulting and Ismea, 2020).

On the island, farmers cross bulls of specialised meat breeds with native or non-specialised breed cows (hardy breeds). This technique makes it possible to combine the greater environmental adaptability of hardy breeds with the higher growth capacity of crossbred calves than pure ones (Theunissen et al., 2013). Hardy breeds are often medium-sized; therefore, they have limited maintenance requirements and an excellent ability to exploit the resources of the island pastures (Poussard et al., 2016). These breeds are also characterised by good maternal ability, being able to rear calves excellently until weaning (Le Neindre, 1989).

Autochthonous breeds

As shown in Table 1, the native “Sarda” breed represents approximately 8% of the total beef cattle production in Sardinia. Sarda is an indigenous breed of the island, characterised by its small size and unique horns, which come in a range of shapes, sizes, and orientations. It is also known for its strong and robust jaws, udder with a symmetrical and regular shape

and size, and quarters that are proportionate and balanced (Figure 2). This native cattle breed is a valuable genetic resource for adaptation to production systems, local conditions, surrounding environment (Petretto et al., 2022) and various stresses under which it has evolved (Bovo et al., 2021). Therefore, it represents a significant gene bank that could be critical for managing future climate change or potential disease outbreaks (Zander et al., 2013). In addition, it plays a role in preserving human cultural heritage and connecting it to traditional food products (Carta, 2015).

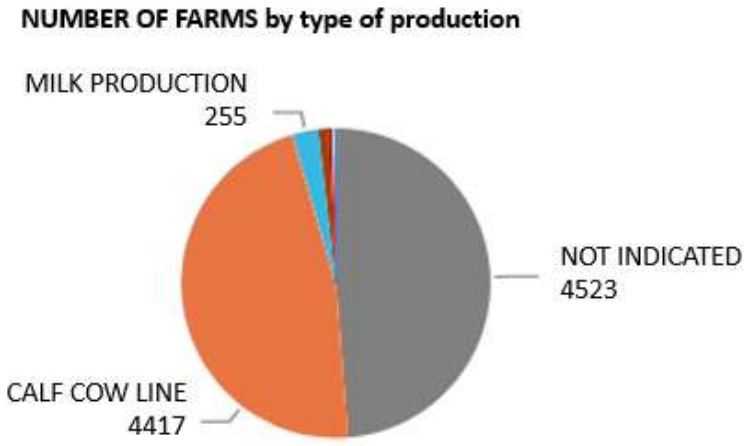


Figure 1. Cattle farm production systems in Sardinia (BDN 2023).

Suckler cow-calf systems

Separating newborn calves from their dams is a common practice on commercial farms, which contrasts with the cow-calf farming operations, where the mother rears the calf (Whalin et al., 2021). Consumers consider this practice controversial (Busch et al., 2017; Ventura et al., 2013). Consequently, an increasing number of breeders have developed rearing systems that allow for extended contact between dams and calves, including suckling (Johnsen et al., 2016; Vaarst et al., 2020). The cow-calf line rearing method is one of the few, if not the only, beef cattle rearing techniques that offer both economic and

environmental sustainability, benefiting individual farms and communities (Whalin et al., 2021). However, water scarcity in agriculture can lead to a lack of animal feed, resulting in a reliance on imported feed for farm operations (FAO, 2009). These conditions have led to the adoption of adapted farming systems, such as the beef cattle farming system in Sardinia, which is primarily based on free-range and extensive grazing management practices (Figure 1). This system relies on suckler cows and calves grazing in mountainous areas with arid and high-hill pastures of medium-low nutritive value (Petretto et al., 2022). Over time, this system has been expanded to more productive areas, as it allows for cost-effective utilisation of pastures and meadows on farms (Sgoifo Rossi et al., 2013). This method reduces herd management costs with low labour costs and fixed investments, as individual animals feed directly by grazing for many years (Lawrence et al., 1999). Generally, natural mating is performed with a ratio of one bull for every 25 to 30 cows, with the cows rarely separated from the bull, except in the case of first-calving heifers (Coizet, 2014). The established cow/calf operation follows the "Autumn-Winter Calving" pattern, with weaning occurring in early summer after 6-7 months (Acciaro et al., 2020). Calves are then sold to fattening centres, primarily located in the "Pianura Padana" region of northern Italy (Acciaro et al., 2021). This livestock production system is similar to others

used throughout the Mediterranean region (Piedrafita et al., 2003; Ligios et al., 2005; Aldai et al., 2012; Humada et al., 2012; Blanco et al., 2017).



Figure 2. Sarda cow and calf on pasture.

Objectives of the research

The Mediterranean region is widely recognised for its remarkable role in global agriculture because of its diverse landscapes and rich cultural traditions (Migliorini et al., 2018). The region's distinctive environmental and sociocultural features have contributed significantly to global food production (CIHEAM/FAO, 2015). However, the Mediterranean region is susceptible to the anticipated changes in its typical temperate climate, which may result in more extreme conditions (Cramer et al., 2022). This emphasises the significance and complexities associated with agricultural systems in this region.

The suckler beef cow-calf system, a prominent feature of livestock production, is well adapted to the environmental and social aspects of the area and is known to contribute to economic proficiency (Ligios et al., 2005). Research has shown that suckler cow–calf

systems, in which calves are reared by cows, are preferred by consumers because of their resemblance to natural behaviour (Mandel et al., 2016). This system is advantageous for calf welfare as it allows for the transfer of passive immunity and essential antibodies through colostrum and milk, as well as the learning of vital survival behaviour through social interactions with the dam on pasture (McGee and Earley, 2019). Nonetheless, compared to artificial rearing, suckling systems are more beneficial for calves, as suckling calves tend to experience higher weight gain than calves reared using other systems (Roth et al., 2009).

However, the behaviour of suckler beef cow-calf systems in the Mediterranean context is a multifaceted subject with implications that span from animal welfare to land management practices. Studying the intake and social behaviour of these animals in their natural environments provides valuable insights into their needs, preferences, and interactions with the ecosystem. By understanding the behaviour of both cows and calves, researchers can identify optimal management strategies that promote animal welfare while enhancing productivity and minimising the environmental impact. Nevertheless, the environmental implications of the suckler beef cow-calf system are a growing concern in relation to climate change, biodiversity loss, and resource scarcity. As the demand for beef continues to increase, it is important to address the environmental impact of beef production. This includes greenhouse gas emissions, which contribute to climate change, as well as the potential loss of biodiversity due to land-use changes and the use of resources such as water and feed. Efforts to improve the technical efficiency of the suckler beef sector could mitigate these environmental implications. By implementing strategies that reduce emissions and optimise resource use, it is possible to minimise the environmental footprint

of beef production while maintaining a sustainable and productive industry.

Despite the importance of studying the behaviour and environmental implications of the suckler beef cow-calf system, several research gaps persist. First, there is a lack of comprehensive studies that integrate suckling behaviour observations with environmental assessments, hindering our understanding of the complex interactions between the behaviour, the surrounding factors, and the impact on the production and reproduction traits of both cows and calves.

Moreover, existing research often neglects the impact of calves on the ingestive and social behaviour of cows in extensive systems, particularly in traditional systems. Despite the significance of this topic, as each stage of calf rearing can affect the energy needs of cows, as evidenced by variations in intake, grazing, and social behaviour, this remains largely unexplored. During the pre-weaning phase, calves rely on their dams in different ways, as previously discussed in terms of colostrum or milk consumption, weight gain, and learning-specific behaviours.

Additionally, the environmental implications of the suckler beef cow-calf system are of great concern in the face of climate change, and the Mediterranean region requires tailored research to assess the impact of this system on the environment.

Overall, studying the behaviour and environmental implications of the suckler beef cow-calf system in the Mediterranean is crucial for ensuring the sustainability and resilience of livestock production in the region, and interdisciplinary approaches have provided valuable insights for informed and sustainable management practices.

These gaps can be covered by a comprehensive review, which will be presented in Chapter 1, in which all aspects related to the suckling phase were addressed. The studies in Chapters

2 and 3 aimed to address this gap by focusing on indigenous breeds in Sardinia and examining the differences in behaviour, and herbage intake according to differences in the calving season or in another term according to differences in the stage of lactation and age of calves. Chapter 4 aimed to address this issue and highlighted the potential of carbon sequestration to mitigate the greenhouse gas emissions from these systems.

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CHAPTER 1

Suckling Behaviour in Beef Cattle under Pastoral Conditions: A Review

Abstract

Monitoring suckling behaviour in beef cow-calf systems under pastoral conditions can provide information on calf performance and on reproduction activity of cows. However, assessing suckling behaviour and the magnitude of its impact is challenging because it is quite difficult to collect objective measurements of behavioural traits, especially in pastoral conditions. Recently, remote-monitoring devices have been introduced in this field. These technologies are promising and may provide opportunities for objective monitoring. However, their validation and applicability require further investigation. Nevertheless, calf suckling behaviour is influenced by a variety of factors including age, weight, sex, genetic type, milk production, and circadian variations. The impact of these variables, in turn, is influenced by geographical and environmental conditions, as well as by monitoring approaches. In addition, management during the suckling phase has been shown to influence the onset of the cow's oestrous cycle and reduce weaning stress. Therefore, the aim of this review was to describe the traits of beef cow-calf suckling behaviour and the factors known to influence it. Current methods of monitoring and assessing behaviour have been reviewed. The effects of suckling behaviour on the production and reproduction traits of cows and calves have also been identified. The objective assessment of beef cow-calf suckling behaviour under pastoral conditions and its impact on cow-calf performance will be beneficial in enhancing management practices in the future.

Keywords: Behaviour monitoring, pastoral system, suckling calf, suckler cow, extensive farming.

Introduction

Suckling is considered the most important behaviour in mammalian species and contributes to offspring growth and survival (Grandinson, 2005). In beef cow-calf operations, the calf interacts with its mother and contacts the dam's udder to ingest milk, accompanied by observable suckling and swallowing (Paranhos da Costa et al., 2006). Additionally, calves engage in various actions to stimulate milk release, such as searching for, butting, and bumping the udder (Lidfors et al., 1994). The diet of young calves depends on maternal milk, particularly during the first phase of life (Cantor et al., 2019). Calves are born without immunity to diseases and rely on their dams' colostrum to obtain passive immunity (Moran, 2012). Therefore, it is helpful for calves to start suckling as early as possible after birth (Hogan et al., 2022). Suckling stimulates physiological processes that facilitate the development of a cow-calf pair bond (Sirovnik et al., 2020). It establishes a distinctive relationship between cows and their offspring during the first week after birth, which continues through the pre-weaning period (Enríquez et al., 2011; Khater et al., 2022). The survival of newborns depends on the formation of a strong and enduring social bond with the dam, particularly under natural conditions (Enríquez et al., 2011). In beef production systems, the initiation and maintenance of suckling bouts, characterised by specific patterns in terms of frequency and duration, are essential for calf nourishment (Von Keyserlingk and Weary, 2011). Suckling bouts are initiated by cows within the first few weeks of life and subsequently maintained by both cows and calves (Lidfors et al., 1994). In a recent study, Hogan et al. (2022) reported that calves may take up to three days postpartum to establish a consistent suckling pattern. Another recent study reported that as calves aged, the pattern of suckling behaviour changed (Kour et al., 2021a).

In addition to the effect of suckling on calf production, suckling is a major regulator of the reproductive performance of beef cows as it affects the postpartum anoestrus period and pregnancy rate (Montiel and Ahuja, 2005). Cows utilise energy to produce milk rather than to reinitiate reproductive functions, which delays their return to cyclic ovarian activity postpartum (Gautam et al., 2010). Previous studies, as mentioned in the review by Montiel and Ahuja (2005), have reported that some management practices, including restricted suckling and temporary separation, influence pregnancy rates. Thus, increasing knowledge of suckling behaviour and its effects on both offspring and dams helps enhance management and initiate the reproductive cycle to produce one calf every year.

Suckling behaviour is influenced by a variety of factors, including genetic (Das et al., 2000), physiological (Álvarez-Rodríguez and Sanz, 2009; Hogan et al., 2022), individual (Kour et al., 2021a; Paranhos da Costa et al., 2006), and environmental factors (Bouissou et al., 2001; Vandenheede et al., 2001). Therefore, concerns have arisen regarding the critical management requirements during the suckling phase, reinforcing the importance of objective monitoring methods for suckling behaviour by selecting an adaptable methodology that depends mainly on experimental conditions, observer experience, and budget. This ensured the collection of unbiased data without disturbing the animals' natural behaviour (Van Der Marel et al., 2022).

Therefore, it is necessary to measure suckling behaviour and evaluate its impact on calf productivity and reproduction in pasture-based systems. A comprehensive review of the literature available in this field can provide valuable insights into the results of previous studies. This study aimed to critically evaluate the literature available on suckling behaviour of beef cattle, while identifying variations in behaviour patterns related to

different factors, determining how to objectively measure suckling behaviour, and demonstrating efficient management strategies used to date to minimise its effect on reproduction in cow-calf operations.

To date, no reviews have explored all aspects concerning beef suckling behaviour under pastoral conditions. This paper presents the first thorough literature review of the monitoring methodologies employed to observe beef calf suckling behaviour under pastoral conditions. Emphasising on the existing methodologies utilised to date, this review specifically highlights the ongoing exploration and validation of automated monitoring methods by researchers. This will provide a clear overview of the subject, help improve future studies, and lead to enhanced management practices during the suckling phase.

Materials and methods

A literature search was carried out using public data search generators that included SCOPUS, Science Direct, and Google Scholar to collect published studies related to beef cattle suckling behaviour under pastoral conditions. The following keywords in different combinations were used to conduct the search process: “suckling behaviour”, “beef calves”, “behaviour monitoring”, “pastoral conditions”, “weaning weight”, “suckler calf”, “suckler cow”, “anoestrus period”, “production traits”, “oestrus cycle”, “cow-calf performance”, resulting in 314 initial search results. To ensure that recent studies were included, the publishing year range was customised between 2000 and 2023, narrowing the results to 252. Subsequently, 55 articles related to dairy science, equine, poultry, and small ruminants were eliminated. Title browsing was conducted, excluding articles related to dairy cattle or animals other than beef cattle, resulting in a set of 40 articles. After reading

the abstracts of these articles, studies that did not meet specific criteria were excluded. The exclusion criteria were studies that were not conducted under pastoral conditions, studies that included housed animals, and studies that focused on behaviour other than suckling. These criteria were applied to ensure the inclusion of only the relevant studies. After applying the exclusion criteria, eight articles were retained. Further analysis through full-text reading and the search was extended beyond the initial year range, resulting in the inclusion of four articles that met all criteria. In total, 12 articles which studied suckling frequency, duration, and/or total suckling duration in beef calves under pastoral conditions were the basis of the review and are shown in Table 1. By examining the references and discussions of the included articles, additional articles were found and included in the review, as they provided information about the effects of various factors on suckling behaviour and contributed valuable insights to the overall review discussion, even though they did not specifically study the suckling parameters of interest.

Monitoring Techniques

1. Direct Observations

Several studies have used direct observation methods to monitor suckling behaviour in beef calves (Martin and Bateson, 2007), including visual observation and time recording of each activity (Louw et al., 1993; Odde et al., 1985; Paranhos da Costa et al., 2006). To ease the monitoring process, many identification methods for cows and calves have been used, such as spray-painted numbers on the cows' sides (Odde et al., 1985) and unified rubber collars for both cows and calves (Odde et al., 1985; Paranhos da Costa et al., 2006). Coloured rubber collars could also help in the detection of allo-suckling and inter-suckling activities when a calf suckles a cow different from its dam, or vice versa. Moreover, binoculars and

supplementary equipment, such as lights (Louw et al., 1993; Paranhos da Costa et al., 2006), night vision scope ("starlight scope"), infrared spotlights during nighttime observations (Day et al., 1987), and video recordings (Hogan et al., 2022), have been used to improve monitoring efficiency.

However, direct observation is considered difficult, labour-intensive, and sometimes inaccurate due to observer bias related to actions' interpretation or the observer's presence affecting animal behaviour (Elischer et al., 2013). In addition, some limitations may encounter the observer, such as monitoring a small number of animals or a shortage in the duration of observations, especially during the night, due to alterations in the observer's capacity to remain alert and focused due to the surrounding environmental conditions (Böhm et al., 2009; Lendvai et al., 2015). Observers' level of expertise in the field, as well as their ability to record data accurately, may affect the reliability and validity of the collected data (Tuyttens et al., 2014; Wark et al., 2019). Rather than assuming observers' proficiency in collecting unbiased data, it is essential to provide them with comprehensive guidance (Marsh and Hanlon, 2004; Tosi et al., 2006). Several methods have been implemented in books and comprehensive guidelines such as those mentioned by Altmann (1974) and Stamp Dawkins (2007). These authors emphasised the importance of providing comprehensive instructions to observers and highlighted various methods to mitigate observer bias. One such method ensures consistency among observers by uniformly recording the same information to enhance interobserver reliability (Torsiello et al., 2024). Additionally, random assignment of observers to different animal groups or ensuring balanced observations across groups helps to minimise biases. Furthermore, employing blind observation techniques, where observers select animals without knowing their

differences, can eliminate inadvertent bias. According to Stamp Dawkins (2007), blinding observers to experimental factors in studies in which specific conditions are being investigated can also mitigate bias. Researchers are advised to critically evaluate their research designs to identify potential confounding factors including observer bias.

In addition, the use of technology to remotely collect behavioural data has been introduced in this field of research. Video recordings along with various types of wireless sensors and transponders have been developed to identify and monitor the behaviour of cattle, horses, and poultry (Ruiz-Garcia et al., 2009).

2. Automatic Data Recording Methods

The most commonly used data loggers for monitoring cattle behaviour are accelerometers that record the direction and speed of movement on 2 or 3 axis data (Elischer et al., 2013). This technology has recently been validated to identify the suckling behaviour of beef cattle (Kour et al., 2018). The authors evidenced that fitting the accelerometer on a neck collar is ineffective for classifying suckling behaviour, whereas data collected from the accelerometer attached to the right side of the calf's halter seems to be more accurate (Figure 1), allowing to distinguish suckling from other behaviour (Kour et al., 2018). Moreover, the authors developed a model based on the recorded acceleration of the x-, y-, and z-axes.



Figure 1. A halter with tri-axial accelerometers fixed on the right side of the monitored calf (Kour et al., 2018).

The model was built using raw data obtained from tri-axial accelerometers and analysed by R statistical software (R studio, version 3.2.2). The model showed clear differences in the x- and y-axes (horizontal and vertical acceleration) between suckling and non-suckling periods, interpreting the calf's head movement from the downward or horizontal position to upward while suckling. The x-values became negative during suckling while remaining positive either in non-suckling periods or between suckling bouts, as reported by (Kour et al., 2018). In this context, it was also revealed that the value of y axis (vertical acceleration) was always less than 1500 during the suckling period. This model did not consider the z transmission values (forward acceleration) since no identifiable change was recorded in the transmission values for forward acceleration. The accuracy of this model for monitoring Belmont Red calves compared to directly monitored records was 98.8% (Kour et al., 2018). More recent studies by the same authors aimed to monitor changes in suckling patterns among Belmont Red calves and have shown the discriminatory ability of triaxial accelerometers in measuring suckling behaviour under pastoral conditions (Kour et al., 2021a, 2021b). Similar results were obtained by McCosker et al. (2023), who aimed to

demonstrate the potential of utilising ear-tag accelerometers to detect calf suckling behaviour. This study introduces a suckling recognition algorithm to infer daily suckling for individual calves that exhibits low computational requirements, indicating its convenient implementation in real-time on ear tags. These findings highlight the viability of integrating ear-tag accelerometer systems into beef calf behaviour monitoring and suggest promising prospects for practical implementation and further advancements in this field.

Nevertheless, since these sensors are wearable, they may cause discomfort to the animal (Fuentes et al., 2020). Therefore, other techniques have been introduced in the field of animal behaviour, including unwearable/non-intrusive sensors, such as radar devices (Gygax et al., 2007), fixed cameras (Porto et al., 2014), and drones (Herlin et al., 2021), which can be an alternative to mitigate these problems. Moreover, research on suckling behaviour has provided useful information, but most studies have involved monitoring a limited number of animals in relatively small paddocks. The applicability of the results of these studies to typical pasture environments has yet to be determined. The adaptation of new methodologies can ensure animal welfare during the monitoring period, eliminate bias in data collection, and provide reliable monitoring of suckling behaviour in typical land environments.

Suckling Parameters

Some observable parameters, including the frequency of suckling bouts or suckling, duration of the suckling bout, and total duration of suckling events during 12 or 24 h, are common in determining calf suckling behaviour.

An observational study was conducted on Polled Hereford and Simmental cows and their calves (aged 1-120 days) in Kansas, United States, using 24-hour continuous direct observations (Odde et al., 1985). The study revealed that the frequency of suckling was five bouts in a 24-hour period, and the total duration of suckling was 46 min. Additionally, this study demonstrated a correlation among calf weight, cow milk production, and suckling patterns. Using 48 h of continuous direct observations, Somerville and Lowman (1979) found that the suckling bout duration of 55-day-old Charolais × Blue × Grey calves (10.7 minutes) was significantly longer than that of 33-day-old Charolais × Hereford × Friesians calves (6.9 min), whereas the frequency of suckling bouts remained the same between the two groups (5.5 bouts per 24 h). The suckling behaviour of 66 cows, consisting of Herefords, Simmentals, or Hereford × Simmental, and their calves, was observed through direct observation by Louw et al. (1993) in South Africa. The cows were differentiated by parity and the calves were differentiated by age. This study revealed that the frequency of suckling is higher in primiparous cows than in multiparous cows. Conversely, the duration of suckling bouts was longer in the multiparous cows. The highest frequency of suckling was found in calves aged less than 45 days (5.8 bouts per 24 hours) compared to 4.4 and 4.6 bouts per 24 hours in calves aged 45-90 days and greater than 90 days, respectively. In a recent study by Hogan et al, (2022) in Queensland, Australia, the suckling behaviour of 17 primiparous Angus, Hereford, and Shorthorn cross heifers was observed using both direct observation and video recordings. The mean nursing bout duration was seven minutes, and the mean daily suckling bout duration was 25.5 minutes. Kour et al. (2018) conducted two separate experiments in Queensland, Australia using triaxial accelerometers and visual observations. The suckling behaviour of 23 Belmont Red

calves in Experiment 1 was monitored. The average suckling duration per bout recorded by accelerometers was 9.7 ± 0.20 min, while visually it was 9.3 ± 0.19 min. In Experiment 2, 21 Brahman calves were fitted using triaxial accelerometers. The average suckling duration per bout recorded by accelerometers was 13.69 ± 1.82 min, while with visual observations it was 12.23 ± 1.77 min. Kour et al. (2021a) collected data from 23 Bos taurus calves reared under pastoral conditions in northern Australia over two periods, when calves had an average of one and four months. Calves suckled less frequently per 24 h at 4 months (8.2 ± 0.66 times) than at 1 month of age (9.8 ± 0.46 times, $p < 0.05$), but had significantly longer suckling durations per bout at 4 months (9.3 ± 0.35 min) than at 1 month of age (8.3 ± 0.31 min, $p < 0.05$). Finally, Kour et al. (2021b) studied 28 Belmont Red cow-calf pairs using triaxial accelerometers. The suckling frequency was 7.72 suckling bouts per day, suckling bout duration was 9.36 min per bout, and the total suckling bout duration per day was 73.27 min per day. Behavioural data were collected by Paranhos da Costa et al. (2006) conducted a study in São Paulo, Brazil, using continuous and direct observations. The study involved pure-bred cows of three breeds, Gir, Caracu, and Nelore, and their calves, which were either pure-bred or crossbred. The research focused on suckling frequency, suckling bout duration, and total suckling bout duration during a 24-hour period, the overall means for these variables were 2.57 (suckling bouts per 12 hours), 9.25 (min) for suckling bout duration, and 23.76 ± 0.47 (min per 12 hours), respectively. Twenty Chinampo cows and their calves were studied by José Espinoza et al. (2011) in Mexico. The animals were observed from day 30 to day 180 of lactation for 24 h each month, and the frequency with which the calves suckle, and duration of each suckling session were recorded. The average frequency of suckling in 24 h was 4.9 ± 0.21, with a total of 66.9 ± 3.1 minutes. Calves

aged 180 days suckle fewer times than those aged below 150 days, and the duration of suckling decreased at 180 days of age ($p < 0.05$). Up to 5 months of age, the frequency and duration of suckling did not vary but were reduced at 180 days.

Factors Impacting Suckling Behaviour

Several factors have been reported to affect suckling behaviour, including calf age and weight, calf sex and genetic type, cow breed, cow body condition score (BCS), and circadian variation. Inconsistency (intended as different findings related to the same factor) among several studies is present due to differences in geographical and environmental conditions, breed variance, monitoring methods, sample size, and duration of observations (Table 1).

Table 1. Literature available on monitoring beef cattle suckling behaviour under pastoral conditions.

Reference	Country	Pasture size (ha)	Breed	Genetic type	Cow-calf pair number	Calves' ages (days)	Monitoring methods *	Monitoring period (hours)	FREQ (bouts/ Monitoring period)	DUR (mins/ bout)	Total DUR (mins/ Monitoring period)
(Somerville and Lowman, 1979)	United Kingdom	-	Cross	*B. taurus*	5	33	DO	48	5.5	6.9	-
(Somerville and Lowman, 1979)	United Kingdom	-	Cross	*B. taurus*	7	55	DO	48	5.5	10.7	-
(Oddeet al., 1985)	Kansas, US	7	Cross	*B. taurus*	227	1- 120	DO	24	5.0	-	46.0

(Day et al., 1987)	US	small pasture	Cross	*B. taurus*	11	52- 167	DO	24	6.3	8.6	52.0
(Day et al., 1987)	US	small pasture	Cross	*B. taurus*	20	17- 80	DO	24	7.8	8.4	60.3
(Louw et al., 1993)	South Africa	-	Pure and cross	*B. taurus*	66	0- 45	DO	24	5.8	9.6	47.4
(Louw et al., 1993)	South Africa	-	Pure and cross	*B. taurus*	66	45- 90	DO	24	4.4	9.6	47.4
(Louw et al., 1993)	South Africa	-	Pure and cross	*B. taurus*	66	>90	DO	24	4.6	9.92	47.4
(Lidfors et al., 1994)	South of Sweden	7	Cross	*B. taurus*	67	1	DO	-	-	14.0	-
(Lidfors et al., 1994)	South of Sweden	7	Cross	*B. taurus*	67	65	DO	-	-	12.0	-
(Lidfors et al., 1994)	South of Sweden	7	Cross	*B. taurus*	67	123	DO	-	-	10.0	-
(Hogan et al., 2022)	Australia	6.1	Cross	*B. taurus*	17	0-5	DO, v. rec.	10	3.5	7.0	25.5
(Kour et al., 2018)	Australia	7	Pure	*B. indicus x B. taurus*	23	3-53	DO	12	-	9.3	-
(Kour et al., 2018)	Australia	7	Pure	*B. indicus x B. taurus*	23	3-53	xyz ACC	12	-	9.7	-
(Kour et al., 2018)	Australia	7	Pure	*B. indicus x B.*	21	17-135	DO	2	-	12.2	-

				taurus							
(Kour et al., 2018)	Australia	7	Pure	*B. indicus x B. taurus*	21	17-135	xyz ACC	2	-	13.7	-
(Kour et al., 2021a)	Australia	7	Pure	*B. indicus x B. taurus*	23	30	xyz ACC	24	9.8	8.3	82.3
(Kour et al., 2021a)	Australia	7	Pure	*B. indicus x B. taurus*	23	120	xyz ACC	24	8.2	9.3	77.2
(Kour et al., 2021b)	Australia	7	Pure	*B. indicus x B. taurus*	28	71-148	xyz ACC	24	7.7	9.4	73.3
(Paranhos da Costa et al., 2006)	Brazil	-	Cross	*B. indicus x B. taurus and B. indicus*	40	0-300	DO	12	2.6	9.3	23.8
(José Espinoza et al., 2011)	Mexico	-	Pure	*B. taurus*	20	30	DO	24	5.6	-	81.5
(José Espinoza et al., 2011)	Mexico	-	Pure	*B. taurus*	20	60	DO	24	5.2	-	65.5
(José Espinoza et al., 2011)	Mexico	-	Pure	*B. taurus*	20	90	DO	24	5.7	-	76.4
(José Espinoza et	Mexico	-	Pure	*B. taurus*	20	120	DO	24	5.6	-	73.0

al., 2011)											
(José Espinoza et al., 2011)	Mexico	-	Pure	*B. taurus*	20	150	DO	24	4.1	-	64.8
(José Espinoza et al., 2011)	Mexico	-	Pure	*B. taurus*	20	180	DO	24	3.3	-	40.6

*FREQ = Frequency; DUR = Duration; DO = Direct observation; v. rec. = Video recording; xyz ACC = triaxial accelerometer.

1. Calf Age and Weight

The age and weight of calves can influence suckling behaviour. Young calves, especially during their first days of life, have a strong instinct to nurse frequently and will actively seek out their mother's udder (Whalin et al., 2021). In contrast, older and heavier calves suckle less frequently because of their greater physical ability to suckle effectively, leading to longer nursing bouts (Odde et al., 1985; Kour et al., 2021a). As calves get older, they become less reliant on their mothers' milk, start to explore their environment (Lynch et al., 2019), and spend more time grazing (Khan et al., 2011). Several studies have reported an increase in suckling bout duration in older calves (Kour et al., 2021a; Nicol and Sharafeldin, 1975; Vitale et al., 1986) and in total suckling duration per 12 h (Paranhos da Costa et al., 2006). In addition, Odde et al. (1985) linked calf weight to its milk-drinking capacity, since these two parameters vary proportionally, thereby reducing suckling frequency per day. This relationship was expressed in the regression equation for calf weight (kg) on suckling incidence, where Y = 6.42 - 0.0083x. Similarly, the equation for the effect of milk production (kg/24 h) on suckling incidence was Y = 6.42 - 0.007x. These

equations suggest that high milk production and higher calf weight are associated with a decreased number of suckling bouts. Otherwise, several studies have indicated that suckling frequency (Lewandrowski and Hurnik, 1983; Somerville and Lowman, 1979), suckling duration (Louw et al., 1993; Reinhardt and Reinhardt, 1981), and total suckling duration (Kour et al., 2021a; Lewandrowski and Hurnik, 1983) are not influenced by calf age. However, the discrepancy between the studies could be due to various factors, including rearing conditions, breeds used, study conditions, health, social dominance, and the surroundings of the calves.

2. Calf Sex and Genetic Type

Concerning the effect of sex, male calves generally have higher milk requirements than female, due to their higher metabolic and growth rates (Goyache et al., 2003; McGee et al., 2016). Asheim et al, (2016) suggests that the general principle of feeding milk to beef calves is approximately 10% of their body weight. Furthermore, Mummed (2013) observed that Ogaden calves suckled 14%, 8%, and 3% of their body weight at the first, third, and sixth months of age, respectively. Consequently, an increase in suckling activity among male calves is expected to satisfy their requirements because male calves generally have a higher body weight than female calves (José Espinoza et al., 2011; Landete-Castillejos et al., 2005; Paranhos da Costa et al., 2006). Paranhos da Costa et al. (2006) reported that male calves exhibited a higher frequency and duration of suckling than female calves, with an average of 2.6 suckling bouts per 12-hour and a duration of 25.05 minutes per 12-hour, compared to the female calves' average of 2.12 suckling bouts per 12-hour period and a duration of 21.5 minutes per 12-hour period. However, several studies have shown that suckling frequency or duration is independent of calf sex (Lidfors et al., 1994; Nicol and

Sharafeldin, 1975; Odde et al., 1985; Reinhardt and Reinhardt, 1981). This difference between the studies could be due to the different rearing conditions and breeds used (Kour, 2018).

In a study by Das et al. (2000) it was observed that Zebu calves had a longer suckling bout duration (11.8 min) than Bos taurus × Mpwapwa crossbred calves (9.4 min). Nicol and Sharafeldin (1975), reported that Friesian × Angus calves had longer single suckling bouts, 35.6 minutes compared to 30.9 minutes in the purebred group ($p < 0.05$) compared to Angus calves, indicating that crossbred calves exhibited a prolonged duration of nursing compared to purebred Angus calves. According to Das et al. (2000), the extended suckling periods exhibited by crossbred calves can be considered an indication of heterosis, which may act as a stimulus for milk production, affecting suckling behaviour trait.

3. Cow breed and milk production

Maternal breed can influence suckling behaviour because of cow milk production and composition. In fact, the suckling behaviour of calves can change in relation to milk composition (in terms of fat or protein level) to meet their nutritional requirements (Kour et al., 2018). According to Das et al. (2000), the total suckling duration was significantly longer in Zebu calves (11.8 ± 0.19 min) than in crossbred Bos taurus × Mpwapwa calves (9.4 ± 0.19 min). Paranhos da Costa et al. (2006) found that the suckling frequency means of Gir and Nelore calves (2.56 ± 0.04 and 2.53 ± 0.03 suckling bouts/12 h, respectively) were higher than the mean of Caracu calves (2.03 ± 0.07 suckling meals/12 h). However, Caracu calves had a higher mean duration than Nelore and Gir calves. Other studies have reported an inversely proportional relationship between cow milk production and suckling frequency (Day et al., 1987; Odde et al., 1985). As milk production increases, calves need

to suckle less frequently to meet their nutritional requirements (Day et al., 1987). These authors reported that the frequency of nursing declined as estimated milk production increased at 52 days of lactation but not at 104 or 167 days of lactation. Das et al. (2000) found that when cows have higher milk yields, calves tended to have longer suckling bouts duration. Additionally, as stated by NRC (2016), the peak of milk production in beef cattle occurs at an average of 8 weeks postpartum. At the same time, several factors can influence milk production, including genetic type, nutritional status, cow age and parity, and environmental conditions such as temperature and humidity. In general, adult cows require a balanced diet with adequate levels of protein, energy, fibres, and other nutrients to produce milk (Sarwar et al., 2009). Factors such as the quality and quantity of forage available, as well as the use of supplements and feed concentrates, can all impact the nutritional status and, therefore, milk production (Hills et al., 2015). Das et al, (2000) showed that high concentrate supplementation improved milk yields and mentioned greater milk production in crossbred cows compared to Zebu cows. Another study reported that the average milk production of different breeds, including Hereford, Charolais, Limousine, Salers, and Obrak was 4.7 kg per day (Bakharev et al., 2021), with the highest milk yield recorded in Charolais cows, the limousine cows had the lowest, and Hereford cows had the most nutritious milk.

Furthermore, different cow breeds can manifest differences in udder size and shape, affecting milk flow rate and the ability of the calf to locate and suck a teat (Beard et al., 2019).

4. Circadian Variation

Calves and cows under pastoral conditions tend to adopt a repeated suckling pattern that occurs during 24 h circadian behavioural rhythms. The suckling activity recorded in most studies occurs at dawn, noon, and late evening (Álvarez-Rodríguez and Sanz, 2009; Hogan et al., 2022; Odde et al., 1985). The results regarding the distribution of suckling activity between the day and night are contradictory. A previous study (Somerville and Lowman, 1979) reported similar suckling periods, whereas other studies (Álvarez-Rodríguez and Sanz, 2009; José Espinoza et al., 2011; Lewandrowski and Hurnik, 1983; Odde et al., 1985) observed more suckling events during the day. Louw et al. (1993) indicated that nocturnal suckling activity was minimal, and generally, no suckling bouts were recorded after midnight. The presence of nighttime suckling might be influenced by the artificial lighting, such as flash or car lights, used to facilitate observations.

In addition, some authors (Edwards, 1982) have reported that calves may have been more lethargic in extreme weather. In cold, high temperature and humidity, rain, or strong winds, animals become behaviourally inactive and seek shelter. Another important factor is the social structure of the herd, impacting maternal behaviour and the ability of the cow-calf pair to have access to water and feed, which may affect suckling behaviour (Bouissou et al., 2001). Furthermore, despite the low probability of twin calving incidence in beef cattle, when it occurs, it may have an impact on suckling traits, as the dam of twin calves produces only 20–40% more milk than rearing a single calf, which may induce insufficient milk intake (Waltl et al., 1995). Other authors have reported that suckling frequency and duration are higher in multiparous and older cows than in primiparous cows (Vandenheede et al., 2001). In addition, some studies have indicated that first-calf heifers generally produce

lower levels of oxytocin than cows with prior calving experiences (Uvnäs-Moberg et al., 2001). This difference in oxytocin levels may contribute to more efficient maternal behaviour in multiparous cows than in primiparous cows (Vandenheede et al., 2001). Nonetheless, the presence of unknown people in pastures can cause anxiety in cows, which may also lead to an alteration in suckling behaviour (Dodzi and Muchenje, 2011).

Understanding these factors can assist farmers and researchers in creating optimal conditions for cattle, promoting healthy calf growth and development, and ensuring the herd's overall productivity.

Effect of Suckling Behaviour on Reproduction and Production

1. *Postpartum anoestrous period*

Following parturition, there is a period of ovarian inactivity characterised by suppressed ovarian follicular growth, lack of responsiveness to gonadotropin release, and absence of oestrus (Xavier et al., 2018). This phenomenon, also known as postpartum anoestrus, is primarily influenced by suckling behaviour, social cohesion between calves, and nutrition (Marongiu et al., 2002). Numerous studies have confirmed that suckling can affect reproduction. It can prevent the onset of the oestrus cycle and delay the onset of ovulation, as mentioned in a review by Montiel and Ahuja (2005).

Many management practices have been introduced to minimise the effect of suckling behaviour on the postpartum oestrous cycle of cows. Some studies have reported that restricted suckling or temporal weaning ranging from 12 h to 8 days, from 45 d postpartum until the beginning of the breeding season, has been associated with improved reproductive performance (Escrivão et al., 2012, 2009). This practice involves limited calf access to

maternal milk for a specific daily period, while being separated from their dams for the remaining duration (Broucek et al., 2020). It is commonly used in beef production systems to increase the productivity of cows by reducing their energy requirements for milk production and allowing them to regain their body condition during the postpartum period (Galina et al., 2001). Moreover, Martins et al. (2012) compared early weaning, in which calves were separated from their dams at 91 days of age, interval weaning which involved separating the calves from their dams for 48 h, repeated five times at 20-day intervals, and the control group following a single 48-hour calf withdrawal. These authors reported that early weaning cows had higher body weights and tended to have higher body condition scores than interval-weaning and control cows. In the aforementioned study, cows in the early weaning and interval weaning groups resumed cyclicity earlier than the control cows, and interval weaning cows had a higher pregnancy rate. Similarly, Rodríguez and Segura (1995) found that once-daily suckling starting 30 d post-calving increased pregnancy rates in Zebu cross cows without affecting calf growth. Mukasa-Mugerwa et al, (1991) demonstrated that continuous suckling or constant cow-calf interaction extended the postpartum anoestrus interval. Quintans et al, (2010) observed that temporary suckling restriction improved reproductive performance in cows with moderate body condition. Das et al, (2000) mentioned that increasing nutrition in restricted suckling cows improved the anoestrous period and led to higher live-weights and daily weight gains in calves. Moreover, these authors reported that crossbred cows (*Bos taurus* × Mpwapwa) had a shorter postpartum resumption interval than did Zebu cows. Reducing the traditional weaning age of Bos indicus cows from 7 to 4 months has been shown to result in significant advancement of the next calving date by up to 39 days (Schottler and Williams, 1975). In

addition, Reis et al. (2023) have reported that for every seven days that weaning is brought forward, the pregnancy odds of cows increase by 14%. However, these practices require careful monitoring of the nutritional status and health of both cows and calves, as well as appropriate management of the separation and reunification processes to ensure the welfare of the animals and success of the practice (Orihuela and Galina, 2019).

Suckling management can also facilitate the weaning process by gradually reducing the dependency of calves on their dam's milk (Acevedo et al., 2005). In fact, the loss of access to the udder, and thus suckling behaviour, and the break of the bond with the mother causes emotional discomfort to the calf (Fernandez-Novo et al., 2020). Weaning distress was reviewed by Enríquez et al. (2011), who provided valuable insights into diverse weaning techniques aimed at mitigating the challenges associated with weaning beef calves. Their study evaluated the outcomes of various methods, including the use of nose-flaps, fence-based separation, and practices such as "creep feeding", which involves allowing calves access to supplemental feed or concentrate while still in contact with their mothers, facilitating the gradual replacement of milk with solid food, and encouraging independence from the mother. Similarly, "creep grazing", which entails providing calves with access to high-quality pasture before complete separation, promoting the early consumption of solid food. These authors reported that six to seven-month-old calves conditioned to hay through creep feeding exhibited longer feeding periods and reduced behavioural distress during weaning. These practices have demonstrated positive outcomes, highlighting their potential to ease the transition of beef calves by stimulating solid food consumption and minimising stress-related behaviours (Enriquez et al., 2011).

2. Effect of suckling behaviour on calves' production

In beef suckler cow-calf systems, calf weight at weaning is a crucial factor in determining production efficiency (Taylor et al., 1955; Walmsley et al., 2018). Weight gain during the pre-weaning period and weaning weight of beef calves are generally associated with advantages for animal welfare and profitability (Minick et al., 2001), which is typically achieved when calves are allowed to exhibit their natural behaviours and gradually transition from their dam's milk to solid by increasing the time spent grazing and ruminating (Whalin et al., 2021). As mentioned previously, the weight gain of beef calves is influenced by their suckling behaviour, as their primary nutritional intake is milk (Beal et al., 1990). Studies have found that suckling cows with high milk production have greater weaning weight and post-weaning growth than suckling cows with low milk production (Casasús et al., 2002; Clutter and Nielsen, 1987; Wright et al., 2019). A positive correlation was found between daily weight gain and milk sucked by Ogaden beef calves at the first and third months of age in a study conducted by Mummed (2013). This researcher also reported that weight at three months showed a significant correlation with weight at four, five, and six months of age, highlighting the influential role of milk in determining weaning weight. Moreover, many studies have found that cow parity has a significant impact on calf production, as mentioned in the meta-analysis by Sapkota et al. (2020). These authors reported a 14.8% higher milk yield in multiparous cows than that in primiparous cows. Moreover, some researchers have reported that calves born to multiparous cows are heavier and have higher average daily weight gain than those born to primiparous cows (Goyache et al., 2003; Linden et al., 2014; Toušová et al., 2014). Other authors (Nelsen and Kress, 1981) have also pointed out that male calves are heavier at weaning and grow faster than

females. In the Asturiana de los Valles breed, the average weaning weight and average daily weight gain of males were 8% higher than those of females (Goyache et al., 2003).

Thus, literature has demonstrated the factors that influence the production traits of calves during the suckling period. This knowledge can be used to improve beef cattle selection methods and to enhance management practices during the suckling phase.

Conclusion

In conclusion, the monitoring of suckling behaviour of beef calves on pasture is an important aspect concerning both the welfare and productivity of the animals. The knowledge gained in this review will help to optimise management practices during the suckling phase. By adopting appropriate monitoring techniques, researchers can ensure accurate and unbiased data while maintaining animal welfare. Understanding the factors that influence suckling behaviour can help in beef cattle selection and enable practitioners to provide an optimal environment for both cows and calves. Moreover, understanding the impact of suckling on the postpartum oestrus cycle facilitates the enhancement of management strategies such as short-term separation, restricted suckling, or temporary weaning, thereby improving reproduction and facilitating the weaning process.

The use of remote technologies for monitoring cattle behaviour in grazing environments is promising for eliminating the constraints of direct observation regarding the number of animals or monitoring in limited spaces. However, several knowledge gaps still need to be addressed. It is important that future research studies consider the effects of indirect factors influencing calf behaviour, such as environmental conditions, pasture quality, and water and shelter availability, and their relationship with direct factors that were detailed in this

review. Additionally, it is important to investigate the relationship between calf health, productivity, and suckling behaviour, as well as their impact on cow performance.

Overall, this review highlights the importance of obtaining more information on the suckling phase of calves in pasture-based herds. Further research is needed to better understand beef cattle suckling behaviour on pasture, to develop practical management strategies that ensure the sustainability and profitability of the beef cattle industry, and to optimise production outcomes.

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CHAPTER 2

Behavioural Evaluation of Beef Suckler Cows in Continuous Grazing: Implications of Calving Season Variations

Abstract

This study explored the behavioural patterns of indigenous beef suckler cows, under continuous grazing conditions, with a specific emphasis on the influence of the calving season. We aimed to offer valuable insights into the behavioural adaptations of these cows within the context of Mediterranean foraging seasonality. Highlighting the significance of behavioural patterns such as grazing, rumination, resting, and walking, this study revealed the ability of cattle to adapt these behaviours to seasonal variations. Using the 1-min scan sampling method for direct observation, two distinct groups of cows, calved in winter (Group W) and spring (Group S), were monitored by one observer on two occasions per day, divided into 10 sub-periods of 30 min each. Statistical analyses confirmed the significant impact of the calving period on certain behaviours, particularly grazing and rumination activity. Group differences in grazing time were observed during the first subperiod, with subsequent periods showing a trend reversal influenced by social facilitation. Group W exhibited significantly higher rumination times (8.9±2.45) compared to Group S (4.4±1.56), with a p-value of 0.0449. No significant differences in rumination activities were observed between the sub-period monitoring sessions (p=0.1504). Circadian rhythms were evident for certain activities, notably grazing. Resting and walking activities were not significantly different between the two groups (p= 0.96 and 0.73, respectively). Furthermore, this study explored the implications of behavioural variations in production traits. Group W exhibited significantly higher body weights (493.4 ± 76.41

kg) and greater weight recovery (0.59 ± 0.06 kg/day) compared to spring-calved cows (body weight: 444.4 ± 36.425 kg; recovery rate: 0.08 ± 0.06 kg/day; $p < 0.001$). This research contributes valuable insights into the relationship between the calving season, cattle behaviour, and production outcomes in pasture-based systems. These insights can assist in making informed decisions regarding the selection of an optimal calving season, thereby enabling more efficient utilisation of available resources.

Keywords: Beef Suckler Cows, Calving Season, Cow Behaviour, Mediterranean, Foraging Seasonality

Introduction

Over centuries, monitoring cattle behaviour has been a fundamental management indicator to satisfy nutritional requirements and design management strategies (Decandia et al., 2017; Estermann et al., 2003; Parnell et al., 2023). This practice provides valuable insights into their spatial distribution, foraging patterns, and animal performance (Parnell et al., 2023). Pasture-based cow–calf operations are critical components of beef production systems worldwide (Greenwood, 2021). The efficacy of this production system is associated with the ability of grazing animals to obtain a diet that meets nutritional requirements for maintenance, growth, and reproduction. The prevalence of pasture-based cow-calf operations in major beef-producing regions and countries further emphasises the significance of understanding the behaviour of grazing animals on pastures (Greenwood, 2021). Consequently, monitoring cattle behaviour is not only valuable but also essential for optimising production outcomes. It provides valuable information on food and water intake (Chapinal et al., 2007), and can improve animal performance, health, and welfare (Charlton and Rutter, 2017; Huzzey et al., 2006; Urton et al., 2005). Braghieri et al. (2011) highlighted the importance of ingestive and social behaviours in determining the degree of adaptation of cattle to their environment.

Emphasising the behavioural patterns in pasture-based systems, Kilgour (2012) showed that beef cattle in pastures primarily engage in grazing, ruminating, and resting, which constitute the majority of their daily activities. Grazing behaviour is particularly important in meeting the nutritional needs of beef cattle for maintenance, growth, and reproduction and ultimately affects their productivity. Napolitano et al. (2011) further elucidated that grazing behaviour is determined by the animals' choice of plant selection

and the duration of their search. Adaptations in grazing time and biting rates reflect their preference for richer forage and their ability to overcome seasonal forage availability (Hejcmanová et al., 2009). According to Ginane et al. (2003), when forage abundance decreases, animals increase grazing time to meet their nutritional requirements. Ponnampalam et al. (2022) reported that grazing time tends to decrease at elevated temperatures and humidity, and animals seek shelter under extreme conditions, such as cold, rain, or intense winds. Other authors (Cazzuli et al., 2023) have highlighted the impact of feed supplements on grazing behaviour, as unsupplemented animals are more likely to graze longer than supplemented animals.

Rumination is of utmost importance in cattle (Kilgour, 2012) because of its role in the digestion and absorption of nutrients (da Silva Santos et al., 2023). Cattle exhibit variations in cud-chewing patterns, including regurgitation, chewing, and swallowing of consumed feed (Napolitano et al., 2011). The duration and frequency of rumination vary depending on diet, particularly with regard to the protein and fibre content of the feed, with longer rumination periods observed in high-fibre diets, stemmy grass species, and young grass with a higher percentage of crude protein (Beauchemin, 2018).

Additionally, resting behaviour, encompassing sleeping and lying activities, serves as an indicator of animals' adaptation and well-being with their environment and surroundings (Fukasawa et al., 2018). Traditional breeds, such as the Sarda breed, are willing to engage in behaviours other than simply meeting nutrient requirements to explore their environment (Hessle et al., 2008). Improving the knowledge of behavioural activities, especially feeding behaviour, is crucial for optimising the use of available plant and animal resources (Parnell et al., 2023). Previous research by Foris et al. (2019) and Gygax et al.

(2010) revealed strong social bonds and hierarchical differences among cattle that typically live in groups in natural environments. Other studies have outlined leadership and follower roles (Harris et al., 2007; Šárová et al., 2010), coordinated time budgets during grazing (Braghieri et al., 2011b), and increased synchronisation of lying between nearby individuals in a group (Stoye et al., 2012). This phenomenon has been studied in various terms, such as social facilitation, allelomimicry, and contagious behaviour, which are fundamental in understanding interactions within social animal groups (Keshavarzi et al., 2020). According to Nicol (1995), social facilitation occurs when the actions of an individual influence behaviours similar to those of others. Living in groups provides advantages, such as better protection against predators and improved foraging for social animals (Rubenstein, 1978). The influence of peers within these groups, observed through interaction and observation, significantly affects decisions related to forage selectivity, access strategies, and predator avoidance (Galef and Laland, 2005; Reader, 2016).

To date, many methodologies have been validated for monitoring beef cattle behavioural activities under extensive system management (Nevard et al., 2022). As mentioned in Chapter 1, these methodologies can be classified into two categories: direct observation and automatic techniques. The choice of the behaviour observation method should be based on many aspects. The effectiveness of the chosen methodology and its suitability for specific research question, sampling sizes, and budget (Altmann, 1974; Van Der Marel et al., 2022). No method can be considered ideal for behavioural sampling, because both direct and automatic monitoring techniques have limitations. Nevertheless, direct observations have been considered the gold standard for behavioural sampling (Poulopoulou et al., 2019). These methods have also been used to validate automated

methods (Gelardi et al., 2020; Rombach et al., 2018). Despite the numerous advantages of automated techniques, some limitations should be considered when selecting the monitoring sampling methods. These limitations are due to short periods of power autonomy, internet access, poor signal strength, and memory shortage (Morrone et al., 2022; Rushen et al., 2012). Among the various direct observation methods employed, scan sampling is the most widely used method. This method involves sampling behaviours from an entire group with predetermined time points and obtaining the proportion of time animals engage in definite behaviours (Van Der Marel et al., 2022). This method was first introduced by Altmann (1974), and has since been adopted by numerous researchers (Leruste et al., 2013; Neisen et al., 2009; Pullin et al., 2017; Schanz et al., 2022).

This study aimed to evaluate differences in the behaviour of suckler cows, characterised by different calving seasons, under a continuous grazing system. This is of particular importance because the suckler cow-calf system is the predominant livestock system used in beef production in Sardinia and other Mediterranean regions. Hence, understanding the behaviour and performance of suckler cows is crucial for more efficient management and sustainable cattle operations.

Material and Methods

1. Experimental Site and Animals

The experiment was performed in compliance with the Italian guidelines on animal welfare (DL No. 116, 27/01/1992) and Directive 2010/63/EU for animal experiments. The study was conducted during spring between May and June 2022 at the experimental farm of Foresta Burgos managed by AGRIS Sardegna (The Agricultural Research Agency of Sardinia), located 850 m above sea level in the inner mountainous area of northwest

Sardinia (latitude 40°25′ N, longitude 8°55′ E). The site is characterised by a typical Mediterranean climate with dry summers and rainy winters, and an average annual rainfall of 816 mm. The environmental data were collected daily from May to June 2022 from the nearest weather station managed by AGRIS Sardegna and included measurements of air temperature, surface temperature, humidity, and precipitation. The maximum air temperature reached 35°C, and the minimum was 2°C, with daily average temperatures ranging from 9°C to 25°C. Surface temperatures showed a maximum of 43°C and a minimum of 4°C, with averages between 11°C and 31°C. Relative humidity remained high, with a maximum of 99% and a minimum of 7%. The highest recorded daily rainfall was 42 mm, while some days had no precipitation.

Ten multiparous Sarda beef cows (aged 8 ± 4.5 years) were selected from a herd maintained at Foresta Burgos and classified into two groups distinguished by distinct calving periods. Group W was composed of five cows calved during the winter season, with an average calving date of 12/12/2021 ± 9 days (mean ± s.d). Group S included five cows that calved during spring, with an average calving date of 07/04/2022 ± 20. The average initial live weight (LW) and average body condition score (BCS; scale of 1 to 5 (Ayres et al., 2009; Lowman et al., 1976)) were 486 ± 69 kg and 3.6 ± 0.6 for Group W and 431 ± 30 kg and 3.25 ± 0.25 for Group S, and were not statistically different. All cows grazed together as one herd on a natural pasture of 12.5 ha, composed of 73.4% grasses, 10% legumes and 16% other species with 19 scattered oak trees (Quercus pubescens L.,). The cows and calves did not receive any supplementary feed during the experiment and water was available from water troughs (ad libitum). Live weight was assessed by individual weighing every three weeks in the morning (0800 CET).

2. Behavioural monitoring data

Behavioural data were collected by direct observation using the 1-min scan sampling method previously adopted by Büchel and Sundrum (2014) and Werner et al. (2018). Two trained observers recorded data. Each group of monitored animals (A and S) was observed by one observer on two occasions per day between the morning (08:00) and dawn (21:00). The individuals in each group were watched cyclically every 30 min, and each 30-mins sub-periods monitoring session was divided into 60 s sample intervals. Behavioural patterns (Table 1) were classified as grazing, ruminating, walking, and resting, as described by Pereira et al. (2020) with modifications. The observed data were documented on a spreadsheet and entered in Microsoft Excel 2018.

Table 1. The behavioural categories used to monitor the animals were adapted and modified from Pereira et al. (2020).

Behaviour	**Definition**
Grazing	Muzzle near the ground, taking frequent bites of forage, and chewing it
Ruminating	moving the jaw in circular motions.
Walking	Continuous movement forwards or backwards
Resting	Lying down or knees bent on the ground

3. Statistical analysis

To compare the effect of calving date on cow behaviour at pasture, the total time allotted

to each activity by the two groups (A and S) during the 10 sub-period monitoring sessions on all experimental days was measured. Statistical analyses were performed using the R software (version 4.0.2, R Foundation for Statistical Computing, Vienna, Austria). To test the effect of calving date on the total time devoted to each activity, a general linear mixed model (GLMM, glmmTMB function of R..) was used, with the time of day at which the monitoring session was carried out (represented by the 10 sub-periods into which the monitoring sessions were divided), groups (A and S), and their interaction as fixed factors, and cattle as a random factor. The estimated means and standard errors (SE) were reported for each monitoring session and group. Post-hoc pairwise comparisons were also conducted (Acciaro et al., 2022), and an F-test was used to determine the differences between treatments. Tukey's multiple comparison test was used to evaluate pairwise comparisons between the treatment group means. Treatment differences with a p-value less than or equal to 0.05 were considered significantly different.

The trend in cow weights in the two experimental groups was analysed using a mixed model (lme procedure of R) with the weight of animals at the beginning of the experimental period as a covariate, treat (Group W or Group S) as fixed factor and animal as a random factor.

The weight recovery rate of cows in the two experimental groups during the experimental period was analysed using a linear model (lm procedure of R), with the weight of animals at the beginning of the experimental period as a covariate and treat (Group W or Group S) as a fixed factor. Further analysis of the calf weight data was conducted using a linear model (lm procedure in R), with the time of calving (group S vs. group W) and sex as fixed effects, and the weight recorded during the first post-calving weighing as a covariate.

Results

The results of the statistical analysis showed that the interaction between treatment effect (the two different calving periods) and the effect of the time of the observation sessions was significant only for the grazing activity. The results regarding this activity are then shown by group for the different sub-periods (Table 2). Statistical differences between the two groups in the time spent grazing were evident only in the first sub-period of the day. In particular, the S group showed greater activity in this sub-period, whereas the reverse occurred in the 2nd and 3rd sub-periods, when group W showed a tendency (P-value = 0.06) for increased grazing activity. Subsequently, the cows in group W, stimulated by the cows in group S (a phenomenon known as social facilitation), began grazing. During the day (sub-periods 4, 7, and 9; Table 2), group S cows showed greater grazing activity, consistent with their presumably greater nutritional needs, but without reaching statistical significance.

The analysis also revealed a significant difference in rumination time between Groups W and S ($\chi^2 = 4.0219$, $p = 0.0449$). Group W ruminated more than Group S (8.909±2.452 vs. 4.406±1.56) (Table 3); however, the effect of the monitoring sub-periods sessions on the time spent ruminating did not reach statistical significance ($p= 0.15$).

However, there were no differences in the time devoted to the two activities of shorter duration (walking and resting) between the two experimental groups (Table 3).

Table 2. Time spent grazing, expressed in minutes, by two groups of cows with different calving seasons for each 30-minute sub-period representing different observation sessions throughout the day.

Subperiods[1]	Group W[2]	Group S[3]	P-value
1	2±2	18±8	0.045
2	18±8	3±3	0.061

3	25±9	6±4	0.068
4	2±2	11±6	0.164
5	2±1	2±2	0.317
6	5±4	2±2	0.465
7	11±6	24±9	0.219
8	22±8	12±7	0.348
9	9±5	21±8	0.206
10	3±3	2±2	0.907

1.Subperiods into which the direct observation sessions were divided 2. Cattle with winter calving period. 3. Cattle with spring calving period

Table 3. Time allocated to various behavioral activities (expressed in minutes spent on each activity during the sub-period of the monitoring session) by two groups of cows with different calving seasons.

Activity	Group W[2]	Group S[3]	P-value
Ruminating	8.909±2.452	4.406±1.56	**0.045**
Walking	0.16±286	0.14±0.247	0.73
Resting	6±2	6±2	0.96

2. Cattle with winter calving period. 3. Cattle with spring calving period

The results of the behavioural measurements demonstrated some effect of the calving date on cow behaviour patterns. Additionally, the time of monitoring sessions shows the circadian rhythm of some activities (e.g., grazing), preferably performed at certain times of the day. (Table 2).

The cows in group S had a significantly lower average body weight than those in group W ($p \leq 0.001$). The final average body weight of the cows in group S was 444.16 ± 36.425, while the final average body weight of the cows in group W was 493.4 ± 76.41 (Figure 1).

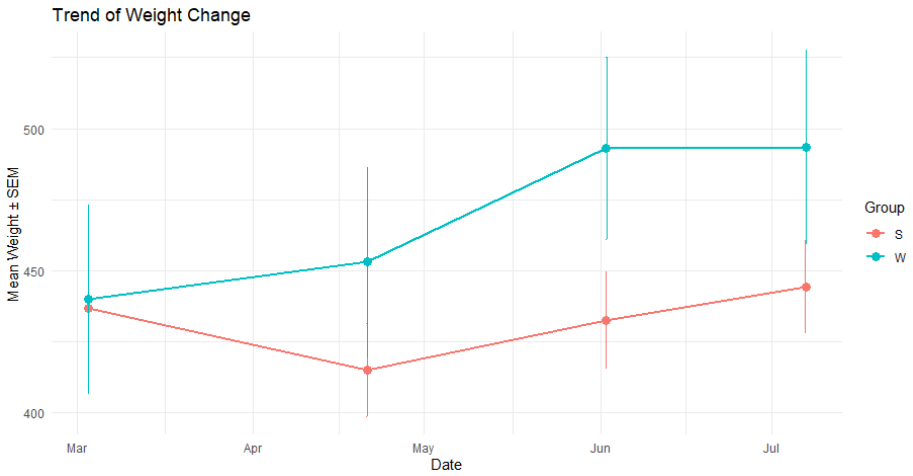


Figure 1. Comparison of cow body weight changes between groups W and S (S.E. are indicated by vertical lines).

Further analysis of the body weight recovery rate of cows revealed a significant difference between groups S and A. Animals in group W had a significantly ($p = 0.0005$) higher recovery rate (0.586 ± 0.058 kg/day, mean ± SE) than those in group S (0.084 ± 0.058 kg/day, mean ± SE) (Table 4).

Table 4. Comparison of body weight and growth rate between the two groups.

		emmean	**SE**	**DF**	**lower.CL**	**upper.CL**	**p value**
Body weight	Group W	478.239	5.915	7	459.7402	496.738	
	Group S	432.161	5.915	7	413.661	450.659	**< 0.001**
Recovery rates	Group W	0.586	0.058	7	0.404	0.769	
	Group S	0.084	0.058	7	-0.099	0.267	**0.0005**

emmean, estimated marginal mean; SE, standard error; DF, degrees of freedom; lower CL, lower confidence limit; upper CL, upper confidence limit.
The effects in bold characters are significant at $p < 0.05$.

The effect of the calving season on calves' growth rates was not statistically significant. The estimated marginal means (emmeans) were 0.85 ± 0.22 for calves born in spring (Group S) and 1.14 ± 0.16 for calves born in winter (Group W). Statistical analysis revealed no significant difference between the two groups ($p = 0.3$) (Table 5).

Table 5. Comparison of calves' growth rates between the two groups.

		emmean	SE	p value
Growth rates	Group W	1.140	0.22	
	Group S	0.85	0.16	0.3

emmean, estimated marginal mean; SE, standard error; The effects in bold characters are significant at $p < 0.05$

Discussion

This study aimed to compare the performance and behavioural activity of two distinct groups of cows that differed in the calving season within continuously grazed typical Mediterranean pastures, where seasonality poses a notable challenge for grazing ruminants (Serrano et al., 2020). Monitoring cow behaviour is an essential aspect of livestock management and provides insights into feed intake, animal performance, and overall health. Monitoring and tracking behavioural activity has been reported to offer valuable information, primarily concerning alterations in feeding quality, voluntary feed intake, grazing dynamics, metabolic processes, weather effects, and the impact on diseases that affect cattle appetite (Omontese et al., 2022). Consequently, farmers can gain valuable insights into determining the optimal calving season by considering the decrease in herbage quality, which may impact animal performance.

Our findings revealed some disparities between the two groups concerning grazing and rumination behaviours as well as notable distinctions in production traits, such as average body weight and recovery rates. Specifically, cows calved in winter exhibited significantly higher weights and recovery rates than spring-calved cows, consistent with observations in Spanish mountain conditions, where the rates were significantly higher in the winter calving herd (0.428 kg/day) than in the spring calving herd (–0.156 kg/day, $p<0.001$) (Casasús et al., 2009). Comparable seasonal effects were evident in Brahman and Belmont

Red Composite breeds, where growth rates during the wet season (+2 kg/day) exceeded those during the dry season (–1.54 kg/day) (González et al., 2014). Carta et al. (2009) underscored in their review the advantages of the winter calving season in Sardegna for sheep, emphasizing the optimal herbage utilization during the growth cycle. Consistent with our results, Iqbal et al. (2022) reported seasonal variations in grazing and ruminating time in dairy cows. In contrast, postpartum cow performance, as noted by Oddoye et al. (1999) was influenced by parity rather than season. A research conducted by Acciaro et al. (2020) under identical conditions as the present study, the aim to evaluate the performance of two groups of cows that calved in early autumn, and another group that calved in winter. The results demonstrated that both groups of cows displayed improved body weights, with no significant differences between the groups.

The results of the current study suggest that the spring-calving group, with higher nutrient requirements because of the coincidence of their early lactation stage with the experiment period, spent more time grazing and subsequently engaged in less time ruminating, which are the primary activities for grazing ruminants (Giovanetti et al., 2017; Iqbal et al., 2022; Pollock et al., 2022). Although, it is hypothesised that the increased grazing activity of the spring-calving group in the early morning hours may be attributed to their greater nutritional and energy demands for milk production. Cows tend to have higher milk yield in the morning (Palmer et al., 1994), emphasizing the importance of satisfying their nutritional needs during this period. Consequently, these higher nutritional requirements may have prompted the cows to start grazing earlier than in the winter-calving group. Calves in group S, being young, have higher nutritional requirements, demanding increased milk production from cows (Lohakare et al., 2012). This is in line with the results obtained

in dairy cattle by Iqbal et al. (2022), who indicated that nutritional requirements, in terms of feed intake, were associated with the lactation stage and milk yield. Cows with lower nutritional requirements may not exhibit an immediate need to graze after night rest, as they have less insistence on meeting their nutritional demands. Therefore, Group S may have allocated more time to grazing than Group W, which had lower nutritional needs and older suckling calves that were already capable of grazing. As mentioned before, the calving period did not have significant effect on "walking," and "resting." Owing to some organizational difficulties, it was not possible to allocate the two groups to separate and similar plots, leading to blurred differences in behaviour due to social facilitation, which stimulates animals in the herd to synchronise their behaviours to some extent (Ginane et al., 2015).

The direct observations also confirmed how cow grazing activity tends to be concentrated in the morning and late afternoon, (Table 2, (Kilgour et al., 2012))

Our findings provide novel insights into the existing literature by shedding light on the circadian variation in beef cattle behaviour. Consistent with the observations reported by Omontese et al. (2022), the lowest ruminating time was observed in the afternoon. In the present study, the grazing time of spring-calved cows was longer than that of winter-calved cows. These results were expected, as spring-calved cows may lengthen their grazing time to recover their postpartum body weight and adapt to the degradation in pasture quality that typically occurs in the late spring season (further information regarding pasture quality can be found in Chapter 3). Some studies have indicated that grazing duration often increases under conditions of limited forage availability (Ginane and Petit, 2005; Hejcmanová et al., 2009).

Conclusion

In conclusion, this study highlights the role of monitoring cattle behaviour in pasture-based cow-calf operations, emphasising its importance for optimising production outcomes. The observed behavioural patterns, particularly grazing and rumination, provide insights into the nutritional needs and adaptation of beef cattle to their environment. Notably, this study highlights the influence of calving season on the performance of postpartum suckler cows in continuous grazing systems. Cows calved in winter exhibited higher body weights and growth rates than those calved in spring, suggesting potential advantages of winter calving under the studied Mediterranean pasture conditions. These findings offer practical guidance to farmers for selecting calving seasons to enhance animal performance and well-being.

This relationship between cow behaviour and performance with seasonality could be further explored by extending this work, first at the monitoring level, allowing the characterisation of daily behaviour during 24-hour observation periods over 24-hour periods and including observations across different seasons to explore variations in behavioural activities and performance. Additionally, consideration of maternal behaviour in future studies may reveal potential effects on postpartum dam performance.

In summary, these findings contribute to our understanding of cattle behaviour in pasture-based systems and provide practical insights for extensive beef livestock production in Mediterranean conditions, guiding improved management strategies for enhanced productivity and welfare in cow-calf operations.

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CHAPTER 3

Effects of Calving Seasonality on Dry matter Intake and Performance of Beef Suckler Cows

Abstract

To assess the adaptability of indigenous suckler cows to varying lactation requirements under Mediterranean natural pastures, the herbage intake, digestibility, and faecal output were analysed for two groups of cows differing in calving season. The n-alkane technique was used to estimate herbage dry matter intake, herbage digestibility, and faecal output in the two groups, group W, calved in winter, and group S, calved in spring. The experiment revealed significant differences in herbage dry matter intake (expressed as a percentage of body weight; HDMIBW) between the two groups, with spring-calving cows exhibiting higher intake levels. HDMIBW was 3.58 ± 0.21 (estimated mean ± SE) kg DM in spring-calving cows and 2.81 ± 0.21 kg in winter-calving cows. No significant differences were observed in herbage dry matter digestibility (HDMD) between the two groups, with an average digestibility of 63.83 ± 1.81%. Faecal output (FO) was consistent between the groups, averaging 5.02 ± 0.28I kg DM. Herbage chemical composition analysis indicated medium to low energy and protein content and high NDF and ADF content during the experimental period. The results of this study can form the basis for informed and effective pasture management and calving season decisions to optimise beef production systems under Mediterranean conditions by considering factors, such as dry matter intake, digestibility, faecal output, body weight, and recovery rates.

Keywords: Forage Seasonality, Suckler cow, dry matter intake and digestibility, n-alkane technique, Mediterranean climate, pasture management, calving season.

Introduction

Energy intake is the most crucial factor determining the reproductive performance and growth rate of beef cattle (INRA, 2018). In Sardinia, suckler cows primarily belong to indigenous breeds, which are well known for their exceptional capacity to utilise natural forage resources (Acciaro et al., 2022). Therefore, determining the herbage intake and energy requirements of suckler cows is of utmost importance in such extensive systems. Sardinia has a dry Mediterranean climate, where seasonality is a challenge for grazing ruminants (Caballero et al., 2009; Serrano et al., 2020). The Mediterranean climate, with the exception of the alpine regions in Italy and the Euro-Atlantic and Alpine regions in Spain, is typically characterised by a hot and dry summer period, which corresponds to the lowest rainfall, and a cooler and wetter period, which coincides with the lowest temperatures (Caballero et al., 2009). According to Dovrat et al. (2021), climate and rainy season have a significant impact on herbage quantity and quality, particularly in terms of chemical composition and digestibility. During hot and dry seasons, herbaceous plants become senescent, resulting in high fibre levels and low crude protein concentrations (Larsen et al., 2021). According to Braghieri et al. (2011), the availability of herbage mass is the lowest during the dry period under Mediterranean conditions. Despite the acknowledged importance of forage availability, Dargatz et al. (2004) highlighted that only 9.3% of producers prioritised this factor when selecting the calving season, whereas tradition (29.7%) and anticipated weather conditions (39.4%) were the most prominent determinants. Johnson (2023) indicated that severe weather patterns can result in unfavourable health outcomes for both calves and cows during the calving process. Specifically, cows that give birth in hot weather are more susceptible to overheating and

exhaustion, with the likelihood of such occurrences being greater than those that give birth at cooler temperatures. Several studies have reported that even limitations in herbage quality and quantity can have a negative effect on spring-calving cows in several areas, including recovery of cow body reserves, reproductive performance, and calf growth (Acciaro et al., 2020; Hennessy et al., 2001). In fact, during the postpartum period in beef cattle, the energy requirements increase significantly, particularly in lactating cows (Bremer and Jenkins, 2015). Winter calving cows could reach peak lactation when pasture grasses are vegetative and rich in nutrients, allowing calves to initiate grazing and become prepared for weaning before the onset of dry seasons in the summer. Conversely, spring calving cows experience a gradual maturing of the pasture, resulting in a decline in the quality of the grass. The presence of calves, which are not yet ready to commence grazing and depend on their dam's milk for sustenance, necessitates a higher energy intake from the cow to produce milk. Accordingly, winter-calving operations have been determined to be more adequate for suckler herds reared in regions with dry summers (Casasús et al., 2002).

Effective pasture management is essential for optimising suckler cow beef production systems, as it enhances the reproductive and production traits of both cows and calves (Diakité et al., 2023). Management practices involve balancing the requirements of livestock with the potential supply and nutritional value of grass or other forage sources (Samuel and Dines, 2023); the performance of grazing ruminants depends on this balance (Dove, 2010). Thus, there is a critical requirement for accurate analyses to assess forage quality and quantity, implement grazing management strategies, and respond to dynamic pasture conditions (Peters et al., 2023). The efficacy of grassland management also depends

on the digestibility of the consumed herbage, which is influenced by both the biomass and nutritional quality of forage, as indicated by Jin and Hou (2022).The intake and digestibility of forage and pasture, greatly influenced by herbage composition and quantity, affect intake, rumination, and animal performance (Harper and McNeill, 2015). Numerous validated methodologies have been used to determine herbage intake and digestibility in ruminants (Garrett et al., 2020; Pepeta et al., 2022; Smit et al., 2005). These rely on techniques that typically employ the sward cutting technique (Smith et al., 2005), external or internal markers (Decruyenaere et al., 2009), and more recent techniques, such as Near Infrared Spectroscopy (NIRS,(Molle et al., 2023; Peters et al., 2023)), determination of faecal crude protein concentration (Lukas et al., 2021), and climate estimation combined with remote sensing, such as unmanned aerial vehicle methods and artificial intelligence (Jin and Hou, 2022).

Studies have shown that markers, such as acid-insoluble ash (AIA), n-alkanes, indigestible internal markers (INDF), and chromium oxide (Cr_2O_3) can provide highly accurate estimates (Andriarimalala et al., 2021; Dawson et al., 2000; Dove and Mayes, 2005; Pepeta et al., 2020; van Wyngaard et al., 2018). The n-alkane method was introduced in the late 1980s and early 1990s by Mayes et al. (1986) and Dove and Mayes (1991) . These indigestible compounds are naturally found and are the most prevalent saturated hydrocarbons in plant waxes, mostly consisting of odd-chain carbon molecules ranging from C21 to C37 (Bush and McInerney, 2010). These compounds are thought to contribute to the mitigation of water loss through transpiration (Hasanuzzaman et al., 2017). The prevalence of n-alkanes with odd numbers of carbon atoms in pasture species is remarkably high. Among these, C29, C31, and C33 are the most commonly observed alkanes (Mayes

and Dove, 2006). The concentration of alkanes within a forage species can vary depending on factors such as phenological stage, growth stage, and part of the plant (Égüez et al., 2022). There are also notable differences in the concentrations of alkanes among different plant species, with C31 and C33 being dominant in grasses (Bush and McInerney, 2013) and C29 in legumes (Boadi et al., 2002). They have been used as internal markers for estimating digestibility and are most commonly orally dosed in combination with even-chain n-alkanes to estimate feed intake in grazing ruminants (Premaratne et al., 2005). This method has been widely used in cattle studies (Bezabih et al., 2012; Sánchez Chopa et al., 2012; Wright et al., 2019; Wegi et al., 2021). The method for estimating herbage intake is based on separate estimates of faecal output and digestibility of grazed forage (Mayes and Dove, 2000). When a known quantity of indigestible marker is ingested and allowed to reach equilibrium within the digestive system, it is expected that the same amount will be excreted in faeces (Coleman, 2005). According to Pepeta et al. (2022) faecal samples can be effectively collected through either rectal grab or ground pickup methods after a waiting period of 3-6 days following dosing initiation. Moreover, Ferreira et al. (2007) discovered that 3-5 days are sufficient for the administration of commercial alkanes (C32) to become uniformly distributed in faeces. Several studies have reported that the recovery of alkane markers can reach equilibrium four days after dosing in cattle (Molina et al., 2004; Oliván et al., 2007; Genro et al., 2009). Nevertheless, other researchers have reported that the marker dosage should be administered twice daily for more accurate results and to avoid errors (Andriarimalala et al., 2021). Previous research has suggested that the use of consecutive pairs with similar faecal recovery rates in intake calculations may result in inaccuracies owing to the incomplete faecal recovery of alkanes (Azevedo et al., 2014;

Andriarimalala et al., 2021). Other studies have highlighted other important factors, such as carrier material, as mentioned in the meta-analyses by Andriarimalala et al. (2020), including paper pellets, gelatin capsules, and controlled-release devices. According to Smith et al. (2007), the type of carrier material employed can affect the distribution of substances within the gut, which in turn may influence their recovery in faecal samples. The use of paper pellets is the most prevalent method for conducting experiments on cattle (Giráldez et al., 2004). Moreover, the average amount of dosed n-alkanes, frequency of dosage, and faecal sampling play a role in determining the validity of rectal grab samples as representatives of total voided faeces (Sibbald et al., 2000). Thus, the estimation of herbage intake is principally based on the determination of the alkane dose and the concentration of the marker in the herbage and faeces.

This study aimed to estimate the herbage intake, digestibility, and faecal output of two groups of cattle characterised by distinct calving seasons and grazing on natural pastures under Mediterranean conditions, using the n-alkane markers method.

Material and Methods

1. Experimental Site and Animals

This study is a continuation of that presented in Chapter 2. Information related to the Experimental Site and Animals are presented in Chapter 2, Section 2.1 (Experimental site and animals).

2. Herbage sampling

Grazing animals select for some plant species or plant parts with different n-alkane content than the herbage on offer. This behaviour, for the n-alkanes method, could be an important

source of error in the estimation of intake. Hence, the sampling of the herbage actually grazed is probably the most limiting step of the marker-based methodology, alkanes included. During this study, hand-plucked herbage samples simulating grazing behaviour (HS) were collected on alternate days. The method used to observe forage preference involved close observation of randomly selected cows as they grazed on the herd. The cows were monitored by observers, who recorded the time spent on each plant species as the cows grazed (Acciaro et al., 2020; Berry et al., 2002). When animals moved to another plant community, samples were hand-plucked and torn in the same way at the sward to mimic the actual bite size.

The collected herbage samples (HS) were subsequently subjected to a chemical composition analysis and alkane pattern assessment. Analysis of the HS showed that odd-chain n-alkanes were the most abundant, with C29 (234±36 mg/kg DM), C31 (170±20 mg/kg DM), and C33 (27±7 mg/kg DM) representing an average of 96.2% of the total.

3. Intake estimation

The herbage dry matter intake (expressed as a percentage of body weight, HDMIBW) was estimated for both groups (winter-calving cows, W and spring-calving cows, S) using the n-alkane technique (Dove and Mayes, 1991). Over a span of 14 days (from 18/05/2022 to 31/05/2022), cows were orally dosed with known quantities of C32 and C36 through hand-manufactured pellets as external markers, following the methodology proposed by Mayes et al. (1986). Each morning at 0800 CET, the cows were orally dosed with pellets containing 475 mg of C32 and 461 mg of C36. C36 was used to measure faecal output because of its high recovery rate (approximately 0.95) (Bezabih et al., 2012). The estimation of HDMI was based on the C31/C32 and C33/C32 ratios because of their low

discrepancy in faecal recovery, as proposed by Dove et al. (2002 and 2000), and Mayes et al. (1986).

In conjunction with herbage sampling, and starting from day 6 of alkane dosing, faecal grab samples were collected from each individual for the remaining 6 days (Malossini et al., 1996). The faecal grab samples were then bulked and dried for 48 h in an oven at 40 °C for chemical analysis (Kennedy et al., 2011; Vulich and Hanrahan, 1995).

4. Measurements

Herbage intake was estimated from the concentration of n-alkane markers C31 and C33 in the faeces and herbage samples (Mayes et al., 1986), as follows:

$$\mathbf{HI\ (kg\ DM/day)} = \frac{\mathbf{D_j}}{\left(\left(\frac{\mathbf{F_j}}{\mathbf{F_i}}\right) \times \mathbf{H_i} - \mathbf{H_j}\right)} \qquad \text{eq. 1}$$

where D_j is the concentration of even-chain n-alkane dosed to animals (C32, mg/day); F_j is the concentration of even-chain n-alkanes in the faeces (C32, mg/kg DM); F_i is the concentration of natural odd-chain n-alkanes in faeces (C31 or C33, mg/kg DM); H_i is the concentration of natural odd-chain n-alkanes in HS (C31 or C33, mg/kg DM); and Hj is the concentration of even-chain n-alkane in the pasture (C32, mg/kg DM).

Faecal output was calculated using the following equation:

$$\mathbf{FO\ (kg/day)} = \mathbf{D_j / F_j} \qquad \text{eq. 2}$$

D_j: concentration of n-alkane dosed to animals (C36, mg/day), and F_j is the concentration of even-chain n-alkanes in faeces (C36, mg/kg DM).

Consequently, digestibility was calculated as follows:

$$\mathbf{DMD}(\%) = (\mathbf{HI} - \mathbf{FO})/\mathbf{HI} \qquad \text{eq. 3}$$

2.5 Chemical analysis

HS were freeze-dried and milled through a 1-mm sieve. Samples were analysed to determine the dry matter content (DM), ash, ether extract (EE), CP (AOAC, 2016), acid detergent fibre (ADF), and acid detergent lignin (ADL) (Möller et al., 2009). The herbage and faeces grab samples were subjected to alkane pattern analysis through direct saponification, with slight modifications to the procedure described by Dove (1992). Alkanes quantification were performed using a GC Varian 3400 (Palo Alto, California) equipped with a flame ionisation detector (FID), with C34 serving as the internal standard (Mayes et al., 1986).

5. Statistical analysis

Statistical analyses were performed using the R software (version 4.0.2, R Foundation for Statistical Computing, Vienna, Austria). To assess the differences in intake, digestibility, and faecal output between the groups W and S, a linear model (lm function of R.) was used. The treatment groups denoted as 'group S' and 'group W' were considered as fixed factors in the model. The response variables were HDMIBW, HDMD, and FO. The outputs from the analysis were summarised, and estimated means (emmean), standard errors (SE), lower and upper confidence limits (lower.CL, upper.CL), and p-values were obtained for each treatment group. The statistical significance was set at $p < 0.05$.

Results

Across a total area of 12.47 hectares, the pasture contained nineteen trees. The herbage yield was 2210±690 (mean ± s.d) kg of dry matter per hectare and the dry matter content accounted for 18.8±1.5%. The most represented families were Poaceae (73.4%), Fabaceae (10%) and whereas other families represented 16.6%. The botanical composition of grazed herbage consisted predominantly of Gramineae (68.7 ± 23.1%), followed by Legumes

(10.2 ± 11.5%) and other species (21.1 ± 15.6%).

The chemical composition of HS is shown in Table 1.

Table 1. Chemical composition of herbage samples.

	Mean	SD
EE (% of DM)	2.95	0.36
Protein (% of DM)	15.82	3.07
CF (% of DM)	25.37	3
NDF (% of DM)	53.57	6.13
ADF (% of DM)	29.64	2.22
ADL (% of DM)	2.57	1.16
ADIN (% of DM)	1.77	0.68
Ash (% of DM)	11.37	1.01
IVDDM (% of DM)	68.91	5.12

SD, standard deviation; EE, Ether Extract; CF, crude fibre; NDF, neutral detergent fibre; ADF, acid detergent fibre; ADL, acid detergent lignin; ADIN, acid detergent insoluble nitrogen; IVDDM, in vitro dry matter digestibility.

The HDMIBW, HDMD, and FO values of the two experimental groups are listed in Table 2.

Table 2. HDMIBW, HDMD and FO in cows characterised by distinct calving seasons (winter-calving cows, group W and spring-calving cows, group S) estimated using C31/C32 and C33/C32 alkane ratios.

Parameter	Treat	emmean	SE	lower.CL	upper.CL	p value
HDMIBW C31/C32 (kg DM)	Group S	**3.433**	**0.207**	2.809	4.056	
	Group W	**2.741**	**0.207**	2.118	3.364	**0.04**
HDMIBW	Group S	**3.585**	**0.208**	2.958	4.212	**0.03**

C33/C32 (kg DM)	Group W	**2.815**	**0.208**	2.188	3.441	
HDMD C31/C32 (%)	Group S	62.314	3.28	52.421	72.206	0.95
	Group W	62.598	3.28	52.706	72.491	
HDMD C33/C32 (%)	Group S	64.298	2.703	56.146	72.449	0.81
	Group W	63.373	2.703	55.221	71.525	
FO (kg DM)	Group S	5.0224	0.42719	3.73409	6.31071	
	Group W	5.02242	0.42719	3.73411	6.31073	0.99

HDMIBW C31/C32, Herbage Dry Matter Intake as % of Body Weight estimated by C31/C32 pair; HDMIBW C33/C32, Herbage Dry Matter Intake as % of Body Weight estimated by C33/C32 pair; HDMD C31/C32, Herbage Dry Matter Digestibility estimated by C31/C32 pair; HDMD C33/C32, Herbage Dry Matter Digestibility estimated by C33/C32 pair; FO, faecal output.

The effects in bold are significant at $p < 0.05$.

In both estimates of herbage dry matter intake (using C31/C32 and C33/C32 pair of alkanes), group S consistently exhibited a higher intake than group W ($p = 0.03$ and $p = 0.04$, respectively) (Figure 3).

The violin plots in Figure 3 illustrate the distribution of HDMIBW estimated by C31/C32 and C33/C32 in Groups W and S. Each violin plot displays the probability density of the data, providing insights into the spread and central tendencies of the two groups.

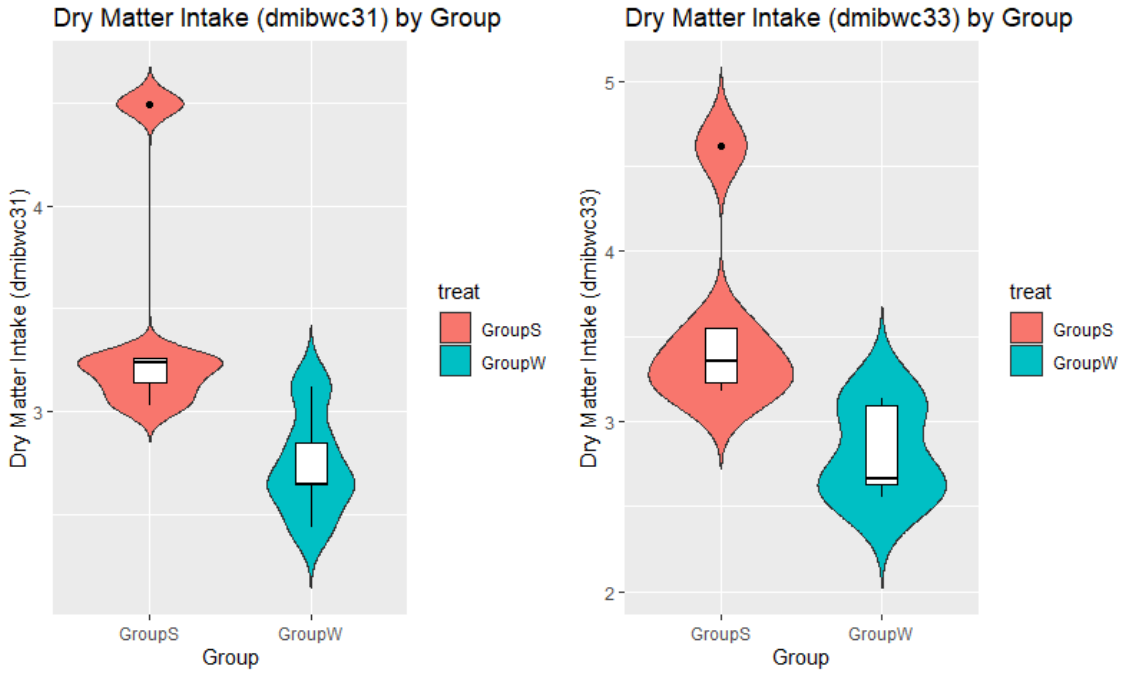


Figure 1. Violin Plots for Herbage Dry Matter Intake (expressed as percentage of body weight and estimated by C31/C32 and C33/C32 pairs) of the experimental groups.

The estimated HDMD expressed as a percentage, did not show a statistically significant difference ($p = 0.95$) between the groups.

The estimated means ± SE of faecal output for both groups (5.022 ± 0.4272 5.02242 ± 0.42719 for Group S and Group W, respectively) were comparable. There was no statistically significant difference between the two groups ($p = 0.99$).

Discussion

The pronounced seasonality of Mediterranean ecosystems affects pasture availability and quality, resulting in variable production traits for cows and calves, particularly during dry and hot seasons. Beef suckler herds in Sardinia face challenges related to seasonal variations in nutrient requirements. According to Casasús et al. (2005), the utilisation of Mediterranean forest pastures by suckler cattle is a promising management option, as it

considers both animal production and pasture management. Nonetheless, when it comes to animal production, dry cows with moderate nutrient requirements can experience net recovery of their body reserves, especially when grazing occurs during the favourable season. However, these authors have discussed the challenges faced by lactating cows and their calves under these conditions due to their higher nutrient demands, which were reported in their study by low productivity and reproductive performance. Therefore, concerns about suckler cow-calf production systems in extensive settings should include not only the estimation of nutrient requirements, but also the determination of animals' performance on pasture. This study, which was a continuation of the one in Chapter 2, aimed to estimate the feed intake of two distinct groups of grazing cows that differed in their calving season and to evaluate overall animal performance under typical Mediterranean conditions. Pasture characterisation, herbage yield, herbage family availability, HDMIBW, HDMD, and FO were examined. Moreover, BW and recovery rate (RR) were determined (more details are available in Chapter 2 section "Results"). As shown in Chapter 2, Group W exhibited significantly higher body weight and recovery rate (BW: 478.239 ± 5.915, RR: 0.586 ± 0.058) than Group S (BW: 432.161 ± 5.915, RR: 0.084 ± 0.058). This may be due to the original hypothesis that group W, during the experimental period, is characterised by lower energy needs due to the presence of already developed calves suckling less milk and being able to graze. In contrast, Group S, which had much younger calves, was expected to have higher energy requirements. In this study, the herbage dry matter intake was expressed as dry matter intake as a percentage of body weight, as described by Kubkomawa et al. (2013) as the most suitable method for comparing intake levels across forage and animals. The results indicated that Group S,

which had a lower BW, exhibited higher HDMIBWC31 (3.433 ± 0.207 kg DM/kg BW) and HDMIBWC33 (3.585 ± 0.208 kg DM/kg BW) than Group W (HDMIBWC31 (2.741 ± 0.207 kg DM/kg BW) and HDMIBWC33 (2.815 ± 0.208 kg DM/kg BW). The higher herbage intake in Group S can be explained by their higher energy need due to the early lactation phase and subsequent postpartum recovery of body weight.

In the current study, the chemical composition of herbage samples was estimated during the experimental period which took place during spring between May and June 2022. Comparable findings were found by Sitzia et al. (2012), who reported CP of 14 (% DM), NDF of 48.6 (% DM), ADF of 27.8 (% DM), and ADL of 4.8 (% DM) in northern Sardinia. However, a study conducted by Acciaro et al. (2020) at the same experimental site determined that the average chemical composition during the autumn-winter season was (as % DM): EE) 4.075, CP 21.95, NDF 43.25, ADF 22.875, and ADL 2.75. These values indicated higher energy and protein and lower fiber and lignin content than those observed in the current study. These findings align with the general hypothesis that better forage quality can be achieved during late winter-early spring period than during the period under the conditions examined in this study.

Regarding digestibility and faecal output, no significant differences were observed between the two groups, as expected. The lack of differences in digestibility and faecal output between the two groups could be attributed to logistical considerations. Both groups were grazed on the same pasture. Homogeneity in grazing conditions may have minimised the variables affecting digestion and faecal output. Moreover, Ferreira et al. (2007) reported that the faecal output of cows differs in terms of body size and breed; however, in contrast to the findings of the current study, in which cows belonged to the same breed, we did not

find a significant difference in faecal output.

Conclusion

This study focused on assessing variations in herbage intake, digestibility, and faecal output in two groups of cows with distinct calving seasons, grazing on natural pastures under typical Mediterranean conditions. The n-alkane technique was used to estimate herbage dry matter intake in both groups, revealing significant differences in dry matter intake as a percentage of body weight between the two groups. Group S, calving during spring, exhibited higher dry matter intake than Group W, because of their higher energy requirements. No significant differences were observed in herbage dry matter digestibility or faecal output between groups.

Overall, this research highlights the ability of environmentally well-adapted Sarda grazing cows to modulate herbage intake according to energy needs. The results made it possible to quantify the amount of herbage ingested on pasture by Sarda cows in a Mediterranean silvopastoral environment, a fundamental datum for the rational management of this livestock system. Moreover, the results underscore the importance of effective pasture management to optimise beef production systems, given the calving season and postpartum period for grazing cows. This study also provides insights into the chemical composition of the herbage samples, confirming the medium-to-low energy and protein content of the Mediterranean pasture during the experimental period. Further research and monitoring are recommended to enhance our knowledge of pasture dynamics and develop targeted management strategies for optimising beef production in such environments.

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CHAPTER 4

Net Carbon Footprint of Suckler cow-calf Farming in Sardinia: Implications for Carbon Sequestration

Abstract

The aim of this study was to determine the net carbon footprint (CFP) of ten extensive beef cattle farms in Sardinia that have adopted a suckler cow-calf system. The cattle on these farms included pure and/or crossbreeds belonging to Sardo-Bruno, Sardo-Modicana, Angus, Blu-Belga, Charolais, and Limousine. Animals primarily graze on pastures and are sold to a fattening centre at an average age of 9 months, with an average weight of 307.5 ± 79.77 kg (mean ± standard deviation). Data collection was conducted through surveys, considering the agricultural year from October 1st to September 30th of the past five years as the temporal boundary and the "Cradle-to-farm-gate" as the spatial boundary. Data analysis was performed using the IPCC Tier 1 and Tier 2 levels (2019). The individual contributions were expressed in terms of CO_2 equivalent (CO_{2eq}), considering the 100-year global warming potential or global warming potential (GWP) (27.9 CH_4 273 for N_2O and 1 for CO_2). The sequestration potential of CO_2 was estimated based on data reported in the literature. The functional unit (FU) considered was the kilogram live weight (kg LW), which refers to calves, bulls, and culled cows. The results showed that the gross CFP related to the production of 1 kg of LW was 17.31 kg CO_{2eq} and was mainly dependent on enteric methane emissions (70%), followed by feed (18.4%), energy (6.1%), and manure management (5.3%). When considering the CO_2 sequestration potential driven by pasture and other agricultural lands, a net CFP of -78.9 kg CO_{2eq} was derived. In conclusion, the employment of carbon sequestration in the CFP estimation made it possible to offset the carbon footprint of livestock farms by improving their environmental impact. However, it

is essential to implement efficient herd-management practices to enhance sustainability.

Keywords: beef cattle, carbon footprint, methane, suckler cow-calf, life cycle assessment, carbon sequestration.

Introduction

Over the last century, the beef industry has been facing remarkable challenges due to the continuously increasing demand (Pulina et al., 2021). The need to ensure high-quality beef production while considering animal welfare and environmental impacts necessitates the inclusion of better practices and the development of economically viable and sustainable production (Hocquette et al., 2018).

However, greenhouse gas (GHG) emissions from beef production are a major concern. Herrero and Thornton (2013) reported that cattle are responsible for approximately 78% of global livestock GHG emissions, with enteric methane (CH_4) being the primary source of beef GHG emissions, accounting for approximately 35% of total livestock emissions in carbon dioxide-equivalents (CO_{2eq}). Subsequently, beef cattle industries are considered significant contributors to climate change due to their substantial average emissions of atmospheric methane (CH_4) and nitrous oxide (N_2O) (Stackhouse-Lawson and Thompson, 2022).

GHG emissions result from multiple sources, including enteric fermentation, manure management, feed production, transportation, and energy consumption (Chen et al., 2020). The total GHG emissions from beef cattle production can vary due to factors such as feedlot manure handling systems, cattle diets, feed additives, and climate conditions (Guo et al., 2022). In addition, other factors, such as land use change and deforestation caused by the expansion of grazing lands, may also contribute to the increase in emission intensities (Chang et al., 2020; Gerber et al., 2013b). High emissions resulting from the deposition of manure on grasslands due to beef production, which is largely pasture-based, also contribute to this increase (Lombardi et al., 2022). Furthermore, factors such as breeding

overhead, which is linked to relatively low growth rates and fertility, also contribute to an increase in emission intensity (Herrero et al., 2015). Gerber et al. (2013) reported that lower growth rates result in increased age at first calving and the time required for beef cattle to reach slaughter weight, ultimately increasing the emission intensity.

Management strategies, such as land-based carbon sequestration and increased production efficiency, can be used to decrease gross greenhouse gas emissions from beef production (Hristov et al., 2013; Reyes-Palomo et al., 2022). According to Cusack et al. (2021), carbon sequestration on grazed lands and growth efficiency strategies can reduce gross GHG emissions by 62 and 30%, respectively. Pasture quality improvement is also a potential source of mitigation (Bogaerts et al., 2017). Sowing better quality pastures and superior pasture management can enhance forage digestibility and nutrient quality (Chang et al., 2020). Furthermore, intensive grazing management by improving the balance between forage growth/availability and grazing on promoting forage production and soil carbon sequestration has also been assessed as a mitigation strategy (Contosta et al., 2021). This may result in higher animal growth rates and an earlier age at first calving. Moreover, better nutrition can increase cow fertility rates and reduce mortality rates in calves and mature animals, thereby enhancing animal and herd performance (Gerber et al., 2013a). Bonnin et al. (2021) found that farms characterized by efficient production had lower global warming potential emissions in North Italy. Becoña et al. (2014) reported that improving grazing efficiency by optimizing the stocking rate and forage production increased beef productivity by 22% and reduced GHG emissions per kg LWG by 28% compared to "low performance" management in beef cow-calf grazing systems in Uruguay. Gere et al. (2021) reported that low-quality diets increased methane yield, and CH_4 production was higher

for diets with lower digestibility. Lombardi et al. (2021) highlight that dung from feeding management plays a crucial role in mitigating emissions. These authors found that dung from supplemented grazing steers produced significantly lower CH_4 and higher N_2O emissions (1.7 g CH_4-C/m^2 and 0.4 g N_2O-N/m^2) than dung from non-supplemented steers (4.0 g CH_4-C/m^2 and 0.1 g N_2O-N/m^2). In the context of livestock production, enhancing animal health and husbandry practices can contribute to lower mortality rates and higher growth and fertility rates, thus improving herd performance (Gerber et al., 2013a). Preventive measures such as routine vaccinations and stress reduction techniques, such as providing shade and water, can help to reduce disease prevalence and improve animal wellbeing. Furthermore, Hristov et al. (2013) emphasised that improving animal productivity through genetic improvement, nutrition, reproduction, health, and better management of animal operations can lead to a reduction in the number of animals required to produce the same quantity of edible products, thereby reducing the environmental impact of animal agriculture. According to Capper et al. (2009), the reduction of animals number in cattle farming has been identified as the main mitigation strategy for reducing the carbon footprint of the industry in the United States.

Various methodologies have been implemented to evaluate the environmental impact of livestock production systems (Ran et al., 2019). Among these, life cycle assessment (LCA) is the most widely used tool for assessing the environmental impact of any activity, including livestock production systems (Goglio et al., 2023). LCAs have become increasingly popular for assessing the environmental impact of livestock products, as they aim to cover the complete product value chain (De Vries et al., 2015). LCAs also encompass impact dimensions that extend beyond the production phase, such as

transportation, processing, consumption, losses, and reuse, along the product value chain. The international standards for LCA methodology are governed by the ISO 14040 series of standards and are commonly combined with the GHG emission algorithms recommended by the IPCC (Gavrilova et al., 2019).

Analysis of the environmental impact of beef cattle production systems involves four stages (Figure 4). The first stage defines the study objectives, outlines the functions of the product system, and establishes functional units (Putman et al., 2023). The functional units of beef cattle include live weight (Desjardins et al., 2012), carcass (Pishgar-Komleh and Beldman, 2022), meat (Jacobsen et al., 2014), and protein (McAuliffe et al., 2023). In addition, the boundaries of the system include cradle-to-grave (Asem-Hiablie et al., 2019) or cradle-to-gate (Hietala et al., 2021). The second stage involves data collection and computational procedures such as quantifying the inflow and outflow of the produced system (Hauschild, 2018). The third stage assesses the life cycle impacts of the products examined. The emissions and consumption of the production system identified in the previous stage will be converted into potential environmental impacts such as global warming, water and soil acidification, and water eutrophication (Putman et al., 2023). Simultaneously, the processes with the highest environmental impact can be identified. The fourth and final stage involves interpreting the results of the analysis and their contextualisation to provide useful information for designing production methods with lower environmental impact and greater eco-sustainability.

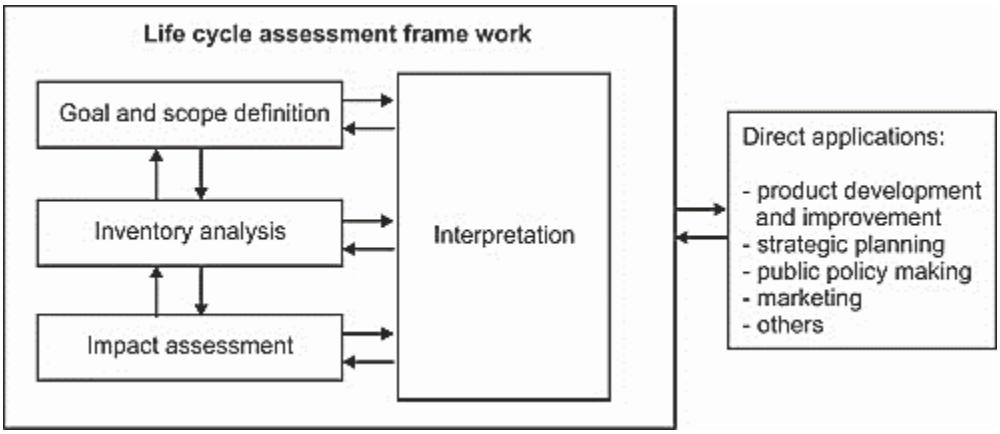


Figure 1. The four stages of LCA were conducted according to the ISO 14040 guidelines.

This study aimed to estimate the environmental impacts of beef production system in Sardinia, with a specific focus on suckler cow-calf farms. Additionally, this study assessed the impact of land-based carbon sequestration in mitigating these impacts.

Materials And Methods

1. *Study sites and animals*

The study was conducted across different beef cattle farms adopting the suckler cow-calf system and situated in Sassari (SS) and Nuoro (NU) provinces in Italy, covering different zones, such as Villanova Monteleone, Nulvi, Sindia, Bonorva, Sorgono, and Macomer. Cattle belong to diverse breeds, such as Sardo-Bruna, Sardo-Modicana, Charolais, Limousine, Angus, and Blue Belgian, as pure breeds and/or obtained by crossbreeding.

2. *Data collection*

Data were collected through on-site surveys, which served as the basis for subsequent analysis. The collected information included the following: the territorial location of the farm was determined by acquiring geographic coordinates, the total area, and more specifically, the utilised agricultural area (UAA) with specific crops and the trees/shrub species present and their use. Moreover, the total number of cattle raised and their distribution by sex and age category, average weight per head and

age category, mode of rearing, distribution of births and mortality rate, average length of stay of cattle on the farm's pastures, and type of supplementary feeding were surveyed. Other data, such as gross saleable production and feed supply categories (on-farm fodder production, purchases, and imported), water consumption and management, diesel fuel consumption for both crop operations and herd management, and agronomic and cultivation data were also included.

3. Spatial and temporal boundaries of the system

The study boundary included all the processes up to the animal farm gate (cradle-to-gate). The analysis accounted for the average agricultural year, covering from October 1st to September 30th, over the past five years. The production system is divided into four main processes: (i) animal husbandry, (ii) manure management, (iii) energy, and (iv) production and purchasing of feed. Electricity consumption and fuel combustion (direct) and production (indirect) were accounted as emissions from energy, while the manufacturing of fertiliser, pesticides, and seeds were accounted as secondary emissions in the category of feed production.

4. Estimating the Carbon Footprint

The CFP was estimated using the guidelines provided by the Intergovernmental Panel on Climate Change (IPCC) given in Volume 4 "Agriculture, Forestry and Other Land Use", Chapter 10 "Emissions from Livestock and Manure Management" of IPCC (2019) (Gavrilova et al., 2019) and Chapter 11 "N_2O emissions from managed soils, and CO_2, emissions from lime and urea application" (Hergoualc'h et al., 2019), regarding Tiers 1 and 2. The emissions of CH_4 from both enteric fermentation and manure management, N_2O from manure management (including those excreted directly during grazing), CO_2 due to energy production and use, and CO_2 related to feed supply were estimated.

5. Estimation of enteric methane emissions

Enteric methane emissions were estimated using GE which represents the gross energy measured in MJ/day (megajoules per day) and was calculated using IPCC Equation 10.16 (Gavrilova et al., 2019):

$$GE = \left[\frac{\left(\frac{NE_m + NE_a + NE_l + NE_{work} + NE_p}{REM}\right) + \left(\frac{NE_g}{REG}\right)}{\frac{DE\%}{100}}\right]$$

Where:

GE = gross energy, MJ/day

NE_m (MJ/day) = net energy required by the animal for maintenance,

NE_a (MJ/day) = net energy for animal activity,

NE_l (MJ/day) = net energy for lactation,

NE_{work} (MJ/day) = net energy for work,

NE_p (MJ/day) = net energy required for pregnancy,

REM = ratio of net energy available in a diet for maintenance to digestible energy consumed,

NE_g (MJ/day) = net energy needed for growth,

REG = ratio of net energy available for growth in a diet to digestible energy consumed,

DE%= digestible energy expressed as a percentage of gross energy.

After obtaining the GE, enteric methane emissions were estimated according to

IPCC Equation 10.21 (Gavrilova et al., 2019):

$$EF = \left[\frac{GE \times \left(\frac{Y_m}{100}\right) \times 365}{55.65}\right]$$

Where:

EF = emission factor, kg CH_4/head/yr

GE = gross energy intake, MJ/head/day

Y_m = methane conversion factor, per cent of gross energy in feed converted to methane

The factor 55.65 (MJ/kg CH_4) is the energy content of methane.

The amount of methane thus obtained was multiplied by the number of animals, and then the emissions obtained from each group were summed in such a way as to obtain methane emitted by

all the animals on the farm, by using the following equation:

$$Kg\ CH_4 = EF\ \times n.of\ animals$$

6. Estimation of methane emissions from manure

The estimation of methane emissions from manure is based on the number of volatile solids (VS) produced by animals, which represents the organic fraction of total solids in manure that oxidises and is driven off as gas. Therefore, they represent the percentage of feed that is not digested by animals. They were estimated by applying IPCC equation 10.24 (Gavrilova et al., 2019):

$$VS\ = \left[GE\ \times \left(1 - \frac{DE\%}{100}\right) + (UE\ \times GE)\right] \times \left[\left(\frac{1 - ASH}{18.45}\right)\right]$$

Where:

VS = volatile solid excretion per day on a dry-organic matter basis, kg VS/day

GE = gross energy intake, MJ/day

DE% = digestibility of the feed in percent,

(UE × GE) = urinary energy expressed as a fraction of GE.

ASH = ash content of manure calculated as a fraction of the dry matter feed intake (0.08 for cattle).

18.45 (MJ/kg) = conversion factor for dietary GE per kilogram of dry matter.

The methane emissions from manure expressed in kg CH/head/yr were calculated using IPCC Equation 10.23 (Gavrilova et al., 2019):

$$EF\ = (VS\ \times 365) \times \left[B_0\ \times 0.67 \frac{kg}{m^3} \times \left(\frac{MCF}{100}\right) \times MS\right]$$

Where:

EF (kg CH_4/animal/yr) = annual CH_4 emission factor

VS (kg dry matter/animal/day) = daily volatile solid excreted

365 = basis for calculating annual VS production, days/yr

B_o (m^3 CH_4/kg) = maximum methane producing capacity for manure produced by livestock, which is 0.18 m^3 CH_4/kg for cattle (Tables 10.16, Vol .4, Chapter 10, IPCC, 2019)

0.67 = conversion factor of m^3 CH_4 to kilograms CH_4

MCF = methane conversion factor for each manure management system and indicates the percentage of manure converted to methane, with a value of 1.5%.

MS (dimensionless) = fraction of livestock manure handled using manure management system in a climate region,

The amount of methane produced by the manure was then multiplied by the number of animals present, and the emissions obtained from each group were summed to obtain the methane emitted by all animals on the farm, which was expressed in kg CH_4/yr.

7. Estimation of nitrous oxide emissions from wastewater

Direct and indirect emissions must be distinguished to estimate the N and O emissions from manure. Direct emissions were estimated from excreted nitrogen (N_{ex}) using IPCC Equation 10.30 (Gavrilova et al., 2019):

$$N_{ex} = N_{rate} \times \frac{TAM}{1000} \times 365$$

Where:

N_{ex} (kg N/animal/yr) = annual N excretion for animals

N_{rate} = default N excretion rate, kg N/ (1000 kg animal mass)/day (Table 10.19, IPCC 2019)

TAM (kg/animal) = typical animal mass

Volatilised nitrogen (N_{vol}) in the form of NH_3 and nitrogen oxides was estimated using the IPCC Equation 10.26 (Gavrilova et al., 2019) as follows:

$$N_{vol} = \frac{N_{ex} \times MS \times FracgasM}{100}$$

Where:

N_{vol} (kg N/yr) = amount of manure nitrogen that is lost due to volatilisation

N_{ex} (kg N/animal/yr) = annual average N excretion per head per year,

MS = fraction of total annual nitrogen excretion

FracgasM (%) = percentage of managed manure nitrogen that volatilises as NH_3 and NO_x (Table 10.22, Vol. Four IPCC 2019, Chapter 10, pg. 10.96)

Direct N_2O emissions from manure were calculated using IPCC Equation 10.25 (Gavrilova et al., 2019):

$$N_2O_D = (N_{ex} - N_{vol}) \times EF_3 \times \frac{44}{28}$$

Where:

EF_3 = emission factor for direct N_2O emissions from manure management systems, where the value considered for manure is 0.004.

44/28 = conversion of $(N_2O\text{-}N)_{(mm)}$ emissions to $N_2O_{(mm)}$ emissions

In contrast, indirect emissions of N_2O_G from the volatilization of nitrogen present

in manure was calculated according to IPCC Equation 10.27 (Gavrilova et al., 2019):

$$N_2O_G = (N_{vol} \times EF_4) \times \frac{44}{28}$$

Where:

EF_4 (kg N_2O-N (kg NH_3-N + NO_x-N volatilised)$^{-1}$ = emission factor for N_2O emissions from atmospheric deposition of nitrogen on soils and water surfaces; default value is 0.01 kg N_2O-N (kg

NH_3-N +NO_x-N volatilised)$^{-1}$, given in Chapter 11, Table 11.3 (Hergoualc'h et al., 2019).

The amounts of N and O obtained from direct and indirect emissions, expressed in kg N_2O/head/yr, were multiplied by the number of animals present, and the emissions obtained from each category were summed to obtain the nitrous oxide emitted by all animals on the farm, expressed in kg N_2O/yr.

8. Estimation of emissions from energy production and use

Regarding emissions from energy use, both emissions from the use of electricity and emissions from the use of fossil fuel, both direct and indirect, were considered. Emissions from electricity use were calculated as follows:

$$CO_{2eq}\ electric\ energy = (consumption\ x\ coef.ISPRA)$$

Where:

CO_2 electric energy is expressed in kg CO_2/yr

Energy consumption is expressed in kWh/yr

The ISPRA coefficient has a value of 0.22 kg CO_2 /kWh (ISPRA, 2021).

The estimated emissions from fossil fuels were calculated as follows:

$$CO_{2eq}\ combustible\ fossil\ fuel\ (KgCO_{2eq}/year) = Kg\ diesel\ \times Coef.of\ emission$$

Where the emission coefficient considered is 2.98 kg CO_2/kg (Bochu et al., 2013).

9. Estimation of emissions from livestock on-farm production and purchased feed.

Feed-related emissions were estimated using two methods: feed produced on a farm and purchased feed. As shown in Table 1, specific emission coefficients were used for each feed type (Serra, 2014). CO_{2eq} emissions from the purchased feed were obtained by multiplying, for each feed type, and kg of DM by the relevant coefficient.

Table 1. Emission factors for purchased feed (Serra, 2014).

Feed type	**Coefficients of emissions, kg of CO_{2eq}/kg of DM**
Corn silage	0.14
Grass silage	0.17
Grass hay	0.23
Mixed hay (grass-legume)	0.20
Alfa-alfa hay	0.07
Corn meal	0.29
Barley meal	0.26
Soybean meal	0.67
Other meal high in protein	0.25
Industrial by-products	0.40
High protein commercial mix	0.70
Commercial mix	0.60
High moisture grain	0.28
Straw	0.04
Pasture	0.06
Mineral and vitamin supplements	0.25
Urea and amino acids supplements	0.25
Fats	0.80
Cereals by-products	0.40
Milk powder	2.00

Regarding the feed produced on the farm, a division was made between primary and secondary emissions, and emissions from the use of nitrogen fertilisers were included. The latter included emissions from the use of phosphate and potassium fertilisers, pesticides, and seeds.

Direct emissions from the use of nitrogen fertilisers were calculated using the following equation:

$$N_2O_D = Kg\ of\ used\ fertilizer\ \times Soil_{EFN20D} \times \frac{44}{28}$$

Where:

N_2O_D is expressed in kg N_2O per year)

$Soil_{EFN2O}$ direct is an emission factor for nitrous oxide emissions derived from N application in soil of 0.056, as given by the CTN-ACE (Centro Tematico Nazionale Atmosfera Clima ed Emissioni, 2022).

The indirect emissions were calculated using the following equations:

$$Total\ emissions\ of\ P = Kg\ of\ fertilizer\ \times 1.026$$

$$Total\ emissions\ of\ K\ =\ Kg\ of\ fertilizer\ \times 0.867$$

$$Total\ emissions\ of\ Pesticides\ or\ Herbicides\ = Kg\ of\ Pest.or\ Herb.\times 22$$

$$Total\ emissions\ of\ seeds\ = Kg\ of\ seeds\ \times 0.3$$

where total emissions are expressed in kg CO_2 per year.

10. Global warming potential and functional unit

The CFP was derived from the sum of the various emission sources: enteric CH_4, CH_4 from manure, N_2O from manure, CO_2 from energy and feed expressed in kg CO_2 equivalent according to the Global Warming Potential (GWP) equivalence scale over a 100-year interval,

Where:

1kg CO_2 = 1kg CO_2 equivalent

1kg CH_4 = 27.9kg CO_2 equivalent

1kg N_2O = 273 kg CO_2 equivalent

Relative to the functional unit, total emissions were related to the kg of live weight (LW) for sale.

11. Carbon sequestration

For the quantification of carbon sequestration, the potential for C removal by areas occupied by natural pastures, meriagos (forests including shrubs), grasslands, and meadows, based on the literature found in Table 2, were considered.

Table 2. Carbon removal potential, expressed in tons CO2/ha/yr, for different land types.

Land Use	**Potential Carbon Removal (t CO_2/ha/yr)**	**Source**
Natural pasture	1.30	Freibauer et al. (2004)
Meriagos	3.89	ISPRA (2023)
Grassland	0.94	ISPRA (2023)
Meadow	1.30	Freibauer et al. (2004)

Results

Table 3 presents the main findings on the distribution and average size of farms surveyed, revealing that the farms have a mean area of 113.50 ± 53.01 hectares (mean of ± standard deviation). The distribution of these hectares is as follows, 38 ± 25.13 ha are represented by natural pastures, 45.7 ± 41.08 ha by Meriagos, 28.5 ± 34.46 by grasslands, and 1.2 ± 3.79 ha by Meadows. On average, 46 ± 22 cows are reared, and 33 ± 19 calves are sold at an average live weight (LW) of 307.5 ± 79.77 kg, with an average weight gain of 1.14 ± 0.1 kg/d. The total LW kg sold was 12,299 ± 5,906, including the sale of calves, bulls, and culled cows.

Table 3. Distribution of farm area and herd stock characteristics (data expressed as mean ± standard deviation).

Category	Media	SD
Area distribution		
Natural pasture, ha	38.00	25.13
Meriagos, ha	45.70	41.08
Grassland, ha	28.50	34.46
Meadow, ha	1.20	3.79
Stocks		
Cows, n	45.7	21.68
Calves born, n	39.5	20.55
Birth weight, kg	37.5	5.53
Annual replacement, n	3.70	2.41
Calves sold, n	32.9	18.81
Calves sold, kg LW	307.50	79.77

Calves age at sale, months	9.1	2.56
Average weight gain kg/d	1.14	0.1
kg live weight from calves sold	9511	5466
kg total live weight sold[1]	12298.5	5906.36

[1]Live weight of calves sold for fattening, bulls, and culled cows sold for slaughter.

Table 4 shows the results for the estimated average farm CFP for both gross and net. Regarding gross CFP, data analysis showed that the total impact related to the production of 1 kg LW was 17.33 ± 6.07 kg CO_{2eq}. Regarding the net CFP, derived from the difference between the CO emitted and that sequestered, and related to the kg of LW sold, the data analysis showed that the net impact related to the production of 1 kg live weight was negative, -78.9 kg CO_{2eq}. The inclusion of carbon sequestration, represented by natural pasture and other lands of the beef cattle farms surveyed, in estimating CFP not only offsets the impact, but also makes it largely negative.

Table 4. Carbon balance, total and net carbon footprints, and main average emission sources for the studied farms.

Category	**Average**	**SD**
Emissions		
Enteric emissions, kg CO_{2eq}/yr	143590	78030
Emissions from manure, kg CO_{2eq}/yr	1679.64	914.32
Emissions from energy, kg CO_{2eq}/yr	12918.73	10262.37
Emissions from purchased feed, kg CO_{2eq}/yr	3003.66	2585.39
Emissions from on-farm feed production, kg CO_{2eq}/yr	46747.42	76892.56
Total emissions, kg CO_{2eq}	217132.3	141851

GROSS carbon footprint, kg CO_{2eq}/ kg LW	17.31	6.07
Distribution of emissions		
Enteric methane, kg CO_{2eq}/ kg LW	11.61	3.44
Manure, kg CO_{2eq}/ kg LW	0.88	0.26
Energy, kg CO_{2eq}/ kg LW	0.94	0.5
Feed purchased, kg CO_{2eq}/ kg LW	0.27	0.23
Feed produced, kg CO_{2eq}/ kg LW	3.61	4.85
Total CO_2 sequestered, kg	981878	816667.9
NET carbon footprint, kg CO_{2eq}/ kg LW	-78.9	99.02

The contribution of each emission source to the overall impact of livestock farming is shown in Figure 5. The plot illustrates the contribution of various emission sources to the overall environmental impact on livestock. Emission sources, such as Enteric Methane, Manure, Energy Production and Use, Feed divided into purchased feed and On-Farm Feed Production, are represented by grouped bars. As shown in the plot, enteric methane emissions accounted for approximately 70% of the total impact of the farms surveyed, followed by emissions associated with on-farm feed production (16.6%), energy (~6%), manure (~5%), and purchased feed (~2%).

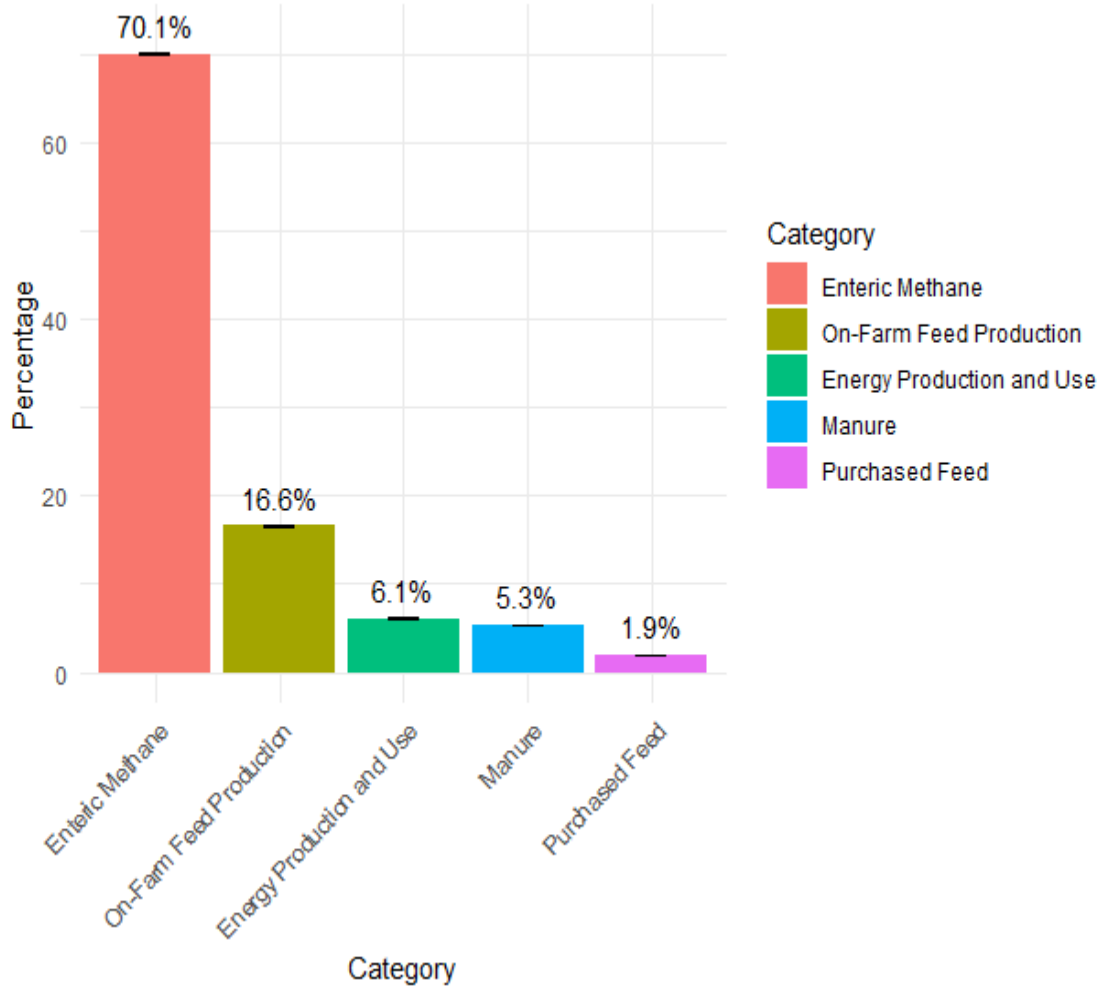


Figure 2. Average contribution (in%) of different emission sources to overall impact.

Further details regarding the carbon-equivalent emissions associated with each category are depicted with the corresponding values, showing the average contribution to the overall emissions (Annex 1). In the enteric methane fermentation plot, the sub-categories, such as methane emissions from cows (179259.35 ± 45842.48), bulls (4132.59 ± 1646.39), and calves (60197.9 ± 31527.39). Similarly, the Manure Management plot revealed the distribution of emissions across the above-mentioned subcategories (927.62 ± 536.52, 47.5 ± 30.23, 704.53 ± 368.98, for cows, bulls, and calves respectively). The Energy-related emissions plot outlines the emissions associated with electricity (236.28 ± 249.89) and fuel consumption (12682.45 ± 10382.46). The Purchased Feed plot highlights the emissions from the different feed categories, such as concentrates (1402.5 ±

1730.54, 586.5 ± 1854.68 for starch and protein respectively), forages (637.5 ± 1347.7), granules (358.68 ± 666.47), silage (18.48 ± 58.44), offering insights into the environmental impact of external feed sources. Finally, the On-Farm Feed Production plot shows the emissions from primary including Direct emissions from N_2O (37254.98 ± 65763.49), indirect emission (2129.71 ± 2109.72, 3144.72 ± 3461.2 from volatile and leaching N respectively), and secondary processes including fertilizers production (3052.27 ± 5576.67), and seed production (1165.74 ± 1027.23), allowing for a detailed assessment of the specific contributors to the emission from on-farm feed production.

Discussion

This study assessed the gross and net carbon footprint of extensive suckler cow-calf farming in Sardinia. Different factors were considered to estimate emissions, including enteric fermentation, manure management, energy consumption, and feed purchase and production. The effect of carbon sequestration on emission mitigation was also assessed. The estimated net CFP was 17.31 kg CO_{2eq}/kg LW. This result corresponds to the previously observed values for grazing cow-calf farms in Uruguay and France, whose gross CFP ranged from 11.4 to 32.2 and 14.8 to 16.8 kg CO_{2eq}/kg LW, respectively (Becoña et al., 2014; Berton et al., 2017). Another study conducted in North Italy, reported a CFP value of 15.0 kg CO_{2eq}/kg LW, with an average global warming potential (GWP) of 15.7 kg CO_{2eq}/kg LW and a range of GWP from 12.1 to 17.6 kg CO_{2eq}/kg LW (Bonnin et al., 2021). In contrast, Toro-Mujica (2021) reported a gross CFP value of 13 kg CO_{2eq}/kg LW for the cow-calf line in Chile. Basarab et al. (2012) also reported a range of carbon footprint in cow-calf systems from 11.63 to 13.22 kg CO_2eq/kg LW, with calf-fed production having a carbon footprint 6.3-7.5% lower than yearling-fed production. Grossi et al. (2020)

estimated the cradle-to-gate CFP of pasture-based beef cattle in a Mediterranean agropastoral system to be 26.9 ± 0.7 kg CO_{2eq}/kg LW. These authors also reported that adopting management practices, such as conservation tillage or high fertilisation, can reduce the CFP of pasture-based beef cattle by 20% to 40%. Ciriello et al. (2020) compared different types of livestock management and found that the CH_4 emission factor for buffalo in a conventional farm was 27.69 kg CH_4/head/yr, while organic and family-based farms had lower emission factors of 22.77 and 21.61 kg CH_4/head/yr, respectively.

In the current study, enteric fermentation accounted for 70% of total emissions. These results are consistent with the findings of Tongwane and Moeletsi (2021), who reported that 64.54% of cattle emissions in South Africa were produced by enteric fermentation. Another study by Beauchemin et al. (2010) revealed that suckler cow herds accounted for approximately 80% of the enteric CH_4 produced within a cow-calf production system in southern Canada.

In the present study, the utilisation of land-based carbon sequestration, considering the potential coefficient sourced from the literature (Table 2), has proven effective in neutralising the carbon footprint. This conclusion aligns with the findings of McPhillips et al. (2022), who, in their investigation of greenhouse gas emissions from a conventional pasture-based cattle production system as opposed to a partial-confinement system, discovered that carbon sequestration from perennial grasslands in the conventional system was sufficiently robust to offset all emissions and CO2. Furthermore, the alternative system exhibited significant carbon sequestration capacity, offsetting 33-72% of system emissions, depending on the Global Warming Potential (GWP) metric employed. O'Brien et al. (2023) reported that the total GWP of an average suckler calf-to-beef system was

40% higher without carbon sequestration. Additionally, these authors highlighted that carbon sequestration was greater in silvopastures than in grasslands. Another study in China found that one hectare of grassland can sequester 1.5 tons of carbon and release 2.8 tons of oxygen per year (Shen, 2023). In contrast, in the UK, woodland planting on arable land has been found to have the greatest sequestration potential (4.02 tC/ha/yr), compared to planting on pastureland (3.75 tC/ha/yr) (Flack et al., 2022).

Conclusions

Our study provides insights into the net environmental footprint of these farms and highlights valuable data for sustainable agricultural policies and practices. Our findings underscore the importance of considering both emissions and carbon sequestration in the comprehensive assessment of beef cow-calf farming systems. The available literature emphasises the importance of considering various management practices and farm types when assessing greenhouse gas emissions from beef cattle farms. This also highlights the importance of considering both soil and vegetation in estimating the overall carbon balance.

In conclusion, the results of this study allowed us to estimate the net carbon footprint of a sample of beef cattle farms in Sardinia that adopted a cow-calf system. Data processing showed that per kg of LW, the net CFP value was 17.33 kg CO_{2eq}/ kg LW, and 70 percent of these were associated with enteric methane emissions. Considering the CO_2 land-based sequestration potential, it was observed that the net CFP was (-) 78.90 kg CO_{2eq}/kg LW. The inclusion of carbon sequestration in CFP estimations has made it possible to offset the carbon footprint of cow-calf farms. However, despite these strategies, the growing global demand for beef will result in an increase in GHG emissions, unless beef consumption

decreases. However, there is a need for further research to assess methane emissions specifically from suckler beef cow-calf lines in the Mediterranean and to develop targeted mitigation strategies for this production system.

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Annexe 1.

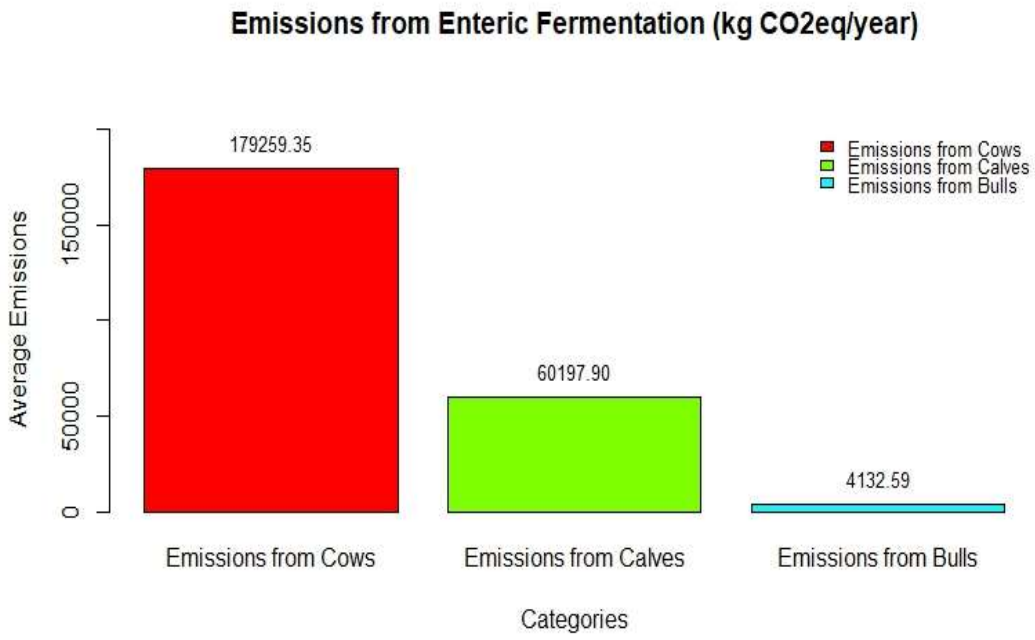


Figure 3. Emissions from Enteric Fermentation.

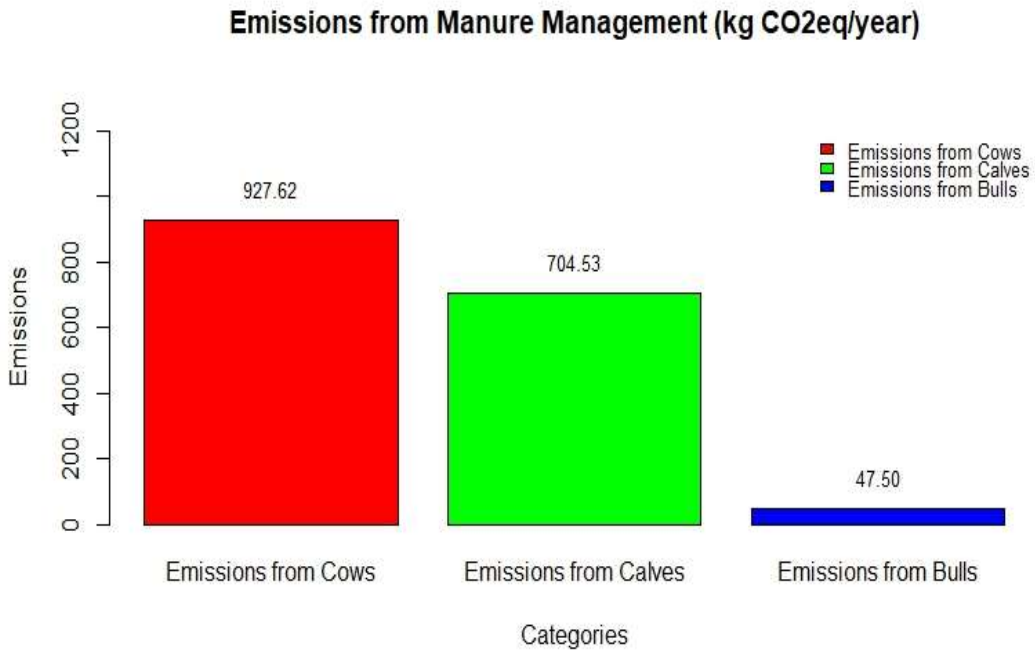


Figure 4. Emissions from Manure management.

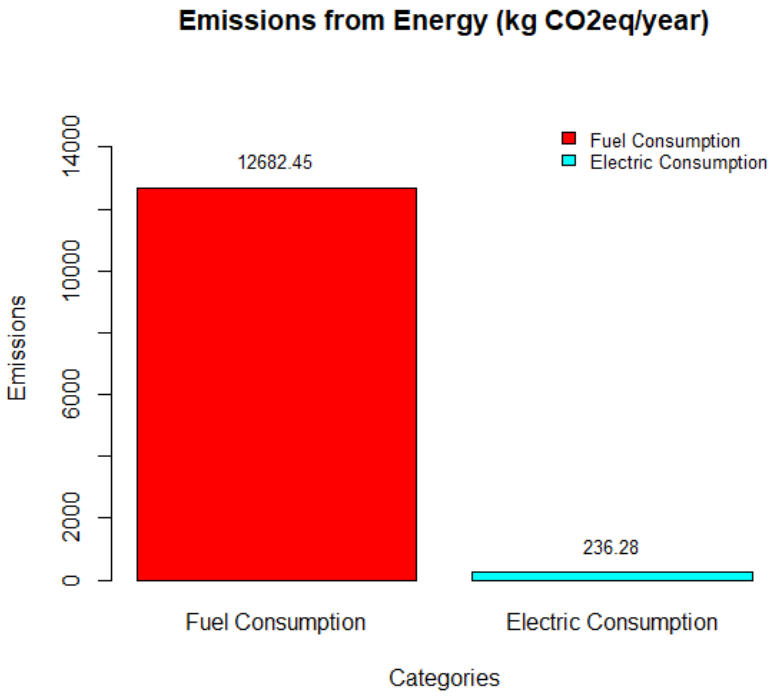


Figure 5. Emissions from Energy.

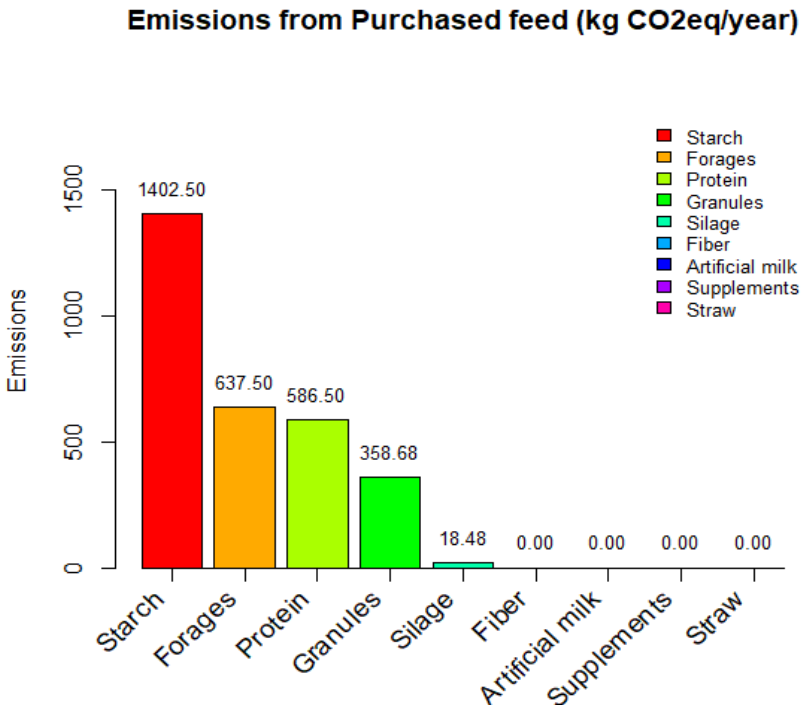


Figure 6. Emissions from Purchased feed.

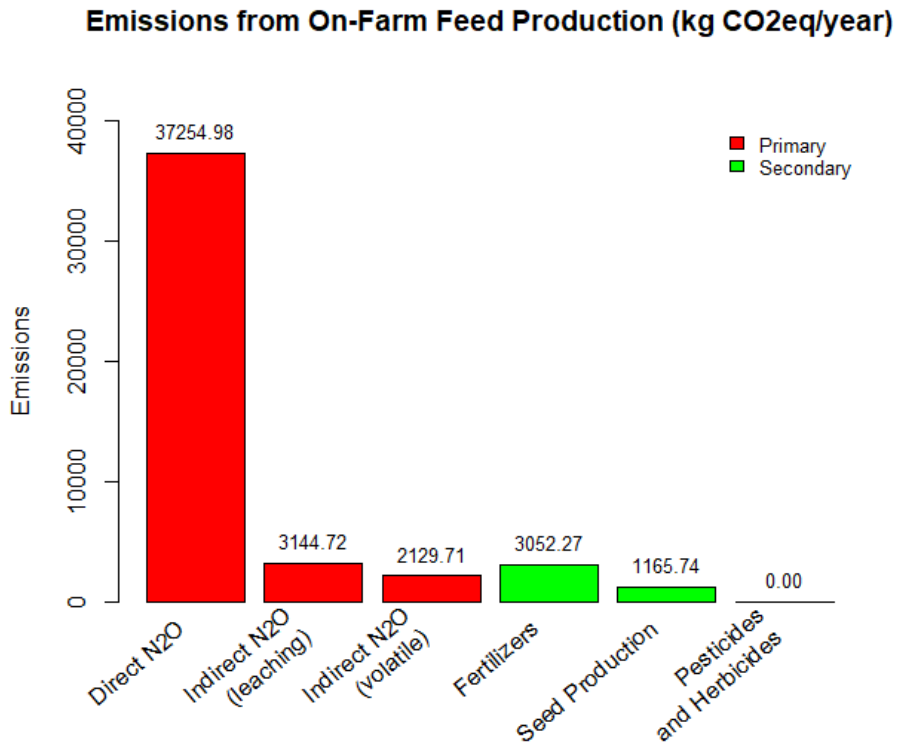


Figure 7. Emissions from On-Farm Feed Production.

General Conclusion

In summary, this dissertation considered the significance of monitoring the behaviour and environmental effects of pasture-based cow-calf systems. However, it also highlighted the impact of calving season on the requirements and performance of postpartum suckler cows in continuously grazed systems.

A review of the available literature on the suckling behaviour of beef calves on pastures is a crucial element for future studies aimed at enhancing the welfare and productivity of suckler cow-calf production systems. This knowledge will help optimize management practices during the suckling phase and improve the postpartum estrus cycle, which in turn will enhance reproduction and facilitate the weaning process. As demonstrated in Chapters 2 and 3, the management of the postpartum period until the calf is weaned has a significant impact on the behaviour and recovery rates of the cow, as well as on reproduction and productivity. Additionally, this management strategy may play a role in reducing the environmental impact of this production system, as discussed in Chapter 4, by reducing greenhouse gas emissions in livestock production systems.

The results obtained in Chapters 2 and 3 support the general hypothesis that cows that calved in winter during the experimental period had lower energy needs owing to the presence of already developed calves that suckled less milk and were able to graze. In contrast, cows that calved in spring, which had much younger calves, were expected to have higher energy requirements. This was reflected in extended grazing time, the initiation of grazing in early morning, and the higher herbage dry matter intake of group S compared to group W. These findings were also reflected in the performance outputs, where cows calved in winter exhibited higher body weights and growth rates than those calved in

spring, suggesting potential advantages of winter calving under the studied conditions.

Chapter 4 provides insights into the net environmental footprint of beef cattle farms in Sardinia that adopted a cow-calf system and highlights valuable data for sustainable agricultural policies and practices. These findings underscore the importance of considering both emissions and carbon sequestration in the comprehensive assessment of beef cow-calf farming systems. The inclusion of carbon sequestration in CFP estimations has made it possible to offset the carbon footprint of cow-calf farms. However, despite these strategies, the growing global demand for beef will result in an increase in GHG emissions unless beef consumption decreases. However, there is a need for further research to assess methane emissions specifically from suckler beef cow-calf lines in the Mediterranean and to develop targeted mitigation strategies for this production system.

In summary, the findings highlighted in this dissertation contribute to our understanding of cattle behaviour in pasture-based systems and provide practical insights for extensive beef livestock production in Mediterranean conditions, guiding improved management strategies for enhanced productivity, welfare, and mitigating the environmental impacts of cow-calf operations. Moreover, the results underscore the importance of effective pasture management to optimize beef production systems, given the calving season and postpartum period of grazing cows.

However, this dissertation has identified several potential directions for future research and has highlighted the importance of incorporating precision livestock farming technologies for monitoring animal behavior, as presented in Chapter 1. Further research is necessary to understand the effects of grazing systems, pasture types, and environmental conditions on suckling behavior in calves. Additionally, studying the impact of varying grazing periods

on cows is essential, as the seasonal variations in the Mediterranean region have been shown to influence both animal behavior and performance, as discussed in Chapters 2 and 3. Otherwise, standardizing the calving season would not only optimize the availability of biomass for grazing but also offer economic benefits by aligning with the optimal selling season for calves. In addition, considering the economic aspects in future studies, including feed costs and labor requirements associated with different management strategies, would be highly valuable for improving system efficiency. Furthermore, research on the effects of managing animals with lower feed intake requirements on pasture conditions and biodiversity, beyond the scope of carbon sequestration that have been considered as mitigation strategies in Chapter 4, would offer additional insights in mitigating the carbon footprint of beef cow-calf production systems.

The Mediterranean region, particularly Sardinia, is characterized by its rustic and diverse agricultural practices, especially in cattle farming. The cow-calf systems in this area have given rise to a distinctive livestock farming model. Native cattle breeds are well adapted to the island's specific terrain and climatic conditions, and the preservation and effective management of these systems are crucial. Such efforts are essential not only for maintaining agricultural biodiversity but also for preserving cultural heritage and supporting the local economy.