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EUROPEAN UNION (EU) GENERAL FARM ANIMAL DIRECTIVE APPLICABILITY IN AFRICA AND BOVINE WELFARE ASSESSMENT IN ITALY

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ABSTRACT

Animal welfare is no longer the concern of a few who care about it, but the subject of international legislation and farm animal agriculture is arguably the most economically important interface between humans and other animals on this planet. The welfare of animals can be very poor because of the design of animal accommodation or of facilities on farms, for transport or for slaughter. Bovine respiratory disease (BRD) is one of the most important challenges in beef cattle, during the adaptation in intensive systems, being a risk factor for the development of poor welfare.

In chapter II, The objectives of the study was to assess the applicability of the European Union (EU) farm animal legislation (Council Directive 98/58/EC) to the context of Africa countries focusing on Ethiopia, to attain the objectives relevant literatures, international organization, regional organization, countries legislations, standards were searched, assessed and reviewed to understand the farm animal welfare situation and legislations in Africa and also Ethiopia, additionally the farm animal legislation of the European union was also reviewed. Finally the articles and annexes of farm animal directive (council directive 98/58/EC) applicability to African context were analyzed. The applicability of the directive that is developed for highly developed countries in European Union (EU) the "farm animal directive'' (Council Directive 98/58/EC) may not be totally applicable in countries like Ethiopia which still have traditional system of livestock production. As a conclusion, the European Union (EU) farm directive, could not be completely implement in African countries like Ethiopia, but it could serve as a good starting point, so that after successful identification of the farm animal welfare critical points which are typical in African countries, the farm animal directive of the European union may help as a starting point with modification to the local situation in the ground.

Chapter III, The study aimed to assess the welfare and health status in the first 15 days after arrival of Limousine bulls imported from France and fattened in a commercial fattening unit in Italy. A total of 264 Limousine bulls with an average age of 11 months were included in the trial. Welfare, biosecurity, and major hazard and warning system were assessed on days 2 (T1) and 15 (T2) after arrival to the unit employing a modified version of the Italian protocol ClassyFarm. At T1, the percentages for welfare, biosecurity, and major hazards and warning systems were, respectively,

79.04% (medium), 63.88% (medium), and 76.47% (medium). At T2, the percentages for welfare, biosecurity, major hazards, and warning systems, respectively, were 74.73% (medium), 63.88% (medium), and 76.47% (medium). At T1 and T2 an inspective clinical examination was performed on all bulls (n=264). The following findings were noted at T1: 1.51% of integument lesions, 0.75% of lameness, 0.75% of diarrhoea, and 27.65% of signs of respiratory disease. Skin lesions (44.69%) and lameness (1.15%) significantly increased in T2 (p-value<0.05), while diarrhoea (0%) and individuals with respiratory disease (31.81%) did not change significantly. At T1 and T2 blood samples were collected from 88 randomly chosen bulls for a complete blood cells count and fibrinogen. There was a statistically significant difference (p-value≤0.05) in leucocytes, neutrophils, monocytes, lymphocytes, eosinophils, basophils, platelets, and fibrinogen counts between T1 and T2. The leucocytes, neutrophils, monocytes, lymphocytes, eosinophils, basophils, and platelets were increased at T2, while the fibrinogen decreased. The haematological changes indicate that the bulls were under higher stress in T2 when compared with T1, most likely associated with a difficult adaptation response to the fattening unit. A multi-factorial approach that integrates the indicators of the checklist and the clinical and haematological findings of animals can be a useful method to deepen the assessment of welfare in beef cattle.

Chapter IV, the study aimed to evaluate the effect of different treatments for BRD on health and welfare in fattening bulls. A total of 264 bulls were enrolled. Welfare was assessed on day 2 (T0) and day 15 (T1) after arrival, showing a worsening. All bulls were inspected clinically at T0 and T1 revealing an increase of skin lesions and lameness in T2. In both periods, a high incidence of respiratory disease was observed. A prevalence of 79.55% and 95.45% of *Mycoplasma bovis* using RT-PCR and culture at T0 and T1 respectively was observed. Blood samples were collected for hematology at T0 and T1. At T0, 36 animals were individually treated for BRD with an antimicrobial (IT), 54 received a metaphylactic treatment with tulathromycin (M), 150 received a metaphylactic treatment with tulathromycin plus a second antimicrobial (M+IT) whereas 24 were considered healthy and therefore not treated (NT). Additionally, 128 were treated with a non-steroid anti-inflammatory (NSAID). Neutrophils of M+IT were significantly higher than groups NT and M and the lymphocytes of M+IT were significantly lower than that of IT. White blood cells, neutrophils and N/L ratio of animals treated with an NSAID was significantly higher than that not treated. Lung inspection of 172 bulls at the abattoir indicated that 92.43% presented at

least one lung lesion. A statistically significant effect of the NSAID treatment on the lung lesions was observed. Our findings indicate that BRD was a major welfare and health concern and evidence the difficulties of antimicrobial treatment of *M. bovis*.

Chapter V, the objective of the study was to clarify what is meant by "navel healing" and to provide strong elements for reaching a consensus. The navel healing and effect on transport the navels of 299 dairy calves (55 males, 244 females) aged 0–90 days were examined and scored 1 to 5 according to their healing status, so as to see which group is feet for transportation based on the navel healing. The result shows that a completely dry and shriveled navel stump entails a high risk of transporting too young calves, whilst the presence of a scab covering the umbilical wound could be considered acceptable for short journeys, as the risk of transporting calves that are too young is low. "Navel healing" should be defined as the scarring of the umbilical wound, which occurs no earlier than 3–4 weeks of life. In transporting calves with a completely healed navel should be considered best practice because it ensures that calves that are too young are not transported and therefore guarantees higher animal welfare standards.

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INTRODUCTION

General Introduction and Objectives

Animals and people have always existed together and lived for centuries. Humans use animals for hunting, food source, for labor, study and recreation. The animal depends on its owners in some capacity for all of these functions, and the owner reciprocally bears responsibility to the animals. The more restrained the animal, the more accountable humans are (Harris, 2005). Over the past 30 years, advances in animal welfare science have been made at a rapid pace thanks to a better understanding of animal motivation, cognition, and the complexity of social behavior (Broom, 2011a). Animals are sentient, which means they have feelings. Therefore, it is important that their basic biological, behavioral, and affective needs be met. This principle underpins both the practical and moral concerns of caring for animals and point outs the need for a high standard of care, including humane way of killings when intended for food (Doyle et al. 2021). Animal agriculture is not only a theoretical interface between humans and other species, but also an economic endeavor; it functions primarily as a business, and the ones who have the greatest impact on animal welfare in this sector are those who work in the livestock industry (Sinclair et al., 2019). In many parts of the world, the livestock production system is changing significantly. The rise in livestock demand in developing nations is a result of rising consumer demand for animal products. The demand for animal products in industrialized nations seems to be at its height, despite the fact that many production processes are becoming more effective and environmentally sustainable. The demand for livestock products has evolved historically primarily as a result of population growth, wealth growth, and urbanization. Production responses in various livestock systems have been correlated with advances in science and technology as well as the growth in animal populations (Thornton, 2010).

The methods employed in breeding, shipping, and slaughter are frequently the subject of public interest and activism because it is an industry that has consistently witnessed fast development and intensification in most parts of the world (Sinclair et al., 2019). Animal health is a crucial aspect of animal welfare. Good animal health and welfare are closely linked to good productivity (FAWC, 2016). A farm animal is considered to have good welfare status if it is healthy, fed properly, cared

for, included with appropriate housing, and able to display its typical behavioral patterns. Poor housing conditions make animals more prone to illness and increase pathogen shedding, which could have a negative impact on public health (FVE, 2018). Despite the varying definitions and concepts of animal welfare that result in greatly changing views on this complex topic, there is a growing belief that farm animal welfare should be protected and improved (Alonso et al., 2020).

Continually ranging from extremely good to very bad, welfare may be precisely measured. The status of an individual welfare is determined by how well they are able to adapt to their surroundings. An accurate assessment of welfare can be made along a continuum from very good to extremely poor. Animal welfare studies can provide information on the circumstances that might promote excellent wellbeing, but it is necessary to weigh the significance of the preference to the individual. When animals experience short- or long-term issues, those issues may be physiological or behavioral, or they may be connected to production or illness. Any measure may suggest poor welfare since individuals have different coping mechanisms, and the lack of evidence for a measure does not imply that there is none welfare problem (Broom, 1988). The design of animal housing or facilities on farms, during transport, or during slaughter can result in extremely poor animal welfare. The welfare of animals is significantly impacted by management practices as well. No matter how well the animal welfare is handled, it won't be beneficial if the structure or management system creates unavoidable issues, as is sometimes the case (Broom, 2009).

The European Union (EU) authorities have been working to successfully secure the protection of farm animals since the last decade of the 20th century, mostly through conventions and horizontal and species-specific legislation. The protection of laying hens in battery cages, transportation regulations, broiler chicken welfare, housing conditions, sheep and goat traceability, etc. are only a few of the linked challenges that Europe has attempted to address in this way (Luca, 2020). In EU laws governing farm animal care apply to all phases of production, including rearing, transporting, and killing. Five directives that set minimum standards apply to farming activities, whereas laws that apply to animal transportation and slaughter set equivalent criteria for all member states. The set of EU law improves the living circumstances for various types of farm animals and the sustainability of the EU food chain (Simonin and Gavinelli, 2019).

There is a rising global need for animal products, particularly food for human use, particularly in developing nations in tropical areas. Animal welfare and minimal environmental impact are also becoming more and more significant to both consumers and non-consumers in today's society. Meeting the rising demand for animal products without ignoring societal issues requires improving the efficiency of current animal production systems. Achieving adequate animal welfare might be crucial for increasing output and satisfying customer demand (Hernandez et al., 2022). Perceptions of animal welfare in Africa vary by region, culture, and customs (Qekwana et al., 2019). The majority of African nations have varying levels of legislation, OIE-compliant legal frameworks, and rules governing animal care. It is discovered that laws, norms, and policies are either missing, insufficient, out of date, or not strictly enforced. Despite the fact that all nations have signed the OIE standards, comprehension and adherence to them are frequently limited, partly because there is a lack of implementation capability and a need to establish country and context specific strategies (AU-IBAR, 2017). Ethiopia like many other African nations, has not consistently developed public awareness initiatives of animal welfare and legislation that governs animal welfare is lacking. Even though country has a high population of livestock, however animal care is poorly handled, leading to low productivity and production in the industry (Zekarias and Tesfaye, 2019).

Objectives of the Thesis

Based on the above introduction and background, the dissertation has four objectives that are discussed in the different chapters of the dissertation report; the objectives are:

1) To examine the applicability of the European Union (EU) farm animal regulations (Council Directive 98/58/EC) to the context of Africa focusing; Ethiopia, and to forward recommendations, since the most important farm animal in African including Ethiopia is cattle, so that the review focused on cattle, since cattle is economically important animal in pastoral and mixed crop agricultural farming community.
2) To assess the welfare and health status of bulls imported from France and fattened in Italy in a commercial fattening unit in the first 15 days after arrival by applying a multidisciplinary approach.

3) To evaluate the effect of different antimicrobial and anti-inflammatory treatments protocols for BRD on health and welfare of newly introduced beef cattle in a commercial fattening unit of Limousine bulls affected by the high prevalence of BRD due to *M. bovis.*

4) To contribute to clarify what is meant by "navel healing", since no specific definition is provided by the law, giving raise to different interpretations. The ultimate aim is to provide strong elements for reaching a consensus among farmers and veterinary practitioners in order to comply with the European regulation.

CHAPTER I: STATE OF ART

1.1. Definitions and General Concept of Farm Animal Welfare

Humans believe they have obligations to other individuals, and indeed humans and other social animals have developed characteristics that make us respond to others in our societies in a way that promotes careful preferences and actions (Broom, 2011b). Animal welfare research developed as an interdisciplinary field of research in the 1970s. The impetus for this work was public concern about the welfare of animals kept in what were then new husbandry systems. Early research explicitly addressing animal welfare was largely based in the areas of animal behavior and stress physiology, but the importance of many other areas was quickly recognized. These included veterinary epidemiology, environmental physiology, environmental design, comparative psychology, and studies of animal caregiver behavior, as well as traditional areas such as nutrition and microbiology (Fraser et al., 2013).

In the OIE Terrestrial Animal Health Code, Animal welfare means ……. "the physical and mental state of an animal in relation to the conditions in which it lives and dies. An animal experiences good welfare if the animal is healthy, comfortable, well nourished, safe, is not suffering from unpleasant states such as pain, fear and distress, and is able to express behaviours that are important for its physical and mental state. Good animal welfare requires disease prevention and appropriate veterinary care, shelter, management and nutrition, a stimulating and safe environment, humane handling and humane slaughter or killing. While animal welfare refers to the state of the animal, the treatment that an animal receives is covered by other terms such as animal care, animal husbandry, and humane treatment (OIE, 2022). And Broom (1988) defines welfare of an animal is "its state as regards its attempts to cope with its environment; for each coping system, the environment is that which is external to the system". The UK Farm Animal Welfare Council defines animal welfare in terms of five freedoms: Freedom from hunger or thirst, freedom from physical discomfort, freedom from pain, injury, or disease, freedom to engage in normal behavior, freedom from fear, and distress as illustrated in table 1 (Farm Animal Welfare Council, 1992). Welfare can be interpreted as the physical and mental state of an animal. Ethics provides the philosophical framework within which this well-being is interpreted and applied and is influenced

by the moral views of the individual. Animal rights are the product of a particular philosophical and ethical viewpoint (Doyle et al., 2021).

Table 1. The five freedoms (Source: Farm Animal Welfare Council, 1992).

The Freedoms and provisions		
1.	Freedom from thirst, hunger and malnutrition	by ready access to fresh water and a diet to maintain full health and vigour
2.	Freedom from discomfort	by providing a suitable environment including shelter and a comfortable resting area
3.	Freedom from pain, injury and disease	by prevention or rapid diagnosis and treatment
4.	Freedom from fear and distress	by ensuring conditions which avoid mental suffering
5.	Freedom to express normal behaviour	by providing sufficient space, proper facilities and company of the animal's own kind

The five freedoms shouldn't be viewed as guidelines for perfection but rather as a set of requirements for adhering to recognized welfare principles and as a useful, all-inclusive checklist for evaluating the advantages and disadvantages of any farming system, whether in the context of international production system standards or at the level of the specific farm (Webster, 2011).

There are two conflicting interpretations of the term "animal welfare." The first describes how an animal feels physically and mentally as it tries to satisfy its physiological and behavioral requirements. It is a gauge of the animal's own welfare, which we may learn more about through detailed examinations of animal behavior and the fields of welfare science. Animal welfare as an expression of moral concern is the second notion. It is based on the idea that since animals may feel emotions that people would understand as pain and suffering, we have a responsibility to shield the animals under our care from these things (Webster, 2011). Animal welfare is about the experiences of individual animals, but in huge herds or in some agricultural systems, such as vast fish and poultry farms, where monitoring and group treatment are the norm, this focus on individual animals' experiences and the capacity to handle them might be lost. Another illustration

is that no matter how well the animals are cared for in these systems, the welfare of sociable animals lacking in social interaction or the welfare of confined animals lacking in exercise or grooming opportunities may not always be favorable (Broom, 2009).

A practice or events that affect animal welfare is influenced by the quantity of animals involved as well as the degree, length, and frequency of any pain, discomfort, or suffering. So, when choosing the priority of interventions, it's vital to consider whether it is possible to lessen or prevent pain. Animal welfare has importance for society on both a moral and financial level. Good productivity is directly related to good animal health and wellbeing, and both can have a positive impact on the environment. For instance, increased livestock efficiency can result in lower pollution emissions (FAWC, 2016). The essence of good farm animal husbandry is to provide the resources and management needed to ensure the economic production of food and other goods in a way that does not compromise animal (and environmental) health and welfare (Webster, 2011). A crucial aspect of animal welfare is animal health. Animals in low welfare circumstances are more susceptible to disease, including increased pathogen shedding, which could have a negative impact on public health (FVE, 2018). Animal health and animal welfare are complementary but not synonymous. Without good health there can be no good welfare, but good health alone does not guarantee good welfare. For this reason, animal productivity cannot be a substitute for well-being (Doyle et al., 2021). Science demonstrates the relationship between animal welfare, animal health, and food safety. Therefore, enhancing animal wellbeing has the potential to lower the hazards to food safety on farms, particularly through reducing stress-induced immune suppression, infectious illness incidence on farms, the transmission of human infections from farm animals, and antibiotic use and antibiotic resistance (de Passillé and Rushen, 2005). Animal welfare requires disease prevention and appropriate veterinary treatment, shelter, management and feeding, humane handling and humane slaughter or killing. Animal welfare refers to the condition of the animal; the treatment an animal receives is covered by other terms such as animal care, husbandry and humane treatment (OIE, 2017). Njisane et al (2020) summarized the poor welfare effect on the production animal's figure 1.

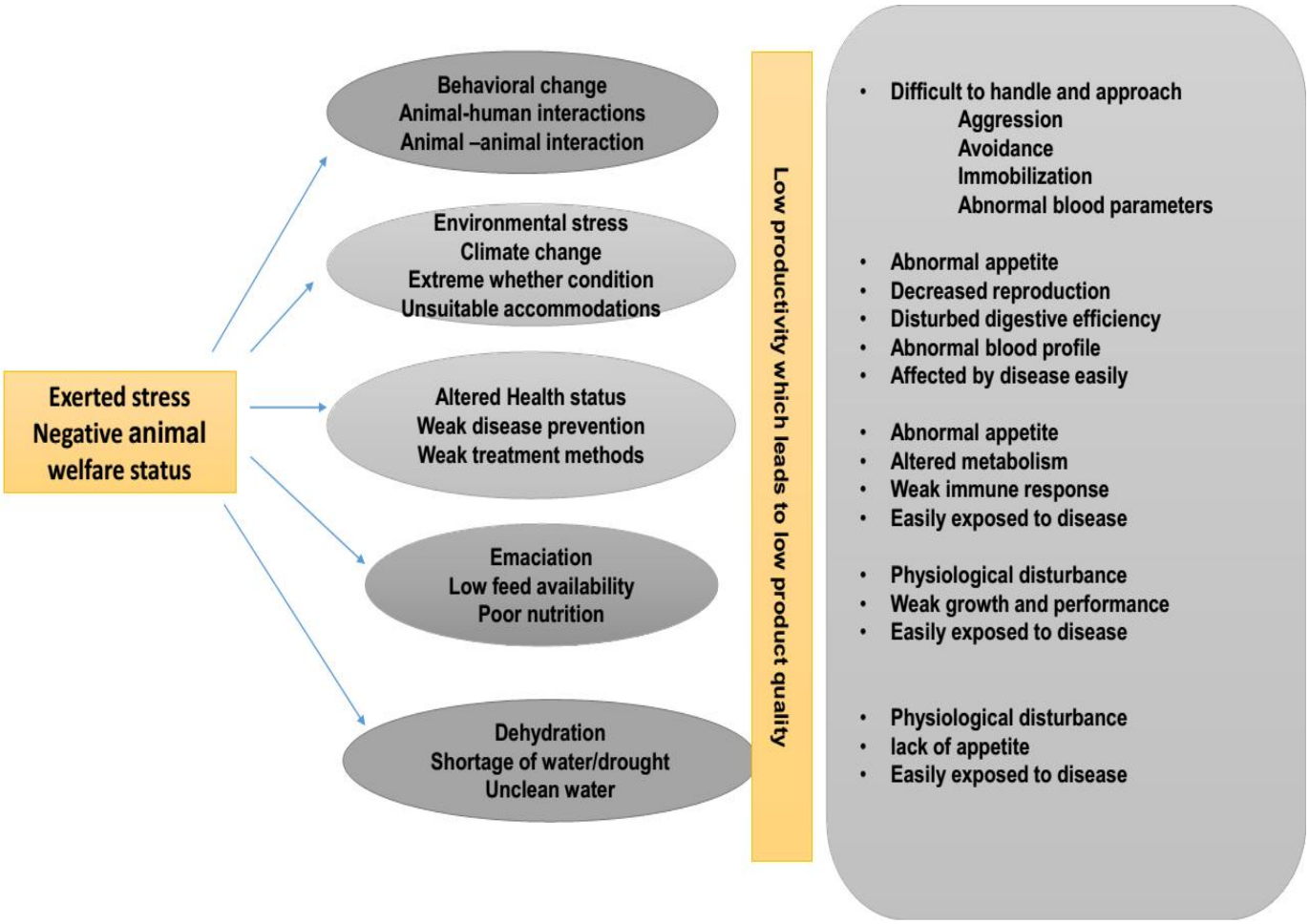

Figure 1: An illustration of poor animal welfare and stress indicators impacting on production and product quality (Adopted, Source: Njisane et al 2020).

To date, many animal welfare research has been conducted in response to concerns about animals in intensive production systems using different assessment protocols that are exclusively developed for intensive livestock production (Ventura et al., 2021; Lawrence et al., 2022). As a result, intensive housing has been the subject of a lot of research, housing and other environmental variables are addressed in many animal welfare standards and reforms. However, a wide range of additional factors, including genetics, diet, disease control, and operator behavior, have a significant impact on animal welfare. Further study is required to understand how each of these elements affects animal welfare in various production systems as well as how to address issues with animal welfare brought on by certain husbandry and feeding practices that are currently in use. On the basis of the belief that animal wellbeing necessitates a complicated match between

genetics, housing, handling, and other aspects of animal husbandry, there is a general need for research that adopts an integrated approach (Fraser et al., 2013).

1.2. Protocols for Welfare Assessment of Farm Animals

Improving animal welfare indicators and more precisely measuring positive and negative animal-related situations is receiving increasing scientific and practical attention as farm animal welfare becomes an increasingly significant component of food chain regulation, process or system validation, and product differentiation. On-farm animal welfare assessment is a rapidly developing and crucial component of the numerous industrial, private, and retail assurance programs that have been established recently to promote quality food markets and address consumer concerns about farming practices. These programs are found in the agri-food and retail industries (Roe et al., 2011). There is a growing understanding that farm animal wellbeing should be safeguarded and improved, despite the various definitions of animal welfare that lead to wildly divergent perspectives on this difficult subject. A growing number of customers view animal-friendly products as being healthier, safer, tastier, more hygienic, more authentic, more ecologically friendly, and more traditional. Animal welfare indicators are increasingly regarded more highly than other qualitative attributes. By giving enough information on the management and housing circumstances of various farm animal species, it may be possible to encourage people to be willing to pay the price increases that greater levels of farm animal welfare may entail (Alonso et al., 2020).

In order to comprehend and improve animal wellbeing, it is necessary to assess it. The idea of five domains offers a framework for this assessment. Because they directly reflect how well an animal is doing in its environment, animal-related indicators should be the main focus of our analyses when trying to understand animal welfare. Comprehensively identifying the areas where actions need to be made to address or mitigate animal welfare issues can be done by incorporating management and resource assessments with the animal-related measurements within the five domains (Doyle et al., 2021). Numerous techniques, including behavioral, physiological, and psychopathological evaluations, longevity tests, and production efficiency, can be used to assess animal welfare. All measures of animal welfare have some limitations and are unreliable when employed as a sole way of evaluation. It is commonly acknowledged that using a collection of

indicators rather than a single parameter for measuring animal wellbeing can lead to better findings (Islam et al., 2020).

Scientists who study animal welfare and those who study human welfare generally agree that the concept of welfare refers to the measurable state of the individual on a scale from very good to very bad (Broom, 2009). An accurate assessment of welfare can be made along a continuum from very good to extremely poor. Studies of preferences can provide insight into the circumstances that might promote excellent welfare, but it is necessary to weigh the significance of each desire to the individual. When animals experience short- or long-term issues, measures of how poorly they are doing may be physiological or behavioral in origin, or they may have something to do with their own production or illness. Any one indicator may suggest poor welfare because people have different coping mechanisms, and the lack of evidence on one measure does not imply that there is no welfare problem (Broom, 1988). The assessment of resource management and provision, in-person animal observation, and a review of farm records serve as the foundation for determining the welfare of animals on farms. Assessment of animal health, physical condition, and behavior can be used to directly infer the impact of housing and management on these animals' welfare, while environmental examination can point to the possibility of specific welfare issues (Islam et al., 2020).

Health, comfort, behaviors, and other aspects of welfare are all interrelated. Therefore, a variety of criteria must be evaluated in order to appraise it overall. The requirements must be complete (nothing must be left out), minimal (just what is absolutely necessary), accepted by all parties involved, and readable (a limited number of criteria) (Botreau et al., 2007). The ability to engage in motivated activities, bodily well-being, the absence of hunger and disease, and other factors are all included in welfare, which covers both physical and mental health. Individuals may place differing levels of priority on certain aspects of animal care. A multidisciplinary approach should be used to evaluate animal wellbeing since doing so can yield the most thorough analysis of an animal's welfare in a particular system (Welfare Quality®, 2009).

Scientific thinking regarding farm animal welfare has changed in the past decade, mainly due to the recognition that animals are sentient beings. Previously, welfare was assessed using measures

of biological functioning related to health and to meat, milk, fiber, or egg outputs. Although such measures are still used, attention now focuses on the following scientifically-supported understanding: Animal welfare states reflect what animal's experience i.e., their emotional or affective states and these experiences may be negative or positive. The acceptability of production systems is now judged not only by inputs such as their design but also by animals' welfare-related responses to them, validated measures of negative welfare states are focused on established physiological, clinical, and/or behavioral responses of animals to adverse conditions, and these measures guide preventative and remedial actions, some behavior-based indices of positive welfare states are well validated and in current use, and science- based support is being sought for others, The negative-positive experiential balance reflects an animal's quality of life such that a net negative balance represents a poor quality of life and Human-animal relationships can have marked effects on animal welfare. Good welfare-related knowledge, skills, and attitudes towards animals by stockpersons enhance the welfare and productivity of livestock (IFC, 2014).

Animal welfare is receiving more attention, which has resulted in the development of numerous distinct animal welfare assessment methodologies. There have already been various (prototype) monitoring systems created in Europe. These include the Freedom Food programs in the United Kingdom (Royal Society for the Prevention of Cruelty to Animals), the ethical account in Denmark, the TGI35L animal welfare index in Austria and the related TGI200 in Germany, a decision support system for the overall assessment of sow welfare in the Netherlands, and specific tools for dairy cows in France and Italy. The majority of these systems are predicated on selected observations of the animals, or performance measures, which are believed to provide information about the internal health of the animals, and observations of the environment, or design measures, which are assumed to have an impact on animal welfare (Blokhuis, 2008). Animal welfare research is well developed in many countries in Europe and elsewhere. Animal welfare is multidimensional and cannot be measured directly, but is derived from external parameters.

The Welfare Quality® (WQ®) method is a more contemporary and popular system for assessing the welfare of farm animals. Development of scientifically valid and practical on-farm animal welfare evaluation methods with a primary focus on animal-related measures was one of the key objectives of Welfare Quality® (WQ®) (Blokhuis et al., 2003). The Welfare Quality ® project,

which focused on integrating animal wellbeing into the food quality chain and created protocols to quantify, among other things, dairy cow welfare at the farm level, was one of the greatest instruments for evaluating farm animal welfare (Ventura et al., 2021). Concerns regarding animal welfare are the focus of Welfare Quality ®, which also seeks to facilitate open dialogue about the welfare of animals and product profiling. In order to build a connection between animal husbandry techniques and the knowledgeable display and purchasing of animal products, the latter is obviously essential. Animal welfare research knowledge has been merged with consumer/citizen perception and attitude analyses by Welfare Quality® to highlight 12 problem areas that should be effectively addressed in measuring systems (Blokhuis, 2008). These are presented in Table 2 as welfare criteria, where the direction for maximizing welfare is indicated. Each criterion covers a separate aspect of good animal welfare and the list was chosen to encompass all potential areas of concern while at the same time keeping the total number of criteria to a minimum. To further reduce the number of items and ease the understanding, they are grouped into 4 classes, called principles in the table, corresponding to the questions: Are the animals properly fed and supplied with water? Are the animals properly housed? Are the animals healthy? Does the behaviour of the animals reflect optimized emotional states? (Blokhuis, 2008).

Table 2. Set of criteria and sub criteria used in Welfare Quality® to develop an overall welfare assessment. (Source: Blokhuis et al., 2010; Botreau et al., 2007).

Principles	Welfare criteria	Specifications
Good feeding	1. Absence of prolonged hunger 2. Absence of prolonged thirst	
Good housing	3. Comfort around resting	Assessed through behaviour (including rising up and lying down movements) but not injuries (included in 5).
	4. Thermal comfort	
	5. Ease of Movement	Not considering health problems (included in 6, 7, 8) and movements around resting (included in 3).

Good health	6. Absence of injuries	Except those produced by a disease or voluntary interventions (eg mutilations).
	7. Absence of disease	Absence of clinical problems other than injuries
	8. Absence of pain induced by management procedures	Eg mutilations and stunning.
Appropriate behaviour	9. Expression of social behaviours	Balance between negative (eg aggression) and positive (eg social licking) aspects.
	10. Expression of other behaviours	Balance between negative (eg stereotypies) and positive (eg exploration) aspects.
	11. Good human-animal relationship	No fear of humans.
	12. Absence of general fear	Except fear of humans.

The Welfare Quality® system has a number of significant benefits: it is animal-centric, addressing the changing consumer, governmental, and animal welfare advocate concerns about the quality of animal life; it is comprehensive and holistic across time and space of animal production; it is rigorous and scientifically tested; it offers scope for market segmentation based on valid and transferable standards; it is dynamic in that it encourages higher animal welfare per unit of production (Blokhuis et al., 2010).

In Italy currently a protocol to measure the quality of animal welfare on farms is used a system called ClassyFarm. The protocol's objectives include supporting governmental regulations, gathering data, fostering the adoption of animal welfare standards, and educating consumers. The ClassyFarm system was created by the Italian National Reference Center for Animal Welfare (CReNBA), which receives funding from the Ministry of Health, and is the officially acknowledged method in Italy for classifying risks on farms where cattle are raised (www.classyfarm.it). For each sort of production and animal species, the system is based on the

opinions of experts. In line with EU requirements on official control of animal health, animal welfare, food safety, and medicine, the ClassyFarm system is primarily centered on preventive because it fosters improved cooperation between operators and relevant authorities. Data collected by animal welfare assessment checklists can be classified into two categories: resource-based indicators linked to management and structure-related risks, and animal-based metrics (ABM) (Mariottini et al., 2022).

Regardless of which definition one may use to evaluate animal welfare, it is important to recognize that there is a continuum of animal welfare that ranges from negative to positive (Figure 2). Improving animal welfare means ensuring that the animal experience is as positive as possible, which often requires changes in the infrastructure and practices of those responsible for the care and handling of animals (Fernandes et al., 2021).

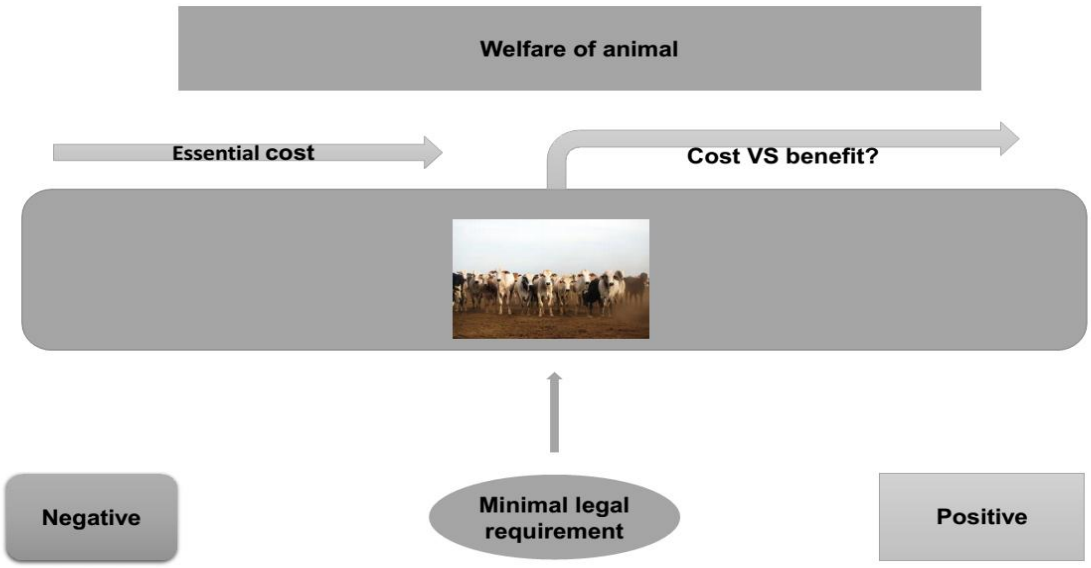

Figure 2: Continuum of animal welfare (Adopted, Source: Fernandes et al 2021).

An animal's welfare might be in either a poor or good state. There is a continuum of animal welfare levels, even if these terms may be viewed as subjective and their meanings may vary from person to person and over time. It is possible to classify the expenses required to ensure animal welfare as essential expenses. It is difficult to estimate the economic benefits of investments aimed at

raising animal welfare levels to more desirable areas of the continuum, but the market protection provided by securing and maintaining public and consumer support may very well outweigh the costs associated with ensuring high levels of animal welfare (Fernandes et al., 2021).

Different protocols developed like to assess the welfare of farm animal's work well in industrialized intensive farming (Example Welfare Quality® protocol). Consequently, emerging assessment protocols are designed to address the problems of these highly mechanized production units, overlooking others, such as traditional production systems that operate under different conditions and face different welfare problems. For example, these protocols are not fully useful in extensive, pasture-based systems and in small, traditional farms in developing countries because of the different characteristics of the production units (Hernandez et al., 2022). For example Ssuna (2021) applied the welfare 'welfare quality® assessment protocol for dairy cows' among farms in Kiruhura District, Uganda and he concluded that not all measures are feasible for on-farm assessment among extensive dairy farms in Kiruhura, district, Uganda.

1.3. General Condition of Farm Animal Welfare in Africa

Animals serve a variety of purposes in Africa, which may help to explain the complicated bond that exists between animals and their owners. Meat, milk, and blood from animals are used to make food; hides are used to make clothing, shoes, and other accessories; and hair and wool are used to make clothing and bedding. Their urine is sterilized and used as medicine, while their excrement is utilized as fuel, fertilizer, and to plaster traditional homes. Venipuncture is the process of drawing blood to utilize as food. Tools like needles and arrows are employed. The ensuing wound is not fatal and is then attended to. Even though it may appear cruel, great care is taken to prevent the animals from suffering unnecessarily. Animals are a symbol of riches and pride and are frequently given as wedding gifts (Masiga, and Munyua, 2005).

Despite the fact that most research on animal welfare concentrate on industrialized nations, mounting evidence suggests that the problem is also one that is starting to emerge in less developed nations. Comparatively speaking, emerging nations have fewer studies on animal welfare. However, elements that affect animal welfare issues, such as economy, culture, and religion,

should also be taken into consideration. For example, the idea of employing developed nation ways to promote animal wellbeing in developing countries should be taken into consideration (Mugenda and Croney, 2019). Animal welfare in developing countries like Africa is not given much attention and is optimal, and it's not a priority due to poverty and other issues like food security (Hernandez et al., 2022). Animal products can be crucial sources of nutrient-dense food in human nutrition because food security is a global concern, particularly in Africa. But in fact, it doesn't seem to be living up to its full potential for a variety of reasons that haven't been sufficiently addressed. One of the main causes of the slow advancement can be attributed to worries about animal welfare. It has been demonstrated through science that the effects have an impact on market access and product quality. However, the idea of animal welfare has not yet fully taken hold in the African population (Njisane et al., 2020). Africa's perceptions on animal welfare differ by region, culture, and customs. Even if everyone has the freedom to exercise their culture or religion, for example there have been disputes between traditional slaughterhouse operators and animal welfare organizations in Africa due to a lack of understanding or tolerance, for instance in South Africa (Qekwana et al., 2019).

In Africa, the livestock production system is majorly extensive system, extensive pastoralism accounts for 25% of the world's land area and supports 200 million subsistence pastoral households. The primary livestock concerns and animal welfare issues that arise in extensive production systems includes; shortage of food in animals that are foraging and these animals may not meet their nutrient requirement that may meet their needs so that animals will become unhealthy and experience chronic hunger, shortage of water, water is frequently one of the most scarce resources in extensive pastoral livestock production system, harmful plants; animals come into contact with a variety of plants that contain PSCs, thermal stress, predators, and other factors. Livestock Animal handling occurs less frequently in extensive systems than it does in intensive ones, which can lead to welfare issues, painful husbandry practices, diseases, and injuries (Temple and Manteca, 2020).

In recent years, concerns over animal welfare have grown in a number of nations around the world, particularly those in Africa. The majority of African nations have varying levels of animal welfare legislation, policy, and regulatory frameworks like the OIE standards. It has been noted that laws,

regulations, and standards are either absent, deficient, out-of-date, or not properly enforced. In a similar vein, despite the fact that all countries have signed the OIE standards, there is frequently little understanding and, as a result, little compliance with the standards, primarily because there is a lack of implementation capacity and a requirement for the development of country- and context-specific measures (AU–IBAR, 2017).

In Ethiopia, smallholder farmers who frequently live as pastoralists in the Lowlands and use communal and harvested land where animals graze freely raise the majority of farm animals. Young children typically are tasked with caring after the animals (shepherds). The animals spend the day in a field or a forest, but at night they are frequently kept in a corral. Small stables are occasionally available for dairy cows. The majority of animals are killed by their owners (not in a slaughterhouse). The few abattoirs that do exist are frequently shabby (birds of prey are usually nearby). Cows function somewhat like a "bank." They are used as working animals for ploughing the fields, and for milk and meat. On special occasions some animals are sold, e.g. if people have a slight surplus of animals (Bracke, 2009). Ethiopia is heavily dependent on agriculture and cattle, but different production systems are not as market-oriented as they might be. The majority of farmers sell their livestock to make money and to cover their family's expenses. Selling animals is typically not the first choice, though. This is due to a variety of factors; for example, in the highlands, cattle are kept as a draft animal for agricultural production, whereas in the lowlands, cattle serve as a social safety net and are highly prized for their reputation (Jerlström, 2013).

The African Union Inter-African Bureau for Animal Resources (AU-IBAR), in 2017, in preparing the Animal Welfare Strategy for Africa, summarized the key issues related to animal welfare, which are as follows: Lack of adequate education and awareness; insufficient stakeholder engagement and participation; lack of domestic science and research; insufficient understanding by stakeholders in the value chain of the value of animal welfare (economic, non-economic, social, etc.) in production systems, trade and health; Inadequate policy framework, strategic directions and action plans, as evidenced by lack of laws or outdated laws, inappropriate regulations and standards and weak or no implementation and enforcement; Inappropriate husbandry practices, both domestic and modern, that lack sufficient knowledge about the impact of good animal welfare practices on production, productivity and quality/health, leading to ignoring animal welfare in

production systems, are same of the main issues that hamper animal welfare discussed in the strategic document (AU–IBAR, 2017). The improvement of the situation of farm animal welfare situation in Africa could increase the productivity of livestock in the region and by doing so help to eliminated absolute poverty, and help in the food security of the region/continent (Hernandez et al., 2022).

1.4. Welfare of Beef Cattle

Beef cattle farming differ widely across the globe, ranging from extensive, extensive feedlots to intensive specialized farms and using different breeds (Nalon et al., 2021). Specific legislation on animal welfare by the European Union (EU) is developed for several species of livestock animals; but there is no specific regulation regarding beef cattle (Cozzi et al., 2009). In the EU, the welfare of beef cattle is covered only by Council Directive 98/58/EC concerning the protection of animals kept for farming purposes, often referred to as 'the General Farm Animals Directive'. This Directive "lays down minimum standards for the protection of animals bred or kept for farming purposes". Article 3 sets out the Directive's core principle. Specifically, it provides that "Member States shall make provision to ensure that the owners or keepers take all reasonable steps to ensure the welfare of animals under their care and to ensure that those animals are not caused any unnecessary pain, suffering or injury" (Council Directive 98/58/EC). In European Union (EU) beef fattening systems may be divided into two main categories: intensive systems, where the calves are reared indoors, and grass-based systems, usually involving winter accommodation. The diversity of beef fattening systems in the EU is influenced by the type of diets (largely related to the climatic environments) and by the different cattle breeds. These breeds may be dairy (primary output milk), dual purpose (producing milk and beef) or beef (primary output beef). The EU dairy herd is dominated by the Friesian/Holstein breed. In contrast, the EU beef herd is very diverse (EFSA, 2012).

Cozzi et al., (2009) reviewed the major welfare problem of beef cattle in Italy as follow: limited space allowance is one of the most important issues impairing animal welfare. Other risk factors for poor welfare related to the housing structures are type of floor, space at the manger, number of water dispensers and lack of specific moving and handling facilities. Microclimatic conditions can

be critical especially during the summer season when cattle can experience heat stress, feeding plan adopted in the Italian beef farms may be another factor negatively affecting the welfare of these animals due to the low content of long fire roughage which increases the risk of metabolic acidosis. The locomotors problems /lameness is also one of the major beef cattle welfare problem, in Italy beef fatting unit this problems are mostly related to feeding strategies that is implemented that is due to provision of high carbohydrate feeds in order to attain the desired weight and these leads to metabolic acidosis which in turn causes laminates and causes locomotors problems (Compiani et al., 2014). Husbandry and management conditions, along with the farmer attitude towards his/her cattle herd, more or less intensive feeding systems and rearing environment represent some of the factors that could negatively affect animal welfare (Cozzi et al., 2009). Most beef cattle diseases have a multi-factorial aetiology. In addition to pathogens and animal-related conditions, other contributing factors include environmental stressors that disturb homeostasis in the animal. These diseases can become chronic when infected animals are not detected and treated early. Chronic pneumonia results in very poor welfare with pain, asphyxiation and ill-thrift. Calves showing severe respiratory distress after multiple treatments should be killed on the farm. To promote effective control of multifactorial infectious diseases, cattle should be kept in environments that minimize physiological and emotional stress (EFSA, 2012).

Studies conducted Kirchner et al., (2014b) in Austria, Germany and Italy on a total of 63 beef bull farms (deep litter or cubicle housing systems) and assessed by Welfare Quality® Assessment protocol for Cattle, shown there are significant areas for improvement of beef cattle welfare, in this study some of the points identified and need improvement in which the scores were less than 50% were related to the criteria 'Absence of disease', 'Expression of social behaviour' and 'Positive emotional state'. Two thirds of the farms achieved the 'Enhanced' level, about one-third was estimated 'Acceptable' and only one farm 'Excellent'. Farm assessment showed poor quality stockman ship to be a very common weak point. The training of stock person is one of the best way to promote human animal interaction in beef farming, by doing so the life of the animals will improve as well as the person (Ceballos et al., 2018).

Beef farmers underestimate the direct and indirect losses created by incorrect feeding and management decisions or the onset of negative human-animal interaction. Producer associations,

public extension services, and other farm advisors should therefore promote specific training courses for stockmen to improve their knowledge and skills in welfare-friendly farm practices (Gottardo et al., 2009). EFSA (2021) hazard analysis for beef cattle identified three major categories of welfare problem attributable to risks associated with housing and management: i) Respiratory diseases: linked to overstocking, inadequate ventilation, and mixing of animals, as well as failure of early diagnosis and treatment ii) Digestive disorders: linked to intensive concentrate feeding, lack of physically effective fiber in the diet. Iii) Behavioral disorders: linked to inadequate floor space, co-mingling in the feedlot and intensive concentrates. Salvin et al., (2020) reviewed the factors affecting beef cattle in Australian feedlot farms which includes such as the inability of cattle to effectively respond to environmental extremes, inability to express their full range of natural behaviours such as grazing, and unsuitable temperaments, as well as management factors, such as stockperson skills in identifying morbidities, comfort of surface conditions for lying, stock-handling methods and yard design, identification and management of pregnancy and mixing of unfamiliar cattle.

Different studies have been performed for the assessment of beef cattle's that are kept in different specialized fatting farms in Italy and different European union countries, and have reported different critical points that affect the welfare of beef cattle (Kirchner et al., 2014b, Tarantola et al., 2020; Diana et al., 2020; Kirchner et al., 2014a, Gottardo et al., 2009; Brscic et al., 2015).

1.5. Welfare of Calves

Many factors are responsible for the welfare of calves in a farm which includes the housing, the environment, the feeding, management of health, the stock person or the farms, the way how transportation is handled, and husbandry managements like dehorning (Stull and Reynolds, 2008). In European Union (EU) there exist a legislation that govern the welfare of calves until six months of age by Council Directive 2008/119/EC (Calves Directive). This directives has an important articles that provides point which help the well-being of calves, some of the provisions of the directive includes: it prohibits on the use of individual pens after the age of eight weeks, it provides the minimum dimension for such pens, and minimum space allowances for calves kept in groups. Additionally, it required the supply of a minimum daily ration of fibrous food for calves older than two weeks (Council Directive 2008/119/EC).

Calves in a farm could suffer from poor welfare due to different factors affecting the welfare of calves includes environment and housing, so that the housing should be able to provide the necessary comfort to express their natural behaviour and should not be cause of stress for the new born (Stull and Reynolds, 2008). Housing calves in a group and pair could relieve stress and be practiced (Costa et al., 2019). Stock person that handle the new born should be trained, trained person is needed to handle and manage (Stull and Reynolds, 2008). In dairy industry calves could be considered as a surplus or unwanted product, if the calf is female, it can be used as replacer or could be sent to veal calf fattening farm, if male can be euthanised immediately after birth also (EFSA, 2012). The fate of each calf varies between countries depending on the dairying system, calf price, and the consumer preference for veal or beef (Haskell, 2020). Especially in countries where pasture based dairy farming is practiced and when there is seasonality of breeding. These unwanted calves will most of the times, their welfare is compromised in the farms or when they are transported (Boyle and Mee, 2021). Transportation of the calves is also a factor that affect welfare of calves since most calves in dairy farm specially are transported with in the short periods after they were born, so that before transportation of the calves necessary measures should be taken like providing sufficient colostrum, providing body protection, checking the navel (Stull and Reynolds, 2008), During long journeys unweaned calves may experience negative welfare consequences such as prolonged hunger and thirst, resting problems, thermal stress and diseases (Velarde et al., 2021). The nutrition and feeding of calves should be also well handled that is proper milk provision and gradual weaning are important factors in the improvement of welfare of a calves (Costa et al., 2019). Health calves is also other factor that should be considered so that the new born should be protected from disease (Stull and Reynolds, 2008). Good colostrum management is still recognised as the single most important factor to preventing calf morbidity and mortality. Only calves fed intensively with colostrum and milk are able to reach their full potential for performance throughout their life (Lorenz, 2012). Use of painful procedures for example dehorning is also a critical animal welfare issue when pain control is withheld; calves show negative behavioral, physiological, and emotional responses during and after dehorning (Costa et al., 2019). EFSA (2012) in their publication on the risk assessment of intensively reared calves identified the major welfare issues which is associated with the housing and management are: iron deficiency anemia which is as a result of inhibition of feed which is for production of

white meat, Digestive and respiratory disorders linked to high intakes of liquid feed and inadequate intake of physically effective fiber, and cross-infection resulting from mixing of calves from multiple sources, discomfort and disturbed resting behaviour linked to inadequate floors and floor space.

Studies were conducted to assess the welfare of calves in a farm by different assessment methods and identified points of compromised welfare for example Bugueiro et al., (2018) in their studies in Spain indicated a low score for "appropriate behaviour" and low score for "good health" indicating an area of improvement. In Canada, Vasseur et al., (2010) in their survey of dairy calf management practices identified the major welfare critical points that hamper the wellbeing of calves, non-existence of calving pens in most of the farms, luck of the disinfection of the navel of calves, luck of colostrum quality control including not checking whether the calf got colostrum or not the passive immunity, dehorning and removal of abnormal teat without use of pain management, harmful weaning, inappropriate housing, and calves housed individually. Moser et al., (2020) in their studies the comparison welfare calves of "Outdoor Veal Calf" Concept and in Conventional Veal Fattening Operations in Switzerland concluded that there is reduced antimicrobial use, and calf health and welfare were improved in "outdoor veal calf" farms in comparison to traditional operations. So that these indicate that alternate calf rearing options should always be investigated so that to improve the welfare of calves.

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CHAPTER II: THE APPLICABILITY OF EUROPEAN UNION (EU) FARM ANIMAL DIRECTIVE (COUNCIL DIRECTIVE 98/58/EC) TO AFRICA

2.1. Summary

Animal welfare is no longer the concern of a few who care about it, but the subject of international legislation and farm animal agriculture is arguably the most economically important interface between humans and other animals on this planet. Since the last decade of the 20th century, the institutions of the European Union (EU) have consistently sought to effectively regulate the welfare of farm animal species, primarily through conventions and horizontal and species-specific legislation. But in the other way even though the relationship between animals and their owner in African conditions is often deep and complex, most African countries are at different levels in terms of animal welfare laws, regulations and policies, such as the OIE standards. Policies, standards and laws are found to be either lacking, inadequate, outdated or poorly enforced. Similarly in Ethiopia animal welfare legislation/proclamation is missing. Therefore the review objective; is to see the applicability of the European Union (EU) farm animal regulations (Council Directive 98/58/EC) to the context of Africa focusing Ethiopia. In order to attain the objective the writer reviewed relevant journal articles, international, national, intergovernmental, governmental, and non-governmental standards, legislations, regulations and codes that focuses in farm animal welfare. So that by doing so to understand the difference in farm animal welfare and production differences between the EU and Africa. Finally the articles and annexes of farm animal directive (council directive 98/58/EC) applicability to African context were compared. The directives that is developed for highly developed countries in European Union (EU), the "farm animal directive'' may not be totally fit with the situation on the ground in countries like Ethiopia which still have traditional system of livestock production, but it could be a starting point, so as after successful identification of the farm animal welfare critical points may help as a starting point with modification to the local situation in the ground.

Key words: Farm animal welfare, legislation, European Union, Ethiopia, Africa

2.2. Introduction

Animal welfare is becoming a topic of international law, not just the concern of a small group of people (Harris, 2005). The interface between people and other animals on this planet that is most economically significant is probably the farming of animals. It has the ability to inflict pain on a vast number of animals over an extended period of time, culminating in a death that might represent that pain in any number of ways (Sinclair et al., 2019). In the OIE Terrestrial Animal Health Code, Animal welfare means the physical and mental state of an animal in relation to the conditions in which it lives and dies. An animal experiences good welfare if the animal is healthy, comfortable, well nourished, safe, is not suffering from unpleasant states such as pain, fear and distress, and is able to express behaviours that are important for its physical and mental state. Good animal welfare requires disease prevention and appropriate veterinary care, shelter, management and nutrition, a stimulating and safe environment, humane handling and humane slaughter or killing. While animal welfare refers to the state of the animal, the treatment that an animal receives is covered by other terms such as animal care, animal husbandry, and humane treatment (OIE, 2022). There are several reasons for the growing need for improvement in animal welfare, which is acknowledged by the worldwide community through policy and regulation. There are now regional animal welfare programs on every continent (Doyle et al., 2022).

The institutions of the European Union (EU) have consistently worked to adequately regulate the welfare of farm animal species since the last decade of the 20th century, principally through conventions and horizontal and species-specific legislation. In doing so, Europe has made an effort to address a number of linked issues, including those involving laying hen protection in battery cages, transportation safety, broiler chicken welfare, living conditions, sheep and goat traceability, etc. (Leone, 2020). The housing, feeding, transportation, and slaughter of billions of farm animals are all protected by some of the strongest animal welfare rules in the world today, which are found in the European Union. The goal of EU regulation on farm animal protection is to make sure that animals are housed and raised while taking into account their bare minimum physiological demands and that painful procedures are kept to a minimum (European commission, 2014). Animals are considered sentient beings by the European Union (EU), and EU members are becoming more aware of the significance of upholding moral norms for their care. The "rules of

the game" for producers, government agencies, and civil society are established by EU animal welfare regulations. They safeguard the wellbeing of farm animals and level the playing field for the domestic market (EU, 2022).

Due to the variety of agroecology on the African continent, which ranges from hot, dry regions to humid, tropical regions and topography that ranges from mountains to lowlands, various animal species can flourish in various settings (Njisane et al., 2020). Animal welfare has risen in importance as a global concern in recent years, particularly in many African nations. Regarding laws, rules, and policies pertaining to animal welfare, such as the OIE standards, the majority of African nations are at various stages. It is discovered that laws, regulations, and policies are either missing, insufficient, out-of-date, or poorly enforced (AU–IBAR, 2017). Although there are international standards for animal care in the industrialized world, most developing regions lack the necessary conditions to adopt such programs, especially among small-scale farmers. These include social class, socioeconomic standing, the resources that are accessible, information dissemination, and monitoring tools (Njisane et al., 2020). Depending on the area, culture, and customs, there are different perspectives on animal care in Africa (Qekwana et al., 2019).

Many Africans depend on their livestock for their livelihood since it provides them with food, income, and other socioeconomic advantages (Masiga and Munyua, 2005). In African settings, the bond between animals and their owners is frequently strong and complicated. For instance, cattle often have names and are kept for longer than is necessary since their owners view them as members of the family. Lack of food and exposure to diseases that can be prevented are the two main factors that contribute to diminished animal welfare, which is frequently correlated with the affluence of their owners. Consequently, ensuring the welfare of animals can also benefit those who own them (Qekwana et al., 2019). The World Organisation for Animal Health's (OIE) animal welfare guidelines, which offer a scientific foundation for management strategies aimed at maintaining an acceptable level of animal care, have made it easier to harmonize animal welfare policies. Even though animal welfare laws are being introduced more frequently around the world, their real enforcement is still lacking, and there are few and inadequately resourced systems to ensure compliance, as is the case in some African nations (Doyle et al., 2022). Despite the fact that Ethiopia and all other African nations have signed the OIE standards, there is frequently little

understanding of the standards and, as a result, little compliance with them. This is primarily because there is a lack of implementation capacity and the need for country- and context-specific measures (AU–IBAR, 2017).

Ethiopia is a storehouse for enormous genetic variation in cattle and possesses the most livestock resources in Africa. The majority of the country's population's needs for meat and milk are met by cattle, making them the most significant species in the national livestock herd (Bekele et al., 2018). Although Ethiopia is growing and the economy is doing well, according to Bimrew (2014), little has been done to improve the welfare of animals. There are currently no regulations in place to safeguard animals against human cruelty. There are, however, a few groups that work to enhance animal welfare, but they mainly focus on displaced or harmed donkeys and other equines. It is notable that whereas efforts for working animals are quite concentrated, those for food animals are not. Draft animals receive more attention than food animals (ICPALD, 2018). In order to build appropriate laws and measures for animal welfare in developing nations like Ethiopia, the techniques should be founded on an awareness of the situation of farm animal handling and management (Bimrew, 2014). Some nations are using or modifying existing laws to combat animal cruelty to strengthen their legislative framework, while others are creating new animal welfare laws that incorporate local and national concerns with global animal welfare principles (Vapnek and Chapman, 2010). Based on the above background, the current literature review objective; is to see the applicability of the European Union (EU) farm animal regulations (Council Directive 98/58/EC) to the context of Africa focusing Ethiopia, and so that finally to forward recommendations.

2.3. Methodology

In order to understand the level and the situation of farm animal welfare, type of farm animal production, to have a clear picture of the animal keepers (farmers, pastoralists) in Africa focusing in Ethiopia; the writer did search and reviews of relevant literatures, relevant governmental, inter-governmental documents standards, non-governmental international organizations documents and standards, societies, agencies to gather and collect information about the situation of welfare of farm animals and legislations. Additionally welfare legislation of the European Union (EU),

specifically focusing on the farm animal directive (Council Directive 98/58/EC) were reviewed. Finally the applicability of the EU farm animal welfare directive (Council Directive 98/58/EC) articles annexes were discussed and conclusion was made.

2.3.1 European Union (EU) Farm Animal Legislation

Over the past 40 years, the European Union (EU) has steadily enacted regulations pertaining to animal welfare. The European Union (EU) has passed a wide range of animal welfare laws since 1974. Animal welfare criteria must be taken into consideration by the EU and Member States when developing and implementing EU policies, such as those pertaining to agriculture or the internal market, because they are recognized as sentient beings under the EU Treaties (Simonin and Gavinelli, 2019). The first piece of law that was put up in the EU in 1974 dealt with controlling animal slaughter, and it was subsequently steadily expanded to cover animal transportation and various forms of livestock production (Pantzer, 2021). At the moment, transport and slaughter processes for all species are covered by EU regulation on the welfare of farm animals, as well as the breeding of chickens, calves, and pigs. This legislation is some of the most cutting-edge ever. The EU has specifically prohibited traditional laying hen cages and mandates group housing for pregnant sows (Simonin and Gavinelli, 2019). Legislation has changed and expanded during the 1970s in response to societal, political, and market needs as well as scientific and ethical advancements (Pantzer, 2021). The EU's unified legal system is a result of the many Treaties that the Member States have ratified. Directives, regulations, and decisions make up European Union legislation, but they must all ultimately flow from the Treaty (Moynagh, 2000). The EU's legal documents on animal welfare, such as the Council of Europe Convention on Animal Welfare from 1976 and Directive 98/58/EC on the protection of food-producing animals, are based on an understanding of animal welfare science that is very different from the advancements that applied ethology, cognitive science, and neuroscience have each brought to light (Leone, 2020).

Article 13 of the Treaty on the Functioning of the European Union (TFEU), generally known as the Lisbon Treaty, recognizes animals as sentient creatures (December 2009). The EU and Member States fully consider the requirements for animal welfare when developing and implementing EU policies on agriculture, fisheries, transport, the internal market, research and technology

development, and space policy (European Commission, 2013). Article 13 of Title II states that: "In formulating and implementing …… policies, the Union and the Member States shall, since animals are sentient beings, pay full regard to the welfare requirements of animals, while respecting the legislative or administrative provisions and customs of the EU countries relating in particular to religious rites, cultural traditions and regional heritage." (Pantzer, 2021). Figure 3 (European Court of Auditors, 2018), illustrates the key events that had taken place in the European Union (EU) related with animal welfare, and animal welfare legislation.

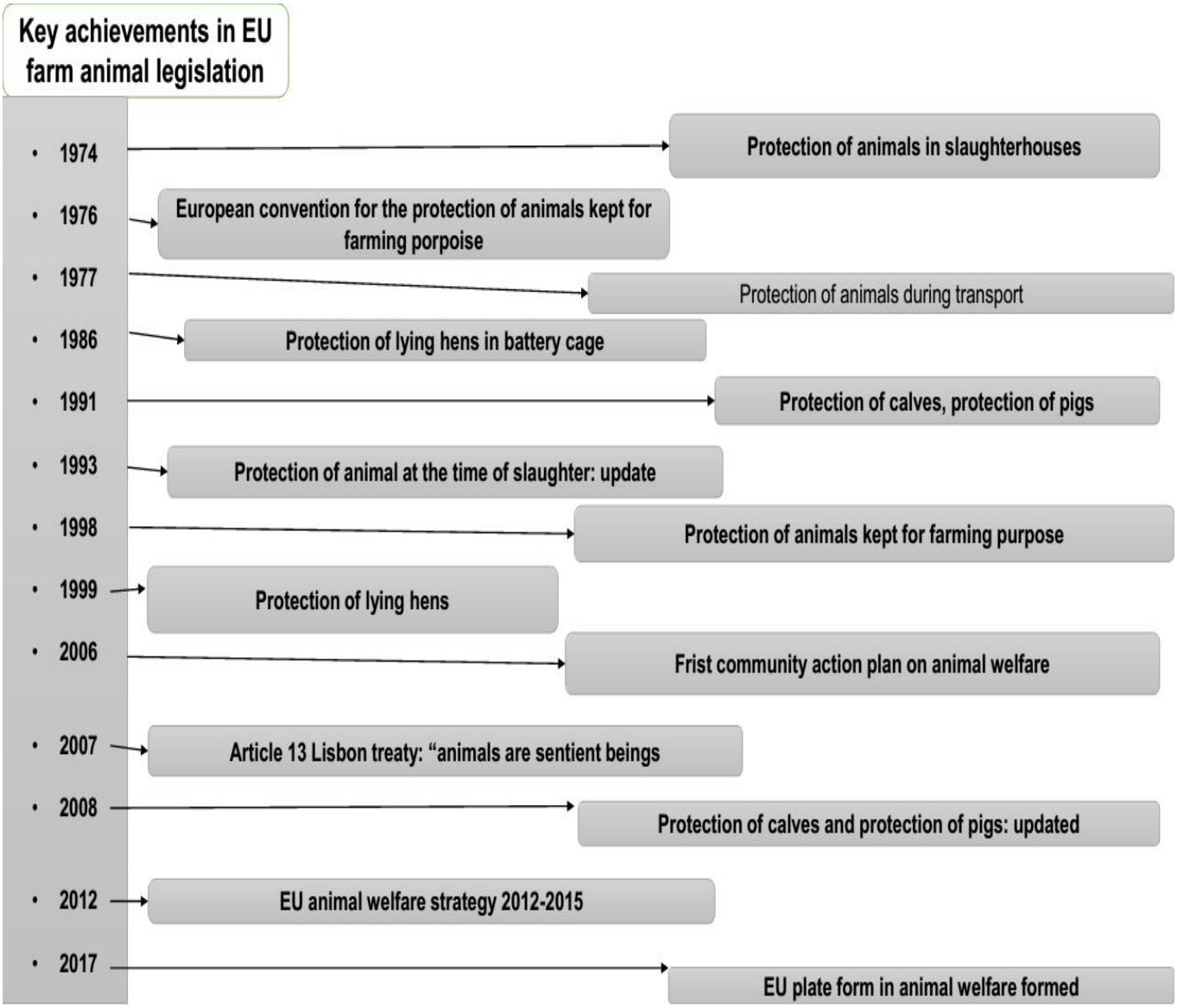

Figure 3: Key EU actions Related to animal welfare. (Adopted, Source: European Court of Auditors. 2018 Available https://www.eca.europa.eu/Lists/ECADocuments/BP_ANIMAL_WELFARE/BP_ANIMAL_WELFARE_EN.pdf. (Accessed on November 30, 2022).

Public opinion is unquestionably the primary motivator for increasing animal welfare. The general wellbeing of animals is in dire need of improvement, according to the public (Moynagh, 2000). Economic variables, however, are also important since they show how improving animal welfare

results in better and safer products for consumers. The EU currently boasts one of the tightest legal systems in the world, and as a result, the greatest welfare requirements are in place to safeguard the billions of animals that are raised, transported, and killed on the soil of its member States. It is now secured that animals are born and live with respect for their physiological needs thanks to the relentless efforts of the EU authorities, and painful and inhumane practices in the slaughter process have almost entirely been abandoned (Zoi et al., 2019). In order to prevent any member State from gaining an unfair advantage, there has been a large degree of legal harmonisation among member states. This harmonisation included basic living standards that would apply across the EU. It also addressed other aspects of decent living standards (Moynagh, 2000).

The protection of animals under EU law includes both general protections as well as the specific measures required to safeguard farm animals (with separate laws covering, in particular, calves, pigs, and laying hens), wild animals, and animals used in farming experiments at various stages of their lives (transport, slaughter, killing, etc.). Nearly much of the EU's animal welfare legislation focuses on agricultural animals, although there is also particular legislation covering companion, laboratory, and wild animals (European court of auditors, 2018). Farm animal protection laws currently encompass every aspect of production, from breeding to transport to slaughter. Five directives establishing minimum standards apply to agricultural activities, whereas regulations establishing uniform requirements for all Member States apply to the transport and killing of animals (Simonin and Gavinelli, 2019). A 1998 Directive on the protection of animals kept for agricultural purposes, which is applicable to all farmed animals and provides them with protection through general principles, serves as the EU's legislative framework for the wellbeing of food-producing animals. Sectoral law is a supplement to this Directive. Four Directives on the conservation of specific species were approved between 1999 and 2008, managing the welfare of laying hens, broilers, pigs, and calves in total, accounting for 48% of the EU's farmed mammals and 80% of its farmed birds. A law on the transportation of animals for commercial purposes was also approved in 2005, and a regulation on the protection of animals during the killing process was adopted in 2009 (European commission, 2020).

- Council Directive 98/58/EC of 20 July 1998 concerning the protection of animals kept for farming purposes, (the “Farm Directive”)

- Council Directive 1999/74/EC of 19 July 1999 laying down minimum standards for the protection of laying hens (the "Laying Hens Directive"),
- Council Directive 2007/43/EC of 28 June 2007 laying down minimum rules for the protection of chickens kept for meat production (the "Broilers Directive"),
- Council Directive 2008/119/EC of 18 December 2008 laying down minimum standards for the protection of calves (the "Calves Directive"),
- Council Directive 2008/120/EC of 18 December 2008 laying down minimum standards for the protection of pigs (the "Pigs Directive"),
- Council Regulation (EC) No 1/2005 of 22 December 2004 on the protection of animals during transport (the "Transport Regulation"), and
- Council Regulation (EC) No 1099/2009 of 24 September 2009 on the protection of animals at the time of killing (the "Killing Regulation"). (European Commission, 2012)

The Commission has committed to reviewing EU animal welfare legislation by 2023 as part of the EU's farm-to-fork strategy in order to ensure a higher level of animal welfare by bringing current regulations into line with the most recent scientific findings, extending their application, and making them simpler to enforce as well as to contribute to a more sustainable food system (European Commission, 2022). Although the European Union is thinking about changing the current farm animal rules, for the time being this is the law that is currently working, and many developing nations can draw valuable lessons from this particular set of laws.

2.3.2. EU General Farm Animals Directive (Council Directive 98/58/EC of 20 July 1998)

The EU also adopted a directive in 1998, which is the oldest and most important directive containing provisions relating to all animals (including fish) raised or kept for food, wool, leather or fur production or other agricultural purposes, usually referred to as the General Farmed Animals Directive. Some of the provisions of this Directive are broadly worded, while others are more specific (Pantzer, 2021). Council Directive 98/58/EC has 12 articles and 21 annexes with different category that provides the general framework for the welfare of farm animals and applies to all animals (including fish, reptiles and amphibians) farmed for food, wool, skin, fur or other farming purposes. Animal owners and keepers are considered responsible for the welfare of animals in

their care and for avoiding unnecessary pain, suffering or injury. Appropriate training is also mandatory for workers involved in the care of animals. The same Directive lays down general requirements for regular inspections, treatment of sick animals, recording of veterinary treatments and mortality, animal buildings, open-air shelters, etc. (Caporale et al., 2005).

One of the most important article in the 1998 Directive is Article 3, which requires EU Member States to:….. "Make provision to ensure that the owners or keepers take all reasonable steps to ensure the welfare of animals under their care and to ensure that those animals are not caused any unnecessary pain, suffering or injury". The other important point that the of the 1998 Directive is "Freedom of movement" and states that: ……"The freedom of movement of an animal, having regard to its species and in accordance with established experience and scientific knowledge, must not be restricted in such a way as to cause it unnecessary suffering or injury. Where an animal is continuously or regularly tethered or confined, it must be given the space appropriate to its physiological and ethological needs in accordance with established experience and scientific knowledge" (Council Directive 98/58/EC). Very general requirements (staff, record-keeping, freedom of movement, housing, equipment, food and water, mutilations and breeding procedures) are listed in the annex to the farm animal guidelines (Council Directive 98/58/EC). These requirements tend to reflect the five freedoms, which were first established in the UK and include freedom from hunger and thirst, freedom from suffering, freedom from pain, injury, and disease, freedom to express normal behavior, and freedom from fear and distress (Pantzer, 2021).

2.3.3. Farm Animal Welfare in Africa

General Situation of Farm Animal Welfare in Africa

Livestock is a major source of sustenance for many Africans, as well as a source of money and other socioeconomic benefits (Masiga and Munyua, 2005). In African settings, the relationship between animals and their owners is frequently complicated and have complex cultural relations. For instance, cattle often have names and are kept for longer than is necessary since their owners view them as members of the family. Lack of food and exposure to diseases that can be prevented are the two main factors that contribute to diminished animal welfare, which is frequently

correlated with the affluence of their owners. Consequently, ensuring the welfare of animals can also benefit those who own them (Qekwana et al., 2019). Njisane et al., (2020) reviewed that about 50% of African households' food and income needs depend on livestock; with the main contributing species being cattle, chickens, sheep and goats. In addition, livestock products can play an important role as nutrient-dense food sources in the human diet, as they are of high quality and readily available for absorption by the human body, thus contributing to food security, which is a current global challenge.

Animal welfare concerns in Africa are frequently complex and related to social, political, religious, cultural, and economic aspects of society. Different viewpoints and interpretations of what "welfare" entails further complicate this. There are variations in practices throughout nations, tribes, and occasionally even amongst practitioners. The variety of cultural customs and the animals involved present a special challenge for African animal welfare laws. Some of the obstacles to animal care in Africa are also caused by a lack of resources or knowledge. In addition, African conditions do not call for the use of contemporary animal welfare guidelines (Qekwana et al., 2019). Some of the food in Africa is produced by a vast number of small-scale farmers and pastoralists who rear enormous quantities of cattle. Small-scale farming also contributes significantly to the rural economy. They frequently reside in isolated or rural places with scant resources and restricted access to knowledge (Njisane et al., 2020).

The majority of African nations have varying levels of legislation, regulatory frameworks, and rules governing animal welfare, such as OIE standards. Similar to this, despite the fact that all nations have signed the OIE standards, understanding and subsequent compliance with them are frequently hindered, mostly because there is a lack of implementation ability and a requirement for developing country and context-specific measures (AU–IBAR, 2017). There are indigenous elements that prevent their acceptance in the majority of developing regions, such as Africa, despite the fact that international animal welfare standards exist in the industrialized world and are a crucial instrument for better management methods (Njisane et al., 2020). Because of this, the region's ability to participate in import and export with the rest of the globe is now limited, which is reflected in its slow contribution to economic progress. Therefore, there is a need for

improvement in the state of animal welfare in Africa from all stakeholders as well as those involved in livestock rearing (Njisane et al., 2020).

Social class, socioeconomic status, cultural norms and practices, resource availability and infrastructure, information dissemination tactics that are ineffective, and a lack of the necessary monitoring instruments are just a few of the variables that may have an impact on animal welfare in Africa (Njisane et al., 2020; Qekwana, 2019). In addition, solving animal welfare issues in Africa is still hampered by poverty, unemployment, and climate change. The recommended techniques for containing, moving, and treating sick animals are frequently expensive and difficult to use. People are consequently compelled to employ substitute techniques that are not considerate of animal welfare. This emphasizes the connection between human and animal wellbeing. Therefore, human welfare must be considered while attempting to address difficulties with animal welfare in Africa (Qekwana et al., 2019). Research on farm animal welfare has largely concentrated on issues that are thought to be prevalent in intensive systems. The welfare of animals raised in extensive systems, in comparison, has gotten far less consideration. But in many regions of the world, extensive animal production systems are crucial (Temple and Manteca, 2020). Many people's livelihoods depend heavily on extensive livestock systems, which in many places are the only means of feeding people. Additionally, these systems are crucial because they support agricultural growth, the preservation of biodiversity, and the preservation of the genetic variety of animal species. However, in order to ensure their long-term social and economic sustainability, efforts must be made to recognise that, although they offer clear advantages over intensive systems in certain welfare areas, they are not without challenges. In addition, research is needed to develop welfare assessment tools that can be used in extensive systems. Many farm animal welfare issues in extensively farmed animals are complex and face multi-factorial challenges that can best be addressed by alternative approaches rather than the traditional top-down, science-to-practice dissemination of knowledge (Temple and Manteca, 2020).

General Situation of Farm Animal Welfare and Legislation in Ethiopia

Ethiopia is classified as a low-income country by the World Bank and ranks 173rd out of 189 countries on the Human Development Index scale and almost 79% of the population lives in rural

areas (Management Entity, 2021). Ethiopia has a highly diverse agricultural sector, and agricultural resources are crucial to the country's economy's rapid expansion. Although the nation is growing and the economy is doing better, little attention has so far been paid to the subject of animal welfare (Jerlström, 2013). Ethiopia have the largest livestock population in Africa. The domestic animal population of Ethiopia includes 70 million cattle population, 42.9 million sheep, 52.5 million goats, 8.1 million camels and including 57 million poultry. According to the Ethiopia central statisical agency that 97.4 percent of the total cattle in the country are local breeds. The livestock sector has contributed significantly to the country's economy and continues to show promise to contribute to the country's economic growth. Livestock products and by-products in the form of meat, milk, honey, eggs, cheese, butter, etc. provide the necessary animal proteins that help improve the nutritional status of the population. The livestock sector also plays an important role in providing export products such as live animals, hides and skins for the country's foreign trade. Livestock also provides a degree of security in case of crop failure as it is a 'quasi-liquid' asset. In addition, livestock provide manure which is commonly used to improve soil fertility and is used as a source of energy (CSA, 2021). A significant portion of the population's livelihoods are supported by livestock, which contributes roughly 45% of the value of all agricultural products. 70 % of households, or more than 14 million, keep animals, many of them being low-income. Three cattle, three goats or sheep, and a small number of hens make up the usual small herd (FAO, 2019).

The most significant livestock subsector in Ethiopia is unquestionably cattle. About 45% of agriculture's value added comes from it (AGDP). Cattle are raised by farmers in a variety of production techniques, the majority of which provide both milk and beef. The mixed crop-livestock, pastoral/agro-pastoral, urban/periurban, commercial dairy, and feedlot production systems are the most common ones (FAO, 2018). Cattle are evenly distributed throughout Ethiopia, with higher density in the highlands. The livestock production system is mainly extensive, with indigenous breeds and low-input/low-output farming practices (Tegegne et al., 2013). The distribution of the cattle population in different production systems includes 77% mixed livestock crop, 14% pastoral/agro pastoral, 7% urban and peri urban, 2% commercial dairy farms (FAO, 2018).

The majority of the intensive management system's animals are housed in peri-urban settings, and are dairy farms are where exotic breeds or crossbred animals are primarily kept for their excellent performance (Tegegne et al., 2013). Poor genetics, poor reproductive performance, poor quality and varying seasonal availability of feed, high disease incidence and parasite challenges, and limited access to services and inputs are just a few of the issues that prevent the livestock sector's productivity from being effective and providing the required productivity (Management Entity, 2021). The primary method of raising livestock in the Ethiopian highlands is mixed crop livestock farming system. Crop wastes serve as feed for the animals, and crops and livestock have interdependent roles in this system, with livestock providing draught power and manure for crop cultivation. In Ethiopia, pastoral and agro-pastoral livestock production is the second most prevalent method, and it is primarily practiced in the south and east of the nation (Tegegne et al., 2013). Most farm animals are raised by smallholders, who are often living as pastoralists in the Lowlands and use communal and harvested land where animals roam freely. Usually, young children have the task of minding the animals (shepherds). During the day the animals live in the open field or bush, but at night they are often placed in a corral. For dairy cows small stables are sometimes available (Bracke, 2009). Livestock is a major source of animal protein, power for crop cultivation, means of transportation, export commodities, manure for farmland and household energy, security in times of crop failure, and means of wealth accumulation (Management Entity, 2021). Cows serve more or less like considered as a ‘deposit’ so in some of the cases they can sold if they are too many in number. They are serve as working animals for ploughing in the crop cultivation, and also are source of food like milk and meat (Bracke, 2009).

There needs to be more investigation into how animals are treated in underdeveloped nations, which has long been a contentious topic. Despite the vast number of animals in the nation, poor welfare conditions result in low productivity and production, which is a prevalent element of the industry (Asebe et al., 2016). In Ethiopia even though most of the community specially the rural and pastoral population livelihood is dependent on their livestock, the limited studies that were performed in the different communities and farm animal related activites including pastoralists, marketing places, during transporting, farming places, slaughtering houses, feeding areas, sheltering areas and watering places are the commonest areas where welfare of farm animals were deprived due to most communities lack of awareness about good animal welfare and animal

handling. In most cases the handling and treatment of livestock is abusive in these areas (Jerlström 2013, Bracke, 2009; Nuguse, 2020; Bekele et al., 2020; Bulitta et al., 2012; Lemma et al 2022; Diro et al., 2021). For Example Most animals are slaughtered by their owners in the back yard. Abattoirs that do exist are often old and dirty usually with wild animal and birds roaming in the compound (Bracke, 2009). Livestock were brought to markets mostly done in Ethiopia by trekking just like most African countries, due to lack of appropriate vehicles and transportation means (Bulitta et al., 2012, Jerlström, 2013).

Limited studies that were conducted in Ethiopia indicate that theirs is lack of awareness about welfare of animals and animals suffer from poor welfare. Lemma et al., (2022) in there study in the rural household of Ethiopia said that community members knew the value of animals, there was limited knowledge of what their animals needed to experience good welfare. When it comes to good animal management practice, there were limitations both due to resource constraints, lack of knowledge, and behavior of owners or caregivers. Their knowledge of diseases and the actual care they give to animals in terms of preventive measures was limited. There was also a knowledge gap regarding nutrition, behavioral and health problems of animals. Diro et al., (2021) in their studies concluded that there was poor handling and stressful situation of beef cattle before slaughtering, which negatively affected the welfare and beef quality. Hence, pertinent proclamations, regulations, and delivery of animal welfare awareness training for different stakeholders are urgently needed.

There are numerous issues with farm animal welfare in Ethiopia that have not been addressed by the relevant parties or even by the non-governmental organizations now working on this topic (Asebe et al., 2016). Animal welfare is not a public or political issue in Ethiopia due to poverty. People do not seem to respect animals and may treat animals in an unfriendly manner. Beating animals is common practice and animals are often malnourished (Bracke, 2009). In most parts of the country, farmed animals are housed outdoors without adequate shelter and exposed to intense direct sunlight and erratic summer rainfall. The animals are forced to stay for long periods of time in the market even for days without water and feed with harsh handling. However, there are several veterinary schools and animal science graduated professionals, yet the health care services and maintenance of animal welfare fall short of expectations (Asebe et al., 2016).

In Ethiopia there is no comprehensive animal welfare legislation, no laws that protect animals from the cruel actions of humans that protect animal rights except may be a single article in the criminal code of federal democratic republic of Ethiopia (Jerlström, 2013). Although there are many proclamations related to animal husbandry, they have not paid attention to animal welfare issues. Except for a few articles in the country's criminal laws presented by the emperor King Minilik II. The country has tried to formulate animal welfare issues since 1889 when the first Italian veterinary mission came to Ethiopia to study the impact of disease in the country (Asebe et al., 2016). Although there is no comprehensive animal welfare regulation in Ethiopia, some of the countries proclamations mention abusive handling of animals and harming of animals is prohibited. For example, Ethiopia's Criminal Proclamation No. 414/2004 states that contamination of water, feed and pasture is considered a crime. Crimes committed through the production and distribution of substances hazardous to animal health, the manufacture, adulteration and sale of feed and products harmful to animals and the scandalous treatment of animals are included, by way of example, in various articles of the Penal Code of the Federal Democratic Republic of Ethiopia. (Proclamation No.414/2004, The Criminal Code Of The Federal Democratic Republic Of Ethiopia), but as far as its know there is no published case in the court of Ethiopia regarding a conviction and punishment of a person due to his abusive action to animals. Federal Negarit Gazeta of Ethiopia under the Proclamation No. 267/2002 stated about the prevention and control of animal diseases, the primary concern is to prevent and control animal diseases in order to maximize the benefits obtained from the extensive livestock resource (Federal Negarit Gazeta of the Federal Democratic Republic of Ethiopia, 2002). Ethiopian Veterinary Drug and Feed Administration and Control Proclamation No. 728/2011 also stated as clinical test shall be conducted with due care to animal welfare requirements (Federal Negarit Gazeta of the Federal Democratic Republic of Ethiopia, 2012).

In 2013 the Ministry of Agriculture (MoA) and the International Livestock Research Institute (ILRI) developed a strategy and vision for animal health in Ethiopia, and in this document the promotion of animal welfare is set as one of the major challenges and as strategic interventions activities have been set which are: Enact and implement animal welfare legislation and guidelines; establish an animal welfare fund to support the implementation of the legislation and guidelines;

encourage good agricultural practices to enhance animal welfare; develop a comprehensive communication action plan to produce and disseminate accurate, useful and timely animal welfare information; and encourage the establishment of animal welfare groups (MoA and ILRI, 2013).

There are well-known NGOs that try to improve animal welfare: like The Brooke Hospital for Animals; International Donkey Protection Trust (IDPT) (Bracke, 2009). However, more attention is paid to working animals mostly equines than to food animals, so equal attention is not given to food animals and working animals. There is a big gap of attention lacking between food animals and working animals. Brooke Ethiopia has participated and had their contribution to OIE working equine standards. In Ethiopia, IGAD/ ICPALD (Intergovernmental Authority on Development in Eastern Africa) (Center for Pastoral Areas and Livestock Development), assessment document of the east African countries on the status of animal welfare stated that the existence of draft Animal Welfare Policy, proclamation in Ethiopia. But still the draft 'Animal health, animal welfare and veterinary public health' proclamation is not yet out (ICPALD/IGAD, 2018). Currently there is a draft proclamation on "A Proclamation to Provide for Animal Health and Welfare" that is going to be sent to the parliament for ratification (Personal communication; the Director for LITS and Animal Welfare (LITAW) at Ethiopian Agricultural Ministry).

Different Livestock Production Systems and Welfare Quality® Criteria

In the EU-research project Welfare Quality® (http://www.welfarequality.net) the five freedoms were used to formulate protocols for on farm assessment of animal welfare for different species. The protocols allow for evaluation of animal welfare measured on the animals themselves. The Welfare Quality project was the largest European project concerning animal welfare (Leenstra, 2013). The aim the Welfare quality project was prepared to develop European standards for welfare assessment on farms. The Welfare Quality project revisited the original concepts of the "Five Freedoms" and developed and extended them into Welfare Quality Assessment Protocols. (Blokhuis et al., 2010). The five freedoms do not directly measure and provide welfare indicators and the EU Welfare Quality® research project found four principles useful in assessing welfare on farms: good feeding, good housing, good health and appropriate behaviour. These correspond to the following questions: Are the animals properly fed and supplied with water? Are the animals

properly housed? Are the animals healthy? Does the behaviour of the animals reflect optimized emotional states? (European Commission, 2014)

Table 3. The five freedoms and resources (Source: European Commission. 2014).

Outcomes	**Resources**
1. Freedom from hunger, thirst and malnutrition	By providing ready access to fresh water and a diet to maintain full health and vigour.
2. Freedom from discomfort	By providing an appropriate environment, including shelter and a comfortable resting area.
3. Freedom from pain, injury or disease	By prevention or rapid diagnosis and treatment.
4. Freedom to express normal behaviour	By providing sufficient space, proper facilities and company of the animals own kind.
5. Freedom from fear and distress	By ensuring conditions and treatment that avoid mental suffering.

In Africa, between 70% and 90% of the livestock are raised in vast, naturally grazing pastoral areas, and they are the main means of subsistence for the indigenous populace. The remaining 10% are produced using intensive and semi-intensive processes. Four criteria are used to categorize production systems in Africa were illustrated figure 4 (Masiga and Munyua, 2005).

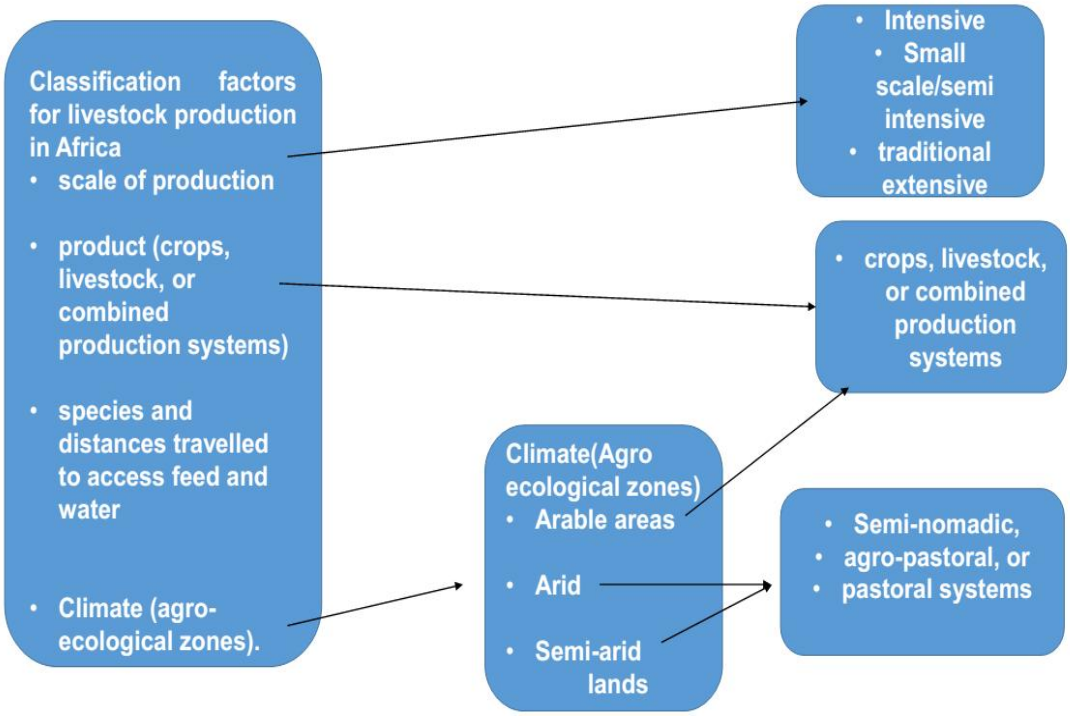

Figure 4: Classification of livestock production in Africa (Adopted, Source: Masiga and Munyua, 2005).

The traditional nomadic and transhumance pastoral systems, which are based on communal grazing and involve the seasonal movement of livestock between mountain and lowland pastures by herders, are well adapted to the fluctuations in annual and seasonal rainfall and vegetation. Animal welfare is significantly influenced by the systems used in livestock production and the societal attitudes of those who raise cattle (Masiga and Munyua, 2005). According to available information no formal protocol has been established to address the needs and welfare issues of tropical production systems (Hernandez et al., 2022). The currently available protocols have been developed for intensive, more or less industrial, systems in developed countries. However, the principles of Quality Welfare® can be used to identify animal welfare issues and risks in all systems (Leenstra, 2013).

When the welfare criteria from Welfare Quality® are addressed by the three production systems, it is not only the system itself that is important, but also many other characteristics such as the quantity and quality of feed, climatic conditions, attention and care of the animals, availability of veterinary care and medicines. All production system have a large variation in those features among themselves, but some general risks can be indicated table 4, a good review of the table 4 by Leenstra, (2013) shows the risk factors associated with different livestock production systems that exist in the world including Africa (Leenstra, 2013). Paul (2021) applied the welfare 'welfare quality® assessment protocol for dairy cows' among farms in Kiruhura District, Uganda and he concluded that not all measures are feasible for on-farm assessment among extensive dairy farms in Kiruhura, district, Uganda. The 12 criteria from the Welfare Quality® protocol can be used as a check list for animal welfare for specific policy options. The Welfare quality® assessment criteria and /or protocols could be a got start up point for the design and development of assessment criteria that suits the tropical livestock production system which is mostly prevalent in Africa including Ethiopia. The Welfare Quality® scheme could be also a good starting point for the evaluation of welfare of farm animals in Africa, and then modification could be done starting from the welfare Quality®.

Table 4. Welfare Quality® criteria and different livestock production system (Sources: Leenstra, 2013).

Welfare criterion	Pastoral systems	Mixed Systems	Industrial systems
1. Absence of prolonged hunger	High danger, frequently hunger and malnutrition	high risk, frequently malnourishment and hunger	Low risk, occasionally limiting feeding to enhance performance
2. Absence of prolonged thirst	Drinking water with a high risk of contamination, is not guaranteed, lack of availability due to drought or extreme heat	Risk and lack of assurance of drinking water quality, may not be available adequately	Low danger; occasionally refused access to drinking water; occasionally polluted water system
3. Comfort around resting	Risk, lack of a specific accommodation, and flexibility in lying	Risk, frequent lack of particular accommodations, and occasionally having the choice of where to lie	Risk, influenced by house design (no bedding, confinement)(some housing system do not comfortable flooring)
4. Thermal comfort	Hypothermia and hyperthermia risks; some behavioral control, pastoral areas are arid	Risk, both for hyper- and hypothermia; some behavioural control, no proper shelter	Limited risk/manageable, dependent on housing conditions; limited behavioural control
5. Ease of movement	No risk, however being able to walk is necessary	Risk, varies with confinement and tethering	Risk, depending on housing design, space allowance
6. Absence of injuries	Injuries might be present (mutilations due to cultural practice, predator attack)	Injuries might be present (mutilations due to cultural practice, predator attack)	Dependent on system (production related injuries may present ex. Hoof lesions (lameness)

7. Absence of disease	Huge burden of infections and/or malnutrition	Huge burden of infections and/or malnutrition	Dependent on management (totally cannot be guaranteed, ex metabolic disorders
8. Absence of pain induced by management procedures	Not guaranteed (mutilations and abuse due to traditional practice)	Not guaranteed (mutilations and abuse traditional practice)	Not guaranteed (painful husbandry procedures without proper pain control mechanism)
9. Expression of social behaviours	Low risk, mostly spend on fields	Limited to high risk, limited due to low land availability	At risk (confinement and not have access to pastor and fields most of the time)
10. Expression of other behaviours	Low risk, livestock stay in open fields	Limited to high risk, may be limited due to confinement and land shortage	At risk (confinement and not have access to pastor and fields most of the time)
11. Good human-animal relationship	Limited and variable relationship, sometimes aversive	Close interaction, sometimes aversive (scavenging animals)	At risk (lack of good stockman ship)
12. Positive emotional state	Close to wild animals	Whole range possible, from wild to very calm, and approachable	At risk, abnormal behaviour, due to inability to express natural behaviour due to confinement, limited space allowance

2.4. Findings

In these section, the writer compares the articles and the annexes of the EU farm animal directives, which articles and annexes could be applied, what can be transcribed from the European Union (EU) farm animal directive (Council Directive 98/58/EC) to the African context specifically for Ethiopia. As we previously discussed in the other sections as methodology in the review the predominant type of livestock production is extensive system comprising of mixed livestock crop production system and pastoralist system.

EU legislation on animal welfare reflects the 'five freedoms' as adopted by the Farm Animal Welfare Council (FAWC) which are listed in table 3. (European Commission, 2014). So in the writer view in the development of any farm animal welfare regulations/directive/laws/ proclamation should be based on the five principles of the Farm Animal Welfare Council (FAWC). The five principles of farm animal welfare council (FAWC) are fundamental, so African countries like Ethiopia, should base their animal legislation/proclamation on the five principles, and local research on the identification of the farm animal welfare problems should be promoted, so that to combine the local situation with the international conditions in developing farm animal welfare legislation/proclamation.

By considering Council Directive 98/58/EC of 20 July 1998 concerning the protection of animals kept for farming purposes, the writer analyzed the articles and annexes that are present in the directives, based on the literatures that are already reviewed and discussed on the pervious sections of these chapter. A table was constructed for this propose that contains the summarized form of the annexes of the directives (European Commission, 2014) and modified so that to indicate which annexes are really can be incorporated/proposed when developing an animal welfare legislation/regulation/proclamation specially focusing on production animals in countries like Ethiopia.

Regarding the articles Article 3 is one of the fundamental provision, of the Directive 98/58/EC it states …. "…that the owners or keepers take all reasonable steps to ensure the welfare of animals under their care and to ensure that those animals are not caused any unnecessary pain, suffering or

injury…….” is one of the main article and could be also used, in the development of animal welfare regulations/ laws/directives and policy of countries like Ethiopia, with appropriate use. Since as we said millions of people livelihoods depend on livestock, so that the owner of the cattle should be the soul responsible to keep his animals from unnecessary suffering, or injury.

Table 5. Major categories and features of Annex 1 of Directive 98/58/EC, and their applicability to the extensive livestock production /system, mixed livestock and pastoral system. (Source: Adopted and modified from; European Commission 2014. Executive Agency for Health and Consumers, (2014). Better training for safer food: animal welfare, Publications Office (https://data.europa.eu/doi/10.2818/16713) and Council Directive 98/58/EC was attached as annexes 1 in these paper).

Annexes	**Major features**	**Mixed crop livestock, and pastoral/agro-pastoral (extensive system), and the applicability of the annexes, to those production systems.**
Staffing	• Animals shall be cared for by a sufficient number of staff who possess the appropriate ability, knowledge and professional competence.	• Cannot be used as legislation straight away. Since farming is almost u a way of life for pastoralists and farmers, stakeholders should encourage appropriate farming practices and the development of farmers' traditional knowledge.
Inspection	• All animals kept in husbandry systems in which their welfare depends on frequent human attention shall be inspected at least once a day. Animals in other systems shall be inspected at intervals sufficient to avoid any suffering. • Adequate lighting (fixed or portable) shall be available to enable the animals to be thoroughly inspected at any time. • Any animal which appears to be ill or injured must be cared for appropriately without delay and, where an animal does not respond to	• Can be taken, with modification especially the owners should make sure that the diseased animals should get appropriate care, treatment, vaccination, deworming etc.

	such care, veterinary advice must be obtained as soon as possible. Where necessary sick or injured animals shall be isolated in suitable accommodation with, where appropriate, dry comfortable bedding	
Record keeping	• The owner or keeper of the animals shall maintain a record of any medicinal treatment given and of the number of mortalities found to each inspection. Where equivalent information is required to be kept for other purposes, this shall also suffice for the purposes of this Directive. • These records shall be retained for a period of at least three years and shall be made available to the competent authority when carrying out an inspection or when otherwise requested.	• Difficult to apply as legislation, since most farmers and pastoralists are illiterate and had no resources.
Freedom of movement	• The freedom of movement of an animal, having regard to its species and in accordance with established experience and scientific knowledge, must not be restricted in such a way as to cause it unnecessary suffering or injury. • Where an animal is continuously or regularly tethered or confined, it must be given the space appropriate to its physiological and ethological needs in accordance with established experience and scientific knowledge.	• Animals in these system has less risk of being avoided their freedom of movement, but it could be also taken as a point, in case of pastoral system animals may walk a long distance with herders in search of feed, pasture.
Buildings And accommodation	• Materials to be used for the construction of accommodation, and in particular for the construction of pens an equipment with which the animals may come into contact, must not be harmful to the animals and must be capable of being thoroughly cleaned and disinfected.	• Cannot be put directly, but these could be customize so that that farmers and pastoralist should be obligated to keep protect their animals from extreme weather and predators,

	• Accommodation and fittings for securing animals shall be constructed and maintained so that there are no sharp edges or protrusions likely to cause injury to the animals. • Air circulation, dust levels, temperature, relative air humidity and gas concentrations must be kept within limits which are not harmful to the animals. • Animals kept in buildings must not be kept either in permanent darkness or without an appropriate period of rest from artificial lighting. Where the natural light available is insufficient to meet the physiological and ethological needs of the animals, appropriate artificial lighting must be provided.	and also they should also be responsible to protect the animals from lesions, trauma, damage that could be caused by the materials in which the accommodation/barns/shelters are constructed.
Animals not kept in buildings	• Animals not kept in buildings shall where necessary and possible be given protection from adverse weather conditions, predators and risks to their health.	• Can be considered greatly and could be applicable, so that it should be the responsibility of farmers and pastoralists to protect their animals from extreme weather and predators
Automatic or mechanical equipment	• All automated or mechanical equipment essential for the health and well-being of the animals must be inspected at least once daily. Where defects are discovered, these must be rectified immediately, or if this is impossible, appropriate steps must be taken to safeguard the health and well-being of the animals.	• In those systems automatic machines are not available almost.

	• Where the health and well-being of the animals is dependent on an artificial ventilation system, provision must be made for an appropriate backup system to guarantee sufficient air renewal to preserve the health and well-being of the animals in the event of failure of the system, and an alarm system must be provided to give warning of breakdown. The alarm system must be tested regularly.	
Feed, water and other substances	• Animals must be fed a wholesome diet which is appropriate to their age and species and which is fed to them in sufficient quantity to maintain them in good health and satisfy their nutritional needs. No animal shall be provided with food or liquid in a manner, nor shall such food or liquid contain any substance, which may cause unnecessary suffering or injury. • All animals must have access to feed at intervals appropriate to their physiological needs. • All animals must have access to a suitable water supply or be able to satisfy their fluid intake needs by other means. • Feeding and watering equipment must be designed, constructed and placed so that contamination of food and water and the harmful effects of competition between the animals are minimised. • No other substance, except those for therapeutic or prophylactic zoo-technical purposes, may be fed unless scientific studies or experience shows that it is not detrimental to health.	• Applicable with modification, Feed and water, should be provided to the animals as much as possible clean water and adequate feed. This article can be modified to these production systems. Since in these systems due to man-made and natural disaster like drought is a risk and animals can suffer from lack of feeds and water., and since they are traditional systems feeding may not be scientific so the provision of enough and clean water and feed should be the priority,

Mutilations	• As laid down by national laws or other EU rules.	• Can be directly applicable, since there are many traditional and cultural practices that could be against the animal welfare, Ex. like branding with hot iron, traditional castration, skin cutting as a treatment etc.
Breeding procedures	• Cannot practice natural or artificial breeding procedures that cause or are likely to cause suffering or injury. Exceptions to this are allowed if the procedure only causes minimal or momentary suffering or injury. • No animal shall be kept for farming purposes unless it can be expected on the basis of its genotype or phenotype to be kept without detrimental effects to its health and welfare.	• These may not be applicable, as legislation but stakeholders should be responsible about breeding practices, and also give the necessary assistance, to improve breeding of the local farmers and pastoralists. • As it is already very understood that almost all the breeding procedures are natural in these systems and farmers and pastoralists could be advised and awareness creation could be there so that they better understand the need of breeding, and making the breeding suitable with welfare of their animal.

It can be observed in the table 5 above, almost all the annexes are designed for intensive/industrial production livestock system, and the annexes' need a major modification so that feet them to the existing conditions of African countries like Ethiopia , the writer considers the some of the annexes and articles could be a good starting point and below suggestion of modification by the writer is provided or proposed so that could be applicable to the majority of the production systems that is found in the Ethiopian context, which are the mixed livestock/crop, pastoral and agro pastoral production system. So below are the articles and annexes that are modified and could be used in the legislation/proclamation/directives development in the writers view:-

- "……that the owners or keepers take all reasonable steps to ensure the welfare of animals under their care and to ensure that those animals are not caused any unnecessary pain, suffering or injury" Article 3, Since as we said millions of people livelihoods depend on livestock, so that the owner of the cattle should be the soul responsible to keep his animals from unnecessary suffering, or injury, so that this article could be adopted.
- Inspection, the owner of animal should be responsible to keep their animals safe, and owners should be responsible to make sure that the animal should get appropriate treatment whenever necessary, or whenever the animal gets injured or diseased, and its responsibility of the farmer to care diseased animals.
- Freedom of movement, farmers and pastoralists should be responsible so that there animals could get enough freedom of movement, even though in the mixed livestock crop/ production system, there may not be risk almost. The other side of the freedom of movement could be that animals in those system freely move, and the herders trek the animals for longer distance so that, during these movement a necessary care for the animals should be provided.
- Animals accommodation/housing and/or animal that are not kept in a house/shelters/open pens, so the farmers and pastoralists should be responsible to protect their animals from extreme weather and predators
- Feed and waters, the farmers and the pastoralists should be responsible to provide their animals the necessary feed and clean water.
- Mutilations, farmers and pastoralists should be prohibited from doing harmful procedures to their animals, since there are many harmful practices culturally present in the pastoralists and farmers community.

2.5. Discussions

There is a global movement underway to improve animal welfare standards. Countries in Africa, Asia, Latin America and the Middle East, which previously provided little or no legislative protection for farm animals, are developing general anti-cruelty/animal welfare legislation and some specific regulations (IFC, 2014). Most of the annexes and articles that are provided in the farm welfare legislation of EU (Council Directive 98/58/EC) cannot be completely and fully incorporated and taken as it is, since there is a major difference in the farm animal production system, especially on those of livestock crop mixed production, pastoralist, agro pastoralist production system. It is very clear and obvious the status of animal welfare keeping is variable between developed and developing world. In general attention for animal welfare increases with increasing wellbeing of the human population and with economic growth (Leenstra, 2013).

Animal welfare challenges in Africa seems to be prominent in the small-scale and pastoralist farming systems and rural areas where access to resources as well lack of knowledge exists. In many African countries, infrastructure, economic, cultural, and political factors, as well as access to veterinary services have a significant impact on animals and their owners. There is a need for raising awareness (Qekwana et al., 2019). Sinclair et al., (2019) in their studies in benefits of improving animal welfare from the perspective of livestock stakeholders across Asia, concluded that improving animal welfare for the sake of the animals is unlikely to be a compelling argument, so in the development of animal welfare regulations this realities should be considered. So that the development of animal welfare regulations, directives and /or proclamations attention should be given in these regard. Community conversations are also an effective way to feedback community voices into planning to build a bottom-up implementation of animal welfare programs (Lemma et al., 2022). Qekwana et al., (2019) stated that countries such as South Africa and Zimbabwe have made significant progress in educating their citizen on animal welfare; however, more still needs to be done to make practitioners and communities aware of animal welfare problems that may arise on farm. In Zambia Njei and Lubungu (2022) in their study in small holder farms, they concluded that more efforts and extension services should also be dedicated to addressing farm animal welfare concerns; furthermore, the policy framework guiding animal welfare is inadequate as particular

welfare concerns such as housing and nutrition are not well covered. This calls for the revision of the policy framework to incorporate all aspects of animal welfare.

In mixed crop livestock system, in pastoral and agro pastoral system the lack of feed/ lack of sufficient feed for animals, the presence of high disease burden or low quality of infrastructure of animal health are some of the factors that negatively affect the welfare of farm animals. At low level of animal production, people also suffer from lack of food, nutrition, and health services (Leenstra, 2013). In one of the community participation study in Ethiopia, the community members stated that the welfare of their animals is affected during drought due to a shortage of feed and water (Lemma et al., 2022). Lemma et al., 2022 in their studies stated how the community members describe animal welfare through community conversations methods……describing animal welfare, community members commonly associated feeding and health with the welfare of animals. They readily identified the biological needs of animals such as health, clean shelters, clean water, and sufficient feed. However, it was not obvious for them to identify the affective state and natural behavior of animals. These components of animal welfare did not come to their mind at first. It was through follow-up probing questions that they started to recognize these components of animal welfare. Alemayehu et al., (2022) in their investigation about 'Animal welfare knowledge, attitudes, and practices among livestock holders in Ethiopia, stated that there is a difference between animal welfare understandings between the mixed crop livestock farmers and pastoralists. They concluded that generally, households practicing mixed crop-livestock farming system had better animal welfare knowledge, attitude, and practice than pastoralist. Mixed crop-livestock farmers had better knowledge on items related to observing the nutrition condition of the animal, animal-human relationship, the importance of water, and health inspection compared to pastoralists. In contrast, pastoralists had better knowledge of items related to natural behavior expression, animal care, and animal suffering than mixed crop-livestock farmers.

African Union Inter-African Bureau for Animal Resources (AU–IBAR) in 2017 when they prepare animal welfare strategy for Africa, they identified seven priority areas that the strategy should focus, some of the strategies that were included are: Training, education and awareness, Policy and Legislation, Research (AU–IBAR 2017). So that to improve the farm animal welfare situation in Africa, in Ethiopia a focus should be given in the creation of awareness of the community

through community conversation methods (Lemma et al., 2022). To attain or improve farm animal welfare in countries like Ethiopia, Africa in general the involvement of all the stakeholders is detrimental, from all population that are involved in the livestock production, from decision makers, governmental officials, and intergovernmental organizations, more over the veterinary professionals of third world countries who are responsible for the provision of service to the community need to understand and update themselves of the welfare science that exist currently in the world (Doyle et al., 2021).

The animal welfare assessment tools that exist in the current days are suitable to evaluate and assesse the welfare conditions of farm animals that are under intensive management system that exist in modern and develop countries, but these assessment tools can be a starting point so that to modify so that it could fit the extensive production system that predominantly dominate the developing countries so as to evaluate and improve the farm animal welfare. Animal welfare needs to be assessed in order to understand and improve it, and the five domain concept provides a framework for this assessment. When trying to understand animal welfare, our assessments should focus on animal-based measures, as they directly identify how well an animal is doing in its environment. Adding the management and resource assessment to the animal-based measures within the five domains identifies overall where actions need to be taken to correct or mitigate welfare problems (Doyle et al., 2021).

2.6. Conclusions

Attention to the farm animal welfare is increasing in the world, and also in the developing countries. The animal welfare problems in African countries like Ethiopia is complex, deep and may not be given attention like the rest of the world due to factors like poverty, luck of knowledge, resource. Livestock is a source of livelihood for most African, including Ethiopian farmers and pastoralists, and due attention should be given to farm animal welfare, so that to improve productivity. There is a difference in the understanding of farm animal welfare among the mixed crop livestock farmers and pastoralists, so creation of awareness, improving their understanding of animal welfare through community engagement conversation, outreach is necessary. So that in the development process of animal welfare legislation, attention to awareness creation to the

different livestock producers (mixed crop livestock producers, pastoralists) should be given. The directives that is developed for highly developed countries in European Union (EU), the "farm animal directive'' may not be totally fit the situation on the ground in countries like Ethiopia which still have traditional system of livestock production, but it could be a starting point, so as after successful identification of the farm animal welfare critical points may help as a starting point with modification to the local situation in the ground.

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CHAPTER III: WELFARE AND HEALTH ASSESSMENT OF BEEF CATTLE DURING THE ADAPTATION PERIOD IN A SPECIALIZED COMMERCIAL FATTENING UNIT

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Health and welfare assessment of beef cattle during the adaptation period in a specialized commercial fattening unit

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ABSTRACT

Beef cattle welfare and health status are influenced by housing and management systems. The present study aimed to assess the welfare and health status in the first 15 days after arrival of Limousine bulls imported from France and fattened in a commercial fattening unit in Italy. A total of 264 bulls were included in the study. Welfare, biosecurity, and major hazard and warning system were assessed on days 2 (T1) and 15 (T2) after arrival to the unit. At T1 and T2 an inspective clinical examination was performed on all bulls. At T1 and T2 blood samples were collected from 88 bulls for haematological analysis. Both at T1 and T2, the welfare, biosecurity, and major hazards and warning systems were classified with a general score of medium but with a decrease on animal-based measurements in T2. At T1 and T2 the clinical examination revealed a significant increase (p-value$\leq$0.05) of skin lesions and lameness in T2. A high incidence of respiratory disease was noticed in both assessed times. Leucocytes and all differentials count, and platelets were significantly increased (p-value$\leq$0.05) at T2, while the fibrinogen was significantly decreased. The haematological changes suggest that the bulls were under higher stress in T2 when compared with T1 linked with a difficult adaptation response to the fattening unit. A multi-factorial approach that integrates the indicators of the checklist and the clinical and haematological findings of animals can be a useful method to deepen the assessment of welfare in beef cattle.

1. Introduction

Animal welfare is the physical and mental state of an animal in relation to the conditions in which it lives (OIE, 2022). Consumers anticipate that their animal-related products, notably food, should be produced with consideration for the animal welfare (Welfare Quality®, 2009). There are various definitions of what constitutes animal welfare, but there is a growing consensus that farm animal welfare has to be safeguarded and enhanced. Recently, compared to other quality traits, there has been an improved recognition of animal welfare criteria. Consumers frequently believe that items that have in consideration animal welfare are more genuine, safer, tastier, and hygienic (Alonso et al., 2020). Scientific data supports the relationship between animal welfare and animal health and, thus, food safety with a strong correlation between excellent animal welfare and good animal health (de Passillé and Rushen, 2005). Housing, transportation, and management practices have a significant impact on the welfare of different species (Broom, 2009; Nannoni et al., 2022; Raspa et al., 2022; Sardi et al., 2020).

Health, comfort, and the expression of species-specific behaviours are indicators related to welfare (Botreau et al., 2007). Therefore, determining welfare requires a multidisciplinary approach through meticulous and trustworthy monitoring of the indicators related to productivity, ethology, endocrine function, immunology, and pathology (Sevi, 2009). In Italy, the farm animal welfare assessment is based on a protocol included in the ClassyFarm system. This protocol aims to support official controls, collect information, advance welfare-level implementation, and inform consumers (Mariottini et al., 2022). Routine application of the protocol offers a promising tool for the improvement of beef cattle welfare and farm profitability, in particular when a welfare certification becomes available (Gottardo et al., 2009). To perform welfare certification, accurate, reliable, and repeatable measures of welfare factors that allow quantification of welfare should be considered (Salvin et al., 2020).

When fattened in intensive systems, beef cattle are more susceptible

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to experiencing poor welfare. The main welfare issues in the beef industry are: bovine respiratory disease (BRD) linked to overcrowding, insufficient ventilation, and animal mixing; digestive problems associated with intensive concentrate feeding; and behavioural issues due to overcrowding and co-mingling (Cozzi et al., 2009; EFSA, 2012). Enhancing farming practices and management that ensure animal health is a step further to improving welfare (Alonso et al., 2020).

In the present study, we aimed to assess the welfare and health status of bulls imported from France and fattened in Italy in a commercial fattening unit in the first 15 days after arrival by applying a multidisciplinary approach.

2. Materials and methods

2.1. Commercial fattening unit and bulls

The study was conducted in a commercial fattening unit in the province of Modena (Italy). All animals fattened in the unit were Limousine bulls imported from France. They came from several French farms distributed over the country, where most of the bulls were kept on pasture or in an indoor free stall system with straw bedding. Before arriving at the unit in Italy, the bulls were kept in transit for one day in a selection centre in France. Here, they were selected based on health status, age, and body weight, to obtain homogenous batches. At the entrance to the fattening unit, the bulls were 11 months old and weighed 380 kg. The bulls arrived once a week at the unit in Italy in numerically heterogeneous batches and were transferred in groups of six to their designated pens. Before the batch's arrival, their assigned pens were pressure washed and disinfected with sodium-P-toluen-N-chlorosulfamide. The dropping pit was emptied every 2 to 3 months.

The unit consisted of four similar barns separated by a 20-m corridor (Fig. 1A). The barns were semi-closed and well-ventilated. Each barn had 44 pens in a free-stall system with a maximum capacity of 6 bulls per pen. The pens were placed 22 × 22 in parallel with the feeder on one side. Each pen had a dimension of 18.4m^2. Each animal had a space of 3.06 m^2 and a feeding front of 45 cm. The feeders were placed on one side along the feeding line. The pens were built adjacent to each other and were separated by iron bars, allowing interaction of animals (Fig. 1B, C). The flooring was slatted and underneath there was a pit for manure collection. Each barn was equipped with 46 automatic water bowls serving 264 animals and at least one automatic water bowl was present per pen. Each barn had a maximum housing capacity of 264 bulls. Therefore, the farm's total housing capacity was 1056 bulls. As each production cycle lasts between 5 and 6 months and the facility allowed the fattening of 2112 animals per year.

The current study had in consideration one barn housing 264 bulls. Bulls arrived in six batches weekly-based over the course of 6 weeks. At arrival, bulls were vaccinated using the live attenuated virus of bovine viral diarrhoea-mucosal disease (Rispoval D-Bvd®) and the live bovine herpesvirus type 1 vaccine (Bovilis IBR®). Ivermectin (Ivomec®, Boehringer Ingelheim Animal Health, Italy) was administrated for the prevention and control of parasites.

2.2. Dietary adaptation

At arrival, the bulls were fed an adaptation diet in order to reduce dietary stressors. The total mixed ration (TMR) was fed ad libitum and fresh clean water was always available. The TMR diet was freshly sampled in different locations (beginning, middle, and end of the feeding line) at T1 and T2 after arrival at the fattening unit. TMR ingredients and proportions are reported in Table 1. Analytical TMR analyses were performed at the University of Bologna feed analysis lab according to the methodology described in previous studies (Mammi et al., 2022).

2.3. Welfare, Biosecurity and Major Hazard and Warning System Assessment

For the welfare, biosecurity and, major hazard and warning system two observations were performed: at T1 and T2. An adapted version of the Italian protocol for the assessment of beef cattle welfare included in the ClassyFarm system (Bertocchi et al., 2020) was applied. The used protocol included a list of 58 items, divided into three main sections: biosecurity (items 1 to 13), welfare (items 14 to 50), and major hazard and warning system (items 51 to 58) (Supplementary TableS1 S1). The welfare section was further subdivided into three areas: A-farm management and staff training (items 14 to 28), B-housing and equipment (items 29 to 40), and C-animal-based indicators (items 41 to 50). For each item, a 2- or 3-point scale scoring system was applied (1 = insufficient; 2 = acceptable; 3 = optimal) (Mariottini et al., 2022). A value for each section was computed by summing the obtained score of each item from each section or area. For welfare, the value was calculated accounting for a contribution of 50% by areas A and B, and 50% by C. The obtained values were further converted into percentages. In particular, a result below 59% indicated a poor status (=low), a result between 60 and 80% a medium status (=medium), and a result over 80% a good status (=high) (Diana et al., 2020).

2.4. Clinical examination

All bulls underwent a clinical examination pen-based at T1 and T2. It consisted of a 10-min observation with the observer standing between the animals in the pen. The following parameters were assessed: mental status, cleanliness, body condition, skin lesions, gait, nasal discharge, ocular discharge, faecal consistency and other possible abnormalities. All data were recorded in a schematic table per pen (Supplementary Table S2). An animal was considered to have BRD if at least two abnormal findings related to the respiratory system were present (i.e., cough and nasal discharge; abnormal breathing and cough; abnormal breathing and nasal discharge).

2.5. Blood investigation

At T1 and T2, blood samples were collected from 88 out of the 264 bulls. At T1, two bulls were randomly selected from each pen, and at T2, the same subjects were re-sampled. Blood samples were collected via coccygeal/jugular venepuncture for haematological investigations. The blood was transferred into vacuum tubes containing EDTA anticoagulant for complete blood count and into citrate tube for fibrinogen analysis. The following set of blood parameters were analysed: erythrocytes (RBC), hemoglobin, hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), erythrocyte distribution width (RDW), platelets (PLT), leucocytes (WBC), neutrophils, monocytes, lymphocytes, eosinophils, basophils and fibrinogen.

2.6. Statistical analysis

Data were entered into a statistics program (JMP Pro 17). Descriptive statistics were generated mean ± standard deviation (S.D.) and/or standard error (S.E.), median and range for continuous data, and count and percentage for categorical data. For continuous variables, normality was tested by Shapiro-Wilk test and non-normally distributed variables were Box-Cox transformed before the analysis. The evaluation of differences between T1 and T2 were undertaken using the Mixed Model Procedure. Each cattle were set as an experimental unit within the arrival group and pen as nested factors. The adaptation time (T1 and T2) was implemented as fixed effect. After the analysis, normal distribution of the data was checked again for the resulting residuals. Means are reported as least square mean and pairwise multiple comparisons were performed using Tukey-test as post hoc test when a significance was

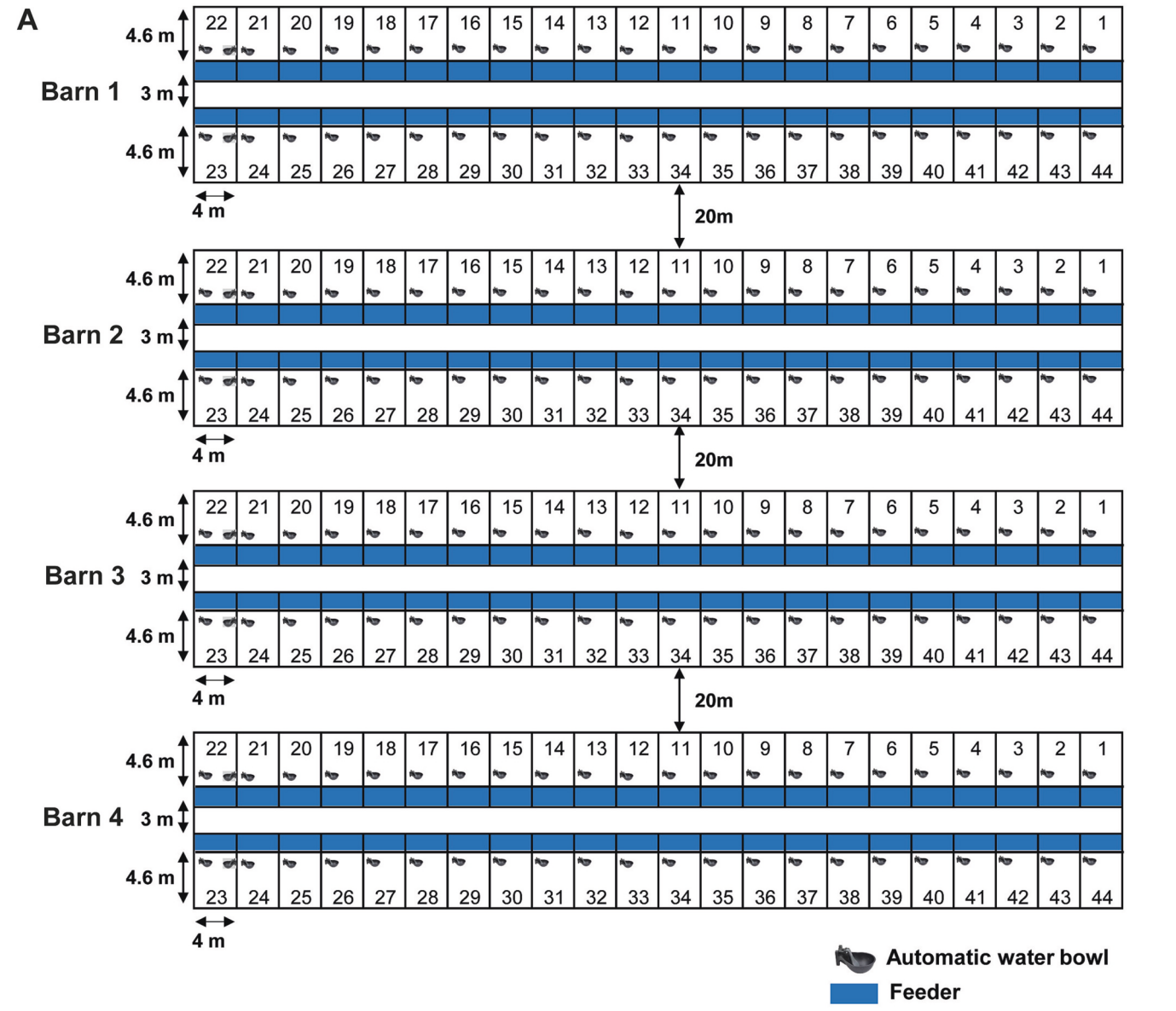

Fig. 1. Beef intensive commercial fattening unit. A, Schematic representation of the unit. B, Image of the barn where the study was performed. C, Image of one pen.

Table 1
Descriptive statistics of the adaptation TMR diet (T1 and T2) and chemical analysis.

TMR	Feed, kg af
Wheat silage	3.5
Meadow hay[a]	1.2
Wheat straw	1.1
Beat pulp	1.3
Corn, finely ground[b]	1.1
Soybean meal	0.5
Cane molasses[c]	0.5
Min and Vit Premix	0.3
	Nutrients, %DM
DM	70.07
UFC	0.81
CP[d]	11.25
Ash	8.78
EE[e]	2.06
Starch	13.57
Sugars	7.37
NDF[f]	39.21
ADF[g]	25.79
ADL[h]	3.45

Abbreviations: [a]the quality of the hay was checked to ensure the absence of molds and spores (Cavallini et al., 2022a, 2022b). [b]the corn was below the EU maxim tolerable level (Girolami et al., 2022). [c] molasses were properly characterized (Palmonari et al., 2021). [d] Crude protein. [e]ether extract. [f]neutral detergent fiber. [g]acid detergent fiber. [h]acid detergent lignin.

detected. Then a nominal logistic model was used for categorical variables using the same discriminant as before mentioned. A p-value≤0.10 was considered a tendency; a p-value≤0.05 was considered statistically significant; and a p-value≤0.01 was considered highly significant.

3. Results

3.1. Welfare, biosecurity and major hazards and warning system assessment

Results of welfare, biosecurity major hazards and warning system assessment at T1 and T2 are presented in Table 2. At T1, the percentages obtained for welfare, biosecurity, and major hazards and warning systems were 79.04%, 63.88%, and 76.47%, respectively. Regarding the welfare at T1, the following results were obtained: 70.45% in area A, 65.17% in area B and 90% in area C. At T2, the percentages obtained for welfare, biosecurity major hazards and warning system were 74.73%, 63.88% and 76.47%, respectively. At T2, although there was an increase in area B (68.57%), a decrease in welfare compared to T1 due to a decrease in score in area C (80%) was noticed. No differences were found between T1 and T2 in terms of biosecurity and major hazards and warning system.

Table 2
Descriptive statistics of welfare, biosecurity and major hazard and warning system of 264 Limousine bulls.

Item	Assessment at T1	Classification at T1	Assessment at T2	Classification at T2
Total welfare	79.04%	Medium	74.73%	Medium
Area A (Farm management and staff training)	70.45%	Medium	70.45%	Medium
Area B (Housing and facilities)	65.17%	Medium	68.57%	Medium
Area C (Animal-based indicators)	90%	High	80%	Medium
Biosecurity	63.88%	Medium	63.88%	Medium
Major hazard and warning system	76.47%	Medium	76.47%	Medium

Abbreviations: T1 = 2 days after arrival to the unit; T2 = 15 days after arrival to the unit.

3.2. Clinical examination

The clinical data is provided in Table 3. At T1, 1.51% of the bulls showed integument lesions, 0.75% lameness, 0.75% diarrhoea, 27.65% signs of BRD. At T2, there was a significant increase in lameness (1.15%, p-value = 0.02) and in integument lesions (44.69%, p-value≤0.01). Most of these were alopecic lesions in the neck. In contrast, no significant changes in the percentage of animals with signs of BRD (31.81%) and diarrhoea (0%) were noticed.

3.3. Blood parameter analysis

Results of the blood analysis at T1 and T2 are presented in Table 4. A significant increase (p-value≤0.05) in platelets, WBC, neutrophils, monocytes, lymphocytes, eosinophil, and basophils was noticed at T2. They were within the normal reference range for bovine species with the exception of monocytes that were higher than the normal range. The fibrinogen values obtained both in T1 and T2 were above the reference range. However, a significant decrease (p-value≤0.05) of fibrinogen was noticed in T2. Indeed, there was an absence of statistically significant difference (p-value>0.05) in the RBC, HGB, HCT, MCV, MCH, MCHC, RDW and N/L between T1 and T2.

4. Discussion

Currently, there are different methods that enable to assess and measure beef cattle welfare (Kirchner et al., 2014a; Mariottini et al., 2022). Welfare assessment methods on farm should be implemented in a consistent modality. With this approach, the results of the assessment are expected to be representative of a longer-term farm welfare status considering that the management practices and housing conditions have not changed. Furthermore, welfare assessment methods must be reasonably free from observer influence (Kirchner et al., 2014b). Firstly, we assessed the welfare using checklist protocol approach. Secondly, we evaluated the clinical and haematological conditions of animals. The methods were applied at T1 and T2 in order to achieve consistency over a critical time. We evaluated the biosecurity and major hazard and warning system obtaining a classification of medium, without significant differences between the T1 and T2. Indeed, a decrease in welfare between T1 and T2 was noticed due to a reduction of animal-based indicators score. The observed welfare decrease could be associated with stress responses to both physical (i.e., transportation, new environment, new feed) and psychological (i.e. social-group mixing) stressors (Bassel and Caswell, 2018).

The clinical examination of animals evidenced a significant increase in integument lesions, which could have contributed to the lowering of welfare score. Crowding, inadequate feed distribution, inadequate space at the manger, mixing social group and poor pen flooring are all detrimental to the welfare of beef cattle, which in turn cause competition and

Table 3
Clinical investigation findings of the 264 bulls at day 2 (T1) and day 15 (T2) after arrival to the farm.

Item	Assessment at T1	Assessment at T2	P-value
Integument lesions (%)	4(1.51%)	118(44.69%)	<0.01
Lameness (%)	2(0.75%)	4(1.15%)	0.02
Diarrhoea (%)	2(0.75%)	0(0%)	0.41
Respiratory disease (%)	73(27.65%)	84(31.81%)	0.54

Table 4
Result of the complete blood analysis at day 2 (T1) and day 15 (T2) after arrival to the farm.

Blood Parameters		Time of assessment			
		T1	T2	*P* value	Reference Range
RBC (M/μL)	Mean ± SD	9.8 ± 1.37	9.88 ± 1.11	0.83	5.1–7.6[a]
HGB (g/dL)	Mean ± SD	12.09 ± 1.25	12.01 ± 1.16	0.62	8.5–12.2[a]
HCT (%)	Mean ± SD	39.55 ± 4.36	39.29 ± 3.96	0.63	22-33[a]
MCV (fL)	Mean ± SD	40.5 ± 2.99	39.9 ± 2.63	0.08	38-40[a]
MCH (pg)	Median [Min.- Max]	12.2 [11.6–13.1]	12.1 [11.6–12.8]	0.05	14-18[a]
MCHC (g/dL)	Mean ± SD	30.6 ± 1.23	30.57 ± 1.28	0.9	34-38[b]
RDW (%)	Mean ± SD	24.11 ± 1.96	24.05 ± 1.68	0.52	15.5–19.4[a]
PLT (K/μL)	Median [Min.- Max]	280 [143–338]	315 [148–510]	<0.01	193-637[a]
WBC (K/μL)	Median [Min.- Max]	8.69 [7.31–9.97]	10.34 [8.53–12.95]	<0.01	4.9-12a
NEU (K/μL)	Median [Min.- Max]	3.37 [2.65–3.97]	3.91 [2.87–5.91]	<0.01	1.8–6.3[a]
MONO (K/μL)	Median [Min.- Max]	1.25 [0.98–1.47]	1.13 [0.88–1.39]	0.04	0–0.6[a]
LYM (K/μL)	Mean ± SD	3.84 ± 1.26	4.46 ± 1.79	<0.01	1.6–5.6[a]
EOS (K/μL)	Median [Min.- Max]	0.08 [0.03–0.2]	0.22 [0.09–0.41]	<0.01	0–0.9[a]
BASO (K/μL)	Median [Min.- Max]	0.06 [0.04–0.07]	0.09 [0.06–0.12]	<0.01	0–0.3[a]
FIBR (mg/dL)	Median [Min.- Max]	985.05 [738.15–1348.2]	778.2 [584.18–1054.35]	<0.01	100-600[b]
N/L ratio	Median [Min.- Max]	0.987 [0.62–1.27]	0.95 [0.64–1.46]	0.42	0.4–2.34[a]

Abbreviations: RBC, Red blood cell; HGB, Hemoglobin; HTC; Hematocrit; MCV, Mean corpuscular volume; MCH, Mean corpuscular hemoglobin; MCHC, Mean corpuscular hemoglobin concentration; RDW, Red blood cell distribution width; PLT, Platelets; NEU, Neutrophils; WBC, white blood cells; MONO, Monocytes; LYM, Lymphocytes; EOS, Eosinophils; BASO, Basophils; FIBR, Fibrinogen; N/L ratio, Neutrophils: Lymphocytes ratio; M/μL,10^6 per microliter; %, percentage; K/μL,10^3 per microliter; g/dL, grams per deciliter; fL, femtoliter; pg, picogram; mg/dL, milligram per deciliter; Min., Minimum; Max., Maximum, [a]George et al. (2010), [b]Cornell University College of Veterinary Medicine, 2023.

stress among pen mates and result in lesions (Cozzi et al., 2009). The clinical investigation revealed that most of the integument lesions developed between T1 and T2 were characterized by alopecia in the neck dorsal region. The development of these lesions, caused by the repeated rubbing of the animals' necks against the iron bars of the feeding structure, suggests the inadequacy of the structure, evidencing a critical point that could be addressed. Furthermore, a significant increase of lameness was noticed contributing to a decrease in welfare. Lameness can have several causes, such as social competitions, poor hygienic level and hoof care, inadequate housing facilities and flooring (e.g, unsuitable grating), and unbalanced feeding (Bertocchi et al., 2020; Nalon and Stevenson, 2019). In addition, animals selected for rapid weight gain, in conditions like those in intensive fattening units, are predisposed to develop metabolic and joint diseases (Compiani et al., 2014). Even though, an adaptation diet was provided, there is a possibility that the bulls received higher amounts of concentrates when compared to the diet provided in France. This could have increase the risk of developing ruminal acidosis that can lead to the development of laminitis and consequenty lameness (Cozzi et al., 2009; Compiani et al., 2014).

In beef cattle BRD, is one of the major health and welfare issues that negatively impacts productivity. A large spectrum of stressors, in particular transportation, contribute to higher disease susceptibility, such as BRD (Chen et al., 2015). BRD, is frequently developed in the first weeks after arrival on the farm (Pratelli et al., 2021; Valadez-Noriega et al., 2022). In particular, even when antimicrobial metaphylactic treatments and vaccines for BRD are administered, the first two weeks following the introduction of cattle to beef-fattening facilities appear to be the most vulnerable time for the development of BRD (Pratelli et al., 2021). In our study, the clinical examination at T2 did not revealed significant change in the number of animals with BRD compared to T1. The first two months after arrival to the farm seem to be the most critical period that impacts negatively on animal health and consequently welfare, and that the signs of BRD can persist during these months (Valadez-Noriega et al., 2022). Finally, our clinical investigation did not evidenced animals with diarrhoea at T2. This finding suggests that an adequate adaptation diet was provided, according to the literature (Fusaro et al., 2022). Ration was provided as total mixed ration (TMR) to promote a synchronized intake of roughage and concentrates which decreases, for example, the risk of the occurrence of ruminal acidosis (Cavallini et al., 2022a, 2022b).

ClassyFarm protocol considers a window of 8 days after arrival during which the welfare assessment should not be performed because all the health and welfare impairments detected during this period may be affected by the stress caused by the transport (Bertocchi et al., 2020). Indeed, immediately after transportation, haematological parameters increase from the baseline due to transportation stress followed by a significantly decreased in haematological parameters after 4 to 7 days, suggesting that animals recover from transportation stress (Zulkifli et al., 2019). In our study, even though mean and median values of the WBC and their differential counts, and platelets were within the reference range for cattle, they were significantly higher 15 days after arrival to the fattening unit when compared to those parameters two days after arrival. We can speculate that the animals two days after transportation were still under transportation stress (Zulkifli et al., 2019). Interestingly, we observed a significant increase of all measured white blood cells and platelets 15 days after arrival suggesting that stressor factors such as new environment, feeding, housing, and management practices have a greater important impact than transportation. Additionally, these increase in white blood cells and platelets observed 15 days after arrival suggest an unsatisfactory adaptation to the fattening unit. In fact, it is well known that increased neutrophil and white blood cell counts are signs of inadequate adaptation (Tarantola et al., 2020). Moreover, ruminants can have increased platelets because of stress and/or inflammatory diseases (Jones and Allison, 2007). Herein, fibrinogen was significantly lower 15 days after arrival at the fattening unit, but it was still higher than the normal range for bovine. Fibrinogen is a marker of acute inflammation and stress in cattle (Ansiliero et al., 2019). In acute inflammatory conditions, fibrinogen reaches the highest peak and then declines, while in chronic inflammatory conditions the fibrinogen generally remains high as long as the disease is present and active (McSherry et al., 1970). Thus, we can speculate that at T1, where the highest values of fibrinogen were observed, the bulls were under an acute inflammation (e.g. associated to BRD) and/or stress. At T2, even though there was a decrease of the fibrinogen it was still higher than the normal range indicating the presence of a chronic inflammation.

Our results suggest that the welfare assessment during the first two week after arrival present several critical aspects but can already evidence some risk factors that can cause welfare issues, if present. The repetition of welfare assessment with a consistent method over time is fundamental to assess the long-term welfare of animal in constant housing and management conditions (Kirchner et al., 2014a).

5. Conclusions

Our study suggests that in the context of beef-intensive fattening systems, in the first 15 days after arrival, adaption to a new environment, feeding and management represents an important challenge for the immune system. Consequently, during this period, a reduction in welfare and health is noticed. White blood cells analysis could be a useful tool as warning sign when measuring the welfare status of beef cattle. Clinical investigations can help to evidence critical points in management and housing system that could threat health and welfare of animals. Protocols for welfare assessment with the integration of different assessment indicators, including health, biosecurity, major hazard and warning system and a complete blood cells count, could provide more information of the welfare status and of the critical points in the housing and management system.

Supplementary data to this article can be found online at https://doi.org/10.1016/j.rvsc.2023.03.008.

Declaration of Competing Interest

None of the authors has any financial or personal relationships that could inappropriately influence or bias the content of the paper.

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Supplementary Table S1. Check-list used for total welfare, biosecurity and major hazard and warning system assessment.

			T1	T2
	AREA	**BIOSECURITY**		
	Items	**Level**		
1.	Rodent and insect control measures	Total absence of control measures		
		Presence of rough and minor control measures (absence of written plans)		
		Presence of defined and effective procedures supported by written manual and recording system		
2.	Contact with other animal species	Yes, the contact is frequent and evident		
		No, contact may occur but is not evident at the time of the visit		
		No, the farm is well protected (fences, etc.); no other animal species are present on the farm perimeter, and no contact with herds of the same species or other animals		
3	General precautions at the entrance of occasional visitors	Total absence of measures		
		Presence of minor procedures (absence of written plans)		
		Presence of defined and effective procedures supported by written and recording system manual		
4	General precautions at the entrance of regular visitors	Total absence of measures		
		All visitors are required to wear disposable footwear before entering the farm or use boots that are on the farm for their exclusive personal use		
		All visitors must pass through a changing area and are required to wear disposable footwear and clothes provided by the farm or use clothing and boots that remain on the farm for their exclusive personal use		
5.	Disinfection of vehicles upon entering the farm	Absence of disinfection facilities		
		Presence of non-specific disinfection facilities or used of disinfection aids only when necessary		
		Presence of specific, fixed and routinely used disinfection facilities		
6.	Possibility of contact between foreign vehicles and farmed animals (< 20 m)	Yes		
		No		
7.	Carcass collection (< 20 m)	Yes, vehicles used to remove the carcasses have direct/indirect contact with cattle (< 20 m distance)		
		No, vehicles used to remove the carcasses are stopped at the border of the farm w (>20m distance)		
8.	Live animal loading (i.e. for sale)	Loading is carried out close to the housing premises where the animals are kept (<20 m)		
		Loading is carried out away from the housing premises where the animals are kept (>20 m)		
9.	Quarantine/Housing management	No quarantine for new entering animals		
		Partial/minor quarantine measures (i.e. designated area not separated from the areas where the other cattle are kept, quarantine is too short, no biological tests)		
		Proper quarantine measures, adequate in time and facilities (i.e. designated area separated from the areas where the other cattle are kept, adequate duration of the quarantine, biological tests)		

10.	Control and prevention of most prevalent infectious diseases	No knowledge of most prevalent infectious diseases or no information of the herd health status		
		Partial knowledge and/or presence of undefined plans (i.e., approximate, random, and not continuous over time)		
		Knowledge of at least three diseases prevalence in the herd; in addition, application of proper operational plans of prevention and control on at least two of them (vaccination plan, plan for dealing of infected animals, eradication plan, etc.).		
11.	Health monitoring activities (Verify the farm's habit of submitting pathological material, fetuses, carcasses, and blood samples to the reference testing laboratory; the farmer must be in possession of an analytical result from the last 12 months)	Absence		
		Presence of analysis on pathological material		
12.	Control and prevention of endo/ectoparasites	No knowledge and absence of prevention/control plans		
		Partial knowledge and/or presence of random control and prevention plans (i.e. approximate, random, and not continuous over time)		
		Knowledge of most prevalent parasites on the farm and prevention performed following laboratory tests		
13.	Control and analysis of water sources	Absence of water analysis		
		Drinking water comes from the central supply system or from other sources and the quality of the water is checked at least once a year		
AREA A FARM MANAGEMENT AND STAFF TRAINING				
Items		**Level**		
14.	Personnel: 1. number of stockpersons	Insufficient number of staff: one operator for more than 800 animals		
		Acceptable number of staff: one operator per 400-800 animals		
		Optimal number of staff: one operator for less than 400 animals		
15.	Personnel: 2. skills and training of stockpersons	Inadequate skills and training: approximate experience of less than 5 years and no beef cattle farming training courses		
		Appropriate skills and knowledge: approximate experience of at least 5 years and no beef farming training (or opposite combination)		
		Optimal skills and knowledge: approximate experience of at least 5 years with relevant degree or training course taken within the last 3 years		
16.	Grouping size and grouping management of animals	More than 40 animals per group and heterogeneous in terms of animal body-weight, sex or age		
		Between 20 and 40 animals per group and homogeneous in terms of body-weight, sex or age		
		Less than 20 animals per group and homogeneous in terms of body-weight, sex or age		
17.	Animal inspection and control: 1. number of inspections	Inadequate: less than 1 inspection/day;		
		Sufficient: at least 1 inspection/day		

		Optimal: 2 or more inspections per day along with written reporting of observations or computerized recording		
18.	Animal inspection and control: 2. treatment of sick or injured animals.	Inadequate: evidence of untrained staff and/or presence of animals that need treatment and have not yet received treatment and/or absence of a veterinarian following the farm		
		Adequate: Presence of trained staff with evidence of any animals in an infirmary or animals receiving treatment in place and the presence of a veterinarian following the farm		
		Optimal: in addition to the criteria for adequacy, the presence of relevant written procedures for handling animals (i.e. plan for management of lameness or enteric and respiratory disease)		
19.	Animal inspection, control and culling (in the event that an animal's condition requires on-farm culling, the methods covered in the specific regulations must be followed - EU regulation 1099/2009)	Inadequate: Failure to use competent personnel and/or absence of instructions and training and/or Use of inadequate and not properly maintained equipment		
		Adequate: Culling carried out by a veterinarian, or personnel with a certificate of fitness for slaughter, or, in the case of culling by farm personnel, the presence of instructions and training of the employees (i.e. presence of a training course attended by those on the farm who are in charge of culling, with specific subject matter covered) and the presence of appropriate equipment that is subject to regular maintenance		
		Optimal: in addition to the criteria for adequacy, the presence of written procedures, indicating responsibilities, tools and periodic audits to facilitate and proper emergency management		
20.	Type of animal Handling	Use of offensive tools (electric prods and/or sharp instruments)		
		Use of non-offensive tools (voice, hands, and/or flexible plastic rods)		
21.	Feed, drinking and other substances: 1. management of feed and the daily ration	Inadequate: presence of a ration that is unsuitable for the animals because it is not adapted to their needs, not calculated		
		Adequate: presence of a ration suitable for the animals because it is specific to each group and consists of healthy foods;		
		Optimum: presence of an optimal ration for the animals, because it is calculated by a nutritionist, reviewed frequently and updated		
22.	Feeding phases	1 feeding phase		
		2 feeding phase (adoption and fattening)		
		More than 2 feeding phases(i.e., adaptation, growing and finishing)		
23.	Feeding, drinking and other substances: 2. type of feeding	Inadequate: Access to feeding at incorrect intervals and it is not guaranteed in 24h		
		Adequate: Access to feed at correct intervals and guaranteed in 24h (concentrates administered at least 2 times)		
		Optimal: Access to food consistently over 24 hours: i.e., feed on wagon available for 24h/day		
24.	Concentrates in the ration (daily dose)	Concentrates greater than 80 % of dry matter and average fiber content less than 6 %		
		Concentrates between 70 % and 80 % of dry matter and average fiber content greater than 6 %		
		Concentrates less than 70% of dry matter and presence of at least 1 kg of straw or hay		
25.	Feeding, drinking and other substances: 3. availability of water and number of troughs	Inadequate: Absence of drinking water (not ad libitum) or unhealthy water for one or more animals with particular regard to animals that are sick or		

		Adequate: Presence of functioning drinking troughs		
		Optimal: in addition to the criteria for adequacy, there must be annual examinations for water potability		
26.	Cleaning of troughs/water point	Presence of dirt on the surface and walls of troughs/water point		
		Presence of food only on the water surface or only on the bottom. The water still remains clear		
		Absence of dirt, clean troughs/water point and clear water		
27.	Storage buildings and rooms: hygiene, cleanliness and management of housing environments and bedding	Inadequate: Dirty, unmanaged and/or animal-harmful housing and bedding environments		
		Adequate: Fairly clean and sufficiently managed housing and/or bedding environments and/or clean grid in almost all groups		
		Optimum: Clean, dry and optimally managed housing and bedding environments with frequent material changes		
28.	Biosecurity	The biosecurity checklist score placed in the lowest percent (0-33 %)		
		The biosecurity checklist score placed in the middle percent (33.1-66 %)		
		The biosecurity checklist score is in the highest percent (66.1-100%).		
	AREA B	**HOUSING FACILITIES AND EQUIPMENT**		
	Items	**Level**		
29.	Buildings: absence of harmful buildings	Inadequate: Presence of pens, environments or equipment harmful to animals: i.e., harmful bedding, paths, accesses, boundaries, attachments and electrical trainers that cause injury to animal		
		Adequate: Presence of suitable premises, environments and equipment that are not harmful to animals		
30.	Type of animal housing	At least one group of animals are tied		
		All animal are in groups are free/ not tied		
		All animals are free/not tied and have access to an exercise and/or grazing area adjacent to the buildings or provided with adequate shelter		
31.	Freedom of movement: surface area available for resting	Inadequate: less than 2.5 m^2/head for animals up to 400 kg (plus 0.5 m^2/head per each extra 100 kg between 400 kg and 800 kg)		
		Adequate: between 2.5 and 4.5 m^2/head for animals up to 400 kg (plus 0.5 m^2/head per each extra 100 kg between 400 kg and 800 kg)		
		Optimal: more than 4.5 m^2/head for up to 400 kg (plus 0.5 m^2/head per each extra 100 kg between 400 kg and 800 kg)		
32.	Buildings: 2. flooring	Unsuitable: presence of floor, solid or cracked, that is unsuitable, smooth and slippery, or that impedes movement; slippery paths due to the presence of mud or presence of natural and/or man-made obstacles that do not allow passage in complete safety or that are a cause of avoidable anxiety or excitement		
		Adequate: presence of floor, solid or cracked, suitable and rough (i.e., due to the presence of suitable grooving, or rubber lining, or slight amount of bedding). For at least most of the surfaces on which the animals walk, animals can walk safely without slipping or suffering injury, anxiety or avoidable excitement		

		Optimum: Only if the animals are stabled free and have a floor - solid or slatted - suitable and rough on all surfaces on which they walk (including the presence of permanent bedding with suitable organic material, that is abundant, nonabrasive, well maintained, and absorbent). Access paths have suitable, non-slippery and non-abrasive surfaces, with no obstacles and/or hazards along their entire length		
33.	Presence of electrical educators/trainers	Presence of electrical trainers		
		Absence of electric trainers		
34.	Feeding, drinking and other substances: number of places available in the feeding trough	Inadequate: For separate rations: less than 100% of the animals can feed at the same time For total mixed rations: less than 70% of the animals can feed at the same time		
		Adequate: For separate ration: 100% of the animals can feed at the same time For total mixed rations: more than 70% of the animals can feed at the same time		
		Optimal: presence of 2 differentiated accesses for feeding, with total number of spaces greater (20% greater) than the number of animals or possibility of access to a suitable pasture with presence of large grazing areas		
35.	Size and operation of troughs/water point	less than 1 functioning water bowl for 13 animals or less than 6 cm of trough per animal		
		1 functioning water bowl for 13 animals or 6 cm of trough per animal		
		more than 1 functioning water bowl for 13 animals or more than 6 cm of trough per animal and different water access points		
36.	Specific handling equipment (It is an assessment of the facilities provided to move animals - group/pen change- within the barn)	Absence of corridors and mobile barriers for animal handling		
		Presence of fixed open-walled corridors for animal handling		
		Presence of corridors and mobile barriers with closed walls for animal handling		
37.	Equipment for capturing of animals	Absence of capture and restraint equipment		
		Presence of non-specific but effective capture systems		
		Presence of specific equipment for capture and immobilization		
38.	Buildings: 3. facilities for sick animal (all groups)	Inadequate: Absence of any specific and identified room/post in which injured or sick animals can be isolated in case of need		
		Adequate: Presence of room/post identified and specially prepared to accommodate sick or injured animals equipped with dry bedding or mat comfortable, where clinical conditions require it		
		Optimal: Free-roaming animals in specific and identified room with dry and comfortable permanent bedding, capable of housing at least 3% of the average number of animals daily on the farm and with plenty of available space (i.e., m^2/head greater than the optimal resting area)		
39.	Buildings: 4. temperature and humidity	Inadequate: the presence of microclimatic conditions harmful to animals: i.e., closed or dusty environments or semi-open stables without adequate ventilation		
		Adequate: the presence of suitable microclimatic conditions for animals: i.e., natural ventilation (open barn) or ventilation/ventilation systems without control systems		

		The higher requirement includes, in addition to the criteria for suitability, the presence of optimal microclimatic conditions for the animals: i.e., presence of facilities for microclimate conditioning with automated control systems / suitable pasture		
40.	Buildings: 5. minimum lighting - light cycle for animals	Inadequate: no or insufficient natural or artificial lighting/no or insufficient rest period		
		Adequate: presence of adequate natural or artificial lighting for at least 8 hours per day / presence of adequate rest period for at least 8 hours per day		
AREA C		**ANIMAL-BASED MEASURES**		
	Items	**level**		
41.	Avoidance test	Difficulty of approach		
		Curious animals on approaching		
		Animals approachable and being touched		
42	Behavior among animals	More than 50% agonistic behaviors out of the total behaviors observed		
		Between 10% and 50% agonistic behaviors out of the total behaviors observed		
		Less than 10% agonistic behaviors out of total behaviors observed		
43.	Nutrition status as measured by body condition score (BCS)	More than 10% of animals with BCS less than 2		
		Between 2% and 10% of animals with BCS less than 2		
		Less than 2% of animals with BCS less than 2		
44.	Cleanliness of the animals	More than 40 % dirty animals		
		Between 10% and 40% dirty animals		
		Less than 10% dirty animals		
45.	Skin lesions	More than 20% of animals with mild skin lesions		
		Between 10% and 20% of animals with mild skin lesions		
		Less than 10% of animals with mild skin lesions		
46.	Lameness	More than 6 % of animals lame		
		Between 2% and 6% lame animals		
		Less than 2% lame animals		
47.	Severe respiratory disease (animals within the first 40 days since the arrival to the fattening unit)	More than 15% of animals (between 8 and 40 days after arrival) with severe respiratory disease		
		Between 5% and 15% of animals (between 8 and 40 days after arrival) with severe respiratory disease		
		Less than 5% of animals (between 8 and 40 days after arrival) with severe respiratory disease		
48.	Mild respiratory and/or enteric diseases (animals within the first 40 days since the arrival to the fattening unit)	More than 40% of animals (between 8 and 40 days after arrival) with mild respiratory and/or enteric disease		
		Between 20% and 40% of animals (between 8 and 40 days after arrival) with mild respiratory and/or enteric disease		
		Less than 20% of animals (between 8 and 40 days after arrival) with mild respiratory and/or enteric disease		
49.	Annual mortality of adult animals	More than 5%		
		Between 2% and 5%		

		Less than 2%		
50.	Mutilations and other practices.	Inadequate: Presence of animals with incisions or with mutilations/castrations performed without adherence to the time and manner required by the regulations		
		Adequate: Presence of animals with mutilation/castration performed in compliance with the time and manner required by regulations		
		Optimal: Presence of all animals intact and showing no incisions or mutilations/castrations		
AREA	**MAJOR HAZARDS AND WARNING SYSTEMS**			
	Items	**Level**		
51.	Origin of the drinking water	only one drinking water source and no storage tank		
		only one drinking water source but presence of a storage tank that guarantees a sufficient water supply in case of disruption of the water source		
		presence of two or more drinking water sources		
52.	Noise	Excessive noise		
		Normal noise		
53.	Lighting for inspection	Absence of artificial lighting for inspection		
		Presence of proper and functioning artificial lighting		
54.	Ventilation system alarm	Inadequate: Absence of an alarm system and replacement to the artificial ventilation system and/or absence of regular checks of the alarm		
		Adequate: Presence of an alarm system and replacement to the artificial ventilation system regularly checked /or the farm does not require a ventilation system (i.e., pasture)		
55.	Fire alarm	Absent		
		Present		
56.	Records of pharmacological treatment	Inadequate: Absence of logbook or records or failure to storage for the stipulated period		
		Adequate: Adequate presence and storage		
57.	Records of loading and unloading register of the animals	Inadequate: Absence of the registry (paper or computerized) or inadequate storage for the stipulated period or evidence of abnormal mortalities Not reported in accordance with Presidential Decree No. 320 of February 8, 1954.		
		Adequate: Presence of the registry, adequate storage and no evidence of abnormal mortalities not reported under Presidential Decree Feb. 8, 1954, n. 320		
58.	Food, drugs and other substances (administration of illicit substances)	Inadequate: Evidence of illicit substance administration		
		Adequate: No evidence of illicit substance administration		

Abbreviations: T1 = Assessment at day 2 after arrival; T2=Assessment at day 15 after arrival.

Supplementary Table S2. Data recording table used for the inspective clinical examination per pen.

Pen Number _______________ **Date** ____

Ear Tag	MS [1]	BCS [2]	CS[3]	SL	LS [4]	RF [5]	N cough [6]	ND [7]	OD [8]	Other
		1 6 2 7 3 8 4 9 5	0 1 2		0 1 2 3			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	

Abbreviations: MS=Mental status; BCS=Body condition score; CS=cleanness scoring; SL=Skin lesions; LS=Locomotion scoring; RF=respiratory findings; ND=nasal discharge; OD=ocular discharge.

[1] the following nomenclatures were used to describe the mental status: alert, depressed, sporous, comatose.

[2] based on Jaymelynn Farney, et al., Guide to Body Condition Scoring Beef Cows and Bulls, Kansas State University, December 2016.

[3] has in consideration the cleanness of flanks including tail and lower hindlimb; score 0 = no dirt or only minor fresh or dried splashing, score 1= an area of dirtiness at least palm size (10 x 15cm), score 2= an area of dirtiness amounting to at least forearm length (40cm) in any dimension; scoring system adapted from AHDB available from https://projectblue.blob.core.windows.net/media/Default/Imported%20Publication%20Docs/Cleanliness%20scorecard%20optimal%20dairy%20systems.pdf

[4] based on Step-Up Beef Cattle Locomotion Scoring System available from http://www.zinpro.com/lameness/beef/locomotion-scoring.

[5] Including the type of breath and respiratory frequency.

[6] Number of spontaneous coughs in an interval of 10 minutes.

[7] the type of nasal discharge was classified as following: absent or present; if present monolateral or bilateral, mucous, hemorrhagic, purulent.

[8] the type of ocular discharge was classified as following: absent or present; if present monolateral or bilateral, mucous, hemorrhagic, purulent.

CHAPTER IV: CROSS-SECTIONAL OBSERVATIONAL STUDY OF DIFFERENT BRD ANTIMICROBIAL AND NON-STEROID ANTI-INFLAMMATORY TREATMENTS ON HEALTH AND WELFARE IN FATTENING BULLS

ARTICLE

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Research in Veterinary Science

Cross-Sectional Observational Study of Different BRD Antimicrobial and Non-Steroid Anti-Inflammatory Treatments on Health and Welfare in Fattening Bulls

--Manuscript Draft--

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Abstract:	Our study aimed to evaluate the effect of different treatments for BRD on health and welfare in fattening bulls. A total of 264 bulls were enrolled. Welfare was assessed on day 2 (T0) and day 15 (T1) after arrival, showing a worsening. All bulls were inspected clinically at T0 and T1 revealing an increase of skin lesions and lameness in T2. In both periods, a high incidence of respiratory disease was observed. A prevalence of 79.55% and 95.45% of Mycoplasma bovis using RT-PCR and culture at T0 and T1 respectively was observed. Blood samples were collected for hematology at T0 and T1. At T0, 36 animals were individually treated for BRD with an antimicrobial (IT), 54 received a metaphylactic treatment with tulathromycin (M), 150 received a metaphylactic treatment with tulathromycin plus a second antimicrobial (M+IT) whereas 24 were considered healthy and therefore not treated (NT). Additionally, 128 were treated with a non-steroid anti-inflammatory (NSAID). Neutrophils of M+IT were significantly higher than groups NT and M and the lymphocytes of M+IT were significantly lower than that of IT. White blood cells, neutrophils and N/L ratio of animals treated with an NSAID was significantly higher than that not treated. Lung inspection of 172 bulls at the abattoir indicated that 92.43% presented at least one lung lesion. A statistically significant effect of the NSAID treatment on the lung lesions was observed. Our findings indicate that BRD was a major welfare and health concern and evidence the difficulties of antimicrobial treatment of M. bovis.

HIGHLIGHTS

- Bovine respiratory disease impacts negatively health and welfare of beef cattle.
- In intensive beef fattening systems, the first 15 days are a critical period.
- In intensively fattened cattle, a high prevalence of lung lesions is observed at the abattoir.
- Non-steroid anti-inflammatory drugs decrease lung inflammation.
- Respiratory disease and antimicrobial use could be reduced through improved management.

Cross-Sectional Observational Study of Different BRD Antimicrobial and Non-Steroid Anti-Inflammatory Treatments on Health and Welfare in Fattening Bulls

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Abstract

Our study aimed to evaluate the effect of different treatments for BRD on health and welfare in fattening bulls. A total of 264 bulls were enrolled. Welfare was assessed on day 2 (T0) and day 15 (T1) after arrival, showing a worsening. All bulls were inspected clinically at T0 and T1 revealing an increase of skin lesions and lameness in T2. In both periods, a high incidence of respiratory disease was observed. A prevalence of 79.55% and 95.45% of *Mycoplasma bovis* using RT-PCR and culture at T0 and T1 respectively was observed. Blood samples were collected for hematology at T0 and T1. At T0, 36 animals were individually treated for BRD with an antimicrobial (IT), 54 received a metaphylactic treatment with tulathromycin (M), 150 received a metaphylactic treatment with tulathromycin plus a second antimicrobial (M+IT) whereas 24 were considered healthy and therefore not treated (NT). Additionally, 128 were treated with a non-steroid anti-inflammatory (NSAID). Neutrophils of M+IT were significantly higher than groups NT and M and the lymphocytes of M+IT were significantly lower than that of IT. White blood cells, neutrophils and N/L ratio of animals treated with an NSAID was significantly higher than that not treated. Lung inspection of 172 bulls at the abattoir indicated that 92.43% presented at least one lung lesion. A statistically significant effect of the NSAID treatment on the lung lesions was observed. Our findings indicate that BRD was a major welfare and health concern and evidence the difficulties of antimicrobial treatment of *M. bovis*.

Keywords: Bovine respiratory disease, beef, cattle, *Mycoplasma bovis*, NSAID, Tulathromycin

1. Introduction

Beef production systems in the European Union (EU) differ in feeding management, housing, and in age and weight at slaughtering. Intensive fattening beef cattle management in Italy consists of indoor housing where cattle are managed more efficiently and fed to gain more weight than in extensive production systems. The fattening period is commonly short (3-10 months), the specialized fattening units are more frequently located in the Po Valley region, and the stocker cattle are often imported from abroad (mainly France). Animals are fed high-energy grain-based diets (Cozzi, 2007). This rearing system is characterized by critical points, such as the health status of the newly received cattle, the risk of digestive disorders during the adaptation phase, and management practices that may impair meat quality. In a short period of time, indeed, young cattle are exposed to several stressors such as weaning, mixing with animals from different farms at the auction market (thus with different immune status and microbial exposure), feed restriction, human interaction, long transport, and new diet and environment in the fattening unit. These stressors predispose the insurgence of illness status in the newly introduced beef cattle (EFSA, 2012).

Bovine respiratory disease syndrome (BRD) is one of the most high-cost disease and risk factor for the development of poor welfare in beef cattle all over the world particularly considering intensive systems (Chai et al., 2022; Cortes et al., 2021; Smith, 2020). It is responsible for increased mortality rates and costs of treatment, reduced feed efficiency, and lower carcass quality (Padalino et al., 2021; Pratelli et al., 2021; Compiani et al., 2014). BRD affects the lower respiratory tract (bronchopneumonia, pneumonia) or/and upper respiratory tract (rhinitis, tracheitis, bronchitis) (Pratelli et al., 2021). It is multi-factorial, with a variety of physical and physiological stressors (Peel, 2020). Transportation, climate change, temperature difference and also a new farm environment play a significant role as predisposing factors for BRD by favouring pathogen transmission and stress-induced susceptibility (Padalino et al., 2021; Smith, 2020). The complexity of the interactions and time between these predisposing

factors make BRD management and control challenging. The most common pathogens associated with BRD in beef cattle are *Pasteurella multocida*, *Mannheimia haemolytica*, *Histophilus somni*, *Mycoplasma bovis*, bovine herpesvirus type 1 (BoHV-1), bovine adenovirus (BAdV), bovine viral diarrhea virus (BVDV), bovine coronavirus (BCoV), bovine respiratory syncytial virus (BRSV), and bovine parainfluenza 3 virus (Jelinski et al., 2020; Cirone et al., 2019). In particular, *M. bovis* infections are associated with chronic pneumonia and polyarthritis syndrome, otitis media, conjunctivitis and meningitis (Prysliak et al., 2011). Indeed, *M. bovis* is an opportunistic bacterium of the respiratory microbiota that can become pathogenic under subsequent stressful situations (Tortorelli et al., 2017). BRD management and control is usually based on the administration of antimicrobials and anti-inflammatory drugs as a metaphylactic treatment and/or for individual treatment of clinically affected animals (Pratelli et al., 2021; Compiani et al., 2020; Moore et al., 2014). However, antimicrobial abuse and the risk of antimicrobial resistance are global issues of great concern for both human and animal health. The necessity to reduce the use of antimicrobials in animal food production sectors is highlighted as they play a significant role in the rise of antimicrobial resistance (Santinello et al., 2022).

Therefore, in the present cross-sectional observational study, we aimed to evaluate the effect of antimicrobial and anti-inflammatory treatments for BRD on health and welfare, on newly introduced beef cattle in a commercial fattening unit of Limousine bulls affected by high prevalence of BRD due to *M. bovis.*

2. Materials and Methods

2.1. Housing, Management and Animals

The observational study was performed in a commercial fattening unit of Limousine bulls imported from France located in the province of Modena (Po valley region, Italy) from

November 2021 until May 2022. This farm had a history of BRD *M. bovis*-related in the last production cycles. The study was conducted in a barn housing 264 animals. The barn was semi-closed and well-ventilated with curtained sidewalls. The barn had 44 pens in a free stall system with a capacity of 6 animals per pen (Supplementary Figure S1). A pen had a dimension of 18.4m^2. Each animal had a space of 3.06 m^2 and a manger front of 45 cm. The feeders were placed on one side along the manger. The pens were built adjacent to each other and were separated by iron bars, allowing interaction of animals in adjacent pens. The flooring was slatted and underneath there was a pit for manure collection. Before placing the animals in their respective pens, it was cleaned with a pressure washer and disinfected.

A total of 264 animals arrived to the fattening unit in numerically heterogeneous groups weekly-based with a total of 6 groups over the course of 6 weeks. The animals came from different French farms located all over the country. At arrival, all animals were vaccinated with live attenuated virus of bovine viral diarrhea-mucosal disease (Rispoval D-Bvd®, Zoetis, Italy) and live vaccine of bovine herpesvirus type 1 (Bovilis IBR Live marker®, MSD Animal Health, Italy) for infectious bovine rhinotracheitis. A vaccine buster was given four weeks after the first administration. Ivermectin (Ivomec®, Boehringer Ingelheim Animal Health, Italy) was given subcutaneously at arrival. No quarantine period was performed.

At the arrival, animals were fed an adaptation diet in order to reduce dietary stressors (Supplementary Table S1). The total mixed ration (TMR) was fed ad libitum and fresh clean water was always available. The TMR diet was fresh sampled in different locations (beginning, middle and end of the feeding line) at day 2 (T0) and day 15 (T1) after the arrival of animals to the fattening unit. Analytical TMR analyses were performed at the University of Bologna feed analysis lab according to the methodology described in previous studies (Mammi et al., 2020).

The production cycle lasted between 5 to 6 months. During this period, 14 bulls were euthanized due to severe BRD and 250 bulls finished the cycle and were slaughtered with 600kg.

2.2. Welfare assessment

The welfare assessment was carried out at T0 and T1 using an adapted version of the Italian protocol for the assessment of beef cattle welfare included in the ClassyFarm system (Bertocchi et al., 2020) as previously described (Masebo et al., 2023).

2.3. Clinical examination

An inspective pen-based clinical examination was performed for all animals (n=264) at T0 and T1. It consisted in a 10 minutes-long observation with the observer standing among the animal in the pen. The following parameters were assessed: mental status, body condition score cleanness score, skin lesions, locomotion score, respiratory findings, nasal discharge, ocular discharge, fecal consistency, and other eventual abnormalities. All data were recorded using a schematic table per pen (Supplementary Table S2). An animal was considered to be affected by BRD if it had at least two abnormal findings associated with the respiratory system (i.e. cough and nasal discharge; abnormal type of breath and cough; abnormal type of breath and nasal discharge).

2.4. Blood analysis

Blood samples from 88 animals were collected for haematological investigation at T0 and T1. Two animals were chosen randomly from each pen at T0, and the same subjects were re-sampled at T1. The samples were transferred into vacuum tubes containing EDTA anticoagulant for a complete blood count (CBC) and then into citrate tube for fibrinogen

analysis. The following parameters were analysed: erythrocyte (RBC), haemoglobin, haematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), red blood cell distribution width (RDW), platelets (PLT), leucocytes (WBC), neutrophils, monocytes, lymphocytes, eosinophil, basophils and fibrinogen.

2.5. RT-PCR for Mycoplasma bovis

Nasal swab in a pool of three samples was obtained from all 264 animals at T0 (total of 88 pools). To collect the nasal swabs, animals were contained and the nostrils cleaned with paper before performing swabbing to avoid contamination. The nasal swabs were stored in dry collection tubes and analysed within 12 hours after sampling. A qualitative RT-PCR for the detection of *M. bovis* was used.

2.6. Culture of Mycoplasma bovis

Nasal swab in a pool of two samples was obtained from 88 bulls at T1 (total of 44 pools). The 88 animals were the same sampled also for haematological investigation. To collect the nasal swabs, animals were contained and the nostrils were cleaned with paper before performing swabbing to avoid contamination. The nasal swabs were stored in dry collection tubes and then immersed into 2 mL of Mycoplasma liquid medium (ML; Mycoplasma Experience Ltd., Bletchingley, UK) and maintained at 4^0C until arrival to the laboratory. Mycoplasma cultivation and isolation were then performed as previously described (Catania et al., 2020).

2.7. BRD antimicrobial treatment

An antimicrobial treatment was started on 240 animals by the local veterinarian at T0 or in the immediately following days. The timing and the type of treatment was decided upon the clinical findings and the *M. bovis* testing. The following antimicrobials were used: 54 received a metaphylactic treatment with tulathromycin (Tulissin®, Virbac, Italy) (M), 150 received a metaphylactic treatment with tulathromycin (Tulissin®, Virbac, Italy) plus another antimicrobial (M+IT), 36 animals were individuality treated with an antimicrobial (IT). Only 24 animals were considered to not need any treatment (NT) (Supplementary Table S3).

2.8. BRD non-steroid anti-inflammatory drugs treatment

At T0 or in the immediately following days, 128 animals were treated with non-steroid anti-inflammatory drugs (NSAID) if deemed necessary by the clinical findings (Supplementary Table S4).

2.9. Lung inspection at abattoir

The period of fattening ranged between 5 to 6 months. 250 bulls were slaughtered with approximately 600 kg of body weight. A lung examination was carried out on 172 bulls. A lung score based on an estimation of the extension of diseased parenchyma was applied as following: no evidence of parenchymal alteration (healthy); inflammatory lesions affecting 1 to 25% of the parenchyma (mild pneumonia); inflammatory lesions affecting 25% to 50% of the parenchyma (moderate pneumonia); inflammatory lesions affecting more than 50% of the parenchyma (severe pneumonia) (Supplementary Figure S2).

2.10. Statistical analysis

Data were entered into a statistics program (JMP Pro 17). Descriptive statistics were generated mean ± standard deviation (S.D.) and/or standard error (S.E.), median and range for continuous data, and count and percentage for categorical data. For continuous variables,

normality was tested by Shapiro-Wilk test and non-normally distributed variables were Box-Cox transformed before the analysis. The evaluation of differences between the use/type of antimicrobial and anti-inflammatory treatment was undertaken using the Mixed Model Procedure. Each animals were set as experimental unit within the anti-inflammatory use, or antimicrobial use, depending on the model tested, arrival group, and pen as nested factors. The use/type of antimicrobial (NT, IT, M, IT+M) or the anti-inflammatory use (Y/N) treatment was implemented as a fixed effect in separate models. The day 2 (T0) was set as a covariate in both models. After the analysis, normal distribution of the data was checked again for the resulting residuals. Means are reported as least square mean and pairwise multiple comparisons were performed using Tukey-test as a post hoc test when a significance was detected. The nominal logistic model was used for categorical variables using the same discriminant as before mentioned. A p-value≤0.10 was considered a tendency; a p-value≤0.05 was considered statistically significant; and a p-value≤0.01 was considered highly significant.

3. Results

3.1. Welfare Assessment

Table 1 shows the results of the welfare assessment at T0 and at T1. At T0 the total welfare was 79.04% (medium). Data obtained for Area A, B and C were 70.45% (medium), 65.17% (medium) and 90% (high) respectively. At T1, a decrease in total welfare was observed (76.47%; medium). Even though there was an increase in Area B (68.57%), a decrease in total welfare was noticed when compared to T1 due to a decrease of Area C score (80%).

3.2. Clinical Examination

At T0 the following clinical findings expressed on percentage of affected animals were recorded as following: 1.51% of integument lesions, 0.75% of lameness, 0.75% of diarrhea,

34% of coughing, 48.86% of nasal discharge, and 6.81% of ocular discharge. At T1 an increase in animals with integument lesions was observed (44.69%). Most of these were alopecic lesions in the neck. In addition, a slight increase in lameness (1.15%) and a moderate increase of coughing (52.65%) was noticed. Contrarily, a decrease in diarrhea (0%) and nasal discharge (41.28%) were observed. More details are presented in Table 2.

3.3. RT-PCR and Culture for Mycoplasma bovis

At T0 70 out of 88 pools (79.55%) were tested positive at RT-PCR for *M. bovis*. At T1 42 out of 44 pools (95.45%) resulted positive at the culture of *M. bovis.*

3.4. Lung lesions at abattoir

Figure 1 depicts the distribution of the lung lesions observed at abattoir. The most prevalent condition was mild pneumonia, observed in 96 animals (55.81%). Moderate pneumonia was observed in 58 animals (33.72%). Severe pneumonia was observed in 5 animals. Only 13 animals (7.55%) could be considered completely normal.

3.5. Effect of the different antimicrobial treatments on blood analysis, clinical findings and lung at abattoir

The effect of the different antimicrobial treatments at T0 on the blood analysis are presented in Table 3. At T1, there was a statistically significant effect (p-value$\leq$0.05) on the neutrophils, lymphocyte counts, and their ratio. The neutrophil count of the group M+IT was significantly higher than that of groups NT and M. Furthermore, the lymphocyte count of M+IT was significantly lower than that of IT. Consequently, the ratio N/L was significantly higher in M+IT compared to the other groups.

The effect of the different antimicrobial protocols at T0 on the clinical findings at T1 (respiratory disease, integument lesions, lameness, diarrhea) are presented in Table 4. No statistically significant effect (p-value≤0.05) was observed. The effect of the different antimicrobial protocols at T1 on the lung lesions observed at the abattoir are presented in Table 5. No statistically significant effect (p-value≤0.05) was observed, too.

3.6. Effect of the anti-inflammatory treatment on blood analysis, clinical findings and lung at abattoir

The effect of NSAID treatment at T0 on blood analysis at T1 are presented in Table 6. At T1, there was a statistically significant effect (p-value≤0.05) of the NSAID treatment on the WBC, neutrophils and N/L ratio. The WBC, neutrophils and N/L ratio of animals that were treated with an NSAID was significantly higher than that not treated with an NSAID. The effect of the NSAID treatment at T0 on the clinical findings at T1 (respiratory disease, integument lesions, lameness, diarrhea) are presented in Table 7. No statistically significant effect (p-value≤0.05) was observed.

The effect of the NSAID treatment at T1 on the lung lesions observed at the abattoir are presented in Table 8. There was a statistically significant effect (p-value≤0.05) of the NSAID treatment on the lung lesions observed at the abattoir.

4. Discussion

This observational study dealt with the assessment of different BRD antimicrobial and NSAID use on health and welfare in newly introduced beef cattle in an intensive fattening system. The first period of the intensive cycle is widely reported to be the more stressful and more susceptible to illness status by young animals (EFSA, 2012). To reduce this stress period an adaption diet is commonly provided (Cozzi, 2007). In the present study the provided diet

was adequate in terms of starch, protein, and fibrous fractions requirements (Fusaro et al., 2022; Fusaro et al., 2021; NRC, 2000). So, we believe that the obtained results are absents from bias due to dietary management.

In our study, for the welfare assessment we used a method based on a modified version of the Italian protocol for beef cattle welfare assessment that is included in the ClassyFarm system (Bertocchi et al., 2020). This method was applied at T0 and T1, in order to achieve consistency over a critical time. From T0 to T1, a decrease in welfare was noticed due to a reduction in Area C Animal-based indicators score. The observed decrease in Area C score might be associated with a stress response to both physical (i.e. transportation, new environment, new feed) and psychological (i.e. maternal separation, social mixing) stressors. Epidemiological research has related a large spectrum of stressors, in particular transportation, as factors contributing to higher disease susceptibility including BRD (Chen et al., 2015). Transportation causes a general immunosuppression that makes it possible for many opportunistic infections to invade the respiratory tract and result BRD (Earley et al., 2017).

In our study, at T0 34% of the animals already presented coughing and 48.86% nasal discharge, and more than 79% of the nasal swab pools for RT-PCR for *M. bovis* were positive indicating a high incidence of BRD at the moment of the introduction in the fattening unit. Moreover, at T1 a moderate increase in coughing (52.65%) and a slight decrease in nasal discharge (41.28%) were noticed, and more than 95% of the nasal swabs for culture of *M. bovis* tested positive. These findings support the increase in BRD incidence in the farm. We could speculate that a certain percentage of animals, although clinically healthy, started their transport to the fattening unit in Italy already infected or alternatively were exposed to pathogens during transportation. They then developed the diseases once in the new location in Italy. Stress factors, such as transportation and social and environmental change, might have a negative influence on the regulation of innate immunity (Chen et al., 2015). This could explain

the occurrence of clinical disease already at arrival at the fattening unit in animals exposed to BRD-pathogens before transportation (Padalino et al., 2021; Cirone et al., 2019). Furthermore, these animals could be active shedders and infect other animals during transportation. Naive animals that are exposed both to pathogens and stress factors are more susceptible to develop the clinical disease (Castillo-Alcala et al., 2012). In addition, intensive systems of housing and rearing animals can also create favourable conditions for the occurrence of BRD (Catania et al., 2020). The housing structure, type and quality of flooring, microclimatic conditions, space allowances/pen size are conditions that may be factors influencing animal health (Cozzi et al., 2009).

In the case of *M. bovis*, Castillo-Alcala et al. (2012) showed an increased prevalence from the day of arrival up to day 15 after arrival similar to the current study. Several studies on the occurrence of BRD in beef cattle transported from France to Italy revealed an increase of the prevalence of BRD-related pathogens (including *M. bovis*) after arrival at the Italian fattening units (Catania et al., 2022; Padalino et al., 2021; Cirone et al., 2019). One of these studies reported that BRD-related pathogens increased from 16% to 82.8 % four days after arrival at the fattening unit (Padalino et al., 2021).

Herein, the effects of the different antimicrobial (NT, M, IT and M+IT) and NSAID treatments for BRD on the blood analysis were investigated. For the antimicrobial treatments, the neutrophil count of the group M+IT was significantly higher than that of groups NT and M, and the lymphocyte count of M+IT was significantly lower than that of IT. In fact, animals included in the M+IT group were clinically affected by BRD. However, there was an absence of statistically significance difference of the clinical findings between M+IT and IT groups. This could be explained by the fact that also animals included in the group IT were clinically affected by BRD. Therefore, our findings suggest that there was no difference between a performing a M+IT or only IT. Moreover, it could be associated with inefficiency of the used

antimicrobials. The number of antimicrobial treatments is linked with a higher probability of bacteria resistant to at least one antimicrobial. In addition, antimicrobial resistance in BRD is higher when using a combination of antimicrobials with different pharmacodynamics. These observations suggest that consideration should be given to antimicrobial pharmacodynamics when selecting drugs for retreatment of BRD (Coetzee et al., 2019). Choosing an ineffective antimicrobial for BRD poses serious risks to both animals and their owners in terms of welfare and financial implications. The decision-making process must take into account all relevant information to select the 'optimal' antimicrobial drug for a given situation, often including the results of bacterial culture and antimicrobial susceptibility testing (Lubbers and Turnidge, 2015). More targeted and selective use of antibiotics in the livestock industry will be required in light of the emergence of antibiotic-resistant pneumonia in feedlot cattle (Earley et al., 2017). For the NSAID treatment, the WBC, neutrophils and N/L ratio of animals that were treated with an NSAID was significantly higher than that not treated. NSAID may have immunomodulatory effects and interfere with the function of neutrophils by increasing cellular immunity that, consequently, decreases the immune response (Curry et al., 2005). The effect of different antimicrobial and NSAID treatments at T0 on the retrieved clinical findings at T1 was also investigated and revealed the absence of a statistically significant effect. The first two weeks after the introduction of cattle in beef fattening units seem to be the most critical period for the development of BRD, even when metaphylactic treatment and vaccination are started (Pratelli et al., 2021). Furthermore, it has been reported that in feedlots, *M. bovis* can be resistant to most of the antimicrobials that are used to treat BRD (García-Galán et al., 2021; Jelinski et al., 2020). Herein, unfortunately, we did not investigate possible antimicrobial resistance. Furthermore, the clinical signs of BRD may not be detected at early stage of the disease and many animals may be undetected so that when detected the disease stage is advanced and the treatment success is less likely. It was suggested that the accuracy of current

approaches for the early detection, prognosis, and diagnosis of BRD is still low, necessitating further study into BRD diagnostics (Chai et al., 2022).

We further observed that more than 90% of the lungs at the abattoir presented at least one lung lesion and the most prevalent category was mild pneumonia. Our findings show a very high prevalence of lung lesions when compared to previous reports (43-72%) (Caucci et al., 2018; Thompson et al., 2006; Wittum et al., 1996). Moreover, the effect of different antimicrobial protocols on the lung lesions at the abattoir showed absence of a statistically significant effect similar to Caucci et al. (2018). Chronic *M. bovis*-associated lung lesions may represent a dynamic situation of bacterial clearance and reinfection with genotypically different *M. bovis* strains. These findings could explain the ineffectiveness of the antimicrobial treatment for chronic pneumonia associated with *M. bovis* (Castillo-Alcala et al., 2012). *M. bovis* involvement in BRD can result in persistent pneumonia that does not respond well to antimicrobial therapy (García-Galán et al., 2021; Jelinski et al., 2020). Contrarily, a significant effect of the NSAID treatment on the lung lesions was observed at the abattoir. The lung lesions from the categories healthy and mild pneumonia were significantly lower in animals that received an anti-inflammatory treatment in the first 15 days after arrival to the farm. These findings suggest that an NSAID treatment for BRD may help to decrease lung inflammation. Compiani et al. (2020) reported that the use of NSAID in beef cattle at arrival to a fattening unit reduces the incidence of BRD.

5. Conclusions

In summary, our observational study revealed a decrease in welfare during the first 15 days after arrival to the farm, in particular considering the score in Area C animal-based indicators. Our findings indicate that the prevalence of BRD, most likely associated with *M. bovis*, in this beef cattle population was already high at the time of arrival to the farm and

increased during the first 15 days after arrival. We further observed an absence of association between different antimicrobial protocols (IT, M, M+IT, NT) started at arrival and the retrieved clinical findings at 15 days after arrival. Moreover, we observed a high prevalence of lung lesions at the abattoir. An absence of association between different antimicrobial protocols (IT, M, M+IT, NT) administered in the first 15 days after arrival and the lung lesions observed at the abattoir was also noticed. In contrast, an association between NSAID treatment and lung lesions was noticed indicating that NSAID treatments for BRD may help to the decrease lung inflammation. Our findings indicate that BRD was a major welfare and health problem in the studied population. Indeed, our findings evidence the difficulties of antimicrobial treatment and the potential efficiency of NSAID treatment of *M. bovis* BRD-associated pneumonia. Therefore, enhancing farming practices, animal health and welfare should primarily be considered to reduce disease prevalence and antimicrobial usage. Furthermore, the use of NSAIDs could represent an optional approach to control BRD and reduce antimicrobial usage but more research should be performed to validate this hypothesis. Our observational study highlights the real challenge in the management of BRD conditions in intensive fattening systems.

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CRediT authorship contribution statement

Naod Thomas Masebo: Conceptualization, Writing – Original Draft, Writing – Review and Editing; **Giovanna Marliani:** Writing – Review and Editing; **Laura Abram:** Investigation, Methodology**; Flavia Shannon Del Re**: Investigation, Methodology; **Damiano Cavallini:** Investigation, Methodology, Writing – Review and Editing; **Marco Di Pietro:** Resources,

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Institutional Review Board Statement

This study did not require official or institutional ethical approval as it was not experimental, but rather part of the routine of clinical and pathological veterinary diagnostics and procedures in a commercial fattening unite. All animals in this study were examined with the consent of their owners and handled according to good ethical standards.

Declaration of Competing Interest

The authors report there are no competing interests to declare.

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Supplementary Materials

Supplementary material associated with this article can be found, in the online version, at **********

Supplementary Figure S1: Structure of the commercial fattening unit. (a) Schematic representation of the structure of the commercial fattening unit, position of feeders and

automatic water bowls, dimension of the pens and corridors. (b) image of the commercial fattening unit.

Supplementary Figure S2: Lung score based on an estimation of the extension of diseased parenchyma. (a) no evidence of parenchymal alteration (healthy); (b) parenchymal inflammatory lesions in 1 to 25% of the lung (mild pneumonia); (c) parenchymal inflammatory lesions in 25% to 50% of the lung (moderate pneumonia); (d) parenchymal inflammatory lesions in more than 50% of the lung (severe pneumonia).

Supplementary Table S1: Descriptive statistics of the adaptation TMR diet (T0 and T1) and chemical analysis.

Supplementary Table S2: Schematic table used for recording the inspective clinical examination per pen.

Supplementary Table S3: Different antimicrobial drugs used for BRD treatment in the fattening unit.

Supplementary Table S4: Different non-steroid anti-inflammatory drugs used for BRD treatment in the fattening unit.

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Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy restrictions. None of the data were deposited in an official repository.

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Figure captions

Figure 1: Lung lesions retrieved at the abattoir.

Table 1: Descriptive statistics of the total welfare, biosecurity and major hazard and warning system of 264 beef cattle from one Italian Herd.

Item	Assessment at T0	Classification at T0	Assessment at T1	Classification at T1
Total welfare	79.04%	Medium	74.73%	Medium
Area A (Farm management and staff training)	70.45%	Medium	70.45%	Medium
Area B (Housing and facilities)	65.17%	Medium	68.57%	Medium
Area C (Animal-based indicators)	90%	High	80%	Medium

Table 2: Clinical findings of the 264 beef cattle.

Item	Assessment at T0	Assessment at T1
Integument lesions (%)	4(1.51%)	118(44.69%)
Lameness (%)	2(0.75%)	4(1.15%)
Diarrhea (%)	2(0.75%)	0(0%)
Coughing (%)	90(34%)	139(52.65%)
Nasal discharge (%)	129(48.86%)	109(41.28%)
Ocular discharge (%)	18(6.81%)	4(1.15%)

Table 3. Effect of the different antimicrobial protocols for BRD at T0 on the blood analysis at T1.

Blood parameters		Days								
		T0				T1				
		Treatment protocol				Treatment protocol				
		IT	M	M+IT	NT	IT	M	M+IT	NT	p
RBC (M/µL)	Mean± SD	9.79±1.44	9.32±1.44	10.06±1.26	8.66±1.38	9.92±1.15	9.79±1.06	9.98±1.09	9.18±1.33	0.76
HGB (g/dL)	Mean± SD	11.55±0.9	12.08±0.82	12.38±1.26	10.72±1.47	11.45±0.81	12.2±0.98	12.18±1.17	11.33±1.61	0.33
HCT (%)	Mean± SD	38.93±3.45	39.61±3.66	40.13±4.43	35.82±5.68	38.46±3.49	40±3.73	39.45±4.06	38.25±4.9	0.68
MCV (fL)	Mean± SD	40.15±3.19	41.72±3.15	40.17±2.87	41.43±3.18	38.91±2.17	40.99±2.76	39.64±2.66	41.78±1.75	0.09
MCH (pg)	Median [Min.-Max]	11.6 [10.9-13.15]	12.8 [11.85-13.5]	12.2 [11.8-12.9]	12.1 [11.8-12.9]	11.35 [10.8-12.78]	12.65 [11.63-13]	12.1 [11.6-12.75]	12.4 [11.73-12.95]	0.7
MCHC (g/dL)	Mean± SD	29.72±1.04	30.55±1.14	30.91±1.19	29.95±1.12	29.88±1.6	30.41±1.45	30.9±1.05	29.62±1.09	0.19
RDW (%)	Mean± SD	24.41±1.61	23.62±2.25	24.22±1.69	23.65±3.89	24.22±1.45	24.12±1.83	24.06±1.55	23.45±2.92	0.41
PLT (K/µL)	Median [Min.-Max]	338 [303-447.5]	289 [192.5-402.5]	227 [128-333]	265.5 [174.5-319.75]	347 [158.75-469.75]	406 [235-524]	296 [133-525.5]	141 [94.5-260]	0.29
WBC (K/µL)	Median [Min.-Max]	8.69 [7.59-11.09]	8.51 [7.07-9.395]	8.79 [7.35-10.28]	7.36 [7.08-8.41]	12.7 [9.1-14.52]	10.15 [8.14-11.69]	10.4 [8.52-12.97]	8.95 [6.82-12.23]	0.13
NEU (K/µL)	Median [Min.-Max]	3.32 [2.43-3.71]	2.94 [2.57-3.77]	3.65 [2.95-4.27]	2.35 [2.00-2.91]	4.27 [3.13-6.7]^{ab}	3.21 [2.62-4.15]^{b}	4.48 [2.93-6.77]^{a}	3.32 [2.23-4.02]^{b}	0.05
MONO (K/µL)	Median [Min.-Max]	1.3 [0.94-1.4]	1.04 [0.84-1.26]	1.29 [1.0275-1.5725]	1.15 [0.9325-1.3625]	1.18 [0.96-1.53]	1.07 [0.83-1.35]	1.12 [0.87-1.4]	1.16 [0.87-1.23]	0.81
LYM (K/µL)	Mean± SD	4.23±0.97	3.94±0.96	3.71±1.43	3.91±0.8	5.87±1.75^{a}	4.68±1.82^{ab}	4.05±1.7^{b}	4.58±1.37^{ab}	0.04
EOS (K/µL)	Median [Min.-Max]	0.13 [0.03-0.33]	0.05 [0.04-0.14]	0.07 [0.02-0.19]	0.12 [0.08-0.23]	0.23 [0.08-0.44]	0.22 [0.09-0.4]	0.23 [0.09-0.43]	0.24 [0.08-0.54]	0.99

BASO (K/µL)	Median [Min.-Max]	0.06 [0.05-0.09]	0.06 [0.05-0.07]	0.06 [0.04-0.08]	0.06 [0.05-0.08]	0.11 [0.09-0.12]	0.06 [0.04-0.11]	0.09 [0.08-0.14]	0.08 [0.06-0.11]	0.06
FIBR (mg/dL)	Median [Min.-Max]	864.6 [694.58-1229.93]	1032 [790.09-1536.98]	1065 [712.95-1441.05]	769.5 [735.45-878.1]	795 [671.7-1077.08]	696.15 [617.25-1098.9]	761.4 [562.35-1033.2]	1111.8 [630.3-1388.33]	0.2
N/L ratio	Median[Min.-Max]	0.6 6[0.6-0.9]	0.85 [0.67-1.01]	0.99 [0.75-1.62]	0.55 [0.48-0.85]	0.91 [0.44-1.11]^{b}	0.72 [0.56-1.32]^{b}	1.12 [0.75-1.93]^{a}	0.69 [0.46-1.07]^{b}	0.02

Abbreviations**:** IT= individual treatment, M=metaphylactic treatment, M+IT=metaphylactic and individual treatment, NT=no treatment, RBC= Red blood cell, HGB= Hemoglobin, HTC=Hematocrit, MCV = Mean corpuscular volume, MCH= Mean corpuscular hemoglobin, MCHC= Mean corpuscular hemoglobin concentration, RDW= Red blood cell distribution width, PLT= Platelets, NEU= Neutrophils, WBC= white blood cells, MONO=Monocytes, LYM=Lymphocytes, EOS=Eosinophils, BASO= Basophils, FIBR=Fibrinogen, N/L ratio= Neutrophils : Lymphocytes ratio, M/µL= 10^6 per microliter ,%,=percentage, K/µL=10^3 per microliter, g/dL= grams per deciliter, fL= femtoliter, pg= picogram, mg/dL=milligram per deciliter, ^{a,b,ab}, denote significant differences among the treatment protocol

Table 4. Effect of different treatment protocols at T0 on the clinical findings at T1.

Clinical findings	Category	Days								p-value
		T0				T1				
		Treatment Protocol				Treatment Protocol				
		IT	M	M+IT	NT	IT	M	M+IT	NT	
Respiratory disease	N	21	45	93	17	21	69	66	24	0.55
	Y	8	9	49	7	8	26	37	13	
Integument alterations	N	36	54	150	24	19	52	59	16	0.31
	Y	0	0	0	0	10	43	44	21	
Lameness	N	36	54	150	24	28	94	101	37	0.59
	Y	0	0	0	0	1	1	2	0	
Diarrhea	N	36	53	149	24	28	94	101	37	0.59
	Y	0	1	1	0	1	1	2	0	

Abbreviations: N=No, Y=yes, IT= Individual treatment, M=Metaphylactic treatment, M+IT=Metaphylactic and individual treatment, NT=No Treatment

Table 5. Effect of different treatment protocols at T1 on the lung lesions observed at the abattoir.

	Category	Days							p-value
		T0				T1			
		Treatment Protocol				Treatment Protocol			
		IT	M	M+IT	NT	IT	M	M+IT	NT
Lung lesions	0	NA	NA	NA	NA	0	4	6	3	0.25
	1	NA	NA	NA	NA	13	23	49	11	
	2	NA	NA	NA	NA	7	11	36	4	
	3	NA	NA	NA	NA	1	0	4	0	

Abbreviations: 0=No evidence of parenchymal alteration (healthy), 1= parenchymal inflammatory lesions in 1 to 25% of the lung (mild pneumonia), 2=parenchymal inflammatory lesions in 25% to 50% of the lung (moderate pneumonia), 3= parenchymal inflammatory lesions in more than 50% of the lung (severe pneumonia), NA=Not applicable, IT= individual treatment, M=metaphylactic treatment, M+IT=metaphylactic and individual treatment, NT=no treatment

Table 6. Effect of the non-steroid anti-inflammatory treatment in blood parameters at T1.

Blood parameters		Days				
		T0		T1		
		AI		AI		
		N	Y	N	Y	p- value
WBC (K/µL)	Median [Max.-Min]	8.3 [7.08-9.97]	8.79 [7.5-10.07]	9.89 [7.88-12.51]	10.54 [8.73-13.02]	0.05
NEU (K/µL)	Median [Max.-Min]	3.19 [254-3.97]	3.46 [2.81-4.01]	3.36 [2.64-4.39]	5.07 [3.02-6.92]	<.01
N/L ratio	Median [Max.-Min]	0.8 [0.58-1.27]	0.92 [0.67-1.36]	0.76 [0.53-1.25]	1.07 [0.76-1.93]	0.01

Abbreviations: NEU= Neutrophils, WBC= white blood cells, N/L ratio= Neutrophils/Lymphocytes ratio, K/µL=10^3 per microliter, AI=non-steroid anti-inflammatory treatment

Table 7. Effect of the non-steroid anti-inflammatory treatment on the clinical findings at T1.

		Days				
		T0		T1		
Clinical findings		AI		AI		
	Category	N	Y	N	Y	p-value
Respiratory disease	N	99	77	107	73	0.47
	Y	30	43	46	38	
Integument alterations	N	NA	NA	79	67	0.16
	Y	NA	NA	74	44	
Lameness	N	NA	NA	151	109	0.75
	Y	NA	NA	2	2	
Diarrhea	N	NA	NA	151	109	0.75
	Y	NA	NA	2	2	

Abbreviations: NA=Not applicable, AI=non-steroid anti-inflammatory treatment, N=No, Y=Yes

Table 8. Effect of the non-steroid anti-inflammatory treatment on the lung lesions observed at the abattoir.

		Days				
		T0		T1		
	Category	AI		AI		
		N	Y	N	Y	P-value
Lung lesions	0	NA	NA	11	2	< .01
	1	NA	NA	56	40	
	2	NA	NA	25	33	
	3	NA	NA	0	5	

Abbreviations: 0= no evidence of parenchymal alteration (healthy), 1= parenchymal inflammatory lesions in 1 to 25% of the lung (mild pneumonia), 2=parenchymal inflammatory lesions in 25% to 50% of the lung (moderate pneumonia), 3= parenchymal inflammatory lesions in more than 50% of the lung (severe pneumonia), NA=Not applicable, AI=non-steroid anti-inflammatory treatment, N=No, Y=Yes

Figure 1

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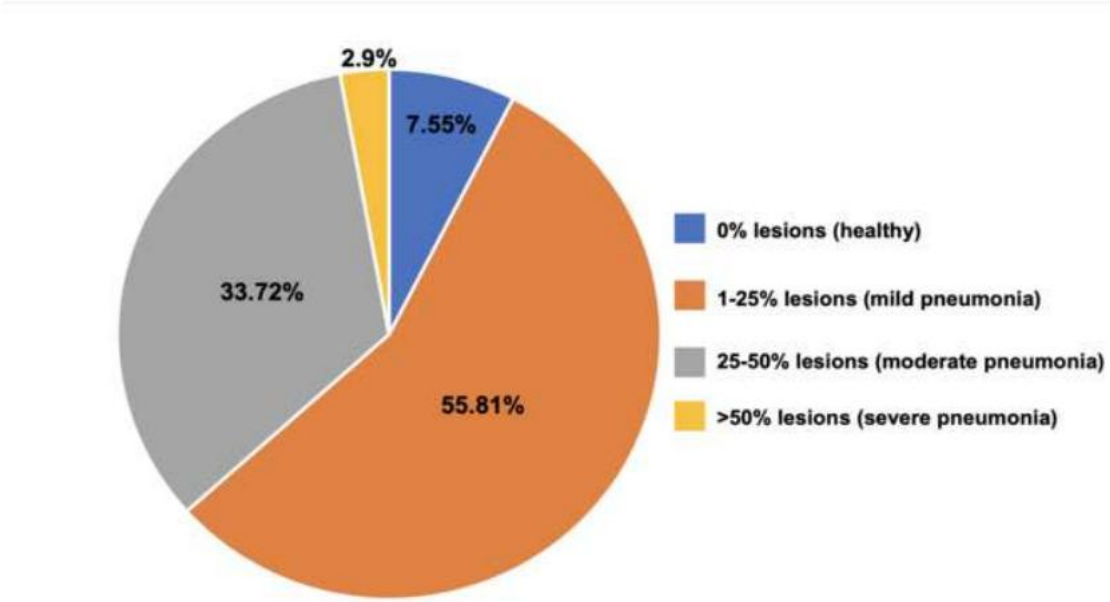


Figure 1: Lung lesions retrieved at the abattoir.

Supplementary Material

Supplementary Table S1. Descriptive statistics of the adaptation TMR diet (T0 and T1) and chemical analysis.

TMR	**Feed, kg af**
Wheat silage	3.5
Meadow hay[1]	1.2
Wheat straw	1.1
Beat pulp	1.3
Corn, finely ground[2]	1.1
Soybean meal	0.5
Cane molasses[3]	0.5
Min&Vit Premix	0.3
	Nutrients, %DM
DM	70.07
UFC	0.81
CP[4]	11.25
Ash	8.78
EE[5]	2.06
Starch	13.57
Sugars	7.37
NDF[6]	39.21
ADF[7]	25.79
ADL[8]	3.45

[1] the quality of the hay was checked to ensure the absence of molds and spores (Cavallini et al. 2022). [2] the corn was below the EU maxim tolerable level (Girolami et al. 2022). [3] molasses were properly characterized (Palmonari et la. 2021). [4] Crude protein. [5] ether extract. [6] neutral detergent fiber. [7] acid detergent fiber. [8] acid detergent lignin.

Cavallini, D., Penazzi, L., Valle, E., Raspa, F., Bergero, D., Formigoni, A., Fusaro, I., 2022. When changing the hay makes a difference: A series of case reports. J Equine Vet Sci. 113, 103940. doi:10.1016/j.jevs.2022.103940

Girolami, F., Barbarossa, A., Badino, P., Ghadiri, S., Cavallini, D., Zaghini, A., Nebbia, C., 2022. Effects of turmeric powder on aflatoxin M1 and aflatoxicol excretion in milk from dairy cows exposed to aflatoxin b1 at the EU maximum tolerable levels. *Toxins* 14, 430. https://doi.org/10.3390/toxins14070430

Palmonari, A., Cavallini, D., Sniffen, C.J., Fernandes, L., Holder, P., Fusaro, I., Giammarco, M., Formigoni, A., Mammi, L.M.E., 2021. In vitro evaluation of sugar digestibility in molasses. Ital. J. Anim. Sci. 20:571-577. DOI: 10.1080/1828051X.2021.1899063

Supplementary Table S2. Schematic table used for recording the inspective clinical examination per pen.

Pen Number ________________ Date ____

Ear Tag	**MS** [1]	**BCS** [2]	**CS**[3]	**SL**	**LS** [4]	**RF** [5]	**N cough** [6]	**ND** [7]	**OD** [8]	**Other**
		1 6 2 7 3 8 4 9 5	0 1 2		0 1 2 3			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	
		1 6 2 7 3 8 4 9 5	0 1 2		1 2 3 4			• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	• Present/Absent • Bilateral/Monolateral • Mucous/Hemorrhagic/ Purulent	

Abbreviations: MS=Mental status; BCS=Body condition score; CS=cleanness scoring; SL=Skin lesions; LS=Locomotion scoring; RF=respiratory findings; ND=nasal discharge; OD=ocular discharge.

[1] the following nomenclatures were used to describe the mental status: alert, depressed, sporous, comatose.

[2] based on Jaymelynn Farney, et al., Guide to Body Condition Scoring Beef Cows and Bulls, Kansas State University, December 2016.

[3] has in consideration the cleanness of flanks including tail and lower hindlimb; score 0 = no dirt or only minor fresh or dried splashing, score 1= an area of dirtiness at least palm size (10 x 15cm), score 2= an area of dirtiness amounting to at least forearm length (40cm) in any dimension; scoring system adapted from AHDB available from https://projectblue.blob.core.windows.net/media/Default/Imported%20Publication%20Docs/Cleanliness%20scorecard%20optimal%20dairy%20systems.pdf

[4] based on Step-Up Beef Cattle Locomotion Scoring System available from http://www.zinpro.com/lameness/beef/locomotion-scoring.

[5] Including the type of breath and respiratory frequency.

[6] Number of spontaneous coughs in an interval of 10 minutes.

[7] the type of nasal discharge was classified as following: absent or present; if present monolateral or bilateral, mucous, hemorrhagic, purulent.

[8] the type of ocular discharge was classified as following: absent or present; if present monolateral or bilateral, mucous, hemorrhagic, purulent.

Supplementary Table S3. Different antimicrobial drugs used for BRD treatment in the fattening unit.

ID	Treatment	Antimicrobial (commercial name and active principle)
1	M+IT	Zuprevo18%® (Tildipirosin)
2	NT	
3	M	
4	M+IT	Nuflor® (Florfenicol), Zuprevo18%® (Tildipirosin)
5	NT	
6	NT	
7	NT	
8	NT	
9	M	
10	M	
11	M	
12	M	
13	M	
14	NT	
15	IT	Nuflor® (Florfenicol)
16	M	
17	M	
18	M	
19	M	
20	M	
21	M	

22	NT	
23	M	
24	M	
25	M+IT	Nuflor® (Florfenicol)
26	M+IT	Nuflor® (Florfenicol)
27	M	
28	M+IT	Nuflor® (Florfenicol)
29	M	
30	M	
31	M	
32	M	
33	M+IT	Nuflor® (Florfenicol)
34	M+IT	Zuprevo18%® (Tildipirosin),Depocillina® (Penicillin G procaine),Valemas10® (Enrofloxacina),Nuflor® (Florfenicol)
35	M	
36	M	
37	M	
38	M	
39	M+IT	Nuflor® (Florfenicol)
40	M	
41	M+IT	Nuflor® (Florfenicol),
42	M	
43	M	
44	M	
45	M	
46	M	
47	M+IT	Nuflor® (Florfenicol),
48	NT	
49	M	
50	M+IT	Nuflor® (Florfenicol),
51	M+IT	Nuflor® (Florfenicol),
52	M	
53	M	
54	M	
55	M	
56	M	
57	M	
58	NT	
59	NT	
60	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
61	M+IT	Zuprevo18%® (Tildipirosin)

62	NT	
63	M+IT	Zuprevo18%® (Tildipirosin)
64	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
65	M+IT	Zuprevo18%® (Tildipirosin)
66	M+IT	Zuprevo18%® (Tildipirosin)
67	M+IT	Zuprevo18%® (Tildipirosin)
68	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
69	NT	
70	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
71	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
72	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
73	M	
74	M	
75	NT	
76	M	
77	NT	
78	NT	
79	M+IT	Nuflor® (Florfenicol)
80	M	
81	M	
82	M+IT	Nuflor® (Florfenicol)
83	M+IT	Nuflor® (Florfenicol)
84	NT	
85	IT	Nuflor® (Florfenicol)
86	M+IT	Zuprevo18%® (Tildipirosin)
87	M+IT	Zuprevo18%® (Tildipirosin)
88	M	
89	M+IT	Nuflor® (Florfenicol)
90	M+IT	Nuflor® (Florfenicol)
91	M	
92	M	
93	M	
94	M	
95	IT	Nuflor® (Florfenicol)
96	NT	
97	IT	Nuflor® (Florfenicol)
98	NT	
99	NT	
100	M	
101	M+IT	Zuprevo18%® (Tildipirosin)
102	M+IT	Zuprevo18%® (Tildipirosin)

103	M+IT	Zuprevo18%® (Tildipirosin)
10	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
105	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
106	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin),Forcyl® (Marbofloxacin)
107	M+IT	Zuprevo18%® (Tildipirosin)
108	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
109	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
110	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
111	IT	Zuprevo18%® (Tildipirosin)
112	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
113	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
114	M+IT	Nuflor ® (Florfenicol),Zuprevo18%® (Tildipirosin)
115	M+IT	Zuprevo18%® (Tildipirosin)
116	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
117	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
118	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin), Vetamplius® (Ampicillin)
119	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
120	IT	Zuprevo18%® (Tildipirosin)
121	IT	Zuprevo18%® (Tildipirosin), Vetamplius® (Ampicillin)
122	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
123	M+IT	Zuprevo18%® (Tildipirosin)
124	M	
125	IT	Nuflor® (Florfenicol)
126	NT	
127	M	
128	NT	
129	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin), Longocillina® (Amoxicillin Trihydrate)
130	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
131	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
132	M+IT	Nuflor® (Florfenicol)
133	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
134	M+IT	Zuprevo18%® (Tildipirosin)
135	M+IT	Zuprevo18%® (Tildipirosin)
136	M+IT	Zuprevo18%® (Tildipirosin)
137	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
138	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
139	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
140	M+IT	Zuprevo18%® (Tildipirosin)
141	M+IT	Zuprevo18%® (Tildipirosin)

142	M+IT	Zuprevo18%® (Tildipirosin)
143	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
144	M+IT	Zuprevo18%® (Tildipirosin)
145	M+IT	Zuprevo18%® (Tildipirosin)
146	M+IT	Zuprevo18%® (Tildipirosin)
147	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
148	M+IT	Zuprevo18%® (Tildipirosin)
149	M+IT	Zuprevo18%® (Tildipirosin),Longocillina® (Amoxicillin Trihydrate)
150	M+IT	Zuprevo18%® (Tildipirosin)
151	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
152	M+IT	Zuprevo18%® (Tildipirosin)
153	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
154	M+IT	Zuprevo18%® (Tildipirosin)
155	M+IT	Zuprevo18%® (Tildipirosin)
156	M+IT	Zuprevo18%® (Tildipirosin)
157	M+IT	Zuprevo18%® (Tildipirosin)
158	M+IT	Zuprevo18%® (Tildipirosin)
159	M+IT	Zuprevo18%® (Tildipirosin)
160	M+IT	Zuprevo18%® (Tildipirosin)
161	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
162	M+IT	Zuprevo18%® (Tildipirosin)
163	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
164	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
165	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
166	IT	Zuprevo18%® (Tildipirosin)
167	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
168	M+IT	Zuprevo18%® (Tildipirosin)
169	M+IT	Zuprevo18%® (Tildipirosin)
170	M+IT	Zuprevo18%® (Tildipirosin)
171	M+IT	Zuprevo18%® (Tildipirosin)
172	M+IT	Zuprevo18%® (Tildipirosin)
173	M+IT	Zuprevo18%® (Tildipirosin)
174	M+IT	Zuprevo18%® (Tildipirosin)
175	IT	Zuprevo18%® (Tildipirosin)
176	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
177	M+IT	Zuprevo18%® (Tildipirosin)
178	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
179	M+IT	Zuprevo18%® (Tildipirosin)
180	M+IT	Zuprevo18%® (Tildipirosin)
181	M+IT	Zuprevo18%® (Tildipirosin)
182	M+IT	Zuprevo18%® (Tildipirosin)

183	M+IT	Nuflor® (Florfenicol), Zuprevo18%® (Tildipirosin)
184	M+IT	Zuprevo18%® (Tildipirosin)
185	M+IT	Zuprevo18%® (Tildipirosin)
186	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
187	IT	Zuprevo18%® (Tildipirosin)
188	IT	Zuprevo18%® (Tildipirosin)
189	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
190	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
191	IT	Zuprevo18%® (Tildipirosin)
192	IT	Zuprevo18%® (Tildipirosin)
193	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
194	M+IT	Depocillina® (Penicillin G procaine)
195	M+IT	Bimoxylla (Amoxicillin),Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
196	M+IT	Zuprevo18%® (Tildipirosin)
197	IT	Nuflor® (Florfenicol)
198	M+IT	Zuprevo18%® (Tildipirosin)
199	M+IT	Longocillina® (Amoxicillin Trihydrate),Nuflor®(Florfenicol),Zuprevo18%® (Tildipirosin)
200	M+IT	Nuflor® (Florfenicol), Zuprevo18%® (Tildipirosin)
201	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
202	M+IT	Nuflor® (Florfenicol)
203	M	
204	IT	Longocillina® (Amoxicillin Trihydrate), Nuflor® (Florfenicol)
205	IT	Nuflor® (Florfenicol)
206	M+IT	Nuflor® (Florfenicol)
207	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
208	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
209	M+IT	Bimoxylla (Amoxicillin),Zuprevo18%® (Tildipirosin)
210	M+IT	Zuprevo18%® (Tildipirosin)
211	M+IT	Zuprevo18%® (Tildipirosin)
212	M+IT	Forcyl (Marbofloxacin),Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
213	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
214	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
215	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
216	M+IT	Zuprevo18%® (Tildipirosin)
217	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
218	M+IT	Vetamplius®(Ampicillin), Zuprevo18%® (Tildipirosin), Forcyl (Marbofloxacin), Longocillina® (Amoxicillin Trihydrate)
219	IT	Zuprevo18%® (Tildipirosin), Depocillina® (Z) (Penicillin G procaine)
220	IT	Nuflor®(Florfenicol),Zuprevo18%® (Tildipirosin)
221	M+IT	Nuflor®(Florfenicol),Zuprevo18%® (Tildipirosin)

222	M+IT	Nuflor®(Florfenicol),Zuprevo18%® (Tildipirosin)
223	M+IT	Nuflor®(Florfenicol),Zuprevo18%® (Tildipirosin), Longocillina® (Amoxicillin Trihydrate)
224	M+IT	Vetamplius®(Ampicillin), Zuprevo18%® (Tildipirosin)
225	M+IT	Zuprevo18%® (Tildipirosin)
226	M+IT	Zuprevo18%® (Tildipirosin)
227	M+IT	Zuprevo18%® (Tildipirosin)
228	IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
229	IT	Zuprevo18%® (Tildipirosin)
230	M	
231	M	
232	NT	
233	NT	
234	NT	
235	M	
236	M	
237	M+IT	Nuflor® (Florfenicol)
238	M	
239	M+IT	Nuflor® (Florfenicol)
240	M+IT	Nuflor® (Florfenicol)
242	IT	Nuflor® (Florfenicol)
242	IT	Nuflor® (Florfenicol)
243	IT	Bimoxylla® (Amoxicillin), Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin), Vetamplius®(Ampicillin),Valemas10® (Enrofloxacina)
244	M+IT	Longocillina® (Amoxicillin Trihydrate), Zuprevo18%® (Tildipirosin), Nuflor® (Florfenicol)
245	M+IT	Longocillina® (Amoxicillin Trihydrate), Zuprevo18%® (Tildipirosin)
246	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
247	M+IT	Depocillina® (Penicillin G procaine), Valemas10® (Enrofloxacina), Zuprevo18%® (Tildipirosin)
248	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
249	M+IT	Zuprevo18%® (Tildipirosin)
250	M+IT	Forcyl® (Marbofloxacin), Longocillina® (Amoxicillin Trihydrate), Nuflor®(Florfenicol),Zuprevo18%® (Tildipirosin)
251	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
252	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
253	M+IT	Zuprevo18%® (Tildipirosin)
254	M+IT	Zuprevo18%® (Tildipirosin)
255	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
256	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
257	M+IT	Zuprevo18%® (Tildipirosin)
258	M+IT	Zuprevo18%® (Tildipirosin)

259	M+IT	Nuflor® (Florfenicol),Zuprevo18%® (Tildipirosin)
260	M+IT	Zuprevo18%® (Tildipirosin)
261	M+IT	Zuprevo18%® (Tildipirosin)
262	M+IT	Zuprevo18%® (Tildipirosin)
263	M+IT	Zuprevo18%® (Tildipirosin)
264	M+IT	Zuprevo18%® (Tildipirosin)

Abbreviations: ID=animal identification, IT= individual treatment M=metaphylactic treatment, M+IT=metaphylactic and individual treatment, NT=no treatment

Supplementary Table S4. Different non-steroid anti-inflammatory drugs used for BRD treatment in the fattening unit.

ID	NSAID treatment at T0 (commercial name and active principle)	NSAID treatment at T0 (commercial name and active principle)
1		
2		
3		
4	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
5		Metacam® (Meloxicam)
6		
7		
8		
9		
10		Metacam® (Meloxicam)
11		
12		
13		
14		
15	Finadyne® (Flunixin meglumina)	Finadyne® (Flunixin meglumina)
16		
17		
18		
19		
20		
21		
22		
23		
24		
25	Finadyne® (Flunixin meglumina)	
26	Finadyne® (Flunixin meglumina)	Finadyne® (Flunixin meglumina)
27		Metacam® (Meloxicam)

28	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
29		
30		
31		
32		
33	Finadyne® (Flunixin meglumina)	Finadyne® (Flunixin meglumina)
34	Finadyne® (Flunixin meglumina)	Dinalgen® (Ketoprofen), Salicil Vet® (Acetylsalicylic acid)
35		
36		
37		
38		
39	Metacam® (Meloxicam)	
40		
41	Finadyne® (Flunixin meglumina)	
42		
43		
44		
45		
46		Finadyne® (Flunixin meglumina)
47	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
48		Finadyne® (Flunixin meglumina)
49		
50	Metacam® (Meloxicam)	
51	Metacam® (Meloxicam)	
52		
53		Metacam® (Meloxicam)
54		
55		
56		
57		
58		
59		
60	Metacam® (Meloxicam)	
61		
62		
63		
64	Metacam® (Meloxicam)	
65		
66		
67		Finadyne® (Flunixin meglumina)

68	Metacam® (Meloxicam)	
69		
70	Metacam® (Meloxicam)	
71	Metacam® (Meloxicam)	
72	Metacam® (Meloxicam)	
73		
74		
75		
76		
77		
78		
79	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
80		
81		
82	Metacam® (Meloxicam)	
83	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina), Metacam®(Meloxicam)
84		
85	Finadyne® (Flunixin meglumina)	
86		
87		
88		
89	Metacam® (Meloxicam)	
90	Metacam® (Meloxicam)	
91		
92		
93		
94		
95	Metacam® (Meloxicam)	Metacam® (Meloxicam)
96		
97	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
98		
99		
100	Finadyne® (Flunixin meglumina)	
101		
102	Metacam® (Meloxicam)	
103		
10	Metacam® (Meloxicam)	
105	Metacam® (Meloxicam)	
106	Finadyne® (Flunixin meglumina), Metacam®(Meloxicam)	Finadyne® (Flunixin meglumina)
107		

108	Metacam® (Meloxicam)	
109	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
110	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
111		Finadyne® (Flunixin meglumina)
112	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
113	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
114	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
115		Metacam®(Meloxicam), Salicil Vet® (Acetylsalicylic acid)
116	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina), Metacam®(Meloxicam)
117	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
118	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
119	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
120		Finadyne® (Flunixin meglumina)
121		Finadyne® (Flunixin meglumina)
122	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
123		Salicil Vet® (Acetylsalicylic acid)
124		Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
125	Metacam® (Meloxicam)	
126		
127	Finadyne® (Flunixin meglumina)	
128		
129	Finadyne® (Flunixin meglumina), Metacam®(Meloxicam)	Dinalgen® (Ketoprofen), Salicil Vet® (Acetylsalicylic acid)
130	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
131	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
132	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
133	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
134		Salicil Vet® (Acetylsalicylic acid)
135		Salicil Vet® (Acetylsalicylic acid)
136		Salicil Vet® (Acetylsalicylic acid)
137	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
138	Finadyne® (Flunixin meglumina)	Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
139	Finadyne® (Flunixin meglumina)	Dinalgen® (Ketoprofen), Salicil Vet® (Acetylsalicylic acid)
140		Salicil Vet® (Acetylsalicylic acid)
141		Salicil Vet® (Acetylsalicylic acid)
142		Salicil Vet® (Acetylsalicylic acid)
143	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
144		Salicil Vet® (Acetylsalicylic acid)
145		Salicil Vet® (Acetylsalicylic acid)

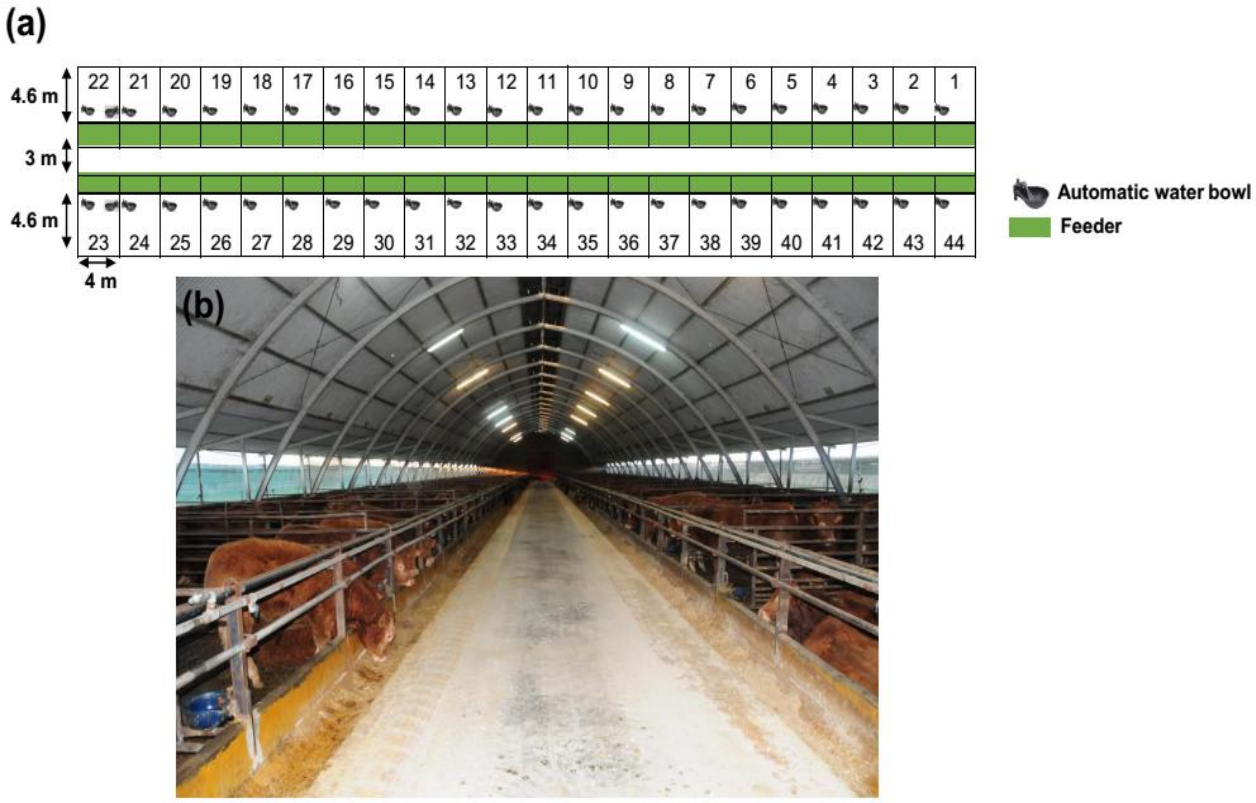
146		Salicil Vet® (Acetylsalicylic acid)
147	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
148		Salicil Vet® (Acetylsalicylic acid)
149		Finadine, Salicil Vet® (Acetylsalicylic acid)
150		Dinalgen® (Ketoprofen), Salicil Vet® (Acetylsalicylic acid)
151	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
152		Salicil Vet® (Acetylsalicylic acid)
153	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
154		Salicil Vet® (Acetylsalicylic acid)
155		Salicil Vet® (Acetylsalicylic acid)
156		Salicil Vet® (Acetylsalicylic acid)
157		Salicil Vet® (Acetylsalicylic acid)
158		Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
159		Salicil Vet® (Acetylsalicylic acid)
160		Salicil Vet® (Acetylsalicylic acid)
161	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
162		Salicil Vet® (Acetylsalicylic acid)
163	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
164	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
165	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
166	Salicil Vet® (Acetylsalicylic acid)	
167	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
168		Salicil Vet® (Acetylsalicylic acid)
169		Salicil Vet® (Acetylsalicylic acid)
170		Salicil Vet® (Acetylsalicylic acid)
171	Salicil Vet® (Acetylsalicylic acid)	
172	Salicil Vet® (Acetylsalicylic acid)	
173	Salicil Vet® (Acetylsalicylic acid)	
174	Salicil Vet® (Acetylsalicylic acid)	
175	Salicil Vet® (Acetylsalicylic acid)	
176	Metacam®(Meloxicam) Salicil Vet® (Acetylsalicylic acid)	
177	Salicil Vet® (Acetylsalicylic acid)	
178	Metacam® (Meloxicam) Salicil Vet®	
179	Salicil Vet® (Acetylsalicylic acid)	
180	Salicil Vet® (Acetylsalicylic acid)	
181	Salicil Vet® (Acetylsalicylic acid)	
182	Salicil Vet® (Acetylsalicylic acid)	
183	Salicil Vet® (Acetylsalicylic acid) Finadyne® (Flunixin meglumina)	
184	Salicil Vet® (Acetylsalicylic acid)	

185	Salicil Vet® (Acetylsalicylic acid)	Dinalgen® (Ketoprofen), Finadyne® (Flunixin meglumina)
186	Metacam® (Meloxicam), Salicil Vet® (Acetylsalicylic acid)t	Dinalgen® (Ketoprofen)
187	Salicil Vet® (Acetylsalicylic acid)	
188	Salicil Vet® (Acetylsalicylic acid)	
189	Salicil Vet® (Acetylsalicylic acid)	Dinalgen® (Ketoprofen)
190	Metacam®(Meloxicam), Salicil Vet® (Acetylsalicylic acid)	Dinalgen® (Ketoprofen)
191	Salicil Vet® (Acetylsalicylic acid)	
192	Salicil Vet® (Acetylsalicylic acid)	
193	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
194	Finadyne® (Flunixin meglumina)	Dinalgen® (Ketoprofen), Finadyne® (Flunixin meglumina), Metacam®(Meloxicam)
195	Finadyne® (Flunixin meglumina)	Dinalgen® (Ketoprofen)
196		Salicil Vet® (Acetylsalicylic acid)
197	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
198		Salicil Vet® (Acetylsalicylic acid)
199		Dinalgen® (Ketoprofen), Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
200	Metacam® (Meloxicam)	Dinalgen® (Ketoprofen), Salicil Vet® (Acetylsalicylic acid)
201	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
202		
203		
204	Finadyne® (Flunixin meglumina)	
205	Metacam® (Meloxicam)	
206		Finadyne® (Flunixin meglumina)
207	Metacam® (Meloxicam)	
208		Finadyne® (Flunixin meglumina)
209		Salicil Vet® (Acetylsalicylic acid)
210		Salicil Vet® (Acetylsalicylic acid)
211		Salicil Vet® (Acetylsalicylic acid)
212	Finadyne® (Flunixin meglumina), Metacam®(Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
213	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
214	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
215	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
216		Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
217	Finadyne® (Flunixin meglumina),Salicil Vet® (Acetylsalicylic acid)	
218	Salicil Vet® (Acetylsalicylic acid)	Finadyne® (Flunixin meglumina)
219		

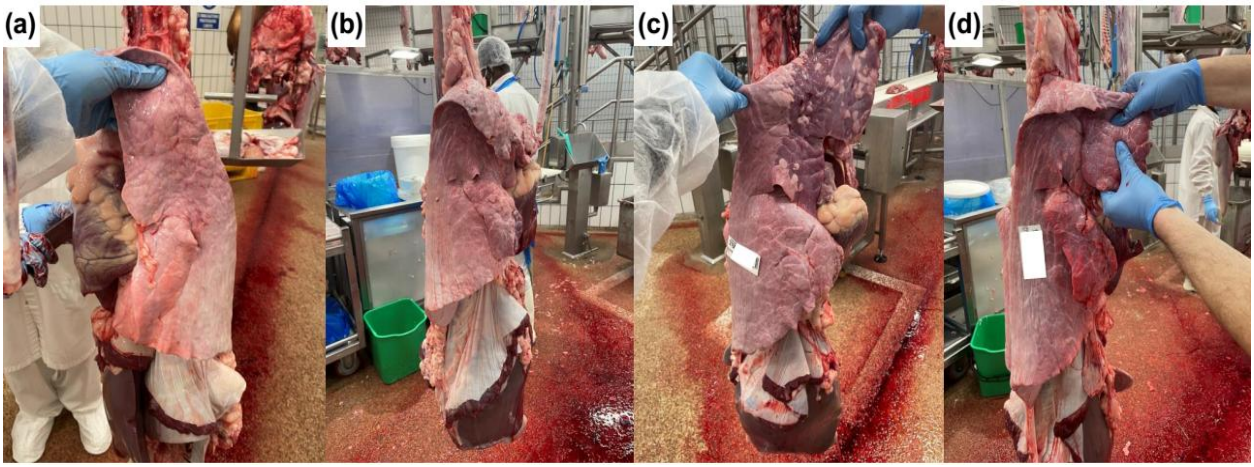
220	Metacam®(Meloxicam) Salicil Vet® (Acetylsalicylic acid)	
221	Metacam® (Meloxicam), Salicil Vet® (Acetylsalicylic acid)	
222	Metacam®(Meloxicam), Salicil Vet® (Acetylsalicylic acid)	Finadyne® (Flunixin meglumina)
223	Metacam®(Meloxicam), Salicil Vet® (Acetylsalicylic acid)	Finadyne® (Flunixin meglumina)
224	Salicil Vet® (Acetylsalicylic acid)	Finadyne® (Flunixin meglumina)
225	Salicil Vet® (Acetylsalicylic acid)	
226	Salicil Vet® (Acetylsalicylic acid)	
227	Salicil Vet® (Acetylsalicylic acid)	
228	Salicil Vet® (Acetylsalicylic acid), Metacam®(Meloxicam)	Finadyne® (Flunixin meglumina)
229	Salicil Vet® (Acetylsalicylic acid)	
230		
231		
232		
233		
234		
235		
236		
237	Metacam® (Meloxicam)	
238		
239	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
240	Metacam® (Meloxicam)	
242	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
242	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
243	Finadyne® (Flunixin meglumina), Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina)
244	Finadyne®, Metacam®(Meloxicam)	
245	Finadyne® (Flunixin meglumina)	Salicil Vet® (Acetylsalicylic acid)
246	Finadyne® (Flunixin meglumina)	Salicil Vet® (Acetylsalicylic acid)
247	Finadyne® (Flunixin meglumina)	
248	Finadyne® (Flunixin meglumina)	Salicil Vet® (Acetylsalicylic acid)
249		Salicil Vet® (Acetylsalicylic acid)
250	Finadyne® (Flunixin meglumina)	Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
251	Metacam®(Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
252	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
253		Salicil Vet® (Acetylsalicylic acid)
254		Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
255	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina),Salicil Vet® (Acetylsalicylic acid)

256	Metacam® (Meloxicam)	Finadyne® (Flunixin meglumina) Salicil Vet® (Acetylsalicylic acid)
257		Salicil Vet® (Acetylsalicylic acid)
258		Salicil Vet® (Acetylsalicylic acid)
259	Metacam® (Meloxicam)	Salicil Vet® (Acetylsalicylic acid)
260		Salicil Vet® (Acetylsalicylic acid)
261		Salicil Vet® (Acetylsalicylic acid)
262		Salicil Vet® (Acetylsalicylic acid)
263		Finadyne® (Flunixin meglumina), Salicil Vet® (Acetylsalicylic acid)
264		Salicil Vet® (Acetylsalicylic acid)

Abbreviations: ID=animal identification, NSAID=non-steroid anti-inflammatory drug.



Supplementary Figure S1. Structure of the commercial fattening unit. (a) Schematic representation of the structure of the commercial fattening unit, position of feeders and automatic water bowls, dimension of the pens and corridors. (b) image of the commercial fattening unit.



Supplementary Figure S2. Lung score based on an estimation of the extension of diseased parenchyma. **(a)** no evidence of parenchymal alteration (healthy); **(b)** parenchymal inflammatory lesions in 1 to 25% of the lung (mild pneumonia); **(c)** parenchymal inflammatory lesions in 25% to 50% of the lung (moderate pneumonia); **(d)** parenchymal inflammatory lesions in more than 50% of the lung (severe pneumonia).

CHAPTER V: NAVEL HEALING AND CALF FITNESS FOR TRANSPORT

ARTICLE

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Navel Healing and Calf Fitness for Transport

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Simple Summary: In the dairy industry, for male calves, the costing and balancing of animal welfare and farmers' interests when determining the optimum age for a calf to leave the farm of origin is a challenge. In the European Union, calves whose navel has not "completely healed" cannot be transported. This study aimed to clarify what is meant by "navel healing", as no specific definition is provided by the law, giving raise to different interpretations. The navels of 299 dairy calves (55 males, 244 females) aged 0–90 days were examined and scored. Our results show that a completely dry and shriveled navel stump entails a high risk of transporting too young calves, whilst the presence of a scab covering the umbilical wound could be considered acceptable for short journeys, as the risk of transporting calves that are too young is low. "Navel healing" should be defined as the scarring of the umbilical wound, which occurs no earlier than 3–4 weeks of life. Transporting calves with a completely healed navel should be considered best practice because it ensures that calves that are too young are not transported and therefore guarantees higher animal welfare standards.

Abstract: Dairy male calves are at risk of welfare compromise as they are usually transported at a very young age. The European Union has set a "completely healed navel" requirement for calf transport; moreover, a minimum age is established for longer journeys. However, this requirement has proven to be prone to misinterpretation. This study aimed to clarify what is meant by "navel healing" and to provide strong elements for reaching a consensus. The navels of 299 dairy calves (55 males, 244 females) aged 0–90 days were examined and scored 1 to 5 according to their healing status. Based on our results, a completely dry and shriveled navel (score 3) would imply a 25.5–38.0% risk of transporting too young calves. Alternatively, the presence of a scab covering the umbilical wound (score 4) would entail a 4.3% risk of transporting calves less than 10 days old and could be considered good practice for transporting calves (except for journeys exceeding 8 h). Conversely, complete navel healing (score 5) guarantees that calves that are too young are not transported; therefore, it should be considered best practice for transporting calves in general and the minimum requirement for transporting calves for journeys exceeding 8 h.

Keywords: calf; navel healing; umbilicus; transport; health; welfare; law

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1. Introduction

Live animal transport poses a risk to animal health and welfare and this risk is particularly relevant when the transported animals are young. The higher sensitivity of young animals, such as calves, to transport stress is due, on the one hand, to the immaturity of their immune system, the incomplete development of the hypothalamic–pituitary axis, and their reduced ability to thermoregulate and, on the other hand, to exposure to a multitude of novel stressful stimuli, such as handling, loading, weighing, regrouping with unfamiliar animals, deprivation of food and water, and fluctuating temperatures [1–4].

Up-to-date statistics on calf transport mortality rates are limited. In Australia, an average mortality rate of 0.64% has been reported for bobby calves (less than one week old) over the period 1998–2000 [5]. Later surveys carried out by the New Zealand Government reported a decrease in mortality rates from 0.68% in 2008, to 0.25% in 2015, and 0.12% in 2016, following farmer education campaigns and the implementation of voluntary initiatives [6]. A study documenting animal transits through the Southern Italian control post from 2010 to 2015 reported a mortality rate of 0.011% for medium-sized calves (<100 kg) and of 0.044% for heavy calves (200 kg), but no information was given regarding the age of the examined animals [7].

Even though few calves die during transport, a strong negative correlation has been shown between age at transport and mortality, which occurs within few weeks after transport due to secondary infections resulting from the impairment of immune system function caused by transport stress [8].

Dairy calves, especially males, are at particular risk of welfare compromise as they are usually transported at a very young age to slaughterhouses (e.g., in Australia and New Zealand) or to white veal production facilities (e.g., in European countries, including Italy) and they often show impaired health conditions upon arrival. Indeed, male gender has been identified as a significant risk factor for increased mortality and unwanted early slaughter in veal production facilities [9], presumably because the nursing care of dairy male calves might be neglected due to their low economic value [10,11].

The correlation between failure of passive transfer of colostral immunoglobulins, morbidity, and mortality is well documented [12,13]. However, studies on the relationship between good passive immunity transfer and infectious disease risk in calves have generated conflicting results regarding umbilical infections, thus suggesting that other management and environmental factors are also important [14,15].

Navel inflammation is one of the most common health problems reported upon arrival at white veal facilities or auction sites, with a prevalence ranging from 20% to 32% [16–19], and it has been associated with an increased risk of mortality in the first three weeks after transport [20]. Umbilical infections are among the main causes for neonatal calf mortality and carcass condemnation, as they often evolve into septicemia with endocarditis, arthritis, hepatitis, meningitis, and, eventually, death [21,22].

Calf age and navel condition are commonly used as indicators for fitness in several regulations for the protection of transported animals. In the European Union, according to the Council Regulation (EC) No 1/2005, calves cannot be transported if their navel "has not completely healed" (Annex I, chapter I). Moreover, "calves of less than 10 days of age" cannot be transported for more than 100 km (Annex I, chapter I) and for journeys that exceed 8 h they must be "older than 14 days" (Annex I, chapter VI) [23]. As another example, according to the Australian and New Zealand regulations, calves can be transported if they are at least 4 full days (96 h) old and their navel cord is wrinkled, withered, and shriveled [24,25]. In Canada, calves that are 8 days of age or less can be transported only once and must not have an unhealed or infected navel [26].

The scientific literature concerning the post-natal evolution of umbilical structures is limited. At birth, the umbilical stump is red-pink, flexible and hydrated. It then undergoes a drying and mummification process; at 5 days of age it turns brown-black and becomes inflexible and shriveled. At about 14 days of age it falls off, leaving a wound that is soon covered by a scab. Around 3–4 weeks of age, the umbilical wound is completely healed [27,28].

Since the healing process of the umbilical wound seems to extend well beyond 10 days of age, what the European legislator means by the term "healed" seems unclear. Contradictory terminology has also been found in the OIE Terrestrial Animal Health Code, where article 7.3.7 states that "animals that are unfit to travel include newborns with an unhealed navel", while article 7.11.7 says that "recently born calves should not be transported until the navel is dry" [29].

The aim of the present study is to contribute to clarify what is meant by "navel healing", since no specific definition is provided by the law, giving raise to different interpretations. The ultimate aim is to provide strong elements for reaching a consensus among farmers and veterinary practitioners in order to comply with the European regulation. In order to do so, a review of the available literature on navel healing and the direct examination of calves in the first weeks of life were performed. A scoring system to help to avoid misinterpretation of the healing status of the navel is presented.

2. Materials and Methods

A total of 299 dairy calves (55 males, 244 females) aged 0–90 days were included in this study. The calves were mainly Holstein breed (n = 201) and crossbreds (n = 98) reared in 5 dairy cattle farms located in Bologna and Modena provinces (Emilia-Romagna region). Calves were housed in ground-level individual igloos (4 farms) or elevated cages (1 farm) with straw bedding up to 2 months of age and then placed in group pens. The hygiene level of the pens and straw bedding was between good and excellent. The pens were located outdoors in a sheltered area, protected from adverse climatic conditions. All calves were identified with two ear tags bearing the calf's identification number and the date of birth. Only data obtained from farms where birth registration was reliable were used. In all farms it was standard practice to treat the navel stump of both male and female calves with a tetracycline-based spray at least once at birth. However, it was not always possible to verify whether this was done. It was also not possible to collect individual information on colostrum administration.

The calves' umbilical stumps were inspected and palpated on a single occasion between January and March 2021. Based on the post-natal evolution of the umbilical structures described in the literature [27,28], a score corresponding to the different stages of umbilical healing was attributed as follows: (1) red-pink color, hydrated, flexible; (2) crimson-purple color, flattened, dry in its distal portion; (3) brown-black color, completely dry and shriveled, inflexible; (4) no umbilical stump, but scab or granulation tissue on the umbilical wound; (5) completely healed umbilical wound (Figure 1). The inspections were carried out by three observers (M.B., A.G., M.R.) with experience in bovine medicine. The first visit was carried out together and the score was agreed upon. Calves showing signs of omphalitis (e.g., enlarged umbilicus, pain upon palpation, draining of purulent material) were excluded from the study.

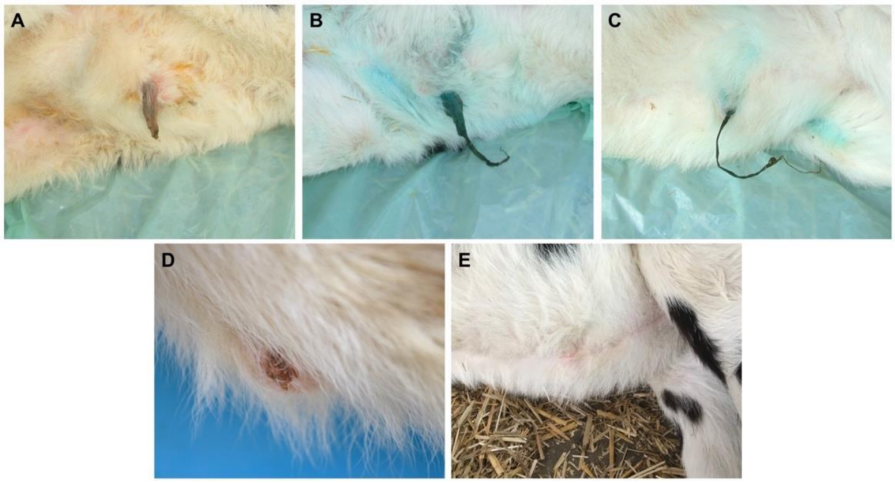

Figure 1. Healing status of the umbilical wound and corresponding score: (**A**) red-pink color, hydrated, flexible (score 1); (**B**) crimson-purple color, flattened, dry in its distal portion (score 2); (**C**) brown-black color, completely dry and shriveled, inflexible (score 3); (**D**) no umbilical stump, but scab on the umbilical wound (score 4); (**E**) completely healed umbilical wound (score 5).

Statistical analysis was performed using Microsoft Excel (v16.16.10, Microsoft, 2018) and XLSTAT (v2016.5, Addinsoft, 2016).

The correlation between calf age and umbilical score was investigated by calculating the Spearman's rank correlation coefficient (the significance level was set at 0.05).

The two variables "calf age" and "umbilical score" were then converted into binary variables and square contingency tables were generated analyzing different scenarios. Regarding calf age, the threshold was either set at 10 days (journeys longer than 100 km), or 14 days (journeys exceeding 8 h), as clearly established by law. Regarding umbilical score, given the lack of clarity on what the European legislator means by "healed navel", three possible cases were considered: in the first case, the threshold was set at score 3 (completely dry and shriveled, inflexible umbilical stump); in the second case, the threshold was set at score 4 (no umbilical stump, but scab or granulation tissue on the umbilical wound); in the third case, the threshold was set at score 5 (completely healed umbilical wound). The probability of finding the relevant observed values was calculated.

3. Results

The age distribution of the calves included in our sample was as follows: 0–9 days (n = 94), 10–14 days (n = 35), 15–20 days (n = 43), 21–30 days (n = 51), 31–40 days (n = 29), 41–50 days (n = 24), and 51–90 days (n = 23).

The age distribution of calves with different umbilical scores is shown in Table 1.

Table 1. Age distribution of calves with different umbilical scores (percentage in brackets).

Umbilical Score	Age Range (Days)							Total
	0–9	10–14	15–20	21–30	31–40	41–50	51–90	
1	16 (100)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	16
2	9 (75)	1 (8)	2 (17)	0 (0)	0 (0)	0 (0)	0 (0)	12
3	63 (47)	28 (21)	22 (16)	13 (10)	4 (3)	4 (3)	0 (0)	134
4	6 (9)	6 (9)	17 (26)	16 (25)	17 (26)	1 (2)	2 (3)	65
5	0 (0)	0 (0)	2 (3)	22 (31)	8 (11)	19 (26)	21 (29)	72

The umbilical score was moderately correlated with calf age (R= 0.604; $p < 0.0001$).

All the calves with umbilical score 1 were less than 10 days old. The majority of calves with score 2 were less than 10 days of age; however, the navel stump was dry only in its distal portion also in a 10-day-old calf and in two 15-day-old calves. The 47% of the calves with a completely dry and shriveled navel stump (score 3) were aged less than 10 days, but the navel stump was still present even in calves over one month old (6%). Conversely, calves with score 4 (no umbilical stump, but scab or granulation tissue on the umbilical wound) were distributed over all the observed age ranges, from 1 day to 2 months old. Finally, in our sample, only two calves less than 3 weeks old (specifically, 19 and 20 days of age) had a completely healed navel, i.e., only 3% of all calves with score 5.

The contingency tables created for the analysis of the different scenarios are shown in Tables S1–S6; the results are summarized in Table 2, which shows the probability that calves with an umbilical score of at least 3 (completely dry and shriveled, inflexible umbilical stump), 4 (no umbilical stump, but scab or granulation tissue on the umbilical wound), or 5 (completely healed umbilical wound) were less than 10 days old (fit for journeys within 100 km), at least 10 days old (fit for journeys longer than 100 km), or older than 14 days (fit for journeys exceeding 8 h).

Table 2. Probability and 95% C.I. (%, in brackets) that calves with an umbilical score of at least 3 (completely dry and shriveled, inflexible umbilical stump), 4 (no umbilical stump, but scab or granulation tissue on the umbilical wound), or 5 (completely healed umbilical wound) were less than 10 days old (fit for journeys within 100 km), at least 10 days old (fit for journeys longer than 100 km), or older than 14 days (fit for journeys exceeding 8 h).

Calf Age	Umbilical Score		
	≥3	≥4	5
<10 d	25.5% (20.6–31.0)	4.3% (2.0–9.2)	0% (0–4.9)
≥10 d	74.5% (69.0–79.4)	95.7% (90.8–98.0)	100% (95.1–100)
>14 d	62.0% (56.1–67.6)	91.3% (85.4–94.9)	100% (95.1–100)

4. Discussion

Calf age is a critical factor affecting transport-related stress, and a strong negative correlation exists between age at transport and mortality [8]. Nevertheless, there is no scientific consensus on the optimal age for transporting calves [30].

Navel inflammation is among the most common diseases and causes of mortality upon arrival to white veal facilities of shortly after in transported calves, with a prevalence of up to 32% [14,16,17,21]. Furthermore, the pain associated with navel inflammation has been identified as a factor reducing calves' lying time during the first 2 weeks after transport, thus interfering with disease recovery [31].

The legislator's response to these challenges has been to develop and implement regulations that limit calf age and health status as well as transport duration and relate to conditions. For example, the European Union has set a "completely healed navel" for calves to be deemed fit for transport; moreover, a minimum age of 10 or 14 days is required for journeys that exceed 100 km or 8 h, respectively.

Apart from a few exceptions [27,28], research on the most authoritative textbooks of veterinary internal medicine, clinical pathology, surgical pathology, and obstetrics failed to provide information on the healing times of the umbilical wound [32–36].

On the basis of the available literature, the navel stump falls at 14 days of age on average. This event is followed by the formation of a firm, thick scab, which takes a few days to occur and precedes healing. As the various umbilical structures heal at different times, it is necessary to ensure that the external umbilicus (i.e., the main entry point for pathogens and the only structure that can be assessed on inspection) is completely healed. The physiological endpoint of mammalian wound repair displays the formation of a scar [37]. Therefore, so-called "navel healing" should be defined as the scarring of the umbilical wound (score 5), which occurs no earlier than 3–4 weeks of life.

The results of our study corroborate this concept. In our sample, 90% of all calves up to 14 days old still had the navel stump, which in most cases was completely dry and shriveled (score 3). Most of the calves with score 4 (no umbilical stump, but scab or granulation tissue on the umbilical wound) were between 15 and 40 days old. However, 18% of calves with score 4 were between 1 and 14 days of age; it was not possible to determine whether this was due to early navel stump detachment or whether it resulted from a rupture of the navel stump at the external umbilicus at birth. The youngest calves with a completely healed navel (score 5) were 19 and 20 days old, whilst the highest percentage of calves with score 5 (31%) was between 21 and 30 days old. We cannot exclude that the navels of the calves over 1-month-old had healed before that age; however, it is highly unlikely that a calf will have a completely healed navel (score 5) before 3 weeks of age.

Our data support the notion that only a completely healed navel (score 5) would prevent calves from being transported too young. In fact, according to our findings, if the minimum requirement were a completely dried and shriveled navel (score 3), there would be a 25.5% risk of transporting calves less than 10 days old for journeys over

100 km, and a 38% risk of transporting calves younger than 14 days old for journeys exceeding 8 h. Alternatively, a score of 4 (no umbilical stump, but scab or granulation tissue on the umbilical wound) as a minimum requirement would still entail a risk of transporting calves that are too young, albeit much lower (i.e., 4.3% risk of transporting calves less than 10 days old for journeys over 100 km and 8.7% risk of transporting calves younger than 14 days old for journeys exceeding 8 h). Conversely, with score 5 it would be reasonably certain that calves are at least 10 days old or older than 14 days.

Consequently, both in score 3 and score 4 scenarios, other elements would be necessary to verify whether the age of the calf complies with the legal requirements concerning transport duration. How, though, can one be sure of the calf's age? Calf age cannot be reliably determined from physiological or physical characteristics. A recent study investigating the accuracy of serum gamma glutamyl transferase and body weight as predictors of age in young calves found only a moderate correlation [38]. Given this limitation, the only element to determine it is documentation. In the European Union, according to Regulation (EC) No 1760/2000 and, since 21 April 2021, the Commission Implementing Regulation (EU) 2021/520 laying down rules for the application of Regulation (EU) 2016/429, with regard to the traceability of certain kept terrestrial animals, bovine animals shall be identified by a physical means of identification approved by the competent authority (e.g., ear tag) bearing a unique identification code, which makes it possible to identify each animal individually along with the holding on which it was born. The means of identification may be applied and registered by the farmer on the national database within a period that shall not exceed 20 days from the date of birth and in any case before the animals are moved from the farm. As a result of this time allowance, it is possible that some calves are falsely registered as having an older age so that they can be transported earlier. Against this background, the element on which competent authorities base their judgement in determining calf fitness for transport is navel status.

The navel healing process may also be affected by different navel care practices. In a study investigating the efficacy of light-emitting diode (LED) phototherapy on navel healing in 57 newborn calves, the umbilical stump of all animals fell off by the 25th day of age; on average, it fell off 3 days earlier in LED-treated calves ($p < 0.01$). At 30 days of age, the umbilical wound had healed in almost all the LED-treated calves (96.4%) but only in 69% of the calves in the control group [39]. In another study, investigating the efficacy of different antiseptic compounds in a sample of 73 Holstein heifer calves, the umbilical stump fell off at 16.3 $\pm$ 7.0 days of age; in calves dipped with 4% chlorhexidine mixed with alcohol (50:50), the umbilical stump detached at an average of 20 days compared with 15.5 days for the other three treatments (7% iodine, dry nisin, liquid nisin), but this difference was not statistically significant. No information was provided for the healing times of the umbilical wound [40].

To the authors' knowledge, there is only one study investigating the drying times of umbilical stumps of dairy calves, whose observations, however, are limited to the first 8 days of life. Nonetheless, it demonstrated that dryness is a poor indicator of age [41].

In the dairy industry, male calves, especially pure-bred dairy calves, represent a cost in terms of feed and space required for raising them; therefore, balancing animal welfare and farmers' interests when determining the optimum age for a calf to leave the farm of origin is a challenge.

Despite the Council Regulation (EC) No 1/2005 establishing a "completely healed navel" as an indicator of calf fitness for transport, this requirement has proven to be prone to misinterpretation and consensus among veterinary practitioners and stakeholders is currently weak.

According to our results, a completely dried and shriveled navel stump (score 3) or the presence of a scab or granulation tissue covering the umbilical wound (score 4) can be observed in calves of highly variable age (from 1 day to 2 months old), and therefore cannot guarantee that calves that are too young will not be transported.

"Navel healing" should be defined as the scarring of the umbilical wound (score 5), which occurs no earlier than 3–4 weeks of life. However, if this were the interpretation intended by the European legislator, it is clearly difficult to associate navel healing with the minimum age thresholds of 10 and 14 days established for longer journeys.

5. Conclusions

In conclusion, the need to address the contradiction in the European Regulation between navel condition and the minimum age at which calves can be transported for longer journeys is evident. Therefore, clarification on what is meant by "navel healing" from an anatomic-physiological point of view is required.

Alternatively, in order to reach a compromise between balancing animal welfare and farmers' interests, good and best practice could be proposed. The presence of a scab on the umbilical wound (score 4) could represent the minimum requirement for transporting calves (except for journeys exceeding 8 h). This could be considered good practice, as the risk of transporting too young calves would be only 4.3%. Complete navel healing (score 5), since it entails 0% risk of transporting too young calves, should be considered best practice for transporting calves in general and the minimum requirement for transporting calves for journeys exceeding 8 h.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/ani12030358/s1, Table S1: Contingency table comparing umbilical score (at least 3—completely dry and shriveled umbilical stump—or less) and calf age (at least 10 days—journeys longer than 100 km—or less), Table S2: Contingency table comparing umbilical score (at least 3—completely dry and shriveled umbilical stump—or less) and calf age (older than 14 days—journeys exceeding 8 h—or less), Table S3: Contingency table comparing umbilical score (at least 4—no umbilical stump, but scab or granulation tissue on the umbilical wound—or less) and calf age (at least 10 days—journeys longer than 100 km—or less), Table S4: Contingency table comparing umbilical score (at least 4—no umbilical stump, but scab or granulation tissue on the umbilical wound—or less) and calf age (older than 14 days—journeys exceeding 8 h—or less), Table S5: Contingency table comparing umbilical score (5—completely healed umbilical wound—or less) and calf age (at least 10 days—journeys longer than 100 km—or less), Table S6: Contingency table comparing umbilical score (5—completely healed umbilical wound—or less) and calf age (older than 14 days—journeys exceeding 8 h—or less).

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Institutional Review Board Statement: This study does not fall within Directive 63/2010/EU of the European Parliament and of the Council on the protection of animals used for scientific purposes and therefore it does not require any authorization from the national competent authorities. It was approved by the Ethics Committee of the University of Bologna (Protocol No. 268584, 19 October 2021). The animals were examined with the owners' consent and in accordance with good veterinary practice.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data presented in this study are openly available in Zenodo at https://doi.org/10.5281/zenodo.5776839 (accessed on 29 January 2022).

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Supplementary Material

Navel Healing and Calf Fitness for Transport

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Table S1. Contingency table comparing umbilical score (at least 3 – completely dry and shriveled umbilical stump – or less) and calf age (at least 10 days – journeys longer than 100 km – or less).

Umbilical score	Calf age		Total
	< 10 d	≥ 10 d	
< 3	25	3	28
≥ 3	69	202	271
Total	94	205	299

Table S2. Contingency table comparing umbilical score (at least 3 – completely dry and shriveled umbilical stump– or less) and calf age (older than 14 days – journeys exceeding 8 hours – or less).

Umbilical score	Calf age		Total
	≤ 14 d	> 14 d	
< 3	26	2	28
≥ 3	103	168	271
Total	129	170	299

Table S3. Contingency table comparing umbilical score (at least 4 – no umbilical stump, but scab or granulation tissue on the umbilical wound – or less) and calf age (at least 10 days – journeys longer than 100 km – or less).

Umbilical score	Calf age		Total
	< 10 d	≥ 10 d	
< 4	88	73	161
≥ 4	6	132	138
Total	94	205	299

Table S4. Contingency table comparing umbilical score (at least 4 – no umbilical stump, but scab or granulation tissue on the umbilical wound – or less) and calf age (older than 14 days – journeys exceeding 8 hours – or less).

Umbilical score	Calf age		Total
	≤ 14 d	> 14 d	
< 4	117	44	161
≥ 4	12	126	138
Total	129	170	299

Table S5. Contingency table comparing umbilical score (5 – completely healed umbilical wound – or less) and calf age (at least 10 days – journeys longer than 100 km – or less).

Umbilical score	Calf age		Total
	< 10 d	≥ 10 d	
< 5	94	133	227
5	0	72	72
Total	94	205	299

Table S6. Contingency table comparing umbilical score (5 – completely healed umbilical wound– or less) and calf age (older than 14 days – journeys exceeding 8 hours – or less).

Umbilical score	Calf age		Total
	≤ 14 d	> 14 d	
< 5	129	98	227
5	0	72	72
Total	129	170	299

6. GENERAL CONCLUSIONS

The current thesis work as it was reported above has four objectives that were discussed in four different chapters. The welfare of farm animal's concept is on the rise in the world and much attention is given especially developed nations like European Union (EU) countries and other in the west nations. In order to improve the welfare of farm animals the European Union developed legislations concerning the farming, transportation, slaughtering of different farm animals; and the European Union (EU) is also working to even improve and update the existing farm animal legislation by 2023 so that to meet the public demands, and attain high level of farm animal welfare. In the less developed nation in Africa like Ethiopia the situation of farm animal welfare may not be even a topic for the wide populations due to different factors related; and most people may not consider in general animal welfare as a priority since the countries are struggling with poverty. Factors like poor economic growth, poverty, poor veterinary and farm infrastructure, shortage of animal feed, high infectious and non-infectious disease burden, lack of awareness among farmers and pastoralists even among the wide consumers about animal welfare were some of the reason that hinders the farm animal welfare issues. Most countries in Africa lack relevant and up-to-date animal welfare policies and laws, in Ethiopia there is no currently working animal welfare legislation and policy. Countries in Africa could learn a lot from European Union animal welfare legislations, but I am not saying the European Union (EU) legislation could be directly applied but with modification and customization it could serve as a starting point rather than starting from a scratch. So, in most African countries there should be proper identification of the main welfare issues through proper risk analysis, welfare assessments and work on the community on awareness creation, work on the farm animal welfare so that by indicating how it can benefit the community even, because solely rising the farm animal issues many not help only for the benefit of the animals but it should be integrated with the benefits that it brings, to the community itself or to the lowland pastoralists or high land farmers.

The regular assessment of farm animal welfare is important so as to improve the well-being of the farming animals so that after identifying the critical points that result in a poor welfare. The welfare and biosecurity assessment tool that we used which is part of ClassyFarm is an important protocol designed to assess welfare of farm animals in Italy. It helps to identified points that should be

improved in handling of the farm animals. We used these protocol to assess the welfare of beef cattle, and in addition we collected blood samples, so as to see the welfare situation of the beef cattle. We can conclude that the first two weeks were difficult periods for beef cattle after they were introduced to the fattening unit. We also observed that the beef cattle's suffers from Bovine respiratory disease (BRD) especially in the first two weeks and there is high circulation of *M. bovis* in the fattening unit that we investigated, but also we could not rule out the possibility of involvement of other probable cause of BRD since we did not test all the probable cause. There is a difficulty in treatment of BRD using antimicrobials in the farm where we performed the study even though the reasons could be a lot. As a conclusion BRD is one of the most serious health and welfare issues of beef cattle; so that detail studies should continue to better understand the disease prevention, avoid the risk factors, develop better treatment options.

Transportation is one of stressful operation in farm animals and it is consider one of factors that hampers welfare of farm animal predisposing them to different disease conditions. In European Union (EU) there is legislation regarding the welfare of claves under age of six months, Council Directive 2008/119/EC (Calves Directive). The transport is regulated under Council Regulation (EC) No 1/2005, and states that ''calves cannot be transported if their navel has not completely healed''. So that there should be a clear cut point when to say the naval is heal so that to transport the calves. Our findings indicate that a fully dry and shriveled navel stump has a high risk of transporting calves that are too young, although a scab covering the umbilical wound may be deemed suitable for short voyages with a minimal risk of doing so. "Navel healing" should be defined as the scarring of the umbilical wound, which occurs no earlier than 3–4 weeks of life. So that transporting of calves with a complete healed navel is best practice since it gives a guarantee not to transport calves that are too young. This work could give a clarification about navel healing and transportation, so that could give a point to improve on the legislation to clarify what it meant to transport a calve with a complete navel healing so that to have a good welfare standards of claves during transportation.

7. APPENDIX

Annex 1: EU farm animal directive (Council Directive 98/58/EC of 20 July 1998 concerning the protection of animals kept for farming purposes (OJ L 221 08.08.1998, p. 23, ELI: http://data.europa.eu/eli/dir/1998/58/oj) (Accessed on November 10, 2022)

COUNCIL DIRECTIVE 98/58/EC
of 20 July 1998
concerning the protection of animals kept for farming purposes

THE COUNCIL OF THE EUROPEAN UNION,

Having regard to the Treaty establishing the European Community, and in particular Article 43 thereof,

Having regard to the proposal from the Commission (1),

Having regard to the opinion of the European Parliament (2),

Having regard to the opinion of the Economic and Social Committee (3),

Whereas all Member States have ratified the European Convention for the Protection of Animals Kept for Farming Purposes (hereinafter called 'the Convention'); whereas the Community has also approved this Convention by Decision 78/923/EEC (4) and has deposited its instrument of approval;

Whereas the Community, as a contracting party, must give effect to the principles laid down in the Convention;

Whereas those principles include the provision of housing, food, water and care appropriate to the physiological and ethological needs of the animals, in accordance with established experience and scientific knowledge;

Whereas it is also necessary for the Community to make further provision for the uniform application of the Convention and its recommendations and for specific rules concerning the application of this Directive;

Whereas the European Parliament, in its resolution of 20 February 1987 on animal welfare policy (5) called on the Commission to make proposals for Community rules covering general aspects of the rearing of livestock;

Whereas Declaration No 24 annexed to the Final Act of the Treaty on European Union calls on the European institutions and the Member States, when drafting and implementing Community legislation, in particular on the common agricultural policy, to pay full regard to the welfare requirements of animals;

Whereas differences which may distort conditions of competition interfere with the smooth running of the organisation of the market in animals;

Whereas there is therefore a need to establish common minimum standards for the protection of animals kept for farming purposes in order to ensure rational development of production and to facilitate the organisation of the market in animals; whereas to that end it is appropriate to take account of animal welfare provisions already laid down in Community rules;

(1) OJ C 156, 23. 6. 1992, p. 11.
(2) OJ C 337, 21. 12. 1992, p. 225.
(3) OJ C 332, 16. 12. 1992, p. 22.
(4) OJ L 323, 17. 11. 1978, p. 12.
(5) OJ C 76, 23. 3. 1987, p. 185.

Whereas a comparative examination of animal welfare provisions applicable in the Community and in certain non-member countries together with an appraisal thereof should be undertaken with a view to determining the nature of future Community initiatives aimed at eliminating distortions of competition,

HAS ADOPTED THIS DIRECTIVE:

Article 1

1. This Directive lays down minimum standards for the protection of animals bred or kept for farming purposes.

2. It shall not apply to:

(a) animals living in the wild;

(b) animals intended for use in competitions, shows, cultural or sporting events or activities;

(c) experimental or laboratory animals;

(d) any invertebrate animal.

3. This Directive shall apply without prejudice to specific Community rules laid down elsewhere, and in particular to Directives 88/166/EEC (6), 91/629/EEC (7) and 91/630/EEC (8), which shall continue to apply.

Article 2

For the purposes of this Directive the following definitions shall apply:

1. 'animal': any animal (including fish, reptiles or amphibians) bred or kept for the production of food, wool, skin or fur or for other farming purposes;

(6) Council Directive 88/166/EEC of 7 March 1998 complying with the judgment of the Court of Justice in Case 131-86, (annulment of Council Directive 86/113/EEC of 25 March 1986 laying down minimum standards for the protection of laying hens kept in battery cages) (OJ L 74 19. 3. 1988, p. 83).
(7) Council Directive 91/629/EEC of 19 November 1991 laying down minimum standards for the protection of calves (OJ L 340, 11. 12. 1991, p. 28). Directive as last amended by Directive 97/2/EC (OJ L 25, 28. 1. 1997, p. 24).
(8) Council Directive 91/630/EEC of 19 November 1991 laying down minimum standards for the protection of pigs (OJ L 340, 11. 12. 1991, p. 33).

2. 'owner' or 'keeper': any natural or legal person or persons responsible for or in charge of animals whether on a permanent or temporary basis;

3. 'competent authority': the competent authority within the meaning of Article 2(6) of Council Directive 90/425/EEC of 26 June 1990 concerning veterinary and zootechnical checks applicable in intra-Community trade in certain live animals and products with a view to the completion of the internal market [1].

Article 3

Member States shall make provision to ensure that the owners or keepers take all reasonable steps to ensure the welfare of animals under their care and to ensure that those animals are not caused any unnecessary pain, suffering or injury.

Article 4

Members States shall ensure that the conditions under which animals (other than fish, reptiles or amphibians) are bred or kept, having regard to their species and to their degree of development, adaptation and domestication, and to their physiological and ethological needs in accordance with established experience and scientific knowledge, comply with the provisions set out in the Annex.

Article 5

1. The Commission shall submit to the Council any proposals which may be necessary for the uniform application of the European Convention for the Protection of Animals Kept for Farming Purposes and, on the basis of a scientific evaluation, any recommendations made under this Convention and any other appropriate specific rules.

2. In addition, every five years and for the first time five years after the date of entry into force of this Directive, the Commission, on the basis of experience acquired since the implementation of this Directive, in particular concerning the measures referred to in paragraph 1 and technical and scientific developments, shall submit to the Council a report, accompanied by any appropriate proposals taking into account the report's conclusions.

3. The Council shall act by qualified majority on these proposals.

Article 6

1. Member States shall ensure that inspections are carried out by the competent authority to check compliance with the provisions of this Directive. Such inspections may be carried out at the same time as checks for other purposes.

2. From a date to be determined in accordance with the procedure laid down in paragraph 3, Member States shall submit to the Commission reports on the inspections required under paragraph 1. The Commission shall submit summaries of those reports to the Standing Veterinary Committee.

3. The Commission shall before 1 July 1999, in accordance with the procedure laid down in Article 9 submit proposals with a view to harmonising:

(a) the inspections required under paragraph 1;

(b) the format, content and frequency of submission of the reports referred to in paragraph 2.

Article 7

1. Whenever uniform application of the requirements of this Directive renders it necessary, veterinary experts from the Commission may, in conjunction with the competent authorities;

(a) verify that the Member States are complying with the said requirements;

(b) make on-the-spot checks to ensure that the checks are carried out in accordance with this Directive.

2. A Member State in whose territory an inspection is made shall provide the veterinary experts from the Commission with any assistance they may require in the performance of their tasks. The outcome of the checks made must be discussed with the competent authority of the Member State concerned before a final report is drawn up and circulated.

3. The competent authority of the Member State concerned shall take any measures which may be necessary to take account of the results of the check.

4. Detailed rules for the application of this Article shall be adopted, where necessary, in accordance with the procedure laid down in Article 9.

Article 8

1. Before 30 June 1999 the Commission shall submit to the Council a report on:

— the comparison between animal welfare provisions in the Community and in non-member countries which supply the Community,

— the scope for obtaining wider international acceptance of the welfare principles laid down in this Directive, and

— the extent to which Community objectives in relation to animal welfare may be liable to be undermined as a result of competition from non-member countries which do not apply equivalent standards.

2. The report referred to in paragraph 1 shall be accompanied by any necessary proposals with the aim of eliminating distortions of competition.

[1] OJ L 224, 18. 8. 1990, p. 29. Directive as last amended by Directive 92/118/EEC (OJ L 62, 15. 3. 1993, p. 49).

Article 9

1. Where the procedure laid down in this Article is to be followed, the matter shall be referred without delay to the Standing Veterinary Committee set up by Directive 68/361/EEC [1], hereinafter referred to as 'the Committee', by its chairman acting either on his own initiative or at the request of a Member State.

2. The representative of the Commission shall submit to the Committee a draft of the measures to be taken. The Committee shall deliver its opinion on the draft within a time limit which the chairman may lay down according to the urgency of the matter. The opinion shall be delivered by the majority laid down in Article 148(2) of the Treaty in the case of decisions which the Council is required to adopt on a proposal from the Commission. The votes of the representatives of the Member States within the Committee shall be weighted in the manner set out in that Article. The chairman shall not vote.

3. (a) The Commission shall adopt the measures envisaged if they are in accordance with the opinion of the Committee.

(b) If the measures envisaged are not in accordance with the opinion of the Committee, or if no opinion is delivered, the Commission shall without delay submit to the Council a proposal relating to the measures to be taken. The Council shall act by qualified majority.

If, on the expiry of a period of three months from the date of referral to the Council, the Council has not acted, the Commission shall adopt the proposed measures and implement them immediately, save where the Council has decided against the said measures by a simple majority.

Article 10

1. Member States shall bring into force the laws, regulations and administrative provisions, including any sanctions, necessary to comply with this Directive not later than 31 December 1999, subject to any different decision taken by the Council in the light of the report referred to in Article 8. They shall forthwith inform the Commission thereof.

When Member States adopt these measures, they shall contain a reference to this Directive or shall be accompanied by such reference on the occasion of their official publication. The methods of making such reference shall be laid down by Member States.

2. However, after 31 December 1999, Member States may, in compliance with the general rules of the Treaty, maintain or apply within their territories stricter provisions for the protection of animals kept for farming purposes than those laid down in this Directive. They shall inform the Commission of any such measures.

3. Member States shall communicate to the Commission the texts of the main provisions of national law which they adopt in the field governed by this Directive.

Article 11

This Directive shall enter into force on the day of its publication in the *Official Journal of the European Communities*.

Article 12

This Directive is addressed to the Member States.

Done at Brussels, 20 July 1998.

For the Council
The President
W. MOLTERER

[1] OJ L 255, 18. 10. 1968, p. 23.

ANNEX

Staffing

1. Animals shall be cared for by a sufficient number of staff who possess the appropriate ability, knowledge and professional competence.

Inspection

2. All animals kept in husbandry systems in which their welfare depends on frequent human attention shall be inspected at least once a day. Animals in other systems shall be inspected at intervals sufficient to avoid any suffering.

3. Adequate lighting (fixed or portable) shall be available to enable the animals to be thoroughly inspected at any time.

4. Any animal which appears to be ill or injured must be cared for appropriately without delay and, where an animal does not respond to such care, veterinary advice must be obtained as soon as possible. Where necessary sick or injured animals shall be isolated in suitable accomodation with, where appropriate, dry comfortable bedding.

Record keeping

5. The owner or keeper of the animals shall maintain a record of any medicinal treatment given and of the number of mortalities found to each inspection.

   Where equivalent information is required to be kept for other purposes, this shall also suffice for the purposes of this Directive.

6. These records shall be retained for a period of at least three years and shall be made available to the competent authority when carrying out an inspection or when otherwise requested.

Freedom of movement

7. The freedom of movement of an animal, having regard to its species and in accordance with established experience and scientific knowledge, must not be restricted in such a way as to cause it unnecessary suffering or injury.

   Where an animal is continuously or regularly tethered or confined, it must be given the space appropriate to its physiological and ethological needs in accordance with established experience and scientific knowledge.

Buildings and accommodation

8. Materials to be used for the construction of accommodation, and in particular for the construction of pens an equipment with which the animals may come into contact, must not be harmful to the animals and must be capable of being throughly cleaned and disinfected.

9. Accommodation and fittings for securing animals shall be constructed and maintained so that there are no sharp edges or protrusions likely to cause injury to the animals.

10. Air circulation, dust levels, temperature, relative air humidity and gas concentrations must be kept within limits which are not harmful to the animals.

11. Animals kept in buildings must not be kept either in permanent darkness or without an appropriate period of rest from artificial lighting. Where the natural light available is insufficient to meet the physiological and ethological needs of the animals, appropriate artificial lighting must be provided.

Animals not kept in buildings

12. Animals not kept in buildings shall where necessary and possible be given protection from adverse weather conditions, predators and risks to their health.

Automatic or mechanical equipment

13. All automated or mechanical equipment essential for the health and well-being of the animals must be inspected at least once daily. Where defects are discovered, these must be rectified immediately, or if this is impossible, appropriate steps must be taken to safeguard the health and well-being of the animals.

    Where the health and well-being of the animals is dependent on an artificial ventilation system, provision must be made for an appropriate backup system to guarantee sufficient air renewal to preserve the health and well-being of the animals in the event of failure of the system, and an alarm system must be provided to give warning of breakdown. The alarm system must be tested regularly.

Feed, water and other substances

14. Animals must be fed a wholesome diet which is appropriate to their age and species and which is fed to them in sufficient quantity to maintain them in good health and satisfy their nutritional needs. No animal shall be provided with food or liquid in a manner, nor shall such food or liquid contain any substance, which may cause unnecessary suffering or injury.

15. All animals must have access to feed at intervals appropriate to their physiological needs.

16. All animals must have access to a suitable water supply or be able to satisfy their fluid intake needs by other means.

17. Feeding and watering equipment must be designed, constructed and placed so that contamination of food and water and the harmful effects of competition between the animals are minimised.

18. No other substance, with the exception of those given for therapeutic, or prophylactic purposes or for the purposes of zootechnical treatment as defined in Article 1(2)(c) of Directive 96/22/EEC [1], must be administered to an animal unless it has been demonstrated by scientific studies of animal welfare or established experience that the effect of that substance is not detrimental to the health or welfare of the animal.

Mutilations

19. Pending the adoption of specific provisions concerning mutilations in accordance with the procedure laid down in Article 5, and without prejudice to Directive 91/630/EEC, relevant national provisions shall apply in accordance with the general rules of the Treaty.

Breeding procedures

20. Natural or artificial breeding or breeding procedures which case or are likely to cause suffering or injury to any of the animals concerned must not be practised.

    This provision shall not preclude the use of certain procedures likely to cause minimal or momentary suffering or injury, or which might necessitate interventions which would not cause lasting injury, where these are allowed by national provisions.

21. No animal shall be kept for farming purposes unless it can reasonably be expected, on the basis of its genotype or phenotype, that it can be kept without detrimental effect on its health or welfare.

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[1] Council Directive 96/22/EC of 29 April 1996 concerning the prohibition on the use in stockfarming of certain substances having a hormonal or thyrostatic action and of beta-agonists (OJ L 125, 23. 5. 1996, p. 3).

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