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Article

Validation of an Animal Welfare Assessment Protocol for Zebu Beef Farms within Pasture-Based Systems under Tropical Conditions

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Simple Summary: Consumers perceive pasture-based livestock production as natural and ethical and, therefore, better for animal welfare. The objective of the study was to validate an animal welfare protocol on 24 commercial zebu cattle farms in tropical pasture systems. The protocol was developed through participatory workshops with producers, academia, and health authorities. The protocol included animal-based, resource-based, and management-based indicators. On-farm validation was carried out through interviews, review of records, direct observation of animal flocks, and individual evaluation in the pastures. Application of the protocol and expert analysis demonstrated that simple measures exist to assess animal welfare in pasture systems. The protocol will help identify opportunities for improvement to strengthen the implementation of animal welfare practices and comply with sanitary requirements.

Abstract: Pasture-based production systems are predominant in major beef-producing countries, however, these systems lack validated protocols to assess animal welfare under commercial conditions. The objective of this study was to validate an animal welfare protocol for fattening Zebu cattle farms in tropical pasture systems. The initial protocol was developed with the participation of producers, professionals, the general public, and the Colombian health authority, through workshops with a participatory approach and collaborative knowledge management. The validation was carried out in 24 pasture-based commercial Zebu cattle farms in the middle Magdalena region of Colombia. Visits were made with an average duration of 2.5 hours, which included the evaluation of 788 fattening cattle. The protocol evaluated animal-based, resource-based, and livestock management indicators through a questionnaire-guided interview to evaluate cattle handling and health, animal-based measurements, and documentation management. A protocol validation process was carried out by selecting indicators that remained unchanged, adjusting those that were feasible to implement, and eliminating inadequate or unnecessary indicators. The application of the protocol demonstrated that there are valid and feasible measures to include in the evaluation protocols of pasture-based fattening systems. Likewise, the active participation of producers is crucial to achieving a greater commitment to the implementation of this protocol.

Keywords: animal wellbeing; beef cattle; good health; welfare assessment

1. Introduction

Intensification of animal production has increased public awareness of environmental conservation, health, and welfare, aimed at promoting food safety, food security, and sustainable food production [1,2]. Consumers perceive pasture-based livestock production as natural and ethical, thereby better for animal welfare, compared to confined systems [3,4]. Likewise, consumers are willing to pay more for milk and meat from pasture-raised cattle [5]. The increasing focus on ensuring that animals have "lives worth living" makes pasture-based production systems essential for cattle to develop positive emotions, favoring their natural behavior [6,7]. Similarly, cattle have a greater preference for natural pasture-based environments [8], in which they develop a more efficient

immune system [9], can exercise more, maintain social cohesion, present less risk of hoof injuries, lameness, culling and mortality, but a higher risk of internal parasitism, biosecurity problems (greater contact with wild animals) [6,10] and thermal comfort [3,4], among other.

Protocols have been proposed to assess the welfare of cattle in confinement (Welfare Quality®, AssureWel, and others), but few are aimed at evaluating extensive or pasture-based Zebu beef cattle farms [11], which address all the welfare challenges that animals must face in these systems. Worldwide, studies show that a large proportion of dairy cows are raised in systems with access to pasture for at least part of the year, as is the case for 90% of cows in France, 95 to 100% in Ireland, 99% in Australia and New Zealand [12,13]; but these systems differ in the management of fattening cattle in tropical systems. Additionally, some of the measures used for animal welfare assessment under confinement conditions (e.g. lameness score, social behavior), are relevant also for grazing cattle [14]; however, it is not feasible to transfer assessment protocols developed for intensive systems to grazing systems, because each system needs a specific protocol and the proposed indicators are not necessarily suitable, relevant, feasible or measurable under grazing conditions [14,15]. Therefore, protocols with an evidence-based approach are needed to assess animal welfare in extensive fattening systems in tropical climates [2].

Pasture-based or forage-based production systems predominate in the main beef-producing countries such as the USA (West), Brazil, Argentina, Australia, New Zealand, Canada, and Uruguay, in some European countries such as France, United Kingdom and Ireland, and in sub-Saharan Africa [16]. Colombian cattle ranching is distributed among 633,841 farms (n= 29,301,392 animals) [17], contributes 6% of the national gross domestic product and generates 810,000 direct jobs. Fattening cattle represents 20% of the inventory and is managed under grazing conditions [18]. Currently, the National Animal Welfare Council and the Technical Committee on Production Animals were formed [17], which, jointly with producers' unions, academia, governmental entities, and citizen participation, prepared the manual and methodology for the evaluation of animal welfare conditions in cattle and buffalo farms (Resolution 0253 of 2020) [19]. However, the protocol has not been validated and implemented under commercial conditions. The objective of this study was to validate the protocol for the evaluation of animal welfare in fattening systems under pasture conditions and to propose other indicators or methodologies to complement the evaluation. The proposed protocol can be an evaluation reference for extensive and pasture-based systems in other countries with similar characteristics.[17].

2. Materials and Methods

2.1. Ethical Consideration

The study was carried out under commercial farm conditions and the researchers participated in the process solely as observers. All procedures related to the use and care of the animals strictly followed the Colombian regulation norm, Resolution 001634-2010 as stated by the Colombian Agricultural Institute [20]. Permission to conduct the study was approved by the Ethics Committee for Animal Experimentation (Act 30/12/2021, -Activities with minimal risk) at the University of Caldas. Farmers were fully informed about the purpose of the study, and they read/listened and signed an informed consent form and authorization to allow us to use the data.

2.2. Development of the Evaluation Protocol

The bovine and buffalo evaluation protocol was developed in its initial phase as an initiative of the Colombian Federation of Cattle Breeders and the National Livestock Fund (FEDEGAN-FNG) with the participation of an international expert in animal welfare. The second phase was developed with the objective of socialization, adjustment, and initial validation of the protocol, through the implementation of "workshops with a participatory approach and knowledge management" [21], which took into account: a) the tacit knowledge acquired through life/work experiences and oral traditions; and b) the explicit knowledge based on the scientific knowledge of validated protocols such as Welfare Quality® [22]. Six national workshops were held, involving professionals and

producers from the departments of Antioquia, Córdoba, Meta, Vichada, Guaviare, Arauca, Casanare, Atlántico, Magdalena, Cesar, Guajira, Bolívar, Sucre, Santander, Tolima, Huila, Cauca, Valle del Cauca, Caldas, Risaralda, Quindío, Boyacá, Cundinamarca and Nariño. In the third phase, the regulatory entity and a committee of experts evaluated and complemented the protocol proposal for its implementation in Colombian bovine and buffalo production [17].

2.3. Farm Sample

The study was conducted in 24 commercial pasture-based Zebu beef farms in the Magdalena Medio region in Colombia, South America, visited between April and July 2022 (Table 1). Herd size ranged from 20 to 1,300 animals, with an average age and weight at slaughter of 2.6 ± 0.1 and 503.3 ± 8.4 kg, respectively.

Table 1. Description of farms evaluated in the Magdalena Medio regions (Colombia, South America).

Farm	Altitud	Livestock numbers	Animals Evaluated	Extension (ha)
1	6°9'26''N	535	40	333
2	6°7'58''N	1100	55	600
3	6°7'58''N	1100	55	600
4	6°7'58''N	500	40	600
5	6°16'4"N	1300	65	750
6	6°7'58''N	1100	55	600
7	6°15'32"N	580	40	370
8	5°56'32"N	620	40	375
9	6°29'37''N	1150	58	523
10	6°15'20"N	398	30	382
11	5°55'34''N	165	30	1200
12	5°59'28''N	36	20	100
13	5°56'4"N	1200	60	1500
14	6°36'10''N	103	30	1200
15	5°42'32"N	76	20	190
16	5°58'34''N	39	20	240
17	5°29'15''N	20	10	300
18	5°54'41"N	20	10	30
19	5°27'41"N	24	10	13
20	5°45'45"N	73	20	2250
21	5°36'11''N	47	20	410
22	5°50'55''N	53	20	100
23	5°59'16''N	76	20	170
24	5°54'15''N	47	20	119

The animals were located in the Magdalena Medio region (low tropic), which corresponds to an extensive mid-Andean valley in central Colombia, formed by the Magdalena River and distributed in the departments of Antioquia, Bolivar, Boyacá, Cesar, Caldas, and Santander. This cattle-grazing area is considered promising for achieving livestock production in harmony with forests and wetlands [18] (Figure 1). Rotational grazing was carried out on improved grass pastures of *Brachiaria decumbens*, *Brachiaria humidicola*, *Dichanthium aristatum* Benth, *Megathyrsus maximus*, among others. In addition, natural grasses, native legumes, and native trees (*Xylopia amazonica*, *Clathrotropis brachypetala*, *Lecythis* sp.). The beef cattle belonged to commercial Zebu cattle crosses and some producers were making F1 crosses with *Bos taurus* breeds. The entire production process was carried out on pasture. The farms raised the beef calves "at full milk" up to 9 m, with average weights of 240 kg. The calves were raised and fattened on nearby farms of the same owners or were

marketed through auctions and livestock markets, to complete their production cycle on the buyers' farms in the same region. The animals were kept in groups without social mixing during the entire fattening process [18].

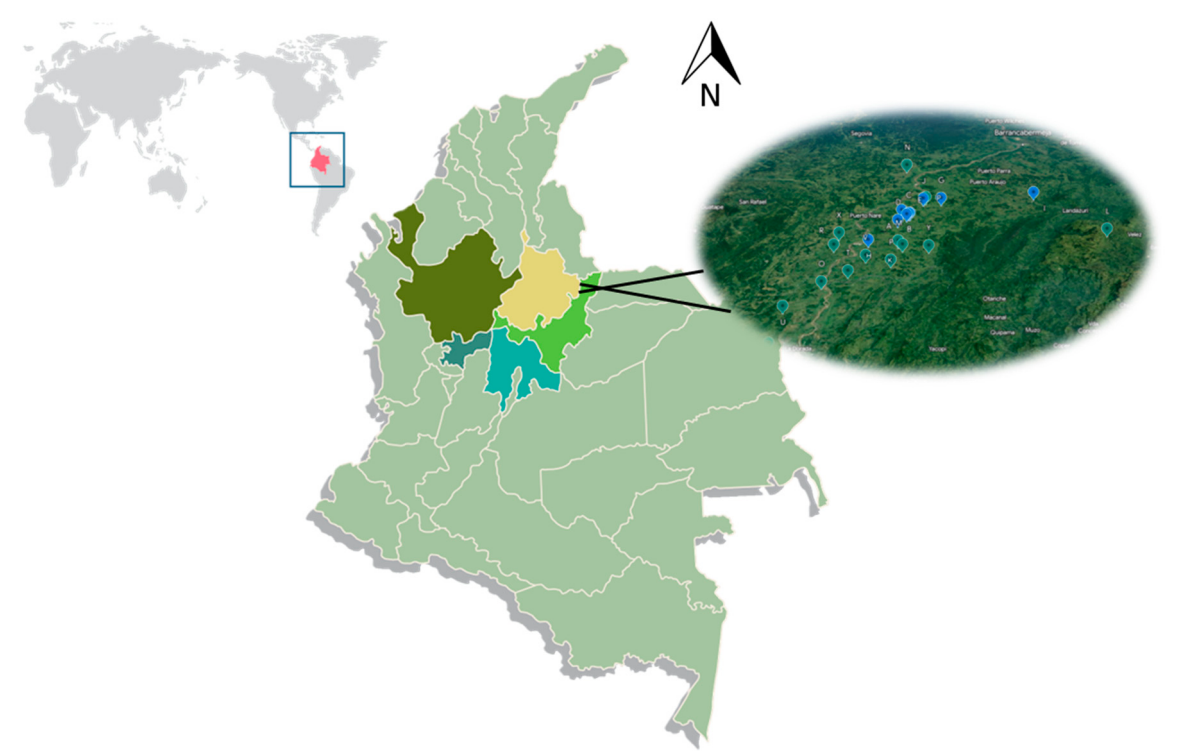


Figure 1. Geographic area evaluated in the study (Magdalena medio, Colombia, South America).

2.4. Animal of Animal Welfare

A trained veterinarian applied the animal welfare assessment protocol to Zebu fattening cattle in pasture-based systems (n = 24 farms). Table 2 presents an overview of the indicators proposed in the original protocol organized into four domains (good nutrition, suitable environment, optimal health, and appropriate behavior).

Table 2. General description of measures assessment in the Colombia beef farms in pasture-based systems protocol.

Principle	Welfare Criteria	Animal welfare measure/indicator	Method of assessment
Good feeding	Absence of hunger	Access and availability of grass	Interview
		Quality and quantity of grass	Direct observation
		Supplementation based on bromatological analysis	Documentary record
		Food storage	Direct observation
	Absence of thirst	Body condition score	Animal-based indicator (animal sampling)
		Access of water	Interview and direct observation
		Availability of water in drinkers and natural sources	
		Ad libitum	
		Water treatment/physicochemical and microbiological analysis	

Appropriate environment	Thermal comfort	Subjective assessment of shade in the paddocks General condition of the paddocks General condition of the facilities and fences Adaptations that provide comfort to animals (scrapers, draught protection, watering and shade feeders)	Interview/ direct observation
	Comfort around resting	Animal rest (clean and dry)	
	Painful procedures	Tagging/disbudding/castration Specify age at painful procedures Procedure and with/without use of analgesia and or anaesthetic Staff expertise	Interview
	Absence of disease	Abrasions/swelling/hairless Presence of biting flies Presence of <i>Dermatobia hominis</i> /ticks/worms Lameness	Animal-based indicators (animal sampling)
Good health		Animals requiring complementary care	Calculation/interview
		Mortality rate Culling rate	
	Use of veterinary medicinal products	Evaluation of the drug storage area and good drug management practices	Direct observation/ records
	Procedures and documentary records	Direct observation/ records Registration of treatment of cattle by a veterinarian and current health plan	
Appropriate Behaviour	Human-animal interaction	Animal reaction to human Human-animal interaction Stereotypes	Animal-based indicators (animal sampling)
		Knowledge and training in animal welfare	Interview - % people with certified training

The information was obtained through documentary evaluation (review of procedures, records), and a structured interview guided by means of a questionnaire with the owner or administrator of the farm to evaluate the health and general management of the cattle. The following, amongst others, were documented: water, feed, and mineralized salt supply; routine management practices; painful procedures (dehorning, castration, hoof trimming), personnel expertise, use of analgesia, anesthesia, age of the cattle when the procedure was performed; vaccination; diseases observed in the animals; mortality (frequent causes); culling (% and causes); average weight and age at slaughter; animals that required special care (%), and handling of sanitary and production records.

A visit was made on horseback (minimum two pastures per farm) to evaluate by direct observation the cattle in the pastures and the environmental conditions, forage supply, access to water and shade, and general condition of the pastures and fences. A proportional random sampling was performed according to the total number of cattle in the herd, to evaluate animal-based measures (body condition, presence of lesions and inflammations, presence of biting flies, ticks, worms, lameness), taking into account the following criteria: (a) herd with 1000 animals, 5% of the population [23]. To evaluate the reaction of the animals to humans, the evaluator observed the behavior of the cattle when the handler entered the pasture on horseback (calm, excitable). The cattle handler was

then asked to interact with a group of cattle in the pastures to evaluate the human-animal interaction (interactions used by the handler and the response of the animals), and the reaction of the animals was generally recorded as positive, negative, or neutral [23].

Visits averaged 2.5 hours in duration. After completing the farm visits, a critical evaluation of the indicators proposed in the protocol was carried out with the participation of two international experts, and the final protocol was adjusted.

Figure 2 presents an overview of the typical grazing conditions in the Colombian Magdalena Medio, the crossbreeds of Zebu cattle in the area, and the interaction of the cattle with the handler.

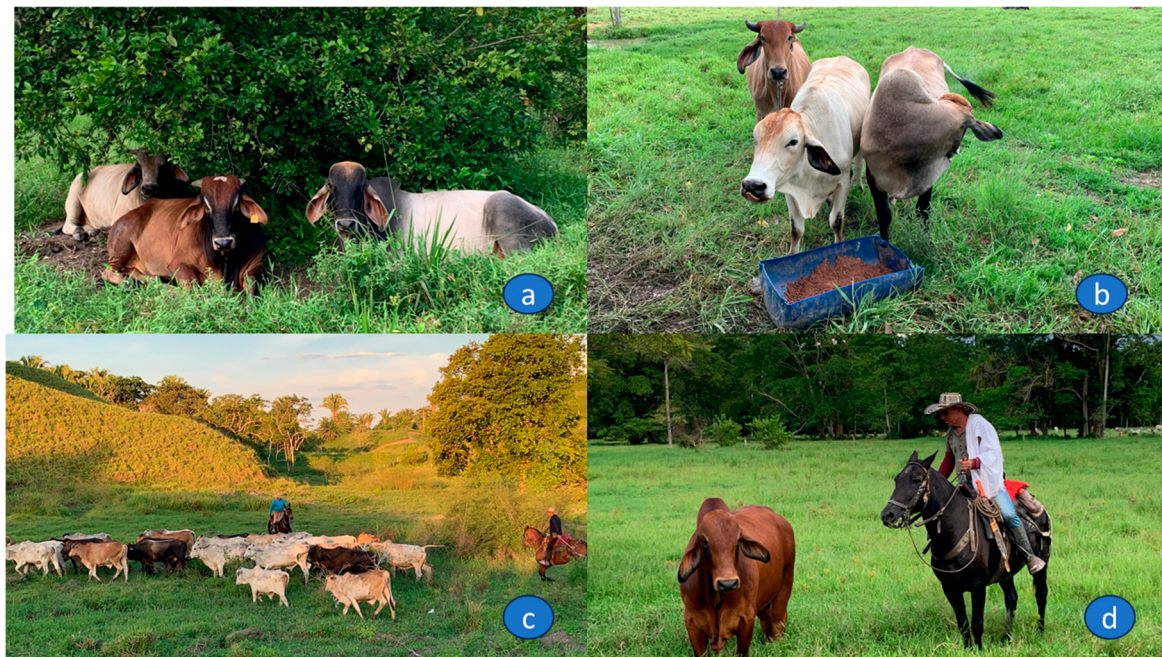


Figure 2. Grazing conditions in the middle Colombian Magdalena. a) Thermal comfort. b) Supply of mineralized salt. c) Reaction of animals to humans. d) Interaction with humans.

2.5. Statistical Analysis

Software Stata Version 13.0 (College Station, Texas, USA) was used for all the statistical analyses. Spearman's coefficient was used to identify measures with a strong association ($\rho \geq 0.8$). A probability level of $P < 0.05$ was chosen as the limit for statistical significance in all tests.

After the visits, the three authors divided the protocol indicators into four groups, according to the feasibility of their application, following the guidelines proposed by Kaurivi et al. [15]: a) indicators that remained unchanged in the final protocol, b) indicators that were not feasible in all farms, but were considered necessary and adequate to be maintained in an adjusted form in the final protocol, c) indicators that were not feasible in all farms or that were considered unnecessary or inadequate; these measures were eliminated from the final protocol and d) indicators significantly correlated with other measures, which could be evaluated by a common indicator.

3. Results

A total of 788 fattening cattle were evaluated in the pastures to validate the animal-based measures. Tables 3–5 describe the reasons why each indicator was classified in each of the proposed categories (included in the protocol without modification, included with adjustments, not included).

Table 3. Measures assessed as feasible for inclusion in the final protocol without change.

Welfare Criteria	Welfare measures/indicator	Method of assessment
Absence of hunger	Access and availability of grass	Subjective assessment of grass in the paddocks (type and availability during the year) as enough or insufficient. Interview and direct observation % thin animals in the herd, based on score ≤ 4 on 1-5 scales
	Body condition score (thin animals)	Categorical scale according to the proportion of animals with score ≤ 4 to assign total score (0-8).
Absence of thirst	Access to water in drinkers and natural sources	Subjective assessment of availability of natural water sources/drinkers as enough or insufficient.
	Quality/ <i>Ad libitum</i> /restriction of water	Interview/direct observation as enough or insufficient.
Thermal comfort	Shade and adaptations that provide comfort to animals (trees)	Subjective assessment of shade in the paddocks (presence of trees, shrubs, galleys).
Presence of hazards	Fence status	Subjective assessment of fence condition in the visited pastures (intact, free from sharp elements or any other conditions that may cause harm or injury to the animals).
Absence of pain from management procedures	Ear tagging, disbudding/castration	Record age and use of local anaesthetic during questionnaire-guided interview.
		At pasture % of cattle with uneven weight-bearing on a limb that is immediately identifiable and/or obviously shortened stride.
Absence of disease and pain	Lameness (animal based-indicator)	Categorical scale: No lameness (0): normal displacement and poise Mild lameness (1): abnormality in displacement or aplombs Severe lameness (2): arching of the back. % cattle with severe lameness ($\leq 5\%$, $> 5\%$, $\leq 10\%$, $> 10\%$) Interview/registers/calculation % Numbers of accidental deaths and deaths/slaughter (either on-farm or sent off-farm) due to disease were combined. (Excellent: $\leq 2\%$, High: 2.1-3%, Medium: 3.1-5%, Low $> 5\%$ or no records)
		Animal-based indicator (sampling) % sick or injured animals not receiving timely treatment and care (herd separation, provision of soft beds, access to water and food, treatment)
	Mortality rate (%)	
Human-animal interaction	Complementary Care	1. Excellent 2 – High 2.1-5% Medium $> 5\%$ Low
	Reaction of animals to humans (Calm/excitable)	Animal sampling (animal-based indicator) Subjective evaluation of beef cattle behavior when the rider enters the environment where the animals are located (reaction and movement of the animals) Categories: Calm-quiet (static animals, with little or no resistance to being approached) Excitable (constant, vigorous movement, attempting to escape, very agitated and frightened) [24].

	Interaction with humans (positive/negative/neutral)	Animal sampling (animal-based indicator) Subjective evaluation of the animal's orientation response to the handler Categories [25]: Positive: the posture of the head, ears and body relaxed, shows interest in the handler. Negative: vigilance, avoidance, flight. Neutral: Head in normal position and ears upright while looking at handler, continuous rumination.
Knowledge and training	Formal training in animal welfare	Interview - % people with certified training in animal welfare (y/n, 100%, < 50%, ≥ 50%)

Table 4. Welfare measures included in the protocol after adjustments, including the rationale for change and the changes that were made.

Welfare Criteria	Welfare measures	Method of assessment	Reason for difficulty	Adjustment of measures
Conforte around resting	HAZARDS	General condition of the paddocks Direct observation/interview	The protocol did not include categorical measurement scales and the presence of other hazards reported in other extensive production systems	Subjective evaluation of paddocks, including the identification of flood-prone areas and potential hazards within pastures such as steep hills, cliffs, gullies, and sinkholes. Also, noting the presence of hazardous objects or debris. Categorical scale required.
Presence of ectoparasites	Presence of biting flies (<i>Tabanus Stomoxys calcitrans</i>)	Animal based indicators/animal sampling Presence of biting flies on head, back, belly and legs (and/n, 50 insects) <i>Dermatobia hominis</i> (y/n, ≥ 5 insects)	These indicators are not practical to measure in pastures, animals are required to be restricted from movement	Interview on the prevalence of ectoparasites, problems caused and control strategies.
	<i>Dermatobia hominis</i> /ticks/worms	% of animals with ticks % of animals with myiasis % of animals with ticks % of animals with horseflies	There is a risk of injury to the evaluator and additional stress to the cattle	
	Presence of gticks	Direct observation of infestation on ears, groin, base of tail and udder (y/n, presence at least two of them)		
	Infestation by fly larvae	Direct observation of the infestation (y/n, presence at least one of them)		
Disease history	Hemoparasites (<i>Babesia sp.</i> , <i>Anaplasma</i>	% of animals with clinical signs, diagnosis and	During the interviews the producers and/or	Interview on the prevalence of blood

Disease history	<i>marginale, Trypanosoma sp.)</i>	treatment for hemoparasites	administrators reported frequent health problems due to blood parasite infestation	parasites, results of diagnostic tests, problems caused and control strategies
	snake bites Poisoning with poisonous plants Presence of predators Photodermatitis	% of cattle that died on the farm during the last 12 m due to snake bites % cattle that died on the farm during the last 12 m due to predators % cattle that died on the farm during the last 12 m due to ingestion of toxic plants % cattle that died on the farm during the last 12 m due to thunderstorms % cattle that died on the farm during or subjected to autanasia the last 12 m due to accidents % cattle who became ill/treated during the last 12 months by photosensitization		

In general, the exclusion or modification of indicators from the protocol was associated with different explanations: the difficulty of measuring them on all farms, animal welfare implications not very applicable to the fattening production system under pasture conditions, time and space limitations, measures requiring specialized evaluations and adjustments to place the indicator in a category, where it was more feasible to evaluate it [15]. Eight indicators were excluded from the protocol due to factors such as, it was not feasible to evaluate them under grazing conditions due to the physical risk to the evaluators, low frequency in this type of system and the measures corresponded to indicators of good farming practices. Table 5 describes each of the indicators and the justification for their elimination.

Table 5. Welfare measures removed from the protocol after feasibility testing on 25 beef farms.

Welfare Criteria	Welfare measures/indicator	Method of assessment	Reason for removal
Absence of hunger	Supplementation based on bromatological analysis	Interview	Producers do not perform this practice routinely because they do not have specialized laboratories in the area and the measurement of the animals' body condition is a more effective measure. Can be considered in systems that use strategic nutritional supplementation
Absence of thirst	Water treatment and laboratory analysis (physiochemical and microbiological)	Interviews/records	It is not a viable practice when working with natural sources.

Comfort	Adaptations that provide comfort to animals (environmental enrichment)	Interview/direct observation	In many regions on the farms there is no supply of drinking water for humans and the water is obtained from wells. Environmental enrichment is not feasible to apply under conditions of natural pastures typical of the tropics, because they are abundant and varied. The protocol does not include any measurements within pens to assess ease of handling. Management of cattle in pens is primarily focused on compulsory vaccination procedures.
	Overall state of facilities designed to facilitate handling and prevent accidents.	Direct observation	
HAZARDS	Absence of disease and pain	Animal-based indicator Direct observation in the standing animal from a distance of no more than 2 m, of the presence of areas of alopecia or scars greater than 2 cm, dividing the animal into three zones: a) head and neck, b) body-trunk; c) front and rear limbs % discard/records/calculation Criteria: discard of the herd through a planned decision, age, slaughter on the farm, euthanasia	Difficulty in measuring and risk to the observer. Low frequency of this type of injury was reported under natural grazing conditions.
		Disposal procedure performed by a veterinarian, humanitarian slaughter ($\leq 15\%$ / $> 15\%$)	Indicator with low frequency in the evaluated farms Slaughtering cattle on the farm is not a routine practice
	Use of veterinary medicinal products	Direct observation/ records Drug Storage Area Assessment Registered with the competent Entity Storage Conditions Validity Veterinary prescription Direct observation/ records	It is part of good primary production practices and no report was found on its use to evaluate animal welfare.
	Procedures and documentary records	Record of treatment of cattle by a veterinarian Written health plan signed with a veterinarian with current professional registration	

Likewise, when performing the correlation analysis of the indicators, it was found that when inquiring about the method used to perform painful practices (castration, tagging, dehorning), these were highly correlated ($q \geq 0.8$; $P < 0.01$) with the use of analgesia and anesthesia, therefore, it would be sufficient to only ask about the method applied for each procedure; but due to the ease of obtaining the information by means of an interview, we decided to keep the indicator in the protocol without modifications (Table 3). There were weak, but statistically significant correlations between handler age ($q = -0.46$, $P = 0.02$), time of experience working with cattle ($q = 0.37$, $P = 0.04$), body condition 1

($\rho = 0.46$, $P = 0.04$), body condition 3 ($\rho = 0.49$, $P = 0.01$), body condition 5 ($\rho = -0.51$, $P < 0.01$), % animals with mild lameness ($\rho = 0.54$, $P = 0.04$) and % animals with obvious lameness ($\rho = 0.491$, $P = 0.04$), associated with the type of interaction observed between handlers and beef cattle (positive, indifferent).

4. Discussion

In the United Kingdom and Germany, the beef industry is at the forefront in adopting welfare outcome measures as part of its farm assurance scheme [28,29]. The Colombian Federation of Cattle Breeders- FEDEGAN- proposed the evaluation protocol under validation in this study. These on-farm assurance program initiatives indicate the commitment of producers to manage their animals according to animal welfare standards, which then allows them to access certain markets [30], sell their products to more demanding retailers [31], change management routines [32], monitor and evaluate changes in practice, target interventions based on results [33], and comply with the requirements of sanitary legislation (Resolution 0253 of 2020) [34]. These evaluation schemes favor active participation and a long-term commitment to improvement by producers and greater adherence to animal welfare audit protocols [29,35].

4.1. Protocols Characteristics

The evaluation of animal welfare in grazing-based bovine herds requires protocols that are valid (adequately measure what they are supposed to measure), reliable (correctly quantify the evaluated characteristic), feasible (applicable under the same conditions), repeatable (obtain the same results in any context), practical to measure in the field and stable over time because they can contribute to improving the quality standards of the productive processes [33,36]. However, there is a high variability in the grazing environment, and it is necessary to use sufficiently flexible protocols that can be adapted to local conditions [37].

4.2. Good Nutrition

This protocol evaluates the criteria of "absence of hunger and thirst" through direct observation of the accessibility, availability, quality and quantity of forage and water in the pastures [18]. In Colombia and other countries with a wealth of natural water, there is an abundant supply of water sources such as rivers, natural wells, and springs, which are used for livestock consumption, an aspect that is present in the Magdalena Medio region. However, despite their ecological importance, cattle ranchers are transforming these ecosystems and have suggested the adoption of water collection, storage, and conduction systems, so that each pasture has a drinking trough, to prevent cattle from entering riverbeds and thus preserve and recover the riverside areas, marshes, wetlands, rivers, micro basins and conserve wildlife and fish [18,38]. The inclusion of this indicator of accessibility to food and water, as described in the original protocol, is suggested because it is supported by scientific evidence [39,40], and because of the difficulty of performing measurements proposed by other authors that we do not consider relevant such as: counting the number of working water points (especially in natural water troughs), flow rate, length of water troughs, classification of water troughs as safe or unsafe (slip hazards, presence of risks, bearing capacity of the soil, etc.), observation of competitive behavior of cattle in front of water sources and cleanliness of water (absence of odors and strange colors) [15,39,41].

Furthermore, other authors have proposed different alternatives: a) evaluation of the distance traveled by the animals to access water, because it has been suggested that if the water supply sources are located more than 250 m away, cattle decrease their water consumption [14], however, the Colombian Magdalena Medio region has a permanent supply of green forage, which decreases fresh water consumption by animals; b) how access to water is managed during grazing [39] and c) feeding strategies to check that the pasture provides sufficient nutrients during grazing (bromatological analysis) [14].

In the interviews conducted for the validation of the protocol, the existence of water treatment systems and bromatological analysis of pasture were inquired about, which are infrequent practices among producers (<5%), taking into account that this region is rich in water sources, producers obtain it from natural sources, as previously indicated. Regarding the bromatological analysis of grasses and legumes to determine supplementation needs, there are no laboratories in the area to perform the tests and there are other indicators such as body condition and daily weight gain of the animals that are more efficient. However, it is important to keep in mind that strategic nutritional supplementation can contribute to reducing deficiencies in the quantity and quality of feed based on pasture or forage and contribute to obtaining higher productive yields, therefore, in a more advanced phase of implementation of the protocols, these criteria could be included [16].

Body condition scoring is an effective measure of medium-term energy balance and is proposed as a unique indicator to assess nutritional performance on dairy farms in New Zealand [40] and considered feasible in this protocol with animal sampling. This procedure was performed on horseback to get close proximity to the cattle and is considered efficient by other authors [14]. Optionally, the measurement of body condition could be performed in the pen during the vaccination process of the animals or by monitoring the batches in the slaughterhouses, through the evaluation of hot and cold carcass yield, as has been proposed in swine in Colombia [42].

4.3. Good Environment

One of the most complex aspects to evaluate quantitatively is the comfort of fattening cattle, because grazing conditions are variable, do not remain stable over time, and can fluctuate due to weather conditions, animal management, pasture quality, and pasture rotation, among others; These aspects, in turn, induce variations in resting areas, feed availability, distances to be covered, forage quality and quantity, soil quality and susceptibility to heat stress, among others [40,43]. One of the most complex aspects to evaluate quantitatively is the comfort of fattening cattle, because grazing conditions are variable, do not remain stable over time, and can fluctuate due to weather conditions, animal management, pasture quality, and pasture rotation, among others; These aspects, in turn, induce variations in resting areas, feed availability, distances to be covered, forage quality and quantity, soil quality and susceptibility to heat stress, among others. Colombia is developing an industry technical standard that establishes environmental requirements for the livestock industry and the creation of an environmental seal in response to market demands for the adoption of sustainable practices through the management of good irrigation practices, planting, land management, waste management, good livestock practices, and social responsibility with employees (NTC 6550/2021, ICONTEC) [44]. This policy is based on the establishment of silvopasture systems and forest conservation on cattle ranches as a strategy and opportunity for environmental offsetting [18].

In this study, comfort was assessed by direct observation of the resources available to help cattle cope with heat or cold stress (e.g., the presence of trees in the pasture, silvopasture systems), which is considered valid in pasture-based systems [40] and is valuable to producers as it provides protection against extreme climates and contributes to wildlife conservation [45]. Other indicators have been suggested such as: a) the evaluation of the cleanliness of the animals' hind quarters and the percentage of dirty animals [41], but these indicators are not very applicable when the animals are free in the pastures, due to the difficulty of observation; however, it is considered valuable because the presence of mud or manure, are risk factors for the presentation of lameness [10,46] b) Use of sensors to evaluate animal behavior during long-term grazing (an option that would be valid, if the evaluation is performed for research purposes); c) evaluation of resting behavior (animals lying outside or inside the resting or shaded area) [41]; d) measurement of clinical signs of heat stress (such as panting) [6], but a single evaluation is not considered representative of the entire pasture [3] and requires additional time for evaluation.

The proposed protocol for beef cattle evaluated comfort around resting through the observation of pasture conditions (presence of floodable areas). In this particular case, the Magdalena Medio region has 70% of hilly areas and the remaining 30% corresponds to floodable forests of the

Magdalena River, rich in native legumes, natural succession trees, and a high biodiversity of wild species [18,47]. The implementation of this indicator is suggested because it is easy to measure and we followed up on the foot lesions of animals from the evaluated farms during post-mortem inspection in two slaughterhouses (unpublished results) and found a high frequency of lesions related to pasture moisture conditions, such as heel erosion and abnormal claw shapes (asymmetric claws and corkscrew claws), results that coincide with those described by Bautista-Fernández et al., [48] in Mexico and by Moreira et al., [46], in Brazil.

These lesions cause pain, decreased feed intake, and significant economic losses [49]. In addition, this indicator is associated with the thermal comfort of the animals, the presence of lameness, and other abnormalities of the hooves. This finding could lead to the strengthening of sanitary programs for the monitoring and treatment of heel erosion and hoof problems, an activity that was routinely carried out in the region, according to the information provided by the interviewees.

The protocol proposed the evaluation of the general resting conditions of the animals in clean and dry places and the adaptations made by the owners to provide greater comfort to the animals (environmental enrichment, and protection against wind, among others), but we considered its evaluation complex due to: a) additional time would be required to visit all the pastures and observe the resting behavior of cattle; b) environmental enrichment strategies and protection against wind are naturally included in the pasture environment [45] and this indicator applies more to stabling conditions.

4.4. Good Health

Information related to the management of painful procedures was obtained by means of an interview, as proposed in the Welfare Quality® protocol, a validated and easy-to-apply methodology [22]. However, the evaluation of the expertise of the personnel to perform painful procedures is not considered feasible, because it is complex to measure in a single visit to the farm. During the protocol validation and implementation process, it was observed that cattle castration and dehorning are common livestock practices that are frequently performed without pain mitigation, results that are consistent with other studies [50]. It has been reported that the use of analgesics, anesthetics, and anti-inflammatory drugs is a more frequent practice in adult cattle than in suckling and newborn calves, according to studies conducted in Brazil [51]. It is suggested the implementation of promotion and continuing education programs by veterinarians, producer organizations, and state institutions, among others, to promote pain mitigation practices; as well as, the development of research to evaluate their efficacy in reducing pain-induced distress and post-treatment responses of cattle (e.g. inflammation after surgical dehorning) [51].

Gross mortality was evaluated as the percentage of animals that died from all causes (disease, accident, no specific cause) during the last year, as indicated by the Welfare Quality® protocol. [22]. Respondents reported percentages below 1% gross mortality, coinciding with results obtained in studies conducted in pasture-based dairy systems [6,52]. It is suggested that the indicator called "complementary care", which corresponds to the % of sick or injured cattle that do not receive timely treatment and attention (separation of the herd, provision of soft bedding, access to water and feed, treatment), be retained without modification because during the visits it was observed that cattle handlers daily check the health conditions of the animals and implement the relevant sanitary measures. In contrast, it is suggested the elimination of the indicator called "% of discarded animals", which is carried out through a planned decision, taking into account age, slaughter on the farm, and euthanasia procedure performed by a veterinarian or humane culling because these are infrequent activities on fattening farms and humane culling on the farm is not a routine practice. This indicator applies more to breeding and milk production.

If an assessment protocol is to be widely used, it must include individual assessments that are practical to measure within the system being assessed, and it must be feasible within a reasonable time frame [40]. The latter is particularly important in a pasture-based system, as many animal-based assessments can only be measured while milking or performing a medical procedure on beef cattle (vaccination, drug administration), as this is the only time when they can be closely and

systematically observed, an aspect that limits their implementation. In this study, the evaluation of animal-based measures (ectoparasite infestation, the presence of inflammations and lesions) was made difficult by the dispersion of animals in the pasture, and even in some cases, it was considered unsafe, to perform an approach to fattening cattle at a distance of 2 m, as is done in dairy cattle [53]. In New Zealand, it has been recommended to obtain information through guided interviews on the prevalence of problems caused by ectoparasites and control strategies (rotational grazing, integrated parasite control, etc.) [15], an aspect that we suggest is feasible to implement.

Several authors indicate that lameness is more frequent in feedlots [2], an aspect that has been widely demonstrated, however, the observation of lameness in pasture-based dairy systems [10] was common in our study because the Magdalena Medio region is rich in water sources and in areas of floodable forest [47], for this reason, this evaluation was included in the protocol. This information can be obtained through interviews and review of records. This can also be evaluated during the visit to the pasture on the day of the audit, by having the handler move the cattle.

Other specific risks for fattening cattle on pasture that could be inquired by interview is the presence of photosensitized animals (frequent in some regions of Colombia) [54], ingestion of toxic plants [55], gastrointestinal parasites (e.g. *Ostertagia ostertagi*) and the strategies used for their control [52], hemiparasites (*Anaplasma marginale*, *Babesia bigemina*, *B. bovis*, *Trypanosoma* sp.) [56,57], predation-related mortality especially for fattening cattle during breeding and rearing [55], and snakebites [27].

Venomous snakebites are a potentially fatal health problem in humans and animals in tropical areas, with cattle and buffalo reported to be the most affected animal species [27]. Studies conducted in Costa Rica, have reported high prevalences due to *Bothrops asper* (family Viperidae) bites (9% in cattle), which are distributed in tropical and subtropical rainforests from northeastern Mexico to the Pacific lowlands of Colombia and Ecuador (altitude between 0 and 1,500 m.a.s.l.) [26]. The proposed indicators were included because of their high reporting in the evaluated area and in other tropical regions of the world.

4.5. *Appropriate Behavior*

Grazing is part of the natural behavior of cattle, which is reflected in greater social cohesion and permanent affiliative interactions, which reduce aggression, stereotypies, and stress in cattle under grazing conditions, compared to those kept in confinement [9,52]. Pastures provide cattle with greater opportunities for exercise, access to varied feed sources, the ability to select feed according to their preferences, and low competition for resources [9,58].

The protocol validated in this study evaluated the quality of human-animal interaction by observing in the paddock the positive behaviors of beef cattle, during interaction with handlers (reaction and interaction), as has been proposed in dairy cattle on pasture [40,41,59]. This study inquired about the practices used by handlers in order to reduce animal fear responses and improve human-animal interaction during routine handling, an indicator that is considered valid, as significant correlations have been revealed between the practices reported by handlers and behavioral responses of beef cattle [60].

In cases where the objective of the evaluation of human-animal interaction on farms is the improvement of management, decrease of the risk of occupational accidents and promote the welfare of beef cattle, we suggest the evaluation of the quality of human-livestock interaction during the routine vaccination process on the farm (official control diseases), as has been done in fattening farms in Brazil [61] and in Zebu fattening cattle in Colombian slaughterhouses and cattle markets [62]. Likewise, studies conducted in beef herds in French farms successfully validated the assessment of human-animal interaction by means of the behavioral test of animals' reactions to humans (an avoidance test) and found a significant association with the practices reported by cattle farmers through a semi-structured interview based on three aspects: (a) general description of farm and herd management practices (grazing period, discarding and selling animals), (b) organization of work with cattle (frequency of herd monitoring, hoof trimming, feeding organization, type of herd

monitoring in pastures) and (c) handler's relationship with cattle (number of accidents with cattle, value of having a good farmer-cattle relationship, methods that facilitate cattle handling) [60].

The evaluation of the presence of stereotypes in this protocol was considered not very applicable to evaluate in grazing cattle because its estimation requires the elaboration of a previous ethogram and a prolonged observation process and these are more useful to evaluate in stabled cattle [63]. On the other hand, it was considered relevant to evaluate the proportion of personnel with certified training in animal welfare and good management practices, because it is a requirement of the Colombian sanitary legislation () directed to all animal handlers in logistic chains (farm, transport, livestock markets); in addition, studies conducted in Brazil in pasture fattening cattle farms found that training cattle handlers through an effective, practical and periodic strategy, promotes positive human-animal interactions [61], improving the quality of life of both handlers and livestock [64,65].

5. Conclusions

The proposed protocol for the evaluation of fattening Zebu cattle under pasture conditions included animal-, resource-, and management-based evaluations, along with record-related evaluations. Although it is considered ideal for protocols to be based primarily on animal-based measurements, which allow for the estimation of actual welfare status in animal behavior, health, and body condition, resource-based and management-based measures are based on science and expert experience, making them feasible to implement. Despite the lack of representativeness of the evaluated farms of all existing types of fattening cattle on pasture, the proposed protocol yielded valid information to propose it as feasible, simple, and representative of animal welfare conditions in these systems. The proposed protocol is expected to be suitable for use at the farm level for comparative evaluation, allowing the monitoring of the progress of each farm and the certification of standards required by sanitary legislation.

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Institutional Review Board Statement: All procedures related to the use and care of the animals strictly followed the Colombian regulation norm, Resolution 001634-2010 as stated by the Colombian Agricultural Institute [20]. Permission to conduct the study was approved by the Ethics Committee for Animal Experimentation (Act 30/12/2021, -Activities with minimal risk) at the University of Caldas.

Informed Consent Statement: Farmers were fully informed about the purpose of the study, and they read/listened and signed an informed consent form and authorization to allow us to use the data.

Data Availability Statement: Data will be made available on request.

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