

**UNIVERSIDADE ESTADUAL PAULISTA - UNESP
CÂMPUS DE JABOTICABAL**

**HUMAN-CALF RELATIONSHIP AND ITS IMPLICATIONS ON
ANIMAL WELFARE**

Maria Fernanda Martin do Amaral Guimarães

Zootecnista

2014

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ANIMAL WELFARE**

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Dissertação apresentada à Faculdade de Ciências Agrárias e Veterinárias – Unesp, Câmpus de Jaboticabal, como parte das exigências para obtenção do título de Mestre em Zootecnia.

2014

G963h Guimarães, Maria Fernanda Martin do Amaral
Human-calf relationship and its implications on animal welfare /
Maria Fernanda Martin do Amaral Guimarães. -- Jaboticabal, 2014
vi, 67 p. : il. ; 29 cm

Dissertação (mestrado) - Universidade Estadual Paulista,
Faculdade de Ciências Agrárias e Veterinárias, 2014
Orientadora: Linda Jane Keeling
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Banca examinadora: Carla Forte Maiolino Molento, Marcelo Simão
da Rosa
Bibliografia

1. Behaviour. 2. Cattle temperament. 3. Human-animal relationship.
I. Título. II. Jaboticabal-Faculdade de Ciências Agrárias e Veterinárias.

CDU 636.2:636.083

Ficha catalográfica elaborada pela Seção Técnica de Aquisição e Tratamento da Informação –
Serviço Técnico de Biblioteca e Documentação - UNESP, Câmpus de Jaboticabal.

CERTIFICADO DE APROVAÇÃO

TÍTULO: HUMAN-CALF RELATIONSHIP AND ITS IMPLICATIONS ON ANIMAL WELFARE

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Data da realização: 03 de junho de 2014.

AUTHOR'S PROFILE

MARIA FERNANDA MARTIN DO AMARAL GUIMARÃES – was born on November 3rd, 1987, in the city of São Paulo, São Paulo State, Brazil. She has a Bachelor degree in Animal Science by the Faculty of Veterinary Medicine and Animal Science, São Paulo State University (FMVZ – UNESP), campus of Botucatu. During her Bachelor, she was a board member of a junior consultancy enterprise specialized in bovines and attended to courses related to the animal behaviour and animal welfare themes. She made her obligatory internship in ETCO Group, located in São Paulo State University (UNESP), campus of Jaboticabal. On March, 2012, she started the Master course in the Animal Science Post-Graduation program in the same university and became a researcher from ETCO group.

“O correr da vida embrulha tudo.
A vida é assim: esquenta e esfria, aperta
e daí afrouxa, sossega e depois desinquieta.
O que ela quer da gente é coragem.”
Guimarães Rosa

To my parents,

To my family,

To life and its beauty.

ACKNOWLEDGMENT

First of all, I would like to thank my parents, Milton and Ana Maria, for all the love, caring, support, teaching and advice. Without them this thesis could never be done. Thanks for all the patience, guidance and helping me to believe in myself; also for being always there on the good and bad moments. I am absolutely proud of being your daughter! I love you!

Special thanks to my fiancé Antonio for all the support and for pushing me across the moments I needed the most. Thanks, my love, for always staying by my side every day and every hour.

To my whole family, specially the aunts: Madi and Carol, the uncles: Zé, Tide and Padi; and the cousins: Fafa, Babaia, Paulinho, Cecé, Lu and Ju. I am sure each one of you played an important role in this achievement. Thanks for being the best family in the world!

To all my beloved friends, that I'm pretty sure each one of them knows exactly who they are. Although being far away, you were always in my heart, helping me to move forward. Thank you for being in my life!!

Special thanks to the examiners Carla Molento and Marcelo Rosa for finding the time to come to Jaboticabal and making great contributions to this thesis.

To my supervisor Dr. Linda Keeling for providing me with one of the best moments of my life, which was the time I spent in Sweden. Thanks for the support, the meetings and the dinners. It was an amazing experience to meet you in person and to be supervised by you. Thank you for all your knowledge and kindness.

To my co-supervisor Dr. Mateus Paranhos da Costa for the opportunity of being part of one of the best animal welfare research groups in Brazil. It was a remarkable experience to work by your side. Thanks for all the teaching, advice and "puxões de orelha". It certainly made me grow personally and professionally.

To ETCO group, in special the milk-part, Livia and Luciana, for the help, lessons and companionship. Thanks to Karen, Maria Camila, Tamara, Jana, Emilia, Nathasha, Arquimedes, Daniel and Mariana for the countless laughs and special moments. You made this journey lighter!! And Aline, Tiago, Adriano, Desiree, Taciana, Franciely, Paola and Monique. Each one of you helped me in a special way! Thank you!

To the Banana people who welcome me so well during the cold Swedish days. Tack så mycket for all the laughs during FIKAs!! Special thanks to Daiana for besides helping me with my work, becoming such a great friend! It was great to

know you better and the time we spent together was wonderful!! 😊 Oh, and Claes, for the beers, laughs and tourism!!

Finally, I would like to thank all the producers that accepted me in their farms; the 12 handlers who allowed me to get into their daily routine, and shared their life with me. And, of course, thanks to the 328 calves that accepted me during their routine really well!!!

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CEUA – COMISSÃO DE ÉTICA NO USO DE ANIMAIS**CERTIFICADO**

Certificamos que o Protocolo nº 014/14 do trabalho de pesquisa intitulado "***The assessment of handler-calf interaction and its implications on animal welfare***", sob a responsabilidade do Prof. Dr. Mateus José Rodrigues Paranhos da Costa está de acordo com os Princípios Éticos na Experimentação Animal adotado pelo Conselho Nacional de Controle de Experimentação Animal (CONCEA) e foi aprovado pela COMISSÃO DE ÉTICA NO USO DE ANIMAIS (CEUA), em reunião ordinária de 03 de fevereiro de 2014.

Jaboticabal, 03 de fevereiro de 2014.



Prof.^a Dr.^a Paola Castro Moraes
Coordenadora - CEUA

HANDLERS' ATTITUDES TOWARD DAIRY CALVES AND THEIR EFFECTS ON CALF WELFARE

ABSTRACT – Dairy calves are usually separated from their dams within few hours after birth, when calves start their relationship with humans. The aim of this study was to assess the handler attitudes toward dairy calves and their effects on calf welfare. Direct observations during milk feeding period were made for three consecutive days, and all actions from the handlers were recorded in ten different commercial dairy farms in the states of São Paulo (9) and Minas Gerais (1). Calf temperament traits were assessed by performing three behavioural tests: flight distance score (FDS); voluntary approach test (VA); and qualitative behaviour assessment (QBA). A questionnaire containing 21 questions was applied for each handler. A total of 12 handlers and 328 calves were assessed. Principal Component Analysis (PCA), Factor Analysis (FA) and Multiple Correspondence Analysis (MCA) were held, using Statistica 7 software package, for shrinking the data dimensionality and understanding the multiple association among the variables. Confirmatory data analyses of variance (ANOVA) were carried out using SAS software program; and the Spearman coefficients of correlation (r_s) were estimated for the non-parametric variables. Significant differences were found among the 10 farms regarding the QBA descriptors ($F = 11.48$, $P < 0.0001$). According to the PCA farms 1, 2, 3 and 5 presented calves with undesirable temperament descriptors. In addition, farms 3 and 5 showed handlers who least cared for the calves according to their answers in the questionnaire. After the FA, it was possible to make a ranking of the 'good' and 'poor' handlers, showing that handlers 1, 2a and 2b were the handlers who performed most positive actions, but were also the ones who performed most negative actions (in lower proportions); handlers 3 and 5 had the most ill calves under their care, and also animals which did not approach the test person. According to the MCA, it was possible to identify two groups of correlated variables: 1) animals which received neutral attitudes were more reactive, did not approach the test person; or vice versa, reactive animals did not approach the test person and received more neutral attitudes; and 2) animals which received more positive attitudes were tamer, thinner; younger and approached within 30 seconds; or the other way around, the tamer, thinner, younger and approachable animals received more positive attitudes. The older calves were also the most reactive ones ($r_s = 0.498$, $P < 0.001$), being the opposite also true. The Spearman correlation coefficient for the type of attitude (positive, negative, neutral and inconsistent) was positively correlated with the flight distance score ($r_s = 0.301$, $P < 0.001$), and positively correlated with the calf's age ($r_s = 0.470$, $P < 0.001$), body condition score ($r_s = 0.255$, $P < 0.001$) and to the latency to approach ($r_s = 0.113$, $P < 0.05$). The latency to approach also showed consistency when tested with de FDS where was shown to be positively correlated ($r_s = 0.352$, $P < 0.001$). Thus, we can conclude that the type of attitude can influence calf temperament and vice-versa, being neutral attitudes the one with the most reactive animals; poor handlers can make the welfare of the calves poorer; and an intense handler-calf relationship can also bring negative attitudes in lower proportions without compromising the welfare of the animals.

Keywords: behaviour, cattle temperament, human-animal relationship

ATITUDES DOS TRATADORES E SEUS EFEITOS SOBRE O BEM-ESTAR DE BEZERROS LEITEIROS

RESUMO – Bezerros leiteiros são separados de suas mães geralmente em algumas horas após o parto e, a partir deste momento, há um aumento expressivo da interação com os seres humanos. O objetivo deste estudo foi avaliar as atitudes dos tratadores para com os bezerros leiteiros, e os efeitos dessas atitudes no bem-estar desses animais. Foram realizadas observações diretas do manejo de aleitamento por três dias consecutivos, e as ações dos tratadores foram registradas em dez diferentes fazendas produtoras de leite nos estados de São Paulo (9) e Minas Gerais (1). Para avaliar o temperamento dos animais, três testes comportamentais foram realizados: escore de distância de fuga (FDS), teste de aproximação voluntária (VA) e avaliação qualitativa do comportamento (QBA). Um questionário contendo 21 questões foi aplicado para cada tratador. Um total de 12 tratadores e 328 bezerros foi avaliado. Para diminuir a dimensionalidade dos dados e entender as múltiplas associações entre as variáveis, foram realizadas: Análise de Componentes Principais (PCA), Análise de Fatores (FA) e Análise de Correspondência Múltipla (MCA), com uso do programa estatístico Statistica 7. Análises de variância confirmatórias foram realizadas usando o programa estatístico SAS; junto com a estimação dos coeficientes de correlação de Spearman (r_s) para as variáveis não paramétricas. Diferenças significativas foram encontradas para os descritores do QBA entre as 10 fazendas avaliadas ($F = 11.48$, $P < 0.0001$). De acordo com a PCA, as fazendas 1, 2, 3 e 5 tiveram os animais com pior temperamento. As fazendas 3 e 5 apresentaram os tratadores que menos se preocupavam com os bezerros, de acordo com as respostas no questionário. Após a FA, foi possível fazer um *ranking* dos ‘bons’ e ‘maus’ tratadores, demonstrando que os tratadores 1, 2a e 2b foram aqueles que mais apresentaram ações positivas em relação aos bezerros, mas também um maior número de ações negativas (em menores proporções). Já os tratadores 3 e 5 foram aqueles com o maior número de bezerros doentes e que não se aproximaram do observador. De acordo com a MCA, foi possível identificar dois grupos de variáveis correlacionadas entre si: 1) animais que receberam atitudes neutras eram mais reativos e não se aproximaram do observador (ou vice-versa, animais reativos não se aproximaram do observador e então receberam mais atitudes neutras por parte do tratador); e 2) animais que receberam atitudes positivas do tratador eram mais mansos, magros, jovens e se aproximavam em até 30 segundos do observador (ou vice-versa). Animais mais velhos eram também os mais reativos ($r_s = 0.498$, $P < 0.001$), sendo o contrário igualmente verdadeiro. A correlação de Spearman para o tipo de atitude (positiva, negativa, neutra e inconsistente) foi positivamente correlacionada com FDS ($r_s = 0.301$, $P < 0.001$), e positivamente correlacionada com a idade do bezerro ($r_s = 0.470$, $P < 0.001$), condição de escore corporal ($r_s = 0.255$, $P < 0.001$) e com a latência para se aproximar do observador ($r_s = 0.113$, $P < 0.05$). A latência para se aproximar apresentou consistência ao ser testada com FDS, onde se mostrou ser positivamente correlacionada ($r_s = 0.352$, $P < 0.001$). Dessa forma, conclui-se que o tipo de atitude pode influenciar o temperamento dos animais e vice-versa, sendo a atitude neutra aquela responsável por animais mais reativos; tratadores que não se preocupam com os animais podem fazer com que o bem-estar de bezerros diminua

e uma intensa relação tratador-bezerro pode acarretar em atitudes negativas em menores proporções sem comprometer o bem-estar dos animais.

Palavras-chave: comportamento, relação ser humano-animal, temperamento de bovinos

CHAPTER 1 – General considerations

1. INTRODUCTION

Animals are an important source of food and materials to humans and animal production has been growing year after year (FAO, 2013). At the same time, there has been an increased concern about the welfare of farm animals, and the public concern with the way animals are being treated (BROOM, 2008). Several attempts have been made to assess farm animal welfare and for trying to standardize it (FAWC, 1992; WELFARE QUALITY PROTOCOL, 2009; OIE, 2012). According to Dawkins (1990) all the concern with animal welfare is related to the attribution of a mental state to animals.

An important part in the farm animal life is the relationship with humans, especially their handlers. If this relationship is not good, it can bring disadvantages not only for the welfare of the animals (LENSINK et al., 2000b), but also for the production (BARNETT et al., 1992; HEMSWORTH et al., 1993; JONES, 1996; BREUER et al., 2003), and for the handlers, due to increased risks of labour accidents because the animals are more difficult to handle (GRANDIN et al., 1987; BOIVIN et al., 1992). It is proven that an early and constant contact with humans improves animal reactions toward people (BOIVIN et al., 1992; BOIVIN et al., 1994; BOIVIN and BRAASTAD, 1996). It is important to give the opportunity for this first contact to happen early in the animal's life, and to be as positive as possible.

In dairy cattle the human-animal relationship is an important issue, since in most commercial dairy farms the calf is separated from the dam immediately, or within a few hours after birth. In this case the first contact with humans is considered to be premature, and can influence the behaviour of the animals in the future (ARAVE et al., 1985; JAGO et al., 1999; LENSINK et al., 2000c; KROHN et al., 2001). Thus, a positive contact is important for trying to assure good animal welfare levels and to make the animals easier to handle for the stockperson.

2. LITERATURE REVIEW

2.1 Animal Welfare

Concern about the welfare of farm animals has been spreading around the world with great speed, leading many producers to perform changes in the management and in the way they treat their animals (BROOM, 2008). This is motivated not only by the growing number of consumers of animal products, who associate the quality of these products with the given condition of animal welfare on farm, during transport and at slaughterhouses; but also by a greater knowledge about animal behaviour and animal feelings nowadays.

The welfare of an individual is defined as “its state as regards its attempts to cope with its environment.”, and it could vary from very good to very poor (BROOM, 1986). Thus, if the animal is allowed to satisfy its needs in a given environment, its welfare is then, considered ideal (BROOM, 1997). Animal welfare is a multifaceted area under discussion, stimulating scientists from different areas, including animal behaviour, animal physiology, nutrition, microbiology, environmental design, studies of the human-animal relationship, veterinary epidemiology, among others, to get involved in research projects, looking for answers to the questions related to this subject (FRASER et al., 2013). For instance, “...how can animal welfare be measured...” and “...how can good welfare standards be applied in farms’ daily management...” (BROOM, 1988; DE JONGE et al., 2000) are important questions to be answered.

One important approach to address animal welfare is the understanding of animal emotions. According to Dawkins (1990) all the concern with animal welfare is closely related to the attribution of a mental state to the animals; meaning the possibility of animals to feel emotions, similarly as we, humans, do (DAWKINS, 2006). Emotions are recognized as a product of various neural and hormonal changes (BROOM, 1998), also known as “bio-regulatory reactions that aim at

promoting, directly or indirectly, the sort of physiological states that secure not just survival but survival regulated into the range that we know as well-being” (DAMASIO, 2004), leading to feelings or not. These “bio-regulatory reactions” are individual’s responses produced by the brain when an object or situation is perceived or recalled from memory (DAMASIO, 2004). Feelings are the internal brain organization of each animal (BROOM, 1998), being also recognized as “the perception of an emotional state, meaning the perception of a body state changing together with the perception of a mental processing” (DAMASIO, 2004). Nowadays, for animals, emotions and feelings are considered the end-product of an evaluative process, not only a pre-programmed reaction (BOISSY, 2013¹).

For Darwin (1871) it was obvious that animals had feelings. In his book, *The Descent of Man*, he describes comparatively how animals show their feelings in ways very similar to humans, besides comparing the mental powers of various species. Since then, research on animal welfare has developed considerably and animal welfare standards have improved. Later in the seventies, the Farm Animal Welfare Council created the “Five Freedoms” (FAWC, 1992), synthesizing what must be considered to assure a good animal welfare condition, as follows: 1) freedom from hunger and thirst; 2) freedom from pain, injuries and diseases; 3) freedom from fear and distress; 4) freedom from discomfort; and 5) freedom to express natural behaviour. For many years, these principles were the basis for the development of many initiatives for understanding animal welfare and for the development of strategies to promote farm animal welfare. More recently, an international project addressing this subject (Welfare Quality Project®, 2009) established four principles (closely linked to the “Five Freedoms”) that should be taken into account to assess farm animal welfare, which were: 1) good housing; 2) good feeding; 3) good health; and 4) appropriate behaviour. The protocols resulting from this project gave directions to practical welfare assessment in commercial farms of beef and dairy cattle, poultry and pigs.

Further in 2012, a group of neuropsychologists and scientists signed the Declaration of Consciousness, which recognized in society, that animals are sentient beings (THE CAMBRIDGE DECLARATION ON CONSCIOUSNESS, 2012).

¹Alan Boissy, “Emotions and cognition: a behavioural approach to assess welfare in farm animals” (lecture). Swedish University of Agricultural Sciences, Uppsala, Sweden, October, 2013.

According to Webster (2001) “sentience implies an awareness of the nature of emotions associated with pleasure and suffering. Many of these emotions are associated with primitive sensations such as hunger, pain and anxiety”. Still in 2012, “10 General Principles for the Welfare of Animals in Livestock Production Systems” were adopted by the 178 member nations of the World Organization for Animal Health, to guide the development of specific standards for various animal species (OIE, 2012). The most important thing in the OIE principles is the comprehensive description of each principle, helping their practical applicability in commercial farms. Besides, they have a complete approach on animal welfare, addressing the subject throughout the assessment on the animals, environment, stockmanship and human-animal relationship. The ten OIE principles are: 1) Genetic selection should always take into account the health and welfare of animals; 2) The physical environment should be suited to the species and breed so as to minimize risk of injury and transmission of diseases and/or parasites to animals; 3) The physical environment should allow comfortable resting, safe and comfortable movement, including normal postural changes, and the opportunity to perform types of natural behaviour that animals are motivated to perform; 4) Social grouping of animals should be managed to allow positive social behaviour and minimize injury, distress and chronic fear; 5) Air quality, temperature and humidity in confined spaces should support good animal health and not be aversive to animals. Where extreme conditions occur, animals should not be prevented from using their natural methods of thermoregulation; 6) Animals should have access to sufficient feed and water, suited to the animals’ age and needs, to maintain normal health and productivity and to prevent prolonged hunger, thirst, malnutrition and dehydration; 7) Diseases and parasites should be prevented and controlled as much as possible through good management practices. Animals with serious health problems should be isolated and treated promptly or killed humanely if treatment is not feasible or recovery is unlikely; 8) Where painful procedures cannot be avoided, the resulting pain should be managed to the extent that available methods allow; 9) The handling of animals should foster a positive relationship between humans and animals and should not cause injury, panic, lasting fear or avoidable stress; and 10) Owners and handlers should have sufficient skill

and knowledge to ensure that animals are treated in accordance with these principles (FRASER et al., 2013).

Unlike humans who have the verbal language to express their feelings, in animals we can try to understand how they feel with behavioural and physiological measures (DÉSIRÉ et al., 2002). Indeed, only with the combination of measures, such as behaviour, physiology, growth rate, health etc., will a trustable assessment of animal welfare be possible to evaluate how good or poor the welfare of an individual is (BROOM, 1987). It is well accepted that more than a single measure is needed for an accurate animal welfare assessment (DAWKINS, 2004). As physiological indicators we have, for example, increased respiration, heart rate and adrenal activity (rapid responses) or reduced immune response after challenge (considered a slower response) which may indicate that the welfare is poorer than in individuals that do not show such changes (MOBERG, 1985). Baldock et al. (1988) for example, measured the heart rate of ewes exposed to different types of handling situations, such as dog approach, introduction to a new flock, visual isolation from companions and transportation. As a result, the authors found that the animals showed elevated heart-rates in the first four situations and a smaller response during transportation. Broom et al. (1986) also used physiological indicators to assess hen welfare after handling and transport. They found that the birds showed higher respiration rates, increased blood corticosterone levels after handling and increased brain levels of noradrenalin, which was measured after slaughter.

Besides physiological measures, behavioural indicators are also systematically used to assess animal welfare. For example the flight distance test and avoidance scores can provide important information about animal feelings towards humans; the highest reaction to move away or escape in the presence of a human being represents the worst status in the welfare of the animal (BROOM and MOLENTO, 2004). Such tests offer the possibility to assess fear responses and also a component of exploratory behaviour (BROOM and MOLENTO, 2004). Each test plays a role in identifying the stress suffered by animals during routine handling and consequent behaviours that may have an economical influence on farms (BURROW, 1997). In most cases, these tests are applied to measure fear, which is assumed to

be an emotional state of suffering (JONES and WADDINGTON, 1993). Actually, measuring fear is not easy, since the responses to the tests vary a lot among individuals who can express fear by being either immobile and apathetic, or by being hyper active, during which time the two most common reactions are flight or fight (BOISSY, 2013¹).

Nowadays it is accepted that not only the absence of negative experiences guarantees that the welfare of a given animal is good, but it is necessary to consider the occurrence of positive emotions, such as happiness and pleasure, in order to draw this conclusion (BOISSY et al., 2007). According to Broom (1988), an excellent way to discover what is good for animals is making preference tests; in which it is possible to observe their preferences and also to know how hard they will work to get a resource or an object, or even to have the opportunity to express a specific behaviour. It is actually a way of asking the animals what is better for them, increasing welfare levels (VON KEYSERLINGK et al., 2009). For example, Dawkins (1983) tested the cage size and flooring preferences of cage-reared hens. The author tested the birds' preference for small *versus* large cages; and litter *versus* wire flooring. The animals spent more time in large cages, and also preferred the litter flooring over the wire one. In another study by Legrand et al. (2009) tested the preference of cows between a free-stall barn and an adjacent pasture area; the authors found that the animals preferred to stay on pasture, but only at night. During the day, due to the warm weather and the solar radiation, they stayed most of the time inside the barn. Thus, if the weather was cooler, they would probably want to stay outdoors also during the day.

Another important thing to consider are "positive mood" behaviours (BOISSY, 2013¹), such as play behaviour (JENSEN and KYHN, 2000); affiliate behaviour (e.g. allo-grooming) (SATO et al., 2003) and vocalizations (e.g. purring in cats) (KNUTSON et al., 2002). The body language such as the position of the ears, tail wagging and neck position, can also tell us a lot about the present feeling of the animal (REEFMAN et al., 2009; BOISSY et al., 2011). In sheep, different ears positions are presented in different situations, as reported by Boissy et al. (2011), who observed that the backward ears position is related to unfamiliar situations; and asymmetric position to

¹ Alan Boissy, "Emotions and cognition: a behavioural approach to assess welfare in farm animals" (lecture). Swedish University of Agricultural Sciences, Uppsala, Sweden, October, 2013.

sudden situations. Thus, animals can show us how they feel; all we need is to learn how to interpret their signs.

2.2 Cattle temperament

Temperament is known to be a complex individual characteristic formed by several different traits, including fear, boldness, reactivity, curiosity, among others. According to these different characteristics, each experience lived by each individual will present singular responses and differences in the way it is witnessed, making every animal respond differently to different types of stimuli, although these responses are usually consistent across experiences (RÉALE et al., 2007). As the difference between temperament and personality has not been maintained consistently in the literature (JONES and GOSLING, 2005), for this study the definition proposed by Réale e Dingemanse (2012) was used which says that temperament is the same as personality; and that “behavioural differences are consistent over time and across situations, becoming predictable”.

Indeed this topic has been intriguing scientists for a long time. Pavlov (1906) started studying different personalities in dogs, and ended by classifying them according to four major characters according to their responses to stressful situations, as follows: 1) Strong excitatory (dogs easily became very excited with moderate levels of stress); 2) Lively (also very responsive to stress, but were not as extreme in their reactions as the strong excitatory animals); 3) Calm imperturbable (had a generally passive response to stress, coping well with it; they neither became particularly excited nor paralyzed); and 4) Weak inhibitory (reacts to stress with extreme passivity in order to avoid tension. High levels of stress led them to a state of virtual paralysis, including inhibition and blocking brain functions). Additionally, Hebb (1946) studying adult chimpanzees discovered that knowing each animal’s “peculiarities” was safer to the handlers when managing them. New staff members were introduced to the animals by the older members and could handle the animals without getting hurt; which could easily have happened if they did not know the temperament of each chimpanzee.

Only in recent years has farm animal temperament started to be studied. Methods for scoring temperament were developed in early 1960s (STRICKLING and KAUTZ-SCANAVY, 1983), and over the past 50 years a number of objective and subjective methods have been created. In most farmed species, these differences are shown with great clarity in the presence of the human; this is why behavioural tests are so frequently used, allowing us to assess these characteristics and qualifying the reaction of the animals toward humans (FORDYCE et al., 1982). Multiple methods are often employed to improve accuracy (BURDICK et al., 2011).

By measuring the reactions of animals with tests, we also get an idea of how animals perceive the human (WAIBLINGER et al., 2006), creating strategies to improve their welfare. Different emotions and motivations are associated with the perception and the reaction of each animal towards the human presence. In some studies, the different flight distances, larger or smaller, can be influenced by negative or positive managements, respectively (BOISSY and BOUISSOU, 1988; MUNKSGAARD et al., 2001). This has a direct influence on the welfare, as some animals may be more likely to feel good emotions, such as happiness and satisfaction, than others (BOISSY et al., 2007).

Factors influencing the temperament according to Burrow (1997) are age (experience), sex, breed, handling, maternal effect, and other factors such as diseases and management procedures considered invasive (e.g. dehorning). Since all these factors are viewed in a different way by each individual, it is expected that the responses are going to be unique for each one of them. Thereby, using temperament studies we can predict the responses of an individual in a given situation, based on responses taken from previous situations (BOISSY et al., 2007), avoiding unnecessary distress. For example, if an animal is known to be aggressive in certain situations, it is possible to create alternatives to handle it in different ways, reducing the risks of accidents to the human, and the stress response for the animal.

Individual differences in behaviour were demonstrated, for example, in pigs by Broom (1987) and Broom and Johnson (2000), when some confined pigs showed stereotyped behaviours, while others remained inactive, with no apparent response to the situation. Many studies with cattle also presented evidence of individual differences in behaviour. For example, Boissy and Bouissou (1995) submitted 14

heifers to four different fear-eliciting situations and found that the animals were consistent throughout the tested situations; the most frightened animals in the first test being also the most frightened in all other tests. In another study, Florcke et al. (2012) assessed individual differences of beef cows 24 hours after calving, regarding calf defence and maternal protective behaviour; the cows presented different types of behaviour: 99% of the cows moved between the threat and their calf to protect it, 13% lowered their heads as a sign of aggression and 78% vocalized. Furthermore, Gibbons et al. (2011) assessed the consistency of dairy cattle temperament in different situations, finding that animals do present a personality that is consistent over time, and over situations.

Despite the environmental components playing an important role in shaping the behaviour of each animal; genetics also has a fundamental function in the expression of temperament (GRIGNARD et al., 2001). When comparing the flight distance of dairy and beef cattle breeds reared in the same conditions, Murphey et al. (1981) found that dairy cows were always closer to humans, while beef animals were more reactive in the presence of humans. Following the same objective, Dodzi and Muchenje (2011) evaluated the temperament of different breeds of dairy cows, and deduced that some types of handling were more aversive to certain genotypes than to others. Temperament has also been considered in beef cattle breeding programs. With this purpose, Sant'Anna et al. (2013) estimated the genetic parameters of four temperament traits (flight speed, crush score, movement score and temperament score) in Nellore cattle, and found that all four traits had "enough genetic variability to respond to selection".

Other examples of how individual differences can influence the production are found in several farm animal species. In studies made with sheep it was found that those animals which were less reactive to humans had higher reproductive success and higher milking production (DIMITROV et al., 2005) compared to the more agitated ones and Sart et al. (2004) demonstrated that calmer sheep produced better milk quality, with higher levels of protein, showing that temperament must be considered when selecting animals for milk production. These studies support the work of Voisinet et al. (1997), working with cattle, who observed that the calmer the

animals, the higher the daily weight gain, when compared to animals which were excitable.

Another interesting way to assess the temperament is through the QBA method (Qualitative Behaviour Assessment). This method uses visual analogue scales to assess the animal's body language and the perception of the observer to provide a basis for assessing the quality of the animal's experience in its environment. It can also be used to measure the health and/or the welfare of animals in a more detailed way, by "observing the animal as a whole" (WEMELSFELDER and LAWRENCE, 2001). This measure can show us how the animal interacts with certain environments, giving the basis to establish individual personality traits, besides being simple and easy to apply on farms (WEMELSFELDER et al., 2000). With that in mind, Sant'Anna et al. (2013) developed a way of assessing different temperament traits in farm animals during handling. The authors assessed 2229 animals of the Nellore breed, associating QBA with other temperament measures such as movement in the crush, crush score, flight speed test and temperament score. The authors confirmed what was already expected, that the QBA can be used as a temperament measure, as it is efficient in the detection of animals' individual differences during handling. This method also works because different characteristics can be combined as temperament elements, for example: confident, nervous, calm and agitated are some of the most commonly used combining elements (WEMELSFELDER et al., 2000; BOTREAU et al., 2009). In another study to identify individual differences in social behaviour, Rousing and Wemelsfelder (2006) video recorded dairy cows kept in free stall barns and then applied the QBA method while watching the videos. They could see the distribution of each cow according to their behaviour, and the behaviours presented when together with the other animals; and concluded that each cow had their own way of interacting with the environment, besides demonstrating that the QBA can be a reliable measure for assessing animal social behaviour.

2.3 Human-animal relationship

The relationship between humans and animals began a long time ago and since animal domestication, this relationship has become increasingly closer (LENSINK, 2002). However, increased investments in technology and the intensification of production (e.g. by using automatic feeders, mechanical milking machines in dairy production etc.) has changed the way the contact among humans and animals happens; usually resulting in a reduction of positive interactions with humans. For example, feed delivery could be recognized by the animal as a positive action from the human, but the use of automatic feeders almost eliminates the opportunity for this positive interaction (RUSHEN et al., 1999).

On the other hand, researchers have shown that many of the daily handling activities performed by the stockperson on farms make the animals more fearful of humans (HEMSWORTH, 2003). For example, Martin Guimarães et al. (2013) measured the flight distance score of dairy calves before and after dehorning, which is a common practice in commercial dairy herds and found a significant increase in the avoidance reaction to the test person after the procedure. Besides, as farm animals were essentially prey animals in nature, they will always try to escape from potentially harmful situations. Thus, humans performing usual handling procedures, with loud noises and sudden and unpredictable movements, could be seen as predators by the animals, increasing avoidance and fear responses (RUSHEN et al., 1999).

Several studies have shown that fearful animals are notoriously more difficult to manage (GRANDIN et al., 1987; BOIVIN et al., 1992), and may cause accidents. For instance, according to Grandin (1994) the management of animals which are not used to humans carries risks for both the safety of the handler and the welfare of the animal. The handler's work also become more difficult when animals show defensive reactions increasing the time spent in that activity, together with the chances of injury (LE NEINDRE et al., 1996), and reducing the worker comfort and time efficiency (BOIVIN et al., 2003). A constant positive contact with humans can make animals less fearful and easier to handle (BOIVIN et al., 1994).

Based on these results, it is often recommended to balance the effect of daily positive interaction with the negative handling, such as catching and restraining for vaccination, medicine administration, transport etc. (RUSHEN et al., 1999). In poultry, for instance, the fear reaction can bring welfare and economical losses, since these animals may panic and show violent escape reactions to human presence, wasting unnecessary energy, and lowering meat and egg production (BARNETT et al., 1992; JONES, 1996). Consequently, the development of a positive relationship can bring beneficial consequences for both, handlers and animals (WAIBLINGER et al., 2006).

The extent to which animals are in contact with humans affects its subsequent behaviour (BOIVIN et al., 1994), making an early life contact better for a less fearful future. This was tested by Boivin et al. (1992), who compared cattle handled by humans at different stages of life. He showed that the animals managed earlier did not present any aggressive reaction to the test person afterwards, unlike those that had a later contact with humans, which were more difficult to handle. In another study, Boivin et al. (1994) compared two groups of calves, reared in the first three months of age in different conditions: one group was reared indoors, being separated from their mothers one day after birth, having a lot of visual and tactile contact with the handlers; and the other was reared outdoors, with their mothers, having little contact with humans. After this period they were both kept under the same conditions, at pasture. At 20 months of age, they were tested, and the animals reared indoors, with more human contact, showed significantly lower heart rates during human approach, were significantly easier to restrain and did not present any aggressive reactions towards humans when compared to the outdoor-reared animals, which kicked and tried to escape more often. Additionally, in the study carried out by Boivin and Braastad (1996), the authors assessed three groups of dairy goat kids handled by humans. The first group was the control, which did not receive any additional contact; the two others were isolated and gently handled twice a day: one was handled for ten days after weaning, and the other at six weeks of age. The group handled earlier stayed closer to humans, approaching and interacting more, and also vocalized less when isolated, compared with the group handled at six weeks of age or not handled at all.

Positive and negative actions from the handlers can also influence the welfare and productivity of the animals. This was confirmed in the results found by Hemsworth et al. (1993), who conducted a study on 25 pig farms. In half of the properties the stockpersons were trained and the negative interactions (e.g. slaps and shouts) were reduced; while in the rest of the properties no training was performed. The authors found significant differences in the handler attitudes towards the animals, decreasing negative interactions with animals after training; they also found changes in pig behaviour, which decreased the expression of fear in the presence of humans. In addition, there was an average increase of 6% in the number of piglets born per sow in trained stockperson farms, against a reduction of 3% in control farms. Additionally, in another study from the same research group (Hemsworth and Barnett, 1991), the authors also demonstrated a decrease in reproductive performance in pigs that showed greater fear reactions toward humans.

In a study with dairy cattle, Rushen et al. (1999) submitted 14 cows to two different handlers: one treated them positively and the other, aversively. When the aversive handler was present during milking, there was an increase of residual milk by 70%. In another study carried out by Breuer et al. (2003), two groups of dairy heifers were positively and negatively handled, the results showed that the negative handled animals were more agitated, and approached less either the familiar or unfamiliar person; they also tended to weigh less. On the other hand, as a positive effect of a good human-animal relationship, Lensink et al. (2000a) assessing the management of 50 veal calves found that positive attitudes of the stockperson were highly correlated with better development of the animals, including higher weight gains. These can support that if a healthy human-animal relationship is not established, there is a risk of damaging performance and welfare of the animals. Actually, most of the time animals react to humans' actions rather than initiating the interaction themselves (WAIBLINGER et al., 2006). An important part of understanding the human-animal relationship therefore is the comprehension of the role of the handler attitudes toward the animals. To realize how the stockperson perceives the animals, it is important to analyse the psychological side of the person, as well as their attitudes and personality traits (LENSINK, 2002). According to Seabrook (1988), the self-confidence of the workers directly influences the decisions

they are going to make. Furthermore, Grommers (1987) identified “the human factors important for managing animals”, which can be divided into: 1) personal (attitudes and personality) and 2) job-related (experience and knowledge). It is possible then, to use these factors as the basis to start assessing the handler’s side of the relationship.

Nowadays, researchers have also been using the Theory of Reasoned Action, proposed by Fishbein and Ajzen (1975) to understand handler attitudes. The theory proposes that the behaviour toward a particular object is approximated by an intention to perform that behaviour, i.e. if we look carefully to someone’s intentions, we will be able predict how that person is going to behave. Reasoned Action predicts that behavioural intent is caused by two factors: 1) the person’s attitudes; and 2) the person’s subjective norms. Attitudes have two components: evaluation and strength of a belief. Subjective norms also have two components: normative beliefs (what the person thinks others would want or expect him/her to do) and motivation to obey (how important it is for the person to do what he/she thinks others expect) (AJZEN and FISHBEIN, 1980). Thus, another way of assessing a stockperson’s behaviour, other than observing it, is by trying to discover about the person’s intentions. Questionnaires and interviews are used with this purpose, to know more about the understanding of the stockperson in their routine activities. For instance, if handlers believe cows are sensitive and smart animals and can easily learn, it is very likely that they have more patience during handling and will make it more carefully (WAIBLINGER et al., 2002). On the other hand, if a handler associates the delay of a pig to move with lack of intelligence and feelings, he will probably make use of negative actions (such as slaps and shouts) for the animals to move quicker, causing the animals to have a constant fear of humans (HEMSWORTH and BARNETT, 1991). Breuer et al. (2000) assessed the attitudes of handlers based on their opinions about the behaviour of cows, and about their own behaviour, using questionnaires. There were questions associating the behaviour of the cows during milking and the handlers’ own attitudes according to the cows’ behaviours. In addition, all the tactile stimuli, both positive (pats, strokes and hand resting) and negative (slaps, pushes and hits), performed by the handler when moving the cows, were recorded. In a similar study, Waiblinger et al. (2002) used three types of

questionnaires to assess the perspectives of the stockperson and his attitudes (beliefs, emotions and behavioural intentions) towards dairy cow characteristics and behaviour; they also observed all tactile and acoustic stimuli presented during handling routines. In both studies, Breuer et al. (2000) and Waiblinger et al. (2002), the authors found a strong correlation between the attitudes of the stockperson and the behaviour and production of the cows, showing that both humans and animals are influenced by each other. This is why it is so important to study both sides, not only animals and not only humans, to have a complete scene of this relationship. As Boivin et al. (2000) once wrote, “The animal behaviour reflects, as a mirror, the human behaviour, and in certain circumstances, the human behaviour reflects the animal behaviour”.

2.3.1 Handler-dairy calf relationship

The relationship between the dairy calf and its handler has some particular characteristics. The dairy industry separates the calf from the dam soon after birth because of a genetic selection over time, which decreased the maternal ability and guaranteed the cow will give milk without its offspring around. This means the first contact with the human happens even earlier than in other species, and as shown in the previous sections, this early life contact can influence animal behaviour in the future, its welfare and its subsequent production.

In a study assessing how handling at different periods after birth can influence the response of dairy calves to humans, Krohn et al. (2001) divided 40 calves into four treatments. The animals were separated from their dams soon after birth, then handled (petting, stroking and talking) and fed by humans for four consecutive days in different ages; except for the control group which was not handled at all. Although all three treatments had a shorter flight distance than the control group, calves handled earlier (1-4 days of age) approached the unknown person quicker and faced people more often than the other groups (6-9 and 11-14 days of age). Herewith, we can conclude that the human contact made in the first days of the calf's life can influence its behaviour in the future.

In another study, Jago et al. (1999) investigated how calves respond to human contact. They separated four groups of dairy calves and submitted each group to different treatments from 3 to 17 days of age, as follows: 1) fed without visual contact with humans and not handled (control); 2) fed without visual contact with humans and handled (stroked); 3) fed with visual contact with humans but not handled; 4) fed with visual contact and handled. After performing behavioural tests with the animals until 62 days of age, the authors found that calves which associated the human presence with food were always quicker to approach. The interesting thing was that there was no significant differences between the four treatments; suggesting that the human contact in the first two days after birth is extremely important for the animal to associate the human with good feelings, such as food delivery.

Furthermore, the implication of the handler's attitudes towards the behaviour of calves in a veal unit was studied by Lensink et al. (2000b). The authors separated two groups of calves: one group received minimal human contact, while calves in the other group were stroked and could suck the handler finger after meals; the study was carried out during 21 weeks. Calves that were handled interacted more often with the test person, stayed closer to humans and defecated less when in the human presence, when compared with the non-handled group. The authors concluded: "Such lower reactivity to people could improve ease of handling, animal performance and welfare", i.e. it would only bring beneficial results. The same research group (LENSINK et al., 2000c) also studied if these beneficial results would influence welfare, productivity and meat quality in the same veal unit, and found that the handled animals were less agitated and had fewer abomasal lesions, which can improve their welfare levels.

Another study made by Magalhães Silva et al. (2007) compared two types of handling. The first was the traditional handling, usually adopted by commercial dairy farms in Brazil, where the animals were kept individually, and receiving milk from teatless buckets, with few opportunities to interact with the handlers. The second handling procedure, called good practices of handling, included group housing, receiving milk from teat-buckets and having the handler closer to the animals while suckling, including talking and stroking the calves. At the end of the study, the authors found a decrease in the mortality rate, a decrease in the antibiotics use and

the animals presented higher weight gains at weaning age. Consequently, a good handler-calf relationship can improve the welfare of the animal, lowering the production costs and making the animal easier to handle in the future.

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CHAPTER 2 – Handlers' attitudes toward dairy calves and their effects on calf welfare

Atitudes dos tratadores e seus efeitos sobre o bem-estar de bezerros leiteiros

1. INTRODUCTION

The human-animal relationship in the dairy industry is one of the closest among all farmed species. The dairy calf is usually separated from the dam within few hours after birth and, due to this early separation it starts a very premature relationship with the handlers, who are responsible for supplying the calf's needs. Several studies have shown that this early relationship can influence animals throughout their whole life (ARAVE et al., 1985; BOIVIN et al., 1992; BOIVIN et al., 1994; BOIVIN and BRAASTAD, 1996). Indeed, a poor relationship is proven to have negative impacts on the animals' productivity (HEMSWORTH and BARNETT, 1991; RUSHEN et al., 1999; BREUER et al., 2003) and leads to lower levels of animal welfare (LENSINK et al., 2000c). On the other hand, a good relationship is shown to increase efficiency and welfare levels (HEMSWORTH et al., 1993; LENSINK et al., 2000b). In dairy calves, the first days after birth were shown to be crucial for their behaviour in the future (JAGO et al., 1999; KROHN et al., 2001; LENSINK et al., 2000b; 2000c). Hence, establishing a healthy human-animal relationship as soon as possible can be worthwhile for the animal that will have higher levels of welfare; for the handler, who will face less risks of labour accidents (GRANDIN, 1994); and also for the producer, earning more money.

Both the animal and the human play an important role in this relationship; as most of the time, animals react to human actions rather than initiating the interaction itself (WAIBLINGER et al., 2006). Thus, we should try to understand how the handler perceives their animals and their own actions towards them. According to Grommers (1987) we should investigate not only the personality traits, but also the job-related

characteristics, such as experience and knowledge for a better understanding of the human-animal relationship. With this purpose, many researchers have used questionnaires and interviews (BREUER et al., 2000; WAIBLINGER et al., 2002) to try to understand more about what is going on inside the handler's mind, usually using the Theory of Reasoned Action (for more information about the theory see FISHBEIN & AJZEN, 1975 and AJZEN & FISHBEIN, 1980).

The aim of this study was to assess stockperson attitudes toward dairy calves and their effects on calf welfare.

2. MATERIAL AND METHODS

2.1. Farms, handlers and animals

Data collection was carried out from April to July 2013 in ten farms located at the states of São Paulo (9) and Minas Gerais (1), southeast of Brazil. In all of them the calves were separated from the dam within 24 hours after birth, starting the relationship with the handler early in their lives. All handlers responsible for taking care of the calves lived on the farms.

Calf birth date, birth weight (when available), past diseases (when available), breed (when available) and body condition score (ranging in a scale from 1 to 5, using a 10 point scale) were recorded. A total of 328 calves were assessed and the number of calves evaluated per farm ranged from 6 to 111. All calves received milk twice daily, except for farm 7 (automatic feeder) and the animals during gradual weaning, that received it once a day. All farms had shaded areas for the animals either kept in groups or individually, and free access to water, concentrate feeding and forage. The description of calves and handlers per farm are presented in Table 1. The number of each farm was given following a chronological order of data collection.

Table 1. Numbers of calves and handlers assessed per farm, and the description of respective housing systems and breeds

Farms	Number of calves	Handler identification	Handler sex (age)	Time working in the property	Housing system	Breeds	Amount of milk (liters)/ Method of delivery	Type of weaning (calf age/weight)
1	49	H1	F (30)	10 years	G	Cb	6L / TB	Gradual (70d/70kg)
2	63	H2a / H2b	F (27) / F (30)	1 y / 3 y	I	Cb	6L / RB	Gradual (70d/70kg)
3	27	H3	F (28)	8 months	I	J	3L / RB	Abrupt (60d)
4	6	H4	M (34)	4 months	I	J	4L / BB	Gradual (70d)
5	11	H5	M (20)	3 years	G	Cb	4L / BB > CT	Abrupt (70d)
6	6	H6a / H6b	M (29) / F (25)	5 y / 10m	I	Cb	4L / BB > RB	Abrupt (70kg)
7	111	H7	M (45)	3 years	I/G	Cb/H	4L / TB > AF	Gradual (70kg)
8	32	H8	F (37)	7 years	I	Cb/H	4L / TB > RB	Gradual (70kg)
9	6	H9	M (24)	1 year	I	H	4L / RB	Gradual (70d)
10	17	H10	M (23)	10 months	I	Cb	4L / RB	Gradual (70d)
Total	328	12						

Where F = Female; M = Male; G = Group housing; I = Individual housing and I/G = Individual housing until certain age, then in groups. Cb = Crossbred; J = Jersey purebred and H = Holstein purebred; Calf age in days, calf weight in kilos. TB = Teat bucket; RB = Regular bucket; BB = Baby bottle; CT = Collective trough; AF = Automatic feeder; ">" = Method of delivery changed at a certain age.

2.2 Description of the farms

Farm 1 was the only one, among the ten farms studied, to adopt the good practices of handling for dairy calves as routine (MAGALHÃES SILVA et al., 2007). It involves keeping the calves in contemporary groups, loose in paddocks and receiving milk in buckets with teats, which allow the calves to express suckling behaviour. Positive tactile stimulation is routinely performed by the handler, by stroking the calves while they suckle. Farms 2, 3, 9 and 10 kept calves individually, held by a two-meter chain attached to a wire fixed on the ground. In the middle of the wire there was a small shaded area, where the animal could escape from direct solar radiation. This system allowed them to walk or run, backwards and forwards, and even to have contact with neighbouring calves. Farm 4 kept calves in individual pens of 4m², inside an open barn, with the possibility for contact with neighbouring calves. Once a day the animals were released outside for the facilities' cleaning, and for about five minutes they could run and walk around. Farm 5 kept calves in group in a paddock (8 x 4 m) outside the milking parlour. Farm 6 kept their calves individually, held by a 2 meter rope to small opened metal houses, with shaded coverage. They did not have any tactile contact to each other. In farm 7 the calves were kept until 20 days of age in suspended cages measuring 1.2 m², with a wooden floor. After that they were loose in paddocks with an automatic calf feeder. On average, one calf feeder was used per 12 calves. Finally, in Farm 8 the calves were kept tied by a 2 meter rope to individual wooden houses. They did not have any tactile contact with other calves. Farm 2, by the time of the data collection, had 175 calves. For a faster collection, 63 calves were randomly selected among the 175 ones.

2.3 Behavioural tests

The calf identification (usually ear tags) was used to control the individual records in each farm. In cases where they did not have a farm identification, each animal was identified with a numbered plaque fixed to their individual housing (farms 3, 6, 8 and 9), or with coloured tapes fixed around their necks (farms 4 and 5). All tests were conducted by the same person and dressed in the same manner, to reduce the

possible influence of different experimenters. The tests were always performed after the morning suckling, to minimize the effect of hunger.

2.3.1 Qualitative behaviour assessment (QBA)

The QBA is a method developed to integrate different aspects of the animal body language in order to assess its welfare state (Wemelsfelder and Lawrence, 2001). In this study, QBA was used to assess the temperament of each calf, according to the adaptation proposed by Sant'Anna et al. (2013), who reduced the number of descriptors from the 20 initially recommended (active, relaxed, fearful, agitated, calm, content, indifferent, frustrated, friendly, bored, positively occupied, lively, inquisitive, irritable, uneasy, sociable, apathetic, happy, distressed) to 14 (active, relaxed, fearful, agitated, calm, tense, playful, frustrated, curious, uneasy, apathetic, comfortable, sociable and indifferent). A Visual Analog Scale (VAS) was used, where each descriptor was represented by a 125 mm long line, and the impression of the observer was marked on this scale, assuming the “minimum” as the absence of the expression of each descriptor, and “maximum”, its most intense expression. Each calf was observed for 30 seconds and then the observer filled the worksheet. The measurements were obtained later for each descriptor, using a ruler and measuring the distance in millimetres from the left edge to the observer's mark. A copy of the worksheet used in the study is presented in Appendix 1.

Again, only one person was responsible for the QBA assessment. She was well acquainted with dairy cattle and was trained in making this assessment before the data collection started.

2.3.2 Flight distance score (FDS)

The standing animal was approached slowly from the front, without eye contact by the test person, who had her arms close to her body until she was at a distance of one stretched arm (one meter) away from the calf. At this point, the test person waited for ten seconds and then stretched out her arm, attempting to touch the animal. The score was given in the moment the calf made the first movement away

from the test person, beginning when she started to approach from a distance of two meters or more. Then, it was given a score from 1 to 5, as follows: 1) The calf accepted to be touched by the test person; 2) The calf accepted the person when she stretched out her hand, but avoided being touched; 3) The calf accepted the test person standing at a distance of 1 m (stretched arm), but avoided her when she stretched out her hand; 4) The calf avoided the approaching person at a distance of between 1 and 2 m; and 5) The calf avoided the approaching person at a distance of more than 2 m.

After the data collection the animals were classified into three categories according to their flight distance score, to facilitate the analysis, balancing the number of animals in each group: 1) Tame animal: score 1; 2) Less reactive animal: scores 2 and 3; and 3) Highly reactive animal: scores 4 and 5.

2.3.3 Voluntary approach test (VA)

A test person (an unknown person for the calves) stood quietly, without making eye contact or any sudden movements for a pre-determined time. During this time, the animals that approached the person until touching her with its nose were identified, and considered to be less fearful of humans. The test had to be adapted from farm to farm, due to differences in the housing systems, but it was always performed in the calves' usual environment.

In the case of group housing (farms 1, 5 and 7), the test person stood quietly in the middle of the calves' paddock, allocating one minute per calf; waiting for the animals to approach or not. We did not isolate each calf from its group because this action influenced their behaviour. In the case of individual housing, when the calf was tied (farms 2, 4, 6, 8, 9 and 10), the test person stood near the individual shaded area of each calf for two minutes and waited for the calf to approach or not. In the cases where calves were in individual stalls (farms 4 and 7), the test person stood just outside, very close and touching the fence, also for two minutes, recording if the animal approached or not.

All tests were video recorded, using a digital camera (SONY DSC-TX5), being later analysed by the same person who had performed the test, using a stopwatch to measure the calves' latency to approach.

For the analysis, the calves were classified into three groups, defined by the calves which: 1) did not approach the test person; 2) approached within 30 seconds; or 3) approached after 30 seconds.

2.4 Assessing the quality of the handler-calf interaction

At the beginning of the study the farm owners were informed about the methodology to assess the handler-calf interactions, but not the handlers. This was done to avoid behavioural changes of the handlers during the observations. The handler was told that the study concerned only the calves' behaviour.

2.4.1 Questionnaire

A questionnaire consisting of 21 questions was completed for each handler, individually. This questionnaire was always the last part of the data collection on the farm and it was always presented by the same person.

The questionnaire was divided in two parts, the first part concerned the professional characterization, with questions about the handler's schooling, life and experience with dairy calves; the second part addressed their knowledge and perception about cattle welfare, and their feelings and behaviours towards the calves (COLEMAN et al., 2000; WAIBLINGER et al., 2002; SIMÃO DA ROSA et al., 2004). All questions had pre-defined answer options, ranging from three to five choices, always going from negative to positive. There were also three "yes or no" questions, used to confirm the pre-defined answers from the questionnaire, and to try to understand more about the handlers' attitudes. The complete questionnaire is presented in Appendix 2. Afterwards, the answers with most variability among handlers were identified and the answers to these questions were linked with the other variables of the study.

2.4.2 *Handler-calf interaction during feeding*

The handler-calf interactions were recorded for three consecutive days during the morning and afternoon feeding times. The observations were always carried out by the same person, who had already been on the farm for at least two days performing behavioural tests with the animals. This previous time before the observations was important for the handler habituation. Vocal and tactile interactions were recorded by direct observation, considering eight categories of handler behaviour. There were three categories for vocal interactions (Talk: talking to the calves with soft voice; Name: calling the calves by their numbers or names with soft voice or giving pet names; and Yell: loud, harsh vocalizations, used to move the animals or stop them when doing something); and five categories for tactile interactions (Stroke: petting the calves with the hand or brush; Soft touch: gentle touch with the hand or resting hand on the animal body or letting the calf suckle the finger; Brusque touch: rough touch with the hand with low use of force; Push: touching the calf with the hands or other parts of the body, using force in a way that results in calf displacement; and Hit: hitting the calves with the hands with high use of force). A copy of the worksheet used to record these data is presented in Appendix 3.

The handlers were classified according to the quality of their actions with the calves under their care, assuming: talk, name, stroke and calm touch, were positive interactions; and yell, brusque touch, push and hit were negative actions. This classification was based on the total number of positive (TPO) and negative (TNE) actions (sum from the three days of observation) and on the average of positive (MPO) and negative (MNE) actions (mean from the three days of observation). In this way four different types of attitudes could be identified, according to their actions towards each of the calves under their care, as follows: Positive: when TPO was greater than MPO and TNE was lower than MNE; Negative: when TNE was greater than TPO; Inconsistent: when TNE was greater than zero and TPO was lower than MPO; or TPO was equal to TNE being both greater than zero; and Neutral: when TPO and TNE were equal to zero; or TPO was greater than zero and lower than MPO being TNE equal to zero. It was rare that a handler had the same attitude towards all the calves in their care so when summed across all calves in their care,

each handler could be described as having a percentage of each type of attitude. This classification was made for each handler to simplify the statistical analysis, so helping to study the associations between the types of actions by the stockperson and the calf's temperament.

For making understanding clearer, in this study, the word "action" will always refer to talk, name, stroke, calm touch, yell, brusque touch, push, hit; and the word "attitudes" to positive, negative, inconsistent and neutral.

2.5 Statistical methods

SAS (version 9.2) and STATISTICA 7 (StatSoft) were used for the statistical analysis. Calves with missing data were excluded from the data file. All variables had their distribution tested, using the Q-Q plots method from SAS.

2.5.1. Exploratory analysis (multivariate analysis)

In general, multivariate analysis refers to all statistical methods that simultaneously analyse variables for each individual or object under investigation. In this study, as most of the variables were connected to each other and interdependent, the multivariate techniques helped in discovering their associations and giving an overview. Three different techniques were used, as described below:

Principal Component Analysis (PCA): The Principal Components Analysis (PCA) is performed with the purpose of simplifying the description of a set of interrelated variables. It is used to reduce the space of variables by creating orthogonal axes that are linear combinations of the original variables, called principal components (STEVENS, 1986). The power of each variable in its respective component is then measured. This allows a reduction of the original variables structure into a new structure, smaller than the original. The principal components are sorted in a descending order according to their variance, i.e., the principal component which contains most information is the first and the component with least information the last. Each principal component is a linear combination of the original variables. QBA

data were analysed through the PCA. It made it possible to know how the calves were distributed according to their scores on each descriptor. Thereafter we could find where each farm was located in the graph, classifying them and relating these results to the answers in the questionnaire.

Factor Analysis: This is an exploratory statistical approach that can be used to analyse inter-relationships contained in groups of variables in terms of indexes or underlying common factors. The aim of this method is to condense the information contained in the original variables into a smaller set, named factors, with a minimum loss of information. So, two or more variables may be highly correlated if they have the same high load factors. It is only applicable to continuous variables (STEVENS, 1986). Thus, all continuous variables (frequency of each interaction - stroking, calm touching, pushing, brusque touching, hitting, shouting, naming and talking; latency to approach and diseases) were tested to show how they might be correlated. The cut-off point was defined for loads from 0.60 and above, as medium and high correlations were desired to better support the results.

Multiple Correspondence Analyses (MCA): The Multiple Correspondence Analysis consists of a combination of multivariate techniques, used for categorical data. It is called correspondence because the lines and columns of a table are transformed into correspondent units. It uses the basic Chi-square concept to standardize the frequencies and create associations between the variables tested (GREENACRE and BLASIUS, 2006). It is a standardized measure that compares a real value with expected values. The total variation of the data is called inertia. The first dimension of the perceptual graph shows the largest quantity of inertia, the second dimension shows the greatest amount of inertia after the first and so on, until the last dimension which is the least important (GREENACRE, 1984). Before the analysis, a Burt matrix is made with the variables and it is this matrix that will be used in the analysis. It consists in comparing two tables given by the software: 1st) observed minus expected frequencies (where only the positive values will be considered); and 2nd) contributions to chi-square (which has the association's values). The highest values from the 'contributions to chi-square' table represent the strongest associations among

variables. By using the MCA, it was possible to associate the calves variables (age, housing, body condition score, avoidance distance score, voluntary approach and breed) with the type of attitudes performed by the handlers (positive, negative, neutral and inconsistent). This analysis can tell us about the associations between these variables, whether they are strong, medium, and weak or are not even associated with each other, by the contributions to Chi-square values. The associations were considered strong if 10 and above; medium if between 10 and 4; and weak if under 4.

2.5.2 Confirmatory data analysis

To confirm the existing differences among farms in the Principal Component Analysis, an ANOVA test was made; and afterwards a Tukey test was used to identify which farms were significantly different from each other.

To confirm the associations found in the Multiple Correspondence Analysis and in the Factor Analysis, the Spearman's Rank-Order Coefficients of correlation were estimated. This is the nonparametric version of the Pearson Correlation Test; and Spearman's Correlation Coefficient measures the strength of association between two ranked variables. It was used to assess the associations between flight distance score (FDS) with latency to approach (LAT); body condition score (BCS) and calf's age. Moreover, to test the association between the two behavioural tests (VA and FDS) and between the type of attitude (positive, negative, neutral and inconsistent) and FDS a Chi-square Test was made. Also a Kruskal-Wallis Test, which is the non-parametric version of ANOVA, was held to know whether the type of attitude was significantly correlated with BCS, LAT and the calf's age.

3. RESULTS AND DISCUSSION

This section is divided into four major points. Firstly an overview of the handler attitudes and calf behavioural tests and how they were distributed among farms are

presented. Secondly, the stockperson's beliefs and intentions are presented. Thirdly, are shown the results of the handlers' attitudes and how these types of attitudes were associated to each other. In the fourth part, the associations between calves' variables and handlers' attitudes are shown, enabling a linked visualization of the data.

3.1 Animals, farms and QBA descriptors

An overview of the attitudes of the handlers and the behaviour of the calves in the flight distance score and voluntary approach tests is presented in Table 2. The handlers were classified according to their attitudes and their proportion of positive and negative actions (see section 2.4.2). Handler 1 was the handler who acted positively most often with the calves, but she also had negative attitudes more often than the other handlers. However, she was predominantly positive in her attitudes (47.0%) towards the calves, presenting negative attitudes to only 12% of her calves and being inconsistent in 41% of the cases. Therefore, just above half of the calves under her care were tame and almost 45% of them approached the test person.

Handlers 2a and 2b, who worked together on farm 2, behaved in a similar way when considering their attitudes towards the calves. Handler 2a was found to have the following distribution of attitudes; 36% positive, 5% negative and 49% neutral, being inconsistent in her attitude to 10% of the calves; and handler 2b had 21% positive, 6% negative and 60% neutral attitudes, with an inconsistent attitude towards 13% of the calves. In this farm about 50% of the calves were tame and 44% of them approached the test person.

Handlers 4, 6a, 6b and 9 were the ones with the least number of calves to take care, only 6 individuals. None of them had a negative attitude toward any of their calves. The calves cared by handlers 4, 6a and 6b were 70% tame and all of them approached the test person, while only 50% of the calves from handler 9 were tame and only 33% approached the test person.

Handler 7 with 111 calves, conversely, was 100% neutral in his attitude to all his calves. The calves under his care were the most reactive ones and only 21% of them approached the test person. Handlers 3 and 5 had to take care of only 27 and 11

calves, respectively, and although they did not have a negative attitude towards any of their calves, they had a neutral attitude to more than 70% of them. Only 25% of the calves handled by handler 3 were tame; and only 27% of the calves handled by handler 5 approached the test person.

Table 2. Summary of the number of calves, percentage of different types of attitude, flight distance score and percentage of the calves that approached the test person, per farm and per handler

Handler	N calves	Attitude (%)			Flight Distance Score (FDS)					Voluntary Approach (VA)		
		Positive	Negative	Neutral	Inconsistent	Average	Mode	Median	Stdev	%React	%Tame	(%)
1	49	46.94	12.2	0.0	40.8	3.76	5	5	1.47	32.7	51.0	44.9
2a	63	36.5	4.8	49.2	9.5	3.64	5	5	1.62	38.1	50.8	44.4
2b	63	20.6	6.4	60.3	12.7	3.64	5	5	1.62	38.1	50.8	44.4
3	27	22.2	0.0	77.8	0.0	2.78	2	2	1.50	59.3	25.9	44.4
4	6	50.0	0.0	33.3	16.7	4.33	5	5	1.21	16.7	66.7	100
5	11	18.2	0.0	72.7	9.1	3.64	5	4	1.43	36.4	45.6	27.3
6a	6	33.3	0.0	66.7	0.0	4.33	5	5	1.21	16.7	66.7	100
6b	6	0.0	0.0	100	0.0	4.33	5	5	1.21	16.7	66.7	100
7	111	0.0	0.0	100	0.0	2.17	2	2	1.26	77.5	8.1	21.6
8	32	25.0	15.6	59.4	6.3	3.78	5	4	1.43	25.0	46.9	37.5
9	6	50.0	0.0	33.3	16.7	3.33	5	3.5	1.86	50.0	50.0	33.3
10	17	17.7	5.9	64.7	11.8	3.65	5	4	1.46	41.2	47.1	41.2

Now regarding the QBA results, the distribution of the 14 different descriptors used in the QBA on the Principal Components 1 (PC1), 2 (PC2), 3 (PC3), 4 (PC4) and 5 (PC5) and the total variance explained by each PC are presented in Table 3. PC1 and PC2 were considered the most important components, since together they explained almost 45% of the data variation.

Table 3. Descriptors loadings in the five first principal components (PC from 1 to 5) of each adjective used in the qualitative behavioural assessment (QBA)

Descriptors	PC1	PC2	PC3	PC4	PC5
Active	-0.463	<u>0.540</u>	-0.272	0.382	-0.219
Relaxed	-0.427	-0.697	0.325	0.044	0.112
Fearful	-0.266	<u>0.513</u>	0.627	-0.333	-0.088
Agitated	-0.257	<u>0.672</u>	-0.077	0.212	-0.332
Calm	-0.327	-0.798	0.168	0.025	0.084
Tense	-0.254	0.411	0.699	-0.325	-0.043
Playful	-0.547	0.328	-0.069	0.408	0.329
Curious	-0.556	0.273	-0.259	-0.149	0.528
Frustrated	<u>0.760</u>	0.267	0.169	0.196	0.310
Apathetic	<u>0.648</u>	0.136	0.276	0.241	0.432
Comfortable	-0.598	-0.506	0.016	0.099	-0.126
Uneasy	-0.137	0.196	0.505	0.439	-0.069
Sociable	-0.569	-0.065	0.242	0.279	0.254
Indifferent	0.420	-0.398	0.287	0.477	-0.305
Eigenvalues	3.187	3.025	1.667	1.202	1.036
Total variance (%)	22.762	21.609	11.909	8.586	7.399

Where bold values have high negative association and underline values have high positive association.

The PC1 presented high positive loadings for the adjectives frustrated (0.760) and apathetic (0.648) i.e. undesirable calf temperament descriptors; as well as high negative loadings for the adjectives playful (-0.547), curious (-0.556), comfortable (-0.598) and sociable (-0.569) i.e. desirable calf temperament descriptors. The PC2 presented high positive loadings for active (0.540), fearful (0.513) and agitated (0.672) i.e. high arousal temperament descriptors; and high negative loadings to relaxed (-0.697) and calm (-0.798) i.e. low arousal temperament descriptors. The adjective uneasy presented a low representation

in both principal components. The descriptors' loading plots on PC1 and PC2 are presented on Figure 1, showing how they are associated or not to each other. Adjectives closer to the graphic centre showed smaller influence in the data variation.

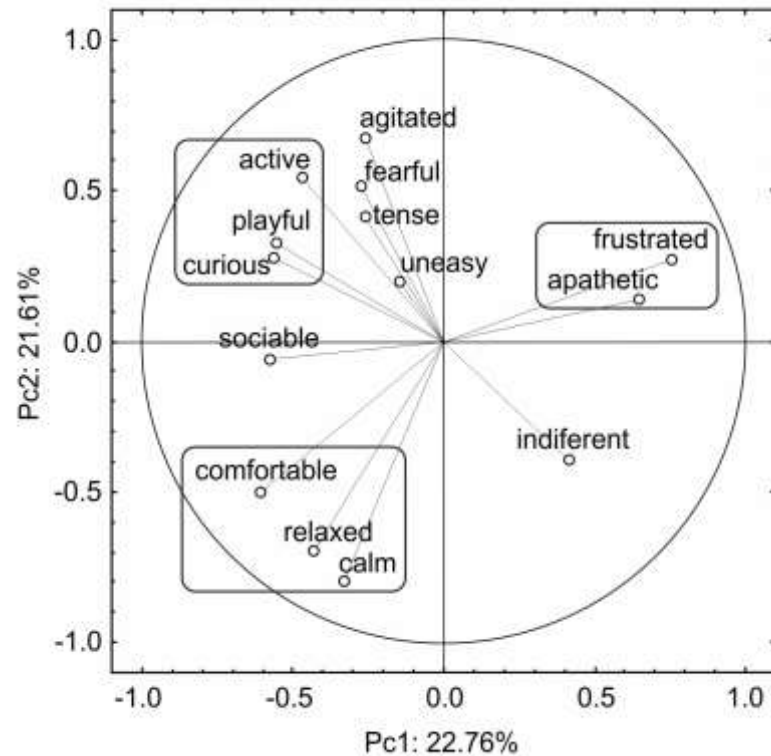


Figure 1. Loading plot for the behavioural based adjectives on the first and second principal components (PC1 and PC2).

The very negative values of PC1 seem to be associated with desirable temperament descriptors for good animal welfare. But these descriptors become less desirable as the PC1 values approach zero, especially if combined with positive PC2 values. Positive values of PC1 are associated with undesirable temperament descriptors from an animal welfare point of view. PC2 seems to be related to temperament descriptors reflecting the level of arousal of the calf. The higher the positive value on PC2 the more aroused the calves whereas they are less aroused when the PC2 value is negative.

The distribution of all calves (represented by circles) in the two principal components (PC1 and PC2) are presented in Figure 2. Based on the visual analysis of Figures 1 and 2, the majority of the calves could be classified as 'comfortable', 'relaxed' and 'calm', and some of them as 'frustrated' and 'apathetic'.

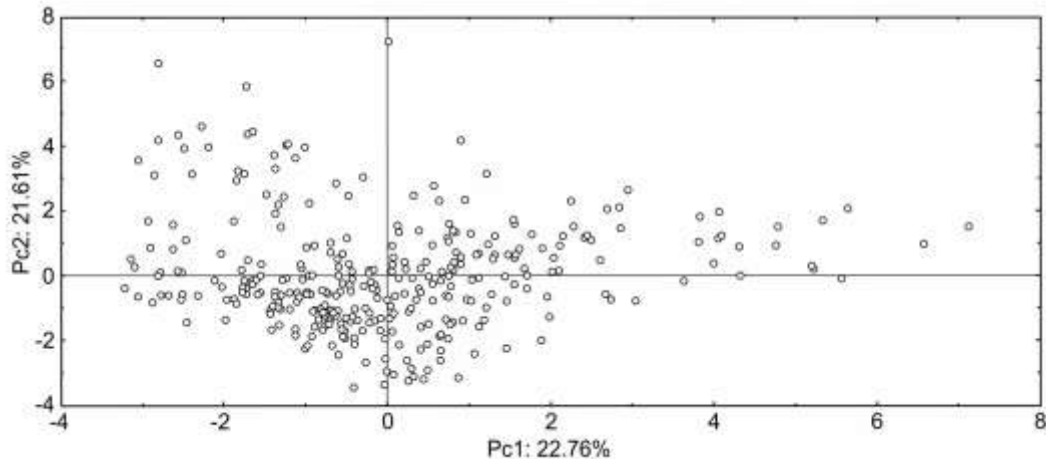


Figure 2. Score plot for individuals on the first and second principal components (PC1 and PC2). Each dot represents one calf.

The means (\pm standard deviation) for each farm on the first and second principal components of PCA are shown in Table 4. There was significant variation on the PC1 ($F = 11.48$, $P < 0.01$), and also on the PC2 ($F = 8.83$, $P < 0.01$) among the farms. On the PC 1, farms 4, 6, 7, 8 and 9 presented the lowest values, indicating that their animals were characterized with more desirable temperament descriptors from an animal welfare point of view. On the other hand, farms 1, 2, 3, 5 and 10 had high values on PC1, indicating more undesirable temperament descriptors for their calves. On the PC2, farms 1, 2 and 7 had the calves with low arousal temperament descriptors, and farms 3, 5, 6 and 10 the ones with higher loadings for the high arousal temperament descriptors, from an animal welfare point of view. It is worth to consider that the farms 1 and 2; 8 and 9; 3, 5 and 10 were not statistically different on both components, showing similarities among each other regarding the calves' temperament.

Table 4. Descriptive statistics for each farm in the QBA for principal components 1 and 2

Farm	N	PC1	PC2	PC1	PC2	PC1	PC2
		Mean \pm SD		Min.	Max.	Min.	Max.
1	49	0.38 \pm 1.79 ^{a,c,f}	-0.30 \pm 1.72 ^{b,c}	-3.04	-3.26	6.60	4.32
2	63	0.97 \pm 1.46 ^a	-0.26 \pm 1.41 ^{b,c}	-1.03	-3.24	7.14	3.11
3	28	0.72 \pm 1.20 ^{a,e}	1.03 \pm 1.52 ^a	-1.73	-1.97	2.95	4.14
4	6	-1.86 \pm 0.71 ^{b,h,i}	0.26 \pm 1.19 ^{a,c}	-2.50	-1.15	-0.94	2.16
5	11	1.43 \pm 1.36 ^a	1.08 \pm 1.05 ^{a,b}	-0.47	-1.45	4.79	2.45
6	6	-1.64 \pm 0.93 ^{b,c,e,g,h,i}	2.56 \pm 1.44 ^a	-2.60	0.78	-0.28	4.41
7	111	-0.27 \pm 1.69 ^{b,e,f}	-0.64 \pm 1.41 ^c	-2.93	-3.47	5.65	3.98
8	32	-1.36 \pm 1.55 ^{d,g,i}	0.47 \pm 2.36 ^{a,b}	-3.04	-2.18	4.33	7.18
9	6	-2.79 \pm 0.56 ^{d,g,h}	-0.02 \pm 0.52 ^{a,c}	-3.19	-0.84	-1.71	0.47
10	17	-0.31 \pm 1.69 ^{a,f,i}	1.88 \pm 1.61 ^a	-2.26	-0.03	4.07	4.55

^{a-i}: means followed by same superscripted letters do not differ according to the Tukey test ($P < 0.05$).

When the means on Table 4 were plotted, it was possible to classify the farms according to their location on the graph and the descriptor which best described that location according to the quadrants on Figure 1. This is illustrated on Figure 3 and Table 5.

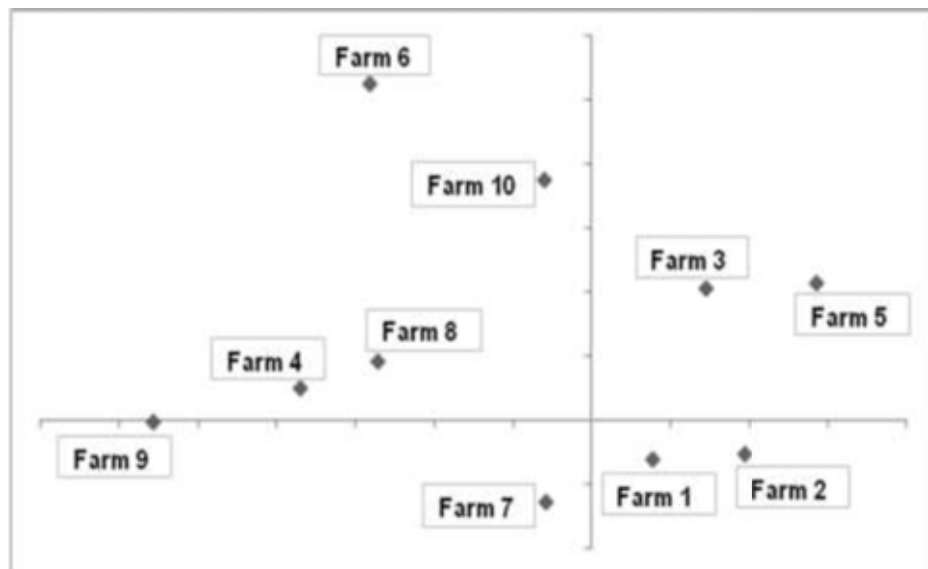


Figure 3. Projection of the 10 farms according to the distribution of their calves in the score plot for individuals.

The means presented on table 4 and the distribution of the QBA terms on Figure 1, suggest that farms 3 and 5 (with positive means on PC1 and PC2) had the most ‘apathetic/frustrated’ calves; farms 6 and 10 had the most ‘fearful/tense’ calves; whereas farms 4, 8 and 9 (negative on PC2) had ‘social/curious/playful’ calves; farm 7 (negative on PC1 and PC2) had ‘calm/relaxed/comfortable’ calves and farms 1 and 2 (positive mean on PC1 and negative on PC2), ‘indifferent’ animals.

Table 5. Calves’ classification for each farm considering its location on the loading plot of the behavioural adjectives assessed through the qualitative behaviour assessment method (QBA)

Farms	Handlers	Calves’ classification
1	1	Indifferent
2	2 and 3	Indifferent
3	4	apathetic, frustrated
4	5	between sociable and curious, playful
5	6	apathetic, frustrated
6	7 and 8	active, between tense, fearful and playful, curious
7	9	calm, relaxed, comfortable - almost indifferent
8	10	between social and curious, playful
9	11	between social and curious, playful
10	12	uneasy, tense and fearful

There is no obvious explanation for the above results according to the characteristics of the farm presented in table 2 (the sex of the handler, housing system and breed) with the exception of the number of calves per handler. The three farms that only have 6 calves (farms 4, 6 and 9) are the three farms with the most negative PC1 values. The three farms with most calves (farms 2, 3 and 7) all have negative values on PC2. This may suggest that the working system (time constraints, number of animals, type of housing, type of handling etc.) that each handler was engaged, seemed to be an important element for the definition of the quality of handling. Moreover, for what we may call good handlers, the number of calves they had to take care was an important influencing factor on how good they could be. “Good handlers” were those who had a lot of positive attitudes towards the calves, represented in this study by handlers 1, 4 and 9 (Table 2). Handler number 1 had to take care of 49 group-housed calves, handlers 4 and 9 only 6 individually-housed

ones. Handlers 4 and 9, because of the small number of calves, did not demonstrate a negative attitude towards any of the calves and only a small percentage of neutral attitudes. Thus, we can say they had time to be good. Handler 1, on the other hand, was never neutral in her attitude, but there was evidence that she was inconsistent and sometimes negative, probably to make the management faster, even though most often she was positive.

In the same way, handlers 2a and 2b, besides having a lot of neutral attitudes, were also good, acting positively with the calves, mainly when teaching them to drink milk from regular buckets. They had to take care of 175 calves, so they also had some negative attitudes for trying to make the management faster. Indeed, the negative attitudes came when the number of animals increased. In this respect, the type of feeding and housing are also important. They had to take care of individually housed animals, but did not use teat buckets (which facilitates the calves' suckling). On the other hand, handler 1 had teat buckets but the calves under her care were kept in groups, making the management slower. Furthermore, handlers 7 and 8 were the most neutral in their attitudes toward their calves. H7 was responsible for 111 group-housed calves and H8, to 32 individually housed ones. They were concerned about the welfare of the animals (being 'really sad' when one of them died, see section 3.2) but because of the high number of animals under their care, it seems they did not have time to perform positive actions that would indicate that this caring about the animals was reflected in the way they actually acted on a daily basis. These results are supported by many authors (Hemsworth and Coleman, 1998; Lensink et al., 2000a; and Seabrook, 2001), who have shown that additionally to the stockpersons' intentions (assessed through the application of questionnaires), other factors, such as working systems (time constraints, number of animals, etc) and motivations (linked to job satisfaction, self-esteem and social recognition) can also modulate the person's behaviour. Indeed, our results corroborate the previous findings from the study of Lensink et al. (2000a), who found, by working with different veal calf units, that the size of the veal unit was a predictor of the amount of positive contact with the calves, describing that in big units, workers had less positive attitudes. According to the authors "a high work load can weaken the link between

positive attitudes and positive behaviour”, as employees on bigger farms have less time to interact with animals individually.

Farms 3 and 5 had calves that were classified as frustrated and apathetic and from table 2 it can be seen that the handlers on these farms (H3 and H5) had among the lowest percentage of positive attitude. The positive attitudes of these handlers were mainly directed to the young calves, usually for teaching them how to drink the milk from regular buckets. After the calves had learnt, the handlers only had neutral attitudes towards them and so, ultimately, their attitudes were among the most neutral handlers (Table 2); even considering that they were in charge (each one) of taking care of six calves. This can show us that careless handlers can negatively affect the welfare of the animals, being an important issue to consider when hiring people to work with animals. According to Hemsworth (2003) “... stockpeople generally have long-standing attitudes and beliefs and corresponding well-established habits in behaviour that have developed over time as a consequence of experiences...”. Thus recommendations on handling alone are unlikely to be effective. According to Boivin et al. (2003) a good way to improve stockmanship other than selecting people carefully, is “...to train them to improve their technical knowledge, working organization and attitudes towards both animals and husbandry practices...”. Another important point is that most handlers nowadays are being overstretched and each day they had to take care of more things in the farm. Most of them had to clean facilities, milk the cows and do a lot of other things rather than just taking care of the calves. Thus, calves can be neglected most of the time, lowering animal welfare levels.

Overall, the most common attitudes observed among the 10 farms assessed were primarily neutral and then inconsistent. The most reactive animals, and also the ones which did not approach the test person, were cared by handlers 3, 5, 7 and 10 who had the highest percentages of neutral attitudes towards their calves (Table 1). Negative attitudes were the least frequent, with highest occurrence being only 16%, for handler 10. This can suggest that some kind of interaction is better than none to reduce the reactivity of the animals and having a less fearful human-animal relationship. This can make the animals stay closer to humans, facilitating their management and improving their welfare. Most calves are often deprived of human

contact. Perhaps calves with the opportunity to interact with humans have there a challenge. Studies by Levine et al. (1956) and Levine (1956) can guide us to understand why this happens. The authors tested how handling during infancy affected rat behaviour in adulthood by comparing the behaviour of four groups of rats: 1) handled in infancy; 2) handled in infancy and receiving an electric shock in every handle; 3) handled in adulthood; and 4) not handled at all. On the contrary of what the author was expecting, the rats handled and handled with electric shock were the best ones in solving the behavioural tests proposed for them. Just like dairy calves with few opportunities to interact with the handler, the rats that were not handled or only handled in adulthood “were susceptible to emotional disturbance because of restricted experience in infancy”. According to Levine (1956) “... when the unhandled rat is subjected to traumatic stimulation in adulthood, greater emotional disturbance occurs, resulting in a disruption of adaptative behaviour...”. Thus, we can say that animals that have to deal with challenges in infancy are better prepared for the future. In addition, findings from Boivin et al. (1994, 1998) also confirm the results above, where the authors demonstrated that an increased animal fear of humans was provoked by a reduced human-animal contact, especially in early ages.

Differently from these findings where neutral attitudes were shown to make animals more fearful, Simão da Rosa (2004) working with lactating dairy cows, found that inconsistent attitudes from the handler were even worse than neutral ones. The cows showed more fear when an inconsistent handler approached, because it was uncertain if his/her behaviour was about to be good or bad. Since his results were found for adult animals, maybe this is why it differed from this study. Also in studies by Hemsworth et al. (1989, 2000); Coleman et al. (1998) and Breuer et al. (2000) it was shown that negative tactile actions such as slaps, pushes and hits were positively associated with cows’ and pigs’ fear of humans, more than neutral interactions. This may be true, but in our study very few negative interactions were performed, preventing us from making this comparison. It is always important to remember that animals’ fear reactions can affect both human and animal safety, besides reducing the worker comfort and time efficiency (BOIVIN et al., 2003), also causing a possible depression on the animal’s productivity and welfare

(HEMSWORTH et al., 2003). This is why we should always try to make animals less fearful of humans as possible.

3.2 Handler beliefs and intentions

The questionnaire was applied for trying to understand the stockperson beliefs and intentions toward the calves. From the original questionnaire (Appendix 2), six questions had the most variation among the handlers; these six questions and their respective answers are presented on Table 6. These questions were related to the handlers overall job satisfaction, the length of time they had worked on the farm, the extent to which they knew the calves' name or number, if they thought it was important to talk and/or brush the calves and how they felt when a calf died.

The results from the handler questionnaire imply that handlers 3 and 5 were least concerned when a calf died, and not interested in brushing or talking to the calves, besides not knowing the number or the name of the calves under their care, confirming the findings above (section 3.1). These two handlers were from farms 3 and 5, which were the farms where most calves showed signs of apathy and frustration. On the other hand, handlers 7 and 8 were most concerned when a calf died, and they also thought it was good to brush the calves and talk to them whenever they could. These handlers came from farms 7 and 8, which had calves that were calm/relaxed and social/curious respectively, also confirming the findings on the last section (section 3.1).

3.3 Quality of stockmanship

The factor analysis was made to reduce the dimensionality of the data, identifying the continuous variables that were associated with each other. Factor 1 explained 26.2% of the data variability; Factor 2 explained 17.8%; and Factor 3, 11.1%. Factor 1 shows that 'stroking' (0.844) and 'calm touching' (0.849) increased together with 'pushing' (0.744) and 'brusque touching' (0.610) as these variables presented the highest positive loadings on the first factor. These four variables seemed to be associated with positive handling, since 'brusque touching' and 'pushing' could be

perceived by the calf as positive actions; so this factor (factor 1) was called 'Good Handling'. On factor 2, 'hitting' (0.708) and 'yelling' (0.655), were the two variables that were above the threshold (>0.6) and had the highest positive loadings; whenever 'hitting' increased, 'yelling' also increased. Since these variables were associated with poor handling, this factor was called 'Poor Handling'. Factor 3 showed that the higher the occurrence of diseases (0.715), the longer was the time for the calf to approach (0.747). A potential name for this factor was less obvious, but we called it 'Calf apathy'. There were some variables that loaded on more than one factor. For example 'yelling' was highly loaded on factor 2, and showed a moderate load on factor 1. 'Diseases' and 'latency to approach' were both highly loaded on factor 3 but also had a medium load to factor 2 (Table 7).

Table 6. Responses presented by each interviewed handler for the six most variable questions asked

Farm Handler	Job satisfaction	Time working in the property	Do you know the			When a calf die
			calves under your care	Talking to the calves	Brushing the calves	
1	unsatisfied	>3years	more or less	it is good, no time	it is good, no time	feel sad
2	unsatisfied	2 to 3 years	well	it is good, I do it	it is good, no time	feel sad
2	satisfied	2 to 3 years	more or less	it is good, no time	it is good, no time	feel really sad
3	unsatisfied	1 to 2 years	no	do not mind	do not mind	do not mind
4	unsatisfied	<1year	more or less	it is good, I do it	it is good, no time	feel sad
5	more or less	2 to 3 years	no	do not mind	do not mind	do not mind
6	more or less	>3years	hardly	it is good, I do it	it is good, no time	feel sad
6	more or less	<1year	more or less	it is good, no time	it is good, no time	feel sad
7	more or less	1 to 2 years	hardly	it is good, I do it	it is good, no time	feel really sad
8	satisfied	>3years	well	it is good, I do it	it is good, no time	feel really sad
9	satisfied	1 to 2 years	hardly	it is good, I do it	it is good, no time	feel sad
10	satisfied	<1year	more or less	it is good, I do it	it is good, no time	feel sad

Table 7. Varimax rotated factor-loading matrix from the factor analysis of continuous variables

Variable	Factor 1	Factor 2	Factor 3
Stroking	0.844	0.083	0.044
Calm touching	0.849	-0.058	-0.057
Pushing	0.744	-0.055	0.082
Brusque touching	0.610	0.092	-0.148
Hitting	-0.095	0.708	-0.010
Yelling	0.406*	0.655	0.066
Diseases	-0.067	0.425*	0.715
Lat (s) to approach	0.072	0.506*	0.747
Naming	0.225	0.549*	-0.016
Talking	0.165	0.296	0.040
Total Variation (%)	26.213	17.752	11.084

Where: bold values correspond to the highest loadings (>0.6), representing which variables may be highly correlated with each other; * indicates medium loadings (>0.4), where probable medium correlations exists among them. Each factor is independent.

Based on the factor analysis, it was possible to make a ranking of the handlers, as shown on Table 8. That is to say, the handler who showed the most stroking, calm touching, pushing and brusque touching was ranked number 1 for 'Good Handling' (factor 1), the next handler in expressing such behaviours was ranked 2, and so on. The same approach was used for hitting and yelling. The handler who showed most of these behaviours was ranked 1 for 'Poor Handling' (factor 2). The handler who's calves had most diseases and/or greatest latency to approach was ranked number 1 for 'Calf Apathy' (factor 3).

Table 8. Ranking of the handlers according to the three independent factor-loadings: good handling, poor handling and calf apathy.

Ranking	Good Handling Str+Ct+Push+Bt	Poor Handling Hit+Yell	Calf Apathy Dis+Lat
1st	Handler 1	Handler 1	Handler 5
2nd	Handler 2b	Handler 2a	Handler 3
3rd	Handler 2a	Handler 2b	Handler 7
4th	Handler 5	Handler 8	Handler 10
5th	Handler 9	Handler 9	Handlers 2a and 2b
6th	Handler 10	-	Handler 9
7th	Handler 8	-	Handler 8
8th	Handler 3	-	Handler 1
9th	Handler 6a	-	Handlers 6a and 6b
10th	Handler 4	-	Handler 4

Good Handling: average of Stroking (Str) + average of Calm touch (Ct) + average of Pushing (Push) + average of Brusque touch (Bt); Poor Handling: average of Hitting (Hit) + average of Yelling (Yell); Calf Apathy: average of diseases (Dis) + average of latency to approach (Lat). Handlers that did not perform any good or bad handling are not shown in the table.

Handlers 1, 2a and 2b who were those showing the highest frequency of positive and the highest frequencies of negative attitudes (Table 2) ranked in the first three places for both good handling and the poor handling in this new analysis. Although handler 5 was the fourth in “good handling” and did not perform any negative interactions, she was the handler who ranked highest for calf apathy. Handler 3, who was ranked second place for calf apathy, also had few positive and no negative interactions. Handler 7 was completely neutral with the calves, showing neither good nor bad handling during daily interactions with the calves, and he was ranked third in the “calf apathy” factor. From table 2 we know that handlers 3, 5 and 7, all had high percentages of neutral attitudes to their calves. Last in the rank for good handling, we find handler 4, with the least number of positive actions, but this handler was best when it came calf apathy since he had the least number of unhealthy calves and calves that had the shortest latency to approach.

Based on the results from the factor analysis and the handlers’ ranking (Table 8), handlers 1, 2a and 2b were the best handlers, but conversely they were also the

poorest ones. According to the factor analysis (Table 7), 'pushing' and 'brusque touching' (initially considered negative actions) were grouped together with the positive interactions 'stroke' and 'calm touching'. One possible interpretation is that calves did not perceive 'pushing' and 'brusque touching' as negative actions; in contrast to 'yelling' and 'hitting' which were grouped together on factor 2, and could be considered as worst actions. Thus, an intense relationship is intense for the two sides, involving mainly positive attitudes, but also bringing negative ones. When a handler is really good and caring for the calves, the animals start to come closer more and more often. This is positive, but can also delay the handler's work, resulting in him/her performing negative interactions e.g. to trying to move the animals away. On the other hand, fearful animals never come closer to the handler so the handler is more likely to develop a neutral attitude towards them. Yet these calves are more difficult to handle and their welfare can be harmed.

3.4 Associations between the stockperson attitudes and the calf behaviour in the presence of an unknown person

By using the Multiple Correspondence Analysis it was possible to combine the calves' variables with the attitudes of the handlers, so decreasing the dimensionality of the data for an easier understanding of how these variables behaved together.

A first trial was made with all variables (attitudes, age, body condition score, housing, behavioural tests and breeds). This analysis showed that positive handling was always given to the younger, tamer and thinner calves, whereas handling was neutral to the older animals. The animals aged less than 28 days were tamer and housed individually, whereas the ones aged 63 days or more were reactive, group housed and fatter. Also, the most reactive animals were highly associated with group housing; and the tamer ones with a quicker voluntary approach, a lower body condition score and individual housing. The behaviour tests showed consistency in the results, as the tamer animals in the flight distance score were also the ones which approached the test person faster in the voluntary approach test. Nevertheless, as many variables had no relation to each other, a second trial was made only with the 'most significant' variables.

In the second analysis only the calf temperament and handler attitudes were tested. The results are presented on Table 9. As age was strongly associated with the body condition score and housing (younger animal were individually housed and thinner, whereas older animal were group housed and fatter) only the body condition score (BCS) was presented in Table 9. A positive attitude was highly associated with the tamer and thinner / younger animals (usually those under 28 days of age); neutral attitudes were shown to be moderately associated with the reactive animals and weakly associated with the fatter/older ones (usually those older than 63 days of age); and the negative and inconsistent attitudes tended to be associated with faster approach and tameness, respectively. The most reactive animals did not approach the test person and were fatter / older. On the other hand, the tamer animals approached the test person within 30 seconds and were usually thinner / younger.

Table 9. Chi-square contributions between variables tested in the Multiple Correspondence Analysis

	positive	neutral	negative	inconsistent	FDS_R	FDS_T	VA_30s
FDS_R	9.07	<u>4.84</u>	0.58	0.38	-	-	-
FDS_T	11.19	7.34	0.44	2.14*	71.2	-	-
VA_n	0.09	0.46	1.81	0.14	<u>6.31</u>	8.02	-
VA_30s	0.22	0.81	1.99*	0.42	10.37	16.53	-
BCS_1	14.23	3.77	2.35	0.12	8.82	13.79	3.23*
BCS_3	4.67	3.72*	0.45	1.51	<u>9.51</u>	6.92	0.29

Where FDS_R= flight distance scores 4 or 5 which indicate reactive calves; FDS_T= flight distance score 1 which indicates tame calves; VA_n= voluntary approach negative; VA_30s= voluntary approach within 30 seconds; BCS_1= body condition score thin; BCS_3= body condition score fat. The only variables presented here, are the ones which had some type of association. **Bold** = strong association; underline = moderate association and * = weak association.

According to the 2D plot of the variables (Figure 4) it is possible to identify two groups of calves may help explain these associations. These are 1) animals who received a neutral attitude from the handler were more reactive, did not approach the test person and were fatter; or vice versa, fatter and reactive animals who did not approach the test person, probably did not approach the handler either and so had received a more neutral attitude from them; and 2) animals who received more

positive attitudes from the handler, were tamer, thinner and approached the test person within 30 seconds; or the other way around, the tamer, thinner and approachable animals received more positive attitudes.

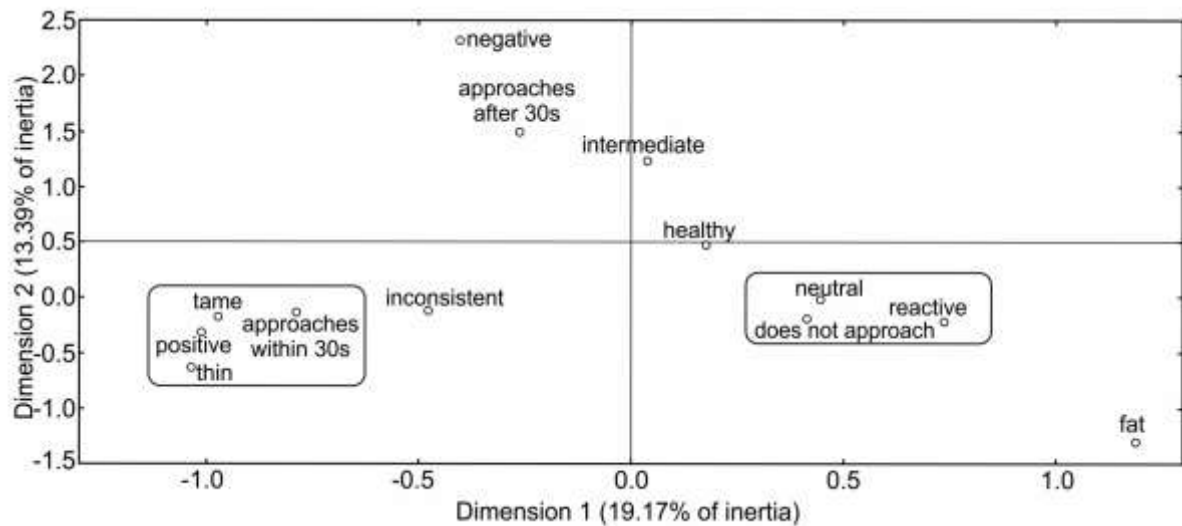


Figure 4. Two dimensions plot of the variables in the multiple correspondence analyses, showing correlated variables inside each one of the two groups.

There are several potential reasons for why on all farms calves under 28 days of age were thinner than the older ones. Firstly, it is known that calves until 28 days of age have higher mortality rates and a greater susceptibility to diseases (DRACKLEY, 2008). Furthermore, the calf is in a transition where it starts by receiving all nutrients from milk, but also needs to start eating some solid food, usually a starter. Most of the farms regulate the milk portion to at least 10% of the calf's body weight. This is proven to be unsatisfying and the animals still present signs of hunger (VIEIRA et al., 2008). Older animals were fatter because they started to rely less on milk and more on concentrate feeding to the nutrients, together with the ending of the critical period for diseases. Most of farms in Brazil adopt the "ad libitum" concentrate diet until weaning; this allows the animals to eat the quantity they want, whenever they want. A possible solution to increase the younger animals' weight would be to increase the number of meals per day, together with increasing the quantity of milk. However, this

would be difficult because farms use a lot of handwork and very few mechanical feeders.

Regarding the confirmatory analysis, the Chi-square Test for the type of handler attitude (either positive, negative, neutral and inconsistent) showed to be significantly correlated with the flight distance score ($\chi^2 = 41.77$, $P < 0.001$). The Kruskal-Wallis Test also for the type of attitude presented significant correlations to the calf's age ($\chi^2 = 92.31$, $P < 0.001$), body condition score ($\chi^2 = 30.23$, $P < 0.001$) and latency to approach ($\chi^2 = 9.45$, $P < 0.05$). Spearman's Rank Correlation coefficients estimated for the latency to approach showed consistency when tested with the FDS, where it was positively correlated ($r_s = 0.352$, $P < 0.001$); i.e. the higher the latency to approach, the higher the FDS. The FDS also showed to be positively correlated with the calf's age ($r_s = 0.498$, $P < 0.001$), confirming the findings in the multiple correspondence analysis, where the older animals were more reactive than the younger ones.

As we could see, the younger calves were the tamer ones, approaching the test person faster. A possible reason for this is that the temperament is still being formed until the animal is mature, so they are in a sensitive period when learning is happening most of the time (MEHTA and GOSLING, 2008). Young calves also highly associate the handler with feeding, making them to approach faster when a human comes closer. This approach behaviour may be why they were also the animals that received many positive interactions and resulted in the handler having a high positive attitude towards them. Another thing that could contribute to the higher number of positive attitudes is because most of the younger calves needed help for learning how to drink the milk and also required more attention concerning health problems. In addition, most handlers let them to suckle their fingers; in this study this was considered a positive action. On the other hand, the older animals did not approach the test person very often, and were the ones with higher neutral attitudes from the handlers. We may say a transition happens during this phase. The new born animals receive a lot of caring, but as they grow older and new calves are born, the handlers does not have time to interact with all of them, or simply does not want to interact with them.

Another point, now concerning the housing, was that animals housed in groups were more reactive and approached the test person less frequently. This can be related to the fact that in most of the farms, young calves were held individually and older ones in groups. These results are the opposite to those found by Boe and Faeverik (2003), where group-housed calves showed less fear of humans than individually isolated ones. Most of the assessed farms adopted the individual housing without isolating the animals. This meant they had more contact with humans compared to group housed ones, besides not having too much contact with other calves. Thus, there is no perfect housing or management. It will always depends on the type and size of the property, number of calves and handlers, type of housing and most importantly what are the owner's objectives.

4. CONCLUSION

We can conclude that the type of attitude can influence the calf temperament and vice-versa. In the case of calf influencing handler, not only the animal temperament will influence the handler attitudes; the working system (time constrain, number of animals, working facilities etc.) can also modulate the handler behaviour towards the calves. Neutral attitudes in the animal infancy make the calves more reactive to people. This is why we should always try to interact with the calves whenever possible. An intense handler-calf relationship is beneficial for the animal when the handler has positive attitudes towards it, and can also bring negative attitudes in lower proportions without compromising the welfare of the animals. On the other hand, poor handlers can make the welfare of the calves worse, as they will not pay attention to the animal, missing the identification of diseases and delaying its treatment, making the calves ill, frustrated and apathetic. Thus, it is important for the farmers to make use of tools for identifying the best handlers for working with their animals.

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APPENDIX 1**QBA**

	Min.	Max.
Active	_____	_____
Relaxed	_____	_____
Fearful	_____	_____
Agitated	_____	_____
Calm	_____	_____
Tense	_____	_____
Playful	_____	_____
Curious	_____	_____
Frustrated	_____	_____
Apathetic	_____	_____
Comfortable	_____	_____
Uneasy	_____	_____
Sociable	_____	_____
Indifferent	_____	_____

APPENDIX 2

QUESTIONNAIRE

Name:

Age:

Farm:

- 1) Until which grade have you studied?
 I have completed high school.
 I have not completed high school.
 I can read, write and do math.
 I know how to write.
 I cannot read, write nor do math.
- 2) Would you change your job?
 I do not change this job for another.
 I only change this job if it is a very good offer.
 What I do, I can continue doing.
 I do the work, but I am looking for another job.
 I do not like what I do.
- 3) Where do you live?
 I live on the farm.
 I live near the farm.
 I live away from the farm.
- 4) How long have you been working in this property?
- 5) How long have you been working with calves here?
- 6) Have you ever worked with calves before? Where and for how long?
 YES NO
- 7) If you had the opportunity, would you swap activities within the farm?
 YES NO
- 8) Knowing the behaviour or the way the calf is helps you during handling?
 YES NO
- 9) Do you know the name or number of the calves under your care?
 know very well the name or the number of them.
 know well the name or the number of them.
 know more or less the name or the number of them.
 hardly know the name or the number of them.
 do not know the name or the number of them.
- 10) Do you worry about the order of the calves when giving milk?

- always in the same order.
- almost always in the same order.
- without worrying about the order.
- almost never in the same order.
- never in the same order.

11) Do you know the amount of milk each calf takes?

- I know very well.
- I know well.
- I know more or less.
- I know little.
- I do not know.

12) Do you know the behaviour or the way of each calf during handling?

- always know.
- almost always know.
- do not pay attention to it.
- almost never know.
- never know.

13) What do you think about yelling at the calves to suckle or while suckling?

- Calves do not mind, nor change.
- I need to yell, otherwise they walk/suckle very slow, and it is good for the calves to become more obedient.
- I need to yell, otherwise they walk/suckle very slow, but it is bad for them, because they get afraid and become more skittish.
- I do not need to yell, I think it is important to manage the calves in silence, or talking softly, so they do not get scared and become more tame.

14) What do you think about hitting the calves when they do something wrong, or do not want to do anything necessary?

- They do not change when are beaten.
- I need to beat, because they learn to do it right, and it is good for the calves to become more obedient and disciplined.
- I need to beat, because they learn to do it right, but I think it is bad for the calves because they get afraid and become more skittish;
- I do not need to hit the calves, I think it is important that management be done without hitting, so they do not feel afraid and become more tame.

15) What do you think about lightweight spanking in the croup, or light scratching on the neck are types of stroking that calves?

- calves do not mind nor change.
- I do not think it is necessary, because they are hardy animals, and I think it is bad because the calves lose their fear of people and become more disobedient.
- I think it is good because they lose their fear of people and become tamer, but I have no time or opportunity of doing this during the day.

It is important and I do it because they lose their fear of people and become gentler and tamer.

16) What do you think about brushing the fur of calves during suckling or at another time?

calves do not mind nor change.

I do not think it is necessary, because they are hardy animals, and I think it is bad because the calves get more skittish.

I think it is good because then they get gentler and easier to handle, but I have no time or opportunity of doing this during the day.

I think it is important and I brush them, because then they get gentler and easier to handle.

17) What do you think about talking to the calves and staying close to them while suckling?

calves do not mind nor change.

I think it is bad because the calves get wary if the handler is close and may become skittish.

I think it is good because they get used to the keeper and gain more confidence, but I have no time or opportunity of doing this during the day.

I think it is important and I do it because they feel more confidence and become tamer.

18) What is the breed of the calves you give milk to?

pure dairy breed (Holstein, Jersey etc.).

criollo.

crossbred or pure meat breed.

pure dairy breed and crossbred.

19) Where do the calves of this property stay until weaning?

in individual pens all the time.

part of the day in individual pens and the other part in paddocks.

until a certain age in individual housing, and then grouped in paddocks.

spend all their time together in paddocks.

20) How do the calves take the milk?

buckets with nipple.

buckets without nipple.

in potties.

other. Which?

21) How do you feel when a calf dies?

I do not mind.

I feel sad, but there was nothing I could do to avoid it, it was supposed to be.

I feel sad and I did everything I could to avoid it.

I feel really sad; they are like children to me.

