Dairy buffalo behaviour and welfare from calving to milking

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Abstract

This review integrates recent scientific findings on the behaviour and welfare of buffalo dams during calving and the subsequent milking phase. These issues are discussed in relation to the level of welfare that buffalo dams and calves experience under different production systems. Key aspects are addressed including welfare issues related to dystocic parturitions and uterine prolapses, the formation of a selective dam–calf bonding, the habituation of inexperienced animals to the milking routine and the appropriate relationship to be developed with stock-people. All these aspects are also discussed in relation to farm profitability, in terms of calf vitality and milk production, and safety of the personnel involved in farm operations.

Keywords: Buffalo dams, Buffalo behaviour, Buffalo welfare, Calving, Dam–calf bonding, Dystocia, Mastitis, Milking behaviour

Review Methodology: The databases were selected because they are the ones with the greatest coverage and relevance in scientific articles in the world and with the aim of identifying the most valuable articles in the subject. Scientific papers have been searched in CAB Abstracts, Scopus and ISI World of Knowledge. The keyword search items were: buffalo cow, maternal behaviour, milking order, *Murrah* buffaloes, *Bubalus bubalis*, water buffalo, dam-calf interaction, calving, parturition, dystocia, calf mortality, environmental stress, temperament, habituation, milking parlour, human-animal relationship, training, heifer, milking order, side preference, buffalo behaviour and buffalo welfare. Of all the items found, the search was exclusively refined to the *Bubalus bubalis*, except for when the article was chosen to complement and support with other ruminant species. The authors also reviewed the sources cited in the articles identified to broaden the search and add relevant materials.

Introduction

Milk production worldwide doubled in recent decades and, significantly, buffalo milk now accounts for approximately 12% of milk produced worldwide. This milk production is concentrated in India and Pakistan (60 and 30% of the world production, respectively) where buffalo milk represents 55 and 75%, respectively of their total domestic milk production, but it also plays an important role in the economy of some Mediterranean countries (e.g. Egypt, Italy) [1].

Due to the increasing competition in the dairy market, interest has grown in incorporating other milk-producing species [2]. In several countries, cattle are being replaced by buffaloes [3] to differentiate dairy production. As a result, the integration of buffaloes into farming units is increasing steadily, mainly because of the high quality of the milk (i.e. high dry matter content), which is often used for transformation in a wide range of local and typical products (e.g. yoghurt, kefir, mozzarella cheese) with particular sensory qualities [4].

Since buffaloes are often replacing cattle in dairy farms, they are treated and handled in the same manner. In addition, as a consequence of the growing economic interest in consuming foods produced by these animals, water buffalo-raising has undergone a process of increasing intensification [5], thus exposing these animals to novel stimuli generated by diverse technologies adapted from dairy cattle [6, 7].

In the current review we will focus our attention on the correlated and critical phases of parturition (including maternal and suckling behaviour) and milking, whose management markedly changed in the last few decades with the adoption of modern techniques such as early mother-young separation, artificial rearing and machine milking, which all involve increased contact with humans. These changes can affect health, behaviour and welfare of buffaloes with potential detrimental effects on mortality rates and growth performances of calves [8] and milk ejection and production of cows [9, 10]. Therefore, we will integrate recent scientific findings on the behaviour of buffalo cows and calves in different production systems during calving and in cases of dystocia, but we will also explore the behaviour of dams and calves in the first hours of life, the effect of separation and the subsequent milking behaviour at the onset of lactation.

Calving

A sign of reproductive and dairy production efficiency is the birth of healthy offspring. After a gestation period of around 310 days (e.g. [11]), riverine buffalo give birth to one calf (or twins on rare occasions). As other gregarious animals, just before parturition the dam separates from the group and gives birth in a protected site at some distance from the herd. This behaviour favours the establishment of the dam–calf bond [6].

The typical postpartum behaviour of dams of ruminant species consists of several common components. Soon after parturition, the dam starts to lick the new-born and also laps up the foetal liquid that spilled onto the ground with the placenta [12]. She consumes all this as part of a cleaning process. Research with ewes demonstrated that they normally find these materials repulsive, but that after birth they are temporarily attractive [13, 14], though this has not been ascertained in the case of buffalo dams. Although it has been suggested that grooming has various functions, few of these are supported today by solid experimental evidence. Generally, in ruminants dams begin to groom their offspring from the head, likely because by removing the foetal membranes they reduce the risk of suffocation. In ruminants grooming immediately after birth is important to dry off the new-born's coat and diminish heat loss, while licking could stimulate the young to seek the dam's teats (e.g. [12]). Grooming and licking also play a fundamental role in the learning process based on the memorization of the odour of the new-born buffalo calf. In ruminants, the dam's behaviour is synchronized with the movements of the new-born animal, as the latter must be able to stand up to locate and reach the teats (e.g. [14, 15]).

In Egyptian primiparous and pluriparous buffalo that are in a standing posture, signs of the onset of parturition include extending the tail, flexing the hip joint and the position of the hind legs. If the dam is lying on the ground, in contrast, she will stretch her neck and extremities [16]. Normal births begin with the dilation of the neck of the uterus and ending with the rupture of the chorio-allantoides in the vagina. At that point, the buffalo calf is visible in the birth canal with the head protruding between the forelegs. Soon after that the calf is expelled, followed by foetal membranes [16]. It is important to note that, as assessed in Egyptian buffaloes, under normal conditions the first phase of the birth process is usually more prolonged in primiparous than pluriparous buffalo [16].

Parturition can be considered an inflammatory process that entails the release of cytokines and prostaglandins. This stimulates the adrenal-pituitary axis which, in turn, increases plasma cortisol concentrations during the immediate pre-partum period. These substances have been utilized as indicators of pain in Egyptian buffaloes during calving [16].

Dystocia and prolapses

Because of the physiological responses it triggers, even the normal (eutocic) birth process is considered a stressful event. However, in the case of abnormal (dystocic) births, additional stressors intensify the normal stress associated with parturition [17]. Although the severity of the stress depends on several factors (e.g. length of parturition, modality of intervention, status of the calf, dimension of the calf, anatomical characteristics of the mother, type of breeding system, etc.), dystocic births in buffalo – though less common than in cattle – can affect both animal welfare and the subsequent milk production, particularly in extensive systems where human intervention is more difficult [18, 19].

One factor that might indicate cases of dystocia is an increase in the duration of the first phase of the calving process, which begins with irregular, intermittent and uncoordinated contractions of the uterine muscles. In Egyptian buffaloes this phase may last as long as 2 h more than the average time required for a normal birth [16]. The release of adrenaline in this first phase in primiparous and pluriparous buffalo is accompanied by significant increases in heart beat and respiratory frequency. In addition, buffalo dams tend to show behavioural changes indicating restlessness, such as kicking the ground, directing their gaze back towards the abdominal region and arching the back [16]. In addition, in the case of dystocic births, Murrah buffaloes show anxiety, muscular activity and pain, which all result in a significantly higher increase of plasma cortisol concentrations on the day of calving, compared to normal births [17]. The average cortisol concentration is generally higher in primiparous than pluriparous buffalo, which seems to indicate that primiparous buffalo respond more acutely to the stress of dystocic births, possibly due to the lower somatic development of these younger animals [16]. In buffaloes, as in other mammal species, two main categories of dystocia were identified: those of maternal origin, and those of foetal origin. Purohit *et al.* [19] found that in Murrah buffalo, maternal dystocia was more common compared to foetal dystocia (59.82 versus 40.17%). Uterine twisting was the most frequent cause of dystocia in buffalo dams (53.57%), whereas less frequent causes consisted of narrow pelvis, too young heifers at their first parturition, pelvic fractures (2.67%), and incomplete cervical dilation (1.78%). However, these values are much lower than in dairy cows [18, 19]. The outcome of a dystocic birth may be the death of the buffalo calf and as a result in Egyptian buffaloes the mortality rate of calves from difficult births is higher during the first 24 h postpartum [16].

In Murrah buffaloes dystocic parturitions may also lead to unexpressed or poorly expressed maternal behaviours, which do not necessarily reflect a lack of maternal ability in terms of caring for neonates. The fact that buffalo dams may not lick or sniff their young, nor nudge them towards the udder, or that buffalo calves may not stand up, approach the teats or attempt to suck during the first 6 h post-partum, might simply be the result of the fatigue that dams experience after suffering through a prolonged dystocic parturition. This deficiency may indicate that the dam may not be in conditions to perform epimeletic behaviour [20].

Dystocic calving may predispose buffaloes to uterine infections. In their study on Murrah buffaloes, Jadon *et al.* [21] identified obligate anaerobes at the moment of eutocic birth, while dams affected by dystocia showed a greater presence of *Arcanobacter pyogenes* combined with other agents. This higher incidence of infections caused by opportunistic microorganisms could be due to either local (e.g. a longer parturition with a consequent open access to pathogens) or systemic factors (e.g. dams suffering acute stress during dystocia, which reduces their immunological response) leaving the uterus susceptible to infections that can compromise the buffalo health status and may later reduce fertility and milk production [21].

Dystocic parturitions activate the hypothalamic-pituitary-adrenal, which triggers an increase in plasma cortisol concentrations in buffaloes. One strategy employed to reduce the concentration of cortisol during dystocic births with release of free radicals and oxygen reactive substances, such as lipid peroxidation, consists of providing a supplement of vitamin E and selenium on the day of the assisted birth and every 24 h during the 2 days subsequent to parturition [17].

In intensive dairy buffalo farming a quite common health problem related to parturition is represented by a high incidence of prolapses [22]. In a sample of 42 Italian dairy Mediterranean buffalo farms, De Rosa *et al.* [22] observed symptoms of recent or remote vulvar or uterine prolapses in 9.3 animal per farm (median value; range: 0–28). According to the thresholds of 2 and 7% set as warning and alarm values, respectively, more than 60% of the farms were above the warning threshold and 40% of the farms were above the alarm threshold [22].

Post-Calving Behaviour

In ruminants, mothers rely on recognition of their own offspring in order to ensure their survival. Thus, they strive to provide only their own new-borns with food and care as they gradually learn key elements of their environment [14, 15]. In buffaloes a long-lasting dam–calf bond develops soon after birth, a relationship promoted by two principle mechanisms: the mother's maternal behaviour and the neonate's capacity for learning [23].

Ruminant dams, including buffalo dams, quickly learn to recognize their own offspring, and will usually reject neonates of other mothers if they approach their udders [14, 15, 24], although some buffaloes, possibly due to the lack of experience, may be tolerant towards alien calves and may allow them to suck [25]. In the ewe, this learning process, defined as imprinting, occurs in a sensitive period under the control of oestrogens and oxytocin, which are abundantly produced at parturition [14, 15, 24]. After a few hours the level of these hormones lowers and mothers become unable to develop an appropriate maternal behaviour towards the new-born. At the same time, the offspring gradually learn about key characteristics of their environment. The young of different mammalian species are born with various degrees of maturity, reflected in such features as their motor or sensory development, or their capacity for thermoregulation. As a result, the dam's behaviour, which needs to be adapted to satisfy the neonate's needs, differs as a function of the degree of maturity or development of the young [13, 14, 26]. The onset of maternal behaviours is propelled by a combination of factors that include neuronal, humoral and sensory elements, it is designed to lead the dam to feed and care for her progeny by performing a range of behavioural patterns whose purpose is to guarantee the viability of the neonate [15, 27, 28].

The calving process constitutes a challenge for the neonate; that is, the sudden need to adapt to extra-uterine life and keep itself alive during the difficulties and hardships of the neonatal period. One ever-present problem in veterinary perinatology is called 'low vitality', a condition that may be affected directly or indirectly by several factors, but that can cause the lack colostrum ingestion and the death of new-born animals. One important determinant of low vitality is foetal hypoxia, an affectation caused by prolonged labour or dystocia in mammals [27, 29]. As assessed in the Egyptian breed, buffalo calves are among the animals that are vulnerable to this condition [30]. Ensuring the vitality and survival of new-born calves entails that buffalo dams optimize the care and feeding they offer their calves. To achieve this, it is indispensable that the animals develop dam–calf bonding, which begins with parturition and continues afterwards. As the buffalo calf passes through the birth canal, it generates cervical-vaginal stimulation that activates the hypothalamus, which releases oxytocin [31]. This hormone acts on the buffalo cow's olfactory bulb which, in turn, triggers the secretion of dopamine. This initiates the period of sensitivity in which the dam identifies her own calf [32]. During this event, observations show that females of the Murrah and Surti breeds of buffalo develop epimeletic behaviour (i.e. care and attention for the neonate). Immediately after birth, the dam stands up [33] and begins to lick and sniff her new-born [20]. Through these actions, she stimulates certain activities in the calf, including its respiratory centre, respiration, circulation, urination and defecation while learning the odour of her offspring [33]. The amount of time devoted to this behaviour is greater in pluriparous than primiparous buffaloes [20], similar to what has been seen in other ruminants, such as ewes [34, 35], possibly due to the lack of experience of younger mothers.

The behavioural patterns of the offspring begin when it raises its head and places itself in a ventral-sternal position, followed by hesitant, sequential efforts to stand up, first with its fore legs, followed by its pelvic limbs. These movements allow it to approach the udder and begin to feed [33]. In a few days, calves start to emit vocalizations with the objective to recall the dam's attention as part of the young's survival strategy [20, 33].

In intensive systems buffalo calves are early separated from their mothers (24–48 h after birth). When separated, both dams and calves display clear behavioural signs of stress, including increased vocalization and locomotion, indicating their motivation to re-join to each other. In addition, due to the frustration of their sucking motivation, these animals, when kept in group, tend to develop cross-sucking behaviour consisting of sucking ears, teats, prepuce and navel of the pen-mates with detrimental effects on the health and behaviour of the calves [36]. In particular, the calves receiving this behaviour showed increased inflammation and infections rates at the navel and teats, whereas those performing it showed increased mortality [37]. Surprisingly though, it has been observed in both cattle and buffaloes [37] that dam-rearing may induce a higher prevalence of allo-suckling in adult lactating animals with a negative impact on milk production [38].

In terms of behavioural changes occurring at the onset of extra uterine life, female neonates tend to be more precocious than males [20]. In addition, observations of Murrah buffaloes in relation to the age of the dams have shown that calves born from primiparous are generally more precocious than those born from pluriparous buffaloes [20].

In extensive systems, buffaloes show a hiding behaviour consisting of keeping their calves hidden for several days postpartum, returning to them at night for feeding [6, 39]. In traditional and extensive conditions buffalo calves are kept with their mothers and are allowed to suckle one or two teats before and/or after milking to facilitate milk ejection [40]. In India they are weaned at up to 6 months and, from a mere animal welfare point of view, this management condition may promote the expression of a fundamental natural behaviour (i.e. suckling) minimizing the stress associated with separation as it occurs close to the natural age of weaning as shown for other farm mammals [41].

In Surti buffaloes, mothers classified as very aggressive and attentive in terms of protection of their calves, seem to be more maternal, make their calves stand up and suck more quickly and devote more time to feeding them, compared to less aggressive dams [33].

Milking

It is well-known that the postpartum period is important for reproduction, milk production and the yield of dairy animals (e.g. [42]). At the onset of milking, the liquid produced by the dam is stored on two levels. The first part is in the dam's cistern, and is called cisternal milk. This milk is easily extracted, simply by applying pressure on the teat. The second portion is found in the alveolar region and lobular ducts, and is called alveolar milk. This milk is more difficult to be extracted and it can only be attained with the release of oxytocin [43]. Buffalo udders have a small cistern and approximately 95% of the milk is stored in the alveolar compartment [44]. Thus, pre-milking stimulation is extremely important for achieving the correct response in terms of milk ejection [32, 45].

In traditional, small-scale production systems, buffalo dams are kept with their calves when they are milked to provide a correct stimulation and achieve prompt and abundant milk flow [32]. In intensive systems, in contrast, stimulation occurs during the sanitizing process with manual washing and massaging of the udder [45]. In both systems – intensive and extensive – the periods and procedures that precede milking must be routinized in order to ensure an adequate milk flow [45].

During milking, buffaloes have been shown to be more sensitive to stressors than cattle [46]. When these animals suffer stress, they secrete adrenaline, which can diminish the supply of oxytocin that is required to keep the milk flowing [9]. Due to the specific anatomy of the buffalo cow's udder and the physiology of milk ejection, even minor changes in the milking routine can make cows uncomfortable and impair milk flow [44]. As a consequence, handlers tend to inject exogenous oxytocin to ensure complete emptying of the udder more often than in other dairy species [47, 48].

Stressors during milking can be either physical, such as those caused by incorrect vacuum level, inappropriate pulsation rate, poorly maintained equipment [9] or psychological, such as those caused by aversive handling and premature calf separation [47].

Mastitis

Mastitis is one of the most prevalent production diseases for dairy ruminants around the world, including buffaloes, and represents a central issue in terms of animal welfare [7]. According to the International Dairy Federation, it may be defined as the inflammation of the udder in response to infectious, traumatic or toxic nature [49]. Mastitis can be divided into contagious and environmental and may also be classified, on the basis of the appearance of clinical signs, into clinical and sub-clinical mastitis. A clinical mastitis refers to an udder infection that can be seen visually (e.g. clots in the milk, hardness and swelling of the udder, etc.), whereas a sub-clinical mastitis pertains to an udder infection that show no external changes. Mastitis leads to a reduction of milk production and in the useful component of bovine milk [50]. As expected, these effects have also been found in buffalo milk [51–53]. As a consequence the economic losses due to mastitis are associated with the drop in quantity and quality of milk production, as well as the costs of antibiotic treatment.

Although mastitis in buffalo is not common as in dairy cattle, it is now becoming an emerging welfare issue. A study conducted on 50 Italian Mediterranean lactating buffalo showed that 9% of the samples were positive for contagious bacteria and 13% were positive for environmental bacteria [54].

The main agents of contagious mastitis are *Staphylococcus aureus*, *Streptococcus agalactiae*, *Coagulase negative staphylococci*, whereas *Streptococcus uberis* and *Escherichia coli* are the main source of environmental mastitis. However, *S. aureus* represents one of the most common aetiological agents in mastitis of bovine, ovine and caprine [55]. In Egypt, on 100 samples of individual buffalo milk the most prevalent contagious bacteria (23%) were represented by *S. aureus* [56].

Antimicrobial agents are usually used in the treatment and control of mastitis. The use of antibiotic may lead to resistance to different diseases by causing bacterial species to become not sensitive to the therapy. An increased resistance to antibiotic of *S. aureus* isolated from dairy cattle and buffaloes with mastitis has been reported [57, 58].

Human–animal relationship

Numerous authors observed in several farm animals that aversive handling is the consequence of a bad quality human–animal relationship [59, 60] and, at least in dairy cattle, it can cause detrimental effects such as augmented cortisol level, heart rates and residual milk, which are all symptoms of reduced animal welfare [61]. Environmental enrichment seems to be a useful strategy that helps animals to cope with stressful conditions, prevent frustration and fulfil their behavioural needs [62]. Although kicking and restlessness in dairy cattle can be due to a number of exogenous factors, including stray voltage [63], recently De Rosa *et al.* [64] observed that the behaviour of dairy buffaloes and the injection of exogenous oxytocin was related to the behaviour of the stock-people directed towards the animals during milking. In particular, these authors noted that a positive stockperson behaviour was negatively correlated with the number of kicks performed by the animals and the number of oxytocin injection. Previous studies reported that kicking was positively correlated with stepping and negatively correlated with positive stockperson interactions, while a positive correlation of stepping and kicking with the prevalence of oxytocin injections was observed [47]. Interestingly, a higher number of negative interactions performed by the stockperson induced a longer avoidance distance from an unknown person [64]. Avoidance distance is considered a reliable indicator of the quality of human–animal relationship [60] both in dairy buffaloes [7] and cattle [59], with higher distances corresponding to worse conditions. Therefore, these results, while indicating that the behaviour of buffaloes in the milking parlour is at least partly dependent on the quality of human–animal relationship, suggest that buffaloes are able to generalize previous negative experience with the stockperson to other unknown people. High numbers of steps and kicks at milking are symptoms of restlessness both in dairy buffaloes [47] and cattle [65], and represent a risk factor for stock-people injuries.

Therefore, at least in cattle, if the routine procedure of herding and handling buffalo cows into the milking parlour implies aggressive or rough interactions with the stock-people, it can cause a loss of milk production and reduce stock-people safety [65], whereas in buffaloes positive interactions such as slow movements and low intensity vocalizations are able to reduce the expression of animal behaviours that reflect anxiety and stress and increase milk production [66]. Hemsworth and Coleman suggest that training programmes should be organized and devoted to the improvement of stockperson attitude and behaviour directed towards farm animals, with beneficial effects on the quality of human–animal relationship and the productivity of farms [60].

Pre-partum habituation to the milking routine

Stress responses may be elicited when young animals, including buffaloes, experience novel stimuli [67]. After calving primiparous cattle and primiparous buffalo are introduced for the first time in the milking parlour and exposed to both a novel environment and a new handling procedure (i.e. the milking routine). This, at least in cattle, can induce stress [68] increase restlessness, the incidence of mastitis [45, 69] and the risk of injury for the stock-people [70]. In dairy cattle, several pre-partum approaches have been suggested to reduce these negative effects, ranging from exposition to positive handling [59, 60], to habituation to the milking parlour [71, 72] and tape-recorded milking parlour noise [68]. As far as we know, only one study has been conducted on primiparous buffalo [73]. In this study animals were exposed to a habituation programme for 10 days before the estimated calving date. The heifers were introduced, once a day, to a 2 × 3 auto-tandem milking parlour and stayed for 10 min in the milking stall, while the udder was washed, wiped and massaged, and teats subject to fore-stripping. Habituated heifers stepped and kicked significantly less than control heifers through the first 20 days of milking, whereas after this period no differences were found between the two groups. Similar results were also observed in cattle, where a period of habituation consisting of udder massaging [71] or introduction into the milking parlour [72] reduced heifer restlessness during milking. In addition, in Polikarpus *et al.*'s study [73] no effect of habituation programme was found on milk quality, milk yield, milk flow and percentage of animals treated with exogenous oxytocin. These results suggest that pre-partum habituation programmes may represent an effective means to reduce restlessness of buffalo heifers during milking [73]. Furthermore, the fact that the number of steps and kicks decreased in the control animals after a longer exposure to the milking routine supports the hypothesis that neophobia is one of the main factors inducing agitation in these animals [73].

Entrance order and side preference

Milking procedures and human interaction may affect animal welfare and small changes in the milking routine of dairy cattle may elicit a stress response even in multiparous cows [61, 74]. One of the factors that must be considered as an example of the importance of having a persistent handling routine is the need to maintain a consistent order of entry of the animals into the milking parlour [75–78]. As also observed in buffaloes [78], evidences from numerous studies indicate that a steady entrance order into the milking parlour is a key feature of the social behaviour of various dairy animals, including cattle [75–77], sheep [79, 80] and goats [81]. It has been reported that milking order can be affected by various factors such as social rank [82], health status [77, 83] and milk yield [81].

In addition, it has been observed that both dairy cattle [75, 84, 85] and buffaloes [78] may show a side preference, as they consistently choose one side of the milking parlour. In cattle, side preference, as for milking order, may be influenced by numerous factors ranging from social behaviour to fear, stress and interaction with humans [84, 86], as well as behavioural lateralization [87]. According to Polikarpus *et al.* [78], buffaloes show stronger consistency than cattle [75] and sheep [79], if the Kendall coefficient of concordance is used as a measure of consistency of the order of entry into the milking parlour.

In Polikarpus *et al.*'s study [78], the percentage of Italian Mediterranean buffaloes showing a preference for a particular side of the milking parlour was higher than that shown by cattle under similar milking conditions [75, 84, 85]. It has been observed that milking the animals in the non-preferred side may increase the stress response in dairy cows [84] and reduce the milk yield in sheep [79]. These findings may suggest, given the sensitivity of dairy buffaloes to stress [23] and the frequent milk withholding observed in the Mediterranean breed [47], handling procedures impairing the expression of the voluntary movements during milking may have a detrimental effect on welfare and, as a consequence, on milk production.

Conclusion

We conclude that much attention should be paid to the peri-partum period as the behaviour expressed by buffalo cows and calves plays a central role in affecting the welfare of the animals and the productivity of the farms in term of calf mortality, udder health and milk production. In particular, preventive measures should be taken to prevent the occurrence of dystocic calving (e.g. genetic selection) and uterine prolapses (e.g. appropriate feeding), while the formation of a selective dam–calf bonding should be promoted soon after birth in the systems where dam-rearing is performed, whereas both in artificially and dam rearing systems the ingestion of colostrum should be ensured. Before parturition inexperienced heifers should be habituated to the milking routine, whereas after the onset of lactation, in order to allow a complete milk ejection, promote personnel safety and sustain buffalo welfare, stock-people should be able to establish a good quality human–animal relationship (e.g. increasing positive interactions with buffaloes) and promote the voluntary choice of side and order of entrance in the milking parlour, while applying appropriate measures to prevent the onset and diffusion of environmental and contagious mastitis, welfare issues both acquiring increasing relevance in buffalo intensive dairy systems.

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