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ORIGINAL RESEARCH

Influence of birth weight, sex and disinfection on the involution of umbilical structures in calves

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Abstract

Background: Umbilical disinfection for calves is often recommended, but scientific studies do not always show a positive effect on disease prevention. In addition, there is little to no metric data on umbilical involution or on factors influencing the size of umbilical structures. Therefore, the aim of this study was to investigate the physiological involution of umbilical structures and the effect of two different umbilical treatment methods.

Methods: After birth, calves' umbilical cord stumps were treated with either an iodine tincture ($n = 43$) or an antibiotic spray ($n = 41$) or were left untreated ($n = 42$). On days 0, 1, 3, 7 and 14 after birth, calves were examined and external umbilical structures were measured. In addition, on day 14 (±2 days), a sonographic examination of umbilical structures was conducted.

Results: The treatment applied had no significant effect on the size of umbilical structures during the first 14 days of life. However, the diameter of the umbilical stalk was larger in male calves and calves with a higher birth weight than in female calves and calves with a lower birth weight.

Limitation: The study was carried out on only one farm, so the generalisability of the findings may be limited.

Conclusion: Umbilical structures showed significant involution over time and were influenced by birth weight and sex.

INTRODUCTION

Umbilical infections are one of the most frequently occurring diseases in neonatal calves worldwide, with incidences up to 29.9%.[1] In addition to local infection and inflammation, bacteria can spread, thereby infecting other organs. In American dairy farms, 1.6% of calves died due to inflammation in the navel or the joints.[2] Umbilical cord infections have a significant impact on the calf's development in their third month due to a decrease in average daily weight gain. Affected calves are known to weigh less than healthy calves.[3]

Umbilical cord care is often recommended as a preventive measure, especially in herds with a high incidence of umbilical infection.[4] To the authors' knowledge, only a few clinical studies have investigated the effect and influence of different disinfection methods on umbilical involution and the incidence of umbilical infection.[5–11] One particular study showed a positive effect of umbilical cord care, using iodine tincture or Navel Guard (SCG-Solutions, McDonough, GA, USA), when compared to a control group.[5] However, another study did not record any positive effects of navel disinfection with a 7% iodine tincture when compared to the control group.[11] A recently published scoping review focusing on umbilical health in intensively farmed cattle summarised papers regarding navel dipping.[12] This review found that few studies have characterised navel dipping in detail, with even fewer studies including a control group for comparison.[12]

In addition to the prevention of umbilical infections by various disinfection methods, the timely observation of the umbilical involution process is crucial for detecting deviations. The external umbilical structures are of great importance, as they allow for the early detection of differences in the involution process or signs of disease. A recently published study has already provided some data on physiological navel regression in male and female calves. In this study, the umbilical structures were investigated with regard to calf- and calving-associated factors (weight, sex and parity of the dam).[13] The mean diameter of the umbilical stalk of 187 male Holstein-Friesian calves was approximately 1.3 ± 0.7 cm between days 7 and 15 postnatum (p.n.).[14] According to Grover and Godden,[5] the mean diameter of the umbilical stalk in the second week (mostly measured on day 11) is 9.8 ± 3.0 mm. The time it takes for the umbilical cord to dry varies between 4 and 7 days p.n.,[13,15–18] and detachment of the umbilical cord is recorded after approximately 14 days.[10,17] However, the length and the girth of the umbilical stump and the involution process have not yet been described.

Different factors that might influence umbilical involution, such as sex or birth weight, have not been fully investigated thus far. However, Watson et al.[19] took the calf's sex into consideration, finding no significant differences between male and female calves in the size of the ultrasonographically visible umbilical stalk, umbilical vein and umbilical arteries. Meanwhile, Konigslow et al.[13] considered the influence of weight and sex on external umbilical structures.

The aim of this study was to evaluate the effect of two different umbilical treatment methods on the involution process compared to a control group and to investigate the physiological involution of the umbilicus in more detail, while placing particular attention on the regression of the outer umbilical structures and the effect of different factors on the size of these structures.

MATERIALS AND METHODS

Within a year, 126 purebred Holstein-Friesian calves were born on a 150 Holstein-Friesian cow farm. All of these calves were included in the study and underwent routine animal control from birth to 14 days p.n. The calving took place in a 4 m × 25 m deep litter area, which could be divided into individual boxes. In most cases, it was possible to house the animals individually at birth. The calving pen was strewn with fresh straw at least every other day.

Three groups, which differed in the method of umbilical cord disinfection, were formed by drawing lots. A total of 30 lots (10 lots for each group) were prepared for that lottery. Then, a lot was taken out of a box for each newborn calf. The taken lots remained outside until the box was empty. This process was repeated completely four times during the trial. A fifth process was started for the last six calves until the trial ended (1 year). There was no blocking by bodyweight and sex or lactation number of the dam.

Group 1: The umbilical cords of 43 calves (21 male/22 female) were dipped in a 10% iodine tincture (10% Vet-Sept-Lösung, aniMedica, Senden-Bösensell, Germany). The dip beaker was filled with iodine tincture up to 75% and the entire umbilical cord was immersed for 15 seconds. After immersion, the rest of the tincture was disposed of and the dip beaker was cleaned with water and set aside to dry.

Group 2: The umbilical cords of 41 calves (20 male/21 female) were sprayed with an antibiotic spray (Engemycin Spray [oxytetracycline hydrochloride; 25 mg/mL], Intervet Productions, Aprilia, Italy).

TABLE 1 Distribution of the calves into the treatment groups depending on the birth weight (kg)

	Animals, n			
	<40 kg	40–45 kg	>45 kg	Total
Group 1	15	13	15	43
Group 2	13	21	7	41
Group 3	12	17	13	42
Total	40	51	35	126

Note: '*n*' denotes total number.

The spray was applied to the umbilical cord for 15 seconds and at a distance of 5 cm, so that the umbilical cord was completely covered by the agent.

Group 3: The umbilical cords of 42 calves (15 male/27 female) were not treated. The group formed the negative control.

The calves were divided into three birth weight groups as follows: less than 40 kg, 40–45 kg and more than 45 kg. The distribution of the calves to the treatment groups depending on the birth weight is shown in Table 1.

The umbilical disinfection was performed in a single application within 2 hours of birth. One person was responsible for the application of the disinfectant and the subsequent examinations. This task was only taken over by trained personnel in a few exceptions. During the 2 hours in the calving pen, each calf was fed at least 3 L of colostrum, usually from its own mother. If the calf did not feed independently, it was drenched using a soft gastric tube (Softdrencher für Kälber, QUIDEE, Homberg Ohm, Germany). The colostrum quality was measured with a digital refractometer (digital hand-held 'pocket' refractometer PAL-S, ATAGO, Tokyo, Japan). If the colostrum's quality was under 18.8%, it was replaced with defrosted colostrum of a high quality (greater than 22.0%) to avoid hypogammaglobulinaemia. In the following 4 days p.n., the calves received 10 L of milk daily, acidified with formic acid and divided into two meals. After that, the milk feeding was subject to two models: a normal model and a restrictive model. The calves were divided into the two feeding regimens according to sex, lactation number of the dam and bodyweight due to another trial that was conducted at the same time. In the restrictive model (total: $n = 32$, group 1: $n = 12$, group 2: $n = 11$, group 3: $n = 9$), the calf received 5.7 L milk replacer per day, and in the normal model, 10 L milk replacer per day (total: $n = 94$, group 1: $n = 31$, group 2: $n = 30$, group 3: $n = 33$). In both groups, the concentration corresponded to 140 g milk replacer powder per 1 L of water. Milk was fed until the 70th day p.n.

The birth weight of the calves was measured using a digital floor scale for calves before the calves were placed in the individual calf pens with a straw bed. Dry straw was added every day. In addition to birth weight, data on the course of parturition as well as colostrum quality of each animal were collected. The course of parturition includes the number of calves born from one cow, the position of the calf during birth, whether obstetric intervention was required and, if it

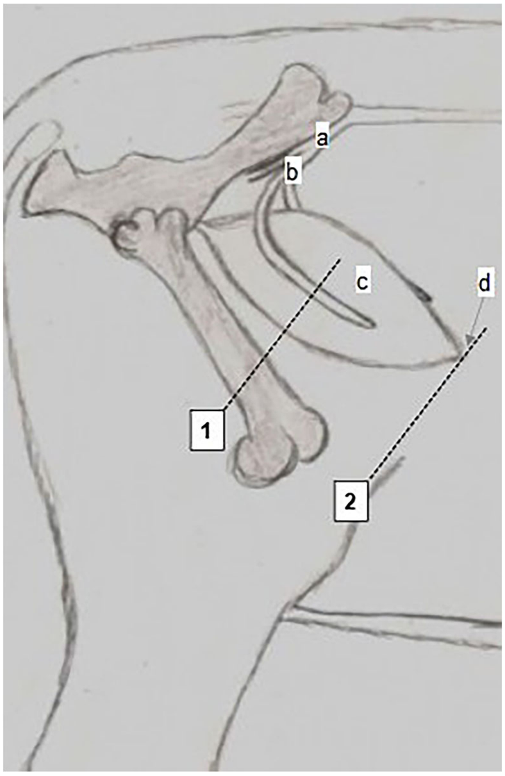

FIGURE 1 Transducer positions for scanning in the caudal abdomen of the calf. 1: umbilical arteries lateral to the urinary bladder, 2: umbilical arteries and urachal remnant on the apex of the urinary bladder. a: arteria iliaca interna, b: arteriae umbilicalis, c: urinary bladder, d: urachal remnant

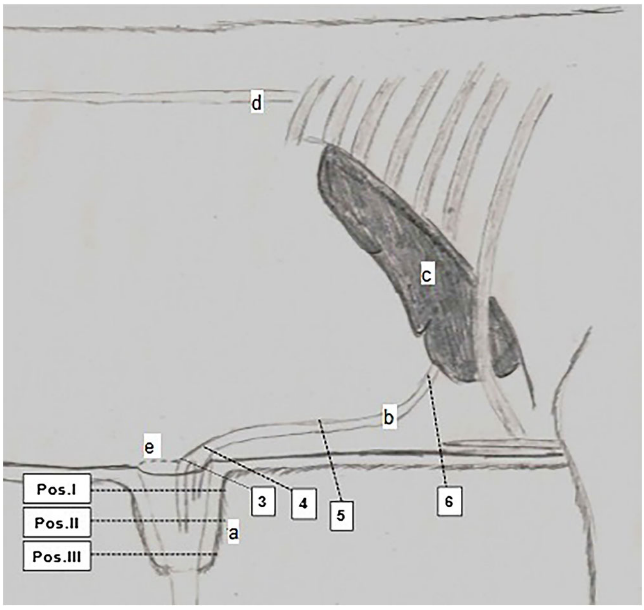

FIGURE 2 Transducer positions for scanning in the cranial abdomen of the calf. I: basis of the umbilical stump, II: midpoint between the basis and apex of the umbilical stump, III: apex of the umbilical stump, 3: umbilical veins at the inner umbilical ring, 4: umbilical vein directly cranial to her merging, 5: middle of the umbilical vein between the umbilical stump and liver, 6: umbilical vein caudal of the entrance into the liver. a: umbilical stump, b: vena umbilicalis, c: liver, d: aorta, e: inner umbilical ring

was required, to what extent (easy extraction, severe extraction or caesarean section).

The calves were examined directly after birth and on days 1, 3, 7 and 14 p.n. The external umbilical structures were measured, and drying of the umbilical cord was noted. The moisture of the umbilical cord was checked by palpation and using cosmetic paper. The umbilical cord was defined as wet if the proximal third of the umbilical cord or at least two-thirds of the umbilical cord were wet and indentable. The measurements were carried out with a 80 mm analogue caliper (CONMETALL, Celle, Germany) and a commercially available 1.5 m tape measure (Wenco, Hennef, Germany). A sonographic examination of the internal umbilical structures was carried out once on day 14 ($\pm$2 days). The examination points of the ultrasonography are shown in Figures 1 and 2. The ultrasound examination was performed on the unsedated, standing calf on the right side of its abdomen. The ultrasound scanner used—MyLabFive (ESAOTE, Genoa, Italy)—was equipped with a 15 MHz linear transducer.

Inflammation of the umbilicus was diagnosed clinically and by ultrasound. Clinically, inflammation of the umbilicus was detected according to the defined signs of inflammation (heat, swelling, pain and secretion). The ultrasound examination relies on the image that is produced, thus allowing for the detection of any pathological deviations. All examinations were performed by one person.

The data were managed, and the figures were produced, with Microsoft Excel 2019. The statistical analysis of the data was carried out using the program IBM SPSS Statistics 20 (Armonk, NY, USA). The results are expressed as arithmetic means and standard deviations. The level of significance was set at a p-value of less than 0.05. Normality of residuals and homogeneity of variance were examined (Shapiro–Wilk test).

In order to analyse the effect of time on the involution process, the parameters that were measured repeatedly (umbilical stump's girth, umbilical stump's length, diameter of umbilical stalk and length of umbilical cord) were analysed by means of a univariate variance analysis with repeated measures and the fixed factors 'treatment' and 'sex' as well as 'treatment' and 'birth weight'.

By analysing the ultrasonographic measuring positions (shown in Figures 1 and 2) that influenced each other (umbilical stump [positions 3–5], umbilical vein [positions 6–8]) using the multivariate variance analysis, the effect of treatment, sex and birth weight on the size of the umbilical structures on day 14 p.n. was tested. In addition, further ultrasound measuring positions (left and right umbilical artery) were analysed by univariate variance analysis to help elucidate the effect of treatment, sex and birthweight on the size of the umbilical structures on day 14 p.n. The intersubject effects (treatment, sex and birth weight) were defined with the sum of type III squares.

The Huynh-Feldt-correction was taken into account in order to identify the effects between time and their interactions. The level of significance was set at $p \leq 0.05$. Chi-square tests were generated to determine differences between treatment groups and birth weight and sex.

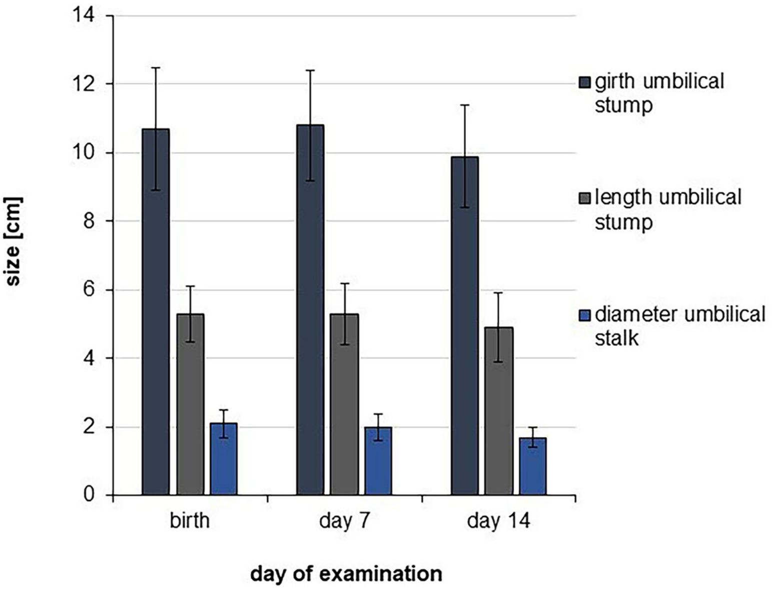

FIGURE 3 Arithmetic mean and standard deviation of the girth and length of the umbilical stump and of the diameter of the umbilical stalk

The possible influence of different factors (calf's position at birth, birth weight and obstetrics) on the umbilical cord's length was investigated by means of correlation analysis. Additionally, correlation analysis was carried out on the length of the umbilical cord in relation to the time of detachment. Parameters such as colostrum quality and birth weight were examined by means of univariate variance in order to detect possible differences between the treatment groups.

RESULTS

The majority of births (81.7%, $n = 103$ out of 126) were unassisted (eutocia) in anterior presentation, while 23 (18.3%) of the births were assisted. Assistance was defined as easy (one person pulling for less than 5 minutes; $n = 17$) or moderate (two people pulling for 5–20 minutes; $n = 6$) extraction in anterior ($n = 18$) or posterior presentation ($n = 5$) of the calf. The mean birth weight was recorded as 42.5 ± 5.8 kg. The male calves were significantly heavier (45.1 ± 6.0 kg) than the females (40.3 ± 4.8 kg) ($p < 0.001$). In group 2, the male calves were lighter than in the other groups, so in total, only seven calves were heavier than 45 kg (Table 1, not significant). There were no differences between the course of birth or bodyweight and the treatment groups.

During the first 2 weeks p.n., none of the calves developed an umbilical infection. Treatment had no significant effect on the size of the umbilical structures during the first 14 days p.n. Figure 3 displays the manually measurable umbilical structures that showed a slight increase in size in the first days after birth and a continuous decrease thereafter.

The data for each parameter on the different days p.n. are shown in Table 2. The girth of the umbilical stump declined by about 0.8 cm by day 14, while the length of the umbilical stump and the diameter of the umbilical stalk both shrunk by about 0.4 cm during this time. The sizes are presented in total, as well as subdivided into groups.

The reduction in umbilical cord lengths is shown in total and divided into the treatment groups in Table S1. The mean length of the umbilical cord was 19.5 ± 9.9 cm immediately after birth, and it shrunk by about 5.0 cm by day 14 p.n. due to drying. It was not influenced by the sex of the calf. The length of the umbilical cord after birth was not correlated with assisted birth (easy extraction, $n = 17$: 17.9 ± 11.8 cm vs. moderate extraction, $n = 6$: 18.0 ± 9.3 cm, $p = 0.922$; $r = 0.009$), birth position (anterior presentation, $n = 121$: 19.2 ± 10.0 cm vs. posterior presentation, $n = 5$: 18.8 ± 15.0 cm, $p = 0.416$; $r = 0.074$) or birth weight ($p = 0.791$; $r = 0.024$). The extra-abdominal umbilical structures shrivelled significantly during the first 2 weeks ($p < 0.001$). The umbilical stump's girth and length did not vary significantly according to sex or birth weight (Tables 2 and 3). However, the male calves' umbilical stalk diameter was significantly larger than that of the female calves. It also varied in calves of different birth weights (Tables 2 and 3), with the umbilical stalk's diameter being significantly larger in calves that were heavier.

On day 1, the umbilical cord of 80.6% (100 out of 124) of the calves was wet, and only in 16.9% (21 out of 124) it was dry. On day 3 p.n., 73.4% ($n = 91$) had dry umbilical cords, increasing to 87.1% ($n = 108$) on day 7 p.n. On day 14, only 12.9% ($n = 16$) of calves still had a wet umbilical cord – 54.8% ($n = 68$) had dry umbilical cords, while in 30.6% ($n = 38$) of the calves the umbilical cord had already fallen off. The time the umbilical cord took to dry out was not significantly different between the treatment groups ($p = 0.711$).

In the ultrasound of the umbilical structures on day 14 (±2 days), the size of the umbilical stalk was measurable in at least 93.7% of the calves (Table S2). On the umbilicus apex (position III), the circumference of the umbilical stalk was 38.1 ± 7.8 mm. A minimal reduction towards the umbilical basis (position I) was visible, and the circumference, close to the ventral

TABLE 2 Measurements of the umbilical stump during the first 14 days postnatum

Parameter	Group	Time: Birth	Day 7	Day 14	p-Values: tm	t	s	tm × t
Girth of umbilical stump	Total	10.7 ± 1.8 (n = 126)	10.8 ± 1.6 (n = 124)[a]	9.9 ± 1.5 (n = 126)	0.953	<0.001	0.790	0.325
	Group 1	10.8 ± 2.0 (n = 43)	11.0 ± 1.3 (n = 42)	9.7 ± 1.4 (n = 43)				
	Group 2	10.9 ± 2.1 (n = 41)	10.7 ± 1.9 (n = 41)	10.0 ± 1.3 (n = 41)				
	Group 3	10.3 ± 1.0 (n = 42)	10.7 ± 1.5 (n = 41)	9.9 ± 1.8 (n = 42)				
Length of umbilical stump	Total	5.3 ± 0.8 (n = 126)	5.3 ± 0.9 (n = 125)[b]	4.9 ± 1.0 (n = 126)	0.181	<0.001	0.756	0.592
	Group 1	5.1 ± 0.8 (n = 43)	5.2 ± 0.8 (n = 42)	4.7 ± 0.7 (n = 43)				
	Group 2	5.4 ± 0.9 (n = 41)	5.3 ± 1.0 (n = 41)	5.1 ± 1.1 (n = 41)				
	Group 3	5.3 ± 0.7 (n = 42)	5.3 ± 0.8 (n = 42)	4.9 ± 1.0 (n = 42)				
Diameter of umbilical stalk	Total	2.1 ± 0.4 (n = 125)[b]	2.0 ± 0.4 (n = 124)[a]	1.7 ± 0.3 (n = 126)	0.321	<0.001	<0.001	0.246
	Group 1	2.1 ± 0.5 (n = 43)	2.1 ± 0.4 (n = 41)	1.7 ± 0.3 (n = 43)				
	Group 2	2.1 ± 0.4 (n = 41)	2.0 ± 0.3 (n = 41)	1.7 ± 0.3 (n = 41)				
	Group 3	2.0 ± 0.4 (n = 41)	2.1 ± 0.5 (n = 42)	1.7 ± 0.4 (n = 42)				

Note: The results are presented as the arithmetic mean ± SD (cm). Group 1: 10% iodine tincture; group 2: Engemycin spray; group 3: negative control. 'n' denotes total number.
Abbreviations: s, sex; t, time; tm, treatment.
[a]Two values are missing.
[b]One value is missing.

TABLE 3 Measurements of the umbilical stump on the first and the 14th day postnatum of calves grouped according to birth weight

Parameter	Day 1: <40 kg	40–45 kg	>45 kg	Day 14: <40 kg	40–45 kg	>45 kg	p-Values: t	bw
Girth of umbilical stump	10.2 ± 1.3 (n = 40)	11.0 ± 2.3 (n = 51)	10.7 ± 1.4 (n = 35)	10.0 ± 2.1 (n = 40)	9.8 ± 1.3 (n = 51)	9.9 ± 1.0 (n = 35)	<0.001	0.528
Length of umbilical stump	5.1 ± 0.8 (n = 40)	5.3 ± 0.9 (n = 51)	5.5 ± 0.8 (n = 35)	4.8 ± 1.0 (n = 40)	4.9 ± 0.9 (n = 51)	5.1 ± 1.0 (n = 35)	<0.001	0.144
Diameter of umbilical stalk	2.1 ± 0.3 (n = 38)[a]	2.2 ± 0.4 (n = 51)	2.4 ± 0.4 (n = 35)	1.6 ± 0.3 (n = 40)	1.7 ± 0.3 (n = 51)	1.8 ± 0.3 (n = 35)	<0.001	0.001

Note: The results are presented as the arithmetic mean ± SD (cm). 'n' denotes total number.
Abbreviations: bw, birth weight; t, time.
[a]Two values are missing.

abdomen, was 36.6 ± 7.5 mm. Tables S9, S10 and S11 describe the size and frequency with which visible vessels occurred in the umbilical stalk on day 14 p.n.

Both umbilical veins were visible on the inner umbilical ring. The larger vein was measurable in 94.4% (n = 119 out of 126) and the smaller vein was measurable in 88.1% (n = 111 out of 126) of the calves (Table S4). The circumference of the larger vein measured 25.8 ± 5.0 mm, whereas the smaller vein only measured 20.8 ± 3.5 mm. Directly cranial to the point of mergence, the umbilical vein's circumference was 27.6 ± 5.1 mm. A reduction in size was visible towards the liver, and the umbilical vein could be shown in 83.3% (n = 105 out of 126) of the calves (Table S5). Both umbilical arteries were nearly equal in size (Table S7). The left artery was visible in 43.4% (n = 54 out of 122) and the right artery was visible in 30.3% (n = 37 out of 122) of the calves. The urachal remnant was not identified in any of the calves.

Overall, treatment had no significant effect on the size of the visible umbilical structures. Only the area of the umbilical stalk at position I was significantly different between the groups (p = 0.046). The umbilical stalk's area on day 14 p.n. was significantly larger in calves in group 3 than in those in groups 1 and 2. The ultrasound results showed that the cross-section of the umbilical stalk and veins was influenced by sex and birth weight (Tables S3 and S6). The sizes of the umbilical stalk and vein(s) were significantly larger in male calves than in females, as well as significantly larger in heavier calves than in lighter ones. Sex and birth weight had no significant effect on the size of the umbilical arteries (Table S8).

DISCUSSION

This study investigated the effects of two different umbilical disinfection methods on the involution process when compared to an untreated control group. In addition, it sheds light on the influence of sex and weight on the size of the umbilical structures.

The mode of administration whereby iodine was administered via a dip while the antibiotic was administered as a spray represents a methodological limitation, with the likelihood being that dipping ensured better coverage than spraying. Furthermore, since calves were allocated to treatment groups by drawing lots, allocation was not truly random, as demonstrated by the weight differences between groups, with group 2 calves being lighter than their counterparts in groups 1 and 3. However, overall, there were no significant differences between the three treatment groups in terms of the umbilical structure's size, detachment or time it took to dry up. As the number of assisted births was low in comparison to unassisted births, the statistical power for identifying differences was poor and should be tested again in a larger number of calves.

Possibly, the optimal calving management and the husbandry of the calves in single, daily straw scattered hutches minimised the risk of pathogens reaching the umbilicus. The most recent study evaluating the effects of navel disinfection also confirms this, with calves with an immediate supply of colostrum having a significantly lower risk of developing an external navel infection.[11] As a result of the optimal farm management in this study, the incidence of navel infection was so low that no effect of umbilical disinfection was evident. No positive effect of navel disinfection on the total incidence of disease in female Holstein-Friesian calves was therefore proven.[20,21] So far, only one study has shown that umbilical disinfection had a positive effect on the prevention of umbilical infections, as the incidence of umbilical infections was reduced in the treatment groups compared to the control group.[5] However, although there were numerical differences between the treatment groups, these differences were not statistically significant.[5] Fordyce et al.[10] found an increased incidence of umbilical infections when using iodine solution and 'liquid nisin' compared to 'dry nisin' or 4% chlorhexidine, but no significant differences were observed in the healing rate and the detachment time of the umbilical cord. In previous studies, the use of the various treatments produced no significant differences with regard to umbilical involution, umbilical cord loss or the presence of umbilical inflammation or hernia.[6–9] In summary, it can be stated that umbilical disinfection for calves is often recommended, but scientific studies do not always show a positive effect on disease prevention.[11,22] Moreover, in studies comparing different disinfection solutions and demonstrating a disease-prevention effect, it was not possible to assign the effect to a particular active substance.[5,10]

In addition to the comparison of treatment, another aim of this study was to gather physiological information about the involution process and the sizes of the umbilical structures. Previous studies, with the exception of Watson et al.[19] and Konigslow et al.,[13] do not provide any information on the effect of birth weight and sex on the size of umbilical structures.[6,8,14,15] This was attributed to the fact that almost exclusively heifer calves were examined. Additionally, in previous studies, only a few structures of the umbilicus were measured, whereas this study evaluated a multitude of parameters. However, Konigslow et al.[13] examined the umbilical structures more frequently than the authors of this study did (14 times compared with five). As already known for the umbilical stalk, all parameters showed a reduction over the time. The large number of parameters measured in our study provided comparative values for estimating the umbilical involution of Holstein-Friesian calves in practice. Compared to the data in the current literature,[8,13,14,23,24] the influence of different examination times as well as the influence of different breeds, in particular, the breed-specific birth weight in relation to the size of the umbilical structures, should be taken into account.

In addition, the data showed a significant effect of sex and birth weight on the size of the umbilical structures, with the umbilical structures of male calves and those with a higher birth weight being larger than those of female calves and calves with a lower birth weight. Since female calves were also significantly lighter than male calves, birth weight or sex seems to be a co-factor. In a regression analysis, the decisive factor could not be found because birth weight had a greater effect on some parameters, whereas sex had a greater effect on others. Further studies with a larger number of animals are needed to clarify which variable is the independent variable.

LIMITATIONS

The study was only conducted on one farm with optimal calf management and calf care. Thus, farm-specific factors may be at play. An effect of umbilical disinfection could perhaps be seen on farms with poorer colostrum supply and poorer birth management.

CONCLUSION

No effect of umbilical treatment on umbilical involution was observed in the first 14 days p.n. The involution of the extra-abdominal umbilical structures showed a significant reduction over the time. The size of the umbilical structures was significantly influenced by the sex and birth weight of the calf.

AUTHOR CONTRIBUTIONS

Study conception and design: Axel Wehrend, Theresa Scheu, Dorothee Lang and Christian Koch. *Data acquisition*: Dorothee Lang, Theresa Scheu and Imke Cohrs. *Data analysis and interpretation*: Theresa Scheu, Imke Cohrs and Dorothee Lang. *Drafting the manuscript*: Dorothee Lang, Theresa Scheu, Imke Cohrs and Axel Wehrend. All authors read, critically revised and approved the final manuscript.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

FUNDING INFORMATION

No funding was obtained for the work reported in this paper.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available in the manuscript and in the tables of Supporting Information.

ETHICS STATEMENT

The calves included in this study were commercial calves kept on a farm in accordance with German regulations and standards for cattle breeding and rearing. The examinations were carried out during the daily routine control.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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