

J. Dairy Sci. 106:9287-9303 https://doi.org/10.3168/jds.2022-23195

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Associations of cow and farm characteristics with cow-level lameness using data from an extensive cross-sectional study across 3 structurally different dairy regions in Germany

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ABSTRACT

The aim of the present study was to evaluate the associations between milk recording data, body condition score (BCS), housing factors, management factors, and lameness in freestall-housed dairy cows in 3 structurally different regions in Germany. These regions substantially vary regarding herd size, breeds, access to pasture, farm management (family run or company owned), and percentage of organic farms. The data used was collected in a large cross-sectional study from 2016 to 2019. A total of 58,144 cows from 651 farms in 3 regions of Germany (North, East, and South) was scored for locomotion and body condition. Additionally, data on milk yield, milk composition, breed, age, as well as information on housing and management were retrieved. One mixed-logistic regression model was fitted per region to evaluate the association of the data with the target variable "lame" and to allow for a comprehensive reflection across different kinds of farming types. In all regions, undercondition (BCS lower than recommended for the lactation stage; North: odds ratio [OR] 2.15, CI 1.96–2.34; East: OR 2.66, CI 2.45–2.88; South: OR 2.45, CI 2.01–2.98) and mid-lactation stage (102–204 d in milk; North: OR 1.15, CI 1.05–1.27; East: OR 1.24, CI 1.17–1.32; South: OR 1.38, CI 1.18–1.62) were associated with higher odds for lameness, whereas overcondition (BCS higher than recommended for the lactation stage; North: OR 0.51, CI 0.44-0.60; East: OR 0.51, CI 0.48–0.54; South: OR 0.65, CI 0.54–0.77) and parity of 1 or 2 was associated with lower odds (parity 1 = North: OR 0.32, CI 0.29-0.35; East: OR0.19, CI 0.18-0.20; South: OR 0.28, CI 0.24-0.33; parity 2 = North: OR 0.51, CI 0.47–0.46; East: OR 0.41, CI 0.39–0.44; South: OR 0.49, CI 0.42–0.57), irrespective of the regional production characteristics. Low energycorrected milk yield was associated with higher odds for lameness in South and North (North: OR 1.16, CI 1.05-1.27; South: OR 1.43, CI 1.22-1.69). Further factors such as pasture access for cows (North: OR 0.64, CI 0.50–0.82; and South: OR 0.65, CI 0.47–0.88), milk protein content (high milk protein content = North: OR 1.34, CI 1.18-1.52; East: OR 1.17, CI 1.08-1.28; low milk protein content = North: OR 0.79, CI 0.71-0.88; East: OR 0.84, CI 0.79–0.90), and breed (lower odds for "other" [other breeds than German Simmental and German Holstein] in East [OR 0.47, CI 0.42–0.53] and lower odds both for German Holstein and "other" in South [German Holstein: OR 0.62, CI 0.43–0.90; other: OR 0.46, CI 0.34 - 0.62) were associated with lameness in 2 regions, respectively. The risk of ketosis (higher odds in North: OR 1.11, CI 1.01–1.22) and somatic cell count (higher odds in East: increased (>39.9 cells × 1,000/mL): OR 1.10; CI 1.03–1.17; high (>198.5 cells \times 1,000/mL): OR 1.08; CI 1.01–1.06) altered the odds for lameness in 1 region, respectively. Cows from organic farms had lower odds for lameness in all 3 regions (North: OR 0.18, CI 0.11–0.32; East: OR 0.39, CI 0.28–0.56; South: OR 0.45, CI 0.29–0.68). As the dairy production systems differed substantially between the different regions, the results of this study can be viewed as representative for a wide variety of loosehoused dairy systems in Europe and North America. The consistent association between low BCS and lameness in all regions aligns with the previous literature. Our study also suggests that risk factors for lameness can differ between geographically regions, potentially due to differences in which dairy production system is predominantly used and that region-specific characteristics should be taken into account in comparable future projects.

Key words: locomotion, dairy cattle, modeling, logistic regression, risk factor analysis

Received December 23, 2022.

Accepted June 16, 2023.

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INTRODUCTION

The optimization of breeding programs, feeding strategies, housing conditions, and management procedures in the dairy sector over the past decades has yielded a type of dairy cow that is capable of producing large amounts of milk with an increased output of milk components such as milk fat and milk protein (Rauw et al., 1998; Wangler et al., 2009; Sundrum, 2015; Britt et al., 2018). On the downside of these productive advancements, modern dairy cows are confronted with a greater susceptibility to production diseases such as metabolic disorders, mastitis, poor fertility, and lameness (Shanks et al., 1978; Rauw et al., 1998; Leroy et al., 2008; Sundrum, 2015). Afflicted animals often undergo premature culling, which has resulted in a large gap between the potential lifespan of cows (20-25 yr) and the current reality in dairy production. In Germany, cows reach a mean lifespan of around 55 (VIT, 2020) to 68 mo (Logue and Mayne, 2014). In an international evaluation, the lifespan of dairy cows averaged 52 mo in Brazil, 75 mo in the Netherlands, 47 mo in Canada, and 59 mo in the United States (Logue and Mayne, 2014; De Vries, 2017).

Lameness is one of the main reasons for involuntary culling in dairy cows (Whitaker et al., 2000; Manske, 2002; Booth et al., 2004; Heise et al., 2016). It has been associated with tremendous financial losses due to decreased milk production, poor reproductive performance, and costs for treatment and prevention strategies (Huxley, 2013; Charfeddine and Pérez-Cabal, 2017; Dolecheck and Bewley, 2018). It is also an animal welfare concern compromising the animals' ability to express its natural behavior (Whay and Shearer, 2017). Afflicted cows display marked behavioral aberrations, such as reduced feeding duration, feeding pace, feeding frequency, rumination behavior, lying duration, and social activity, and are subjected to severe, often chronic pain (Whay et al., 2005; Alawneh et al., 2012; Grimm et al., 2019).

Most cases of lameness originate either from noninfectious claw horn disruption lesions (i.e., sole ulcers, white line disease and sole hemorrhages) or from infectious claw disorders such as digital dermatitis and foot rot. The development of clinical lameness is complex, and has been linked to a vast abundance of factors related to housing conditions and management practices (Sundrum, 2015; Oehm et al., 2019). Additionally, animal-level factors such as milk yield and body condition play an important role (Green et al., 2010; Green et al., 2014; Sundrum, 2015; Randall et al., 2018) and, moreover, interact. The association between daily milk yield and lameness cases or lameness treatments has been subject of several studies and an association of lameness and milk yield has been demonstrated (Green et al., 2002, 2010, 2014). Both before and after treatment of lame cows, the average daily milk yield was found to be reduced both in comparison with cows not being diagnosed lame and with their estimated milk yield based on their previous test-day milk yields, respectively (Warnick et al., 2001; Green et al., 2002). Green et al. (2002) found higher yielding cows to be more likely to become lame than their lower-yielding herd mates. Those lame cows produced a mean increased milk yield of 1.12 kg/d during lactation on the days where lameness did not cause reduced milk production. This has implications for the health of high-yielding dairy cows. They are at greater risk of ketosis (Gröhn et al., 1999) and other health disorders (Hansen et al., 1979), and we can now confirm that they are at greater risk of lameness.

In the light of this background, the aim of the present study was to explore the association of animal-level factors, farm characteristics, and management aspects with cow-level lameness across 3 structurally different regions in Germany using multiple mixed-logistic regression. By fitting one model each for the 3 different regions, we could evaluate for which predictors the association with lameness was stable between regions, and for which predictors an association was not consistently found.

MATERIALS AND METHODS

Farm Selection

Data used in the current study were retrieved in the context of a large cross-sectional study on animal health and housing conditions on German dairy operations from 2016 to 2020 (PraeRi, 2020). Whereas previous work from our group was interested in applying a novel modeling approach to a subset of the current data (Oehm et al., 2022), the present work was able to include the entirety of the available data set and shift the focus toward region-specific differences among risk factors.

In the study providing the data, 3 study regions were defined (Figure 1) as North, covering the federal states of Schleswig-Holstein and Lower Saxony; East, covering the federal states of Mecklenburg Western Pomerania, Brandenburg, Thuringia and Saxony-Anhalt; and South, representing the federal state of Bavaria.

Sampling is presented in detail in PraeRi (2020), also described by Oehm et al. (2022). Sample size (number of farms) was to be calculated to reflect different potential scenarios (i.e., different prevalences of e.g., lameness) given a power of 80% and a significance level of 5%. For the estimation of an expected value (e.g., a

Rittweg et al.: CROSS-REGIONAL RISK FACTORS FOR LAMENESS



Figure 1. Study regions included North (Schleswig-Holstein [DE-SH] and Lower Saxony in yellow), East (Mecklenburg Western-Pomerania [DE-MV], Brandenburg, Thuringia, and Saxony-Anhalt [DE-ST] in blue) and South (Bavaria in green) as defined by PraeRi (2020).

prevalence of 30%), hence a standard deviation of 7 and a precision of 1, 2, 3, and 4 was assumed (Glaser and Kreienbrock, 2011). Based on these scenarios and taking into consideration feasibility (e.g., how many cows can be scored per day, how many farms can be visited during the 3-yr period of the study given a single 1-d visit), the sample size for each study region was determined to be 250 farms. Selection of farms based on administrative district and on herd size (number of cows) within the respective federal state and study region. Using an automated randomization algorithm, farms were randomly sampled based on the national animal information database (Herkunftssicherungs- und Informationssysem für Tiere, HIT; https://www.hi-tier.de/) and on the farm data from the Milchpruefring Bayern e.V. in South, as well as from the state control associations (Landeskuratorium der Erzeugerringe für tierische Veredelung) in North and East. A list of farms within the target population was available and an automated

approach incorporating the information required for sampling (e.g., herd size by regionally differing cutoffs) selected farms to be contacted. A response rate of 30%to 40% was expected. Within each study region, 1,250farms (i.e., 5 times more farms than required) were drawn from the population (all operations that housed dairy cows based on the information provided within HIT) to cover a response rate of at least 20%. Considering farms registered in HIT, region-specific cutoff values for herd size (number of cows) were calculated, to be able to include farms based on herd size and to make sure the broad variety of different herd sizes was represented in the study. A sampling plan was conceived to achieve an evenly distributed selection of included farms regarding herd size. Therefore, farms were classified into small (North: 1–64 cows; East: 1–160 cows; South: 1–29 cows), medium (North: 65–113 cows; East: 161–373 cows; South: 30–52 cows), and large (North: >114 cows; East: >374 cows; South : >53 cows) farms. Cutoffs were chosen, and these cutoffs were the values that divided the targeted population into 3 groups of the same size given the information within HIT. Because response rate was lower than initially expected (<10%), a second sample of farms was drawn and contacted using the sample measure of randomization as outlined above. A complete overview of the total number of contacted and visited farms is provided in Table 1. Letters were sent to the randomly selected farms with information about the study and an invitation to participate. Participation was on a voluntary basis and motivated farmers had to autonomously contact the study team and arrange the date and time of the farm visit. Farms were visited once between December 2016 and August 2019.

The anonymity of the participating farms was guaranteed in alignment with the German and European data protection legislation. For an observational study such as the present, ethics approval was not necessary in Germany at the time the study was initiated.

Data Collection

All farm visits followed a standard operating procedure (PraeRi, 2020). Paper-based questionnaires and data entry forms were used for on-farm data collection. Farm-level information (farming type, i.e., organic vs. conventional farming, full-time vs. part-time farming, presence of pasture access for dairy cows) was retrieved during a standardized interview with the farm manager at the farm visit. Scoring of cows was conducted by trained observers. On large farms, not all cows were scored. Instead, a sample size of cows was chosen for each region that would detect an expected prevalence of 40% with a confidence interval (**CI**) of 95% and a

Rittweg et al.: CROSS-REGIONAL RISK FACTORS FOR LAMENESS

Table 1. Response rate of dair	y farms in the collection of the stud	v data (invited and visited farms)) in each region of Germany

	Sma	ll farms, ¹ n	Medi	um farms, ² n	Larg	ge farms, ³ n	Total n	umber of farms
Study region	Invited	Visited (response rate %)						
North	1,664	83 (5)	674	90 (13)	449	80 (18)	2,787	253 (9)
Schleswig-Holstein	330	70	210	59	110	55 `	650	184
Lower Saxony	1,334	13	464	31	339	25	2,137	69
East	701	83(12)	433	87 (20)	605	83(5)	1,739	252 (9)
Mecklenburg Western Pomerania	189	18	123	26	264	22	576	66
Brandenburg	109	24	103	22	173	19	385	65
Thuringia	247	20	76	13	86	16	409	49
Saxony-Anhalt	156	20	131	26	82	26	369	72
South	1,345	92(7)	2,015	84(4)	1,058	84 (8)	4,418	260(6)
Bavaria	1,345	92	2,015	84	1,058	84	4,418	260

¹Cutoffs for small farms per region: North: 1–64 cows; East: 1–160 cows; South: 1–29 cows.

²Cutoffs for medium farms per region: North: 65–113 cows; East: 161–373 cows; South: 30–52 cows.

³Cutoffs for large farms per region: North: \geq 114 cows; East: \geq 374 cows; South: \geq 53 cows.

standard error of $\pm 5\%$ (power: 80%) as described by Jensen et al. (2022). Therefore, on farms in regions South and North if more than 130 or 213 cows were present, respectively, a sample of 130 and 213 animals was scored. In region East, where the largest farms were located, 2 groups were formed and on farms with 166 to 292 cows, a sample of 166 cows was scored and on farms with more than 293 cows, a sample of 292 cows was scored. Cows were systematically selected across all groups and if cows were held in more than one pen, a similar percentage of cows was selected in each pen. Observers aimed to evenly distribute the selected cows in the herd by scoring and marking (e.g., 4 cows and marking the fifth without scoring it to reach 80% scored animals). Animals were chosen irrespective of whether they were resting, feeding, standing in the alleys or up to any other activity.

Locomotion score (**LS**) and BCS were individually assessed for each sampled cow. The 5-point locomotion scoring procedure suggested by Sprecher et al. (1997) was implemented which is based on characteristics of gait and posture both during locomotion as well as when the animal is standing. A cow was regarded as lame with an LS of ≥ 3 .

The assessment of BCS was conducted using the scoring system by Edmonson et al. (1989) modified by Metzner et al. (1993), which is represented by a 5-point scale with 0.25-increment intervals. Other animal-level information (parity, breed) as well as all milk test-day sampling records from the 12 mo before the farm visit (DIM, milk yield [in kg], milk fat [in %], milk protein [in %], SCC [in number of cells \times 1,000 per mL] and milk urea [in mg/L]) were retrieved from HIT and from the German DHIA after written consent from farm managers. The data from the questionnaires and data

entry forms were manually transferred to a central SQL database (https://ibei.tiho-hannover.de/praeri/pages/69) immediately after each farm visit and supplemented with the data from HIT and DHIA.

Observers (total: n = 22, North: n = 6, East: n =11, South: n = 7) were trained using photos and videos and were subjected to the assessment of interobserver reliability during the time of the study. Before the onset of data collection, all researchers participated in a 3-d workshop including video sessions and group discussions for training and preparation purposes. This preparation of the raters was complemented by continuous telephone conferences between observers and a pilot phase of 3 mo before the actual start of the study which aimed at getting acquainted with the procedures of data collection and farm visits. Throughout the study period, all researchers involved in data collection participated in 3 seminars throughout the study period. The first seminar took place 1 yr after the onset of data collection, the second seminar was held in the middle of the second study year, and the third seminar in the middle of the third study year. All observers participated in the same 3 seminars, which were designed as both group discussions, video sessions, and practical courses on the first day and assessment of interobserver reliability by independently scoring the same cows for BCS and LS without sharing information during the assessments on the second day. This resulted in the assessment of 43, 59, and 60 animals being subjected to body condition scoring and 36, 53, and 54 cows being scored for locomotion at the 3 assessment dates, respectively. The intraclass correlation coefficient, which gives an indication about the variance between measurements of the animals compared with the total variance between all measurements and all animals was calculated for

BCS. Agreement was defined as perfect agreement at the 0.25 increment (i.e., perfect agreement was present when 2 observers assigned the same score. For the comparison of every single observer with the scorings of the other observers, we fitted a variance model including 2 factors. A random effect was modeled for the individual animal so that dependencies between the measurements of different observers could be acknowledged. A fixed effect was modeled for observer. Because all observers scored the same animals, the selection of the observers is fixed and the model claims are to be applicable for this selection. In case an observer significantly deviated in their scoring from the other observers, they received detailed information about the kind of deviation. Values are taken between 0 (poor agreement) and 1 (excellent agreement) with higher values suggesting a better agreement among observers. Agreement between raters for the variable BCS was fair (0.59 during the first seminar) to excellent (during the second and third seminar [i.e., 0.79 and 0.76, respectively]; Barnhart et al., 2007; Hallgren, 2012).

For lameness assessments, global kappa values were estimated and exclusion tests were performed as described by Ruddat et al. (2014). Global agreement among all m observers was calculated for locomotion assessments to indicate inter-rater reliability (Krummenauer, 2005). Due to the ordinal nature of the locomotion scale, kappa values were yielded in a weighted manner using the quadratic weight function by (Fleiss and Cohen, 1973). One observer was continuously compared with a random collective of the other observers and each observer was hence tested repeatedly in a varying collective. To determine disagreeing observers, an exclusion test was performed for each single observer 1, $\dots m$ (Ruddat et al., 2014). Therefore, the expected and observed agreement between a specific observer 1 and the remaining raters (m-1) was assessed. Using this to calculate a weighted kappa coefficient specific for the observer, observer-specific kappa values significantly smaller than global kappa estimates indicated disagreement of the respective observer (Ruddat et al., 2014). In this very context, the minimum and maximum values of the kappa values were yielded including their 95% CI. Cutoffs for kappa values are rather arbitrary (Landis and Koch, 1977). Values of 1.0 indicate perfect agreement, whereas 0 indicates completely random or poor agreement (Landis and Koch, 1977; Viera and Garrett, 2005; Hallgren, 2012). Agreement in this study was moderate across the 3 assessment dates. The agreement was moderate (0.48 [CI 0.33–0.63] to 0.57 [CI (0.43-0.71]) on the first evaluation occasion, moderate on the second occasion (0.56 [CI 0.46–0.66] to 0.63 [CI (0.53-0.74]), and moderate on the third occasion (0.39)[CI 0.25–0.53] to 0.44 [CI 0.30–0.60]).

Journal of Dairy Science Vol. 106 No. 12, 2023

Data Handling and Preparation

Microsoft Excel data sheets (Microsoft Corporation, 2018) were extracted from the SQL database. Plausibility of the data was checked in 2 steps: First, the database included a function to check variable values based on predetermined cutoffs. These cutoffs had been determined based on available literature on the topic and knowledge gained during the farm visits. All researchers involved in the study were part of determination of cutoffs by means of intense discussions. Subsequently, a database internal validation was defined for the variables and values that were not in accordance with the determined cutoffs were presented by the database. Implausible values indicated by the database were checked and corrected, if necessary and possible considering the available data sources such as paper-based data entry forms and questionnaires. If it was not possible to correct implausible values, they were changed to missing. After data export, 5 of the coauthors checked the data for plausibility by assessing the distribution of the variables. If implausibilities occurred, the respective observations were checked within the database to detect potential irregularities during data export as well as in the original paper-based forms to detect introduction of implausibilities during transfer of recorded data into the database.

In a next step, the final data set was created. Out of the 86,355 animals on 765 farms in the original data set, 28,211 animals were excluded either due to missing records (any missing record lead to exclusion from the study), not being of interest in this context (e.g. males, animals in tiestall housing) or not meeting the requirements (e. g. not having enough test-day-milking data sets). Regarding this requirement, all animals had to have at least 3 test-day milkings in the 4 mo before scoring, with an exception of cows in the first 3 mo of lactation. For those cows, the following rule applied (to exclude the milk assessments from the prior lactation, but still include early lactation cows): cows within the first month of lactation were included if they had 1 or 2 assessments in the last 31 d; cows in the second and third month of lactation were included if they had 1 to 3 (2-3 for cows in third month of lactation) assessments within the last 62 or 93 d, respectively. All steps are displayed in Figure 2. As a result, the final data set for analysis contained a total number of 58,144 cows from 651 farms with 17,248 cows in North, 34,413 in East, and 6,483 in South.

Similar to Oehm et al. (2020, 2022), the raw BCS values were categorized into 3 groups (undercondition, recommended, and overcondition) stratified by breed and DIM, because body condition naturally changes during lactation (Souissi and Bouraoui, 2020). The ap-

Rittweg et al.: CROSS-REGIONAL RISK FACTORS FOR LAMENESS

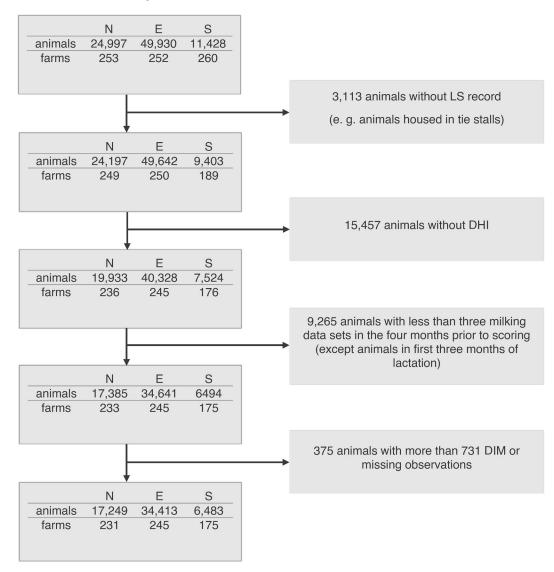


Figure 2. Flowchart displaying the steps of creating the final data sets from the original raw data on German dairy cows. Out of 86,355 animals on 765 farms in the original data set, 3,113 had not been scored for locomotion (e.g., tiestall-housed cattle). Another 15,457 animals had no milk recordings (e.g., males), 9,265 cows had fewer than 3 milking data sets in the last 4 mo before scoring (except for animals in the first 3 mo of lactation), and 376 cows were more than 2 yr in milk (>731 DIM) or were excluded due to missing records. The final data set contained 58,144 cows on 651 farms. (N = North; E = East; S = South; LS = locomotion score; DHI = German Dairy Herd Improvement Association).

plied limits were set as suggested by Kritzinger and Schoder (2009a,b) and Kritzinger et al. (2009) and are displayed in Table 2.

The composition of breeds was categorized for each region into German Holstein (**GH**), Simmental (**SIM**), and others. Herd size was categorized cross-regionally by quantiles (<25%, 25–75\%, <75%) based on the number of lactating and dry cows present on farm at the day of the farm visit into small (<105), medium (105-431) and large (>431). A variable "stage of lactation" was created by transforming DIM values into early lactation (DIM <102), mid lactation (DIM 102-204), and

late lactation (DIM >204) resembling the thirds of a 305-d lactation span.

The processing of the milking data was conducted in several steps. To best assess the milk components of individual cows when evaluating risk of ketosis, we implemented recent recommendations by Glatz-Hoppe et al. (2019a) in our analyses. Accordingly, the metabolic status of dairy cows can be estimated way more accurately when the milking data are evaluated with individual upper and lower limits for milk fat and milk protein per cow, depending on the milk yield (in kg), which is inversely correlated with milk protein and

9292

				1	BCS	
			GH ai	nd BS	SIM and of	ther breeds
Stage of lactation	Phase	DIM	LL	UL	LL	UL
Late lactation or dry	Ι	-60 to -1	3.25	3.75	3.75	4.25
First third of lactation	II1	0 to 29	2.75	3.75	3.30	4.25
	II2	30 to 99	2.50	3.40	3.25	4.00
Middle third of lactation	III	100 to 199	2.50	3.25	3.25	3.75
Last third of lactation	IV	200 to 299	2.75	3.75	3.25	4.25

Table 2. Upper and lower limits of optimal BCS according to different breeds of German dairy cows and the present stage of $actation^1$

¹GH = German Holstein, BS = Brown Swiss, SIM = German Simmental, LL = lower limit, UL = upper limit. Modified after Kritzinger and Schoder (2009a,b), Kritzinger et al. (2009), and PraeRi (2020).

milk fat content (Glatz-Hoppe et al., 2019b). In a first step, we calculated these dynamic limits for both milk protein and milk fat depending on the total milk yield (supplemental materials, Rittweg et al., 2023; https:// /b2drop.eudat.eu/s/fRjGckCjERjLfFF) for each cow and each test milking individually, instead of using the commonly used imposing standard, nondynamic limits by Spohr and Wiesner (1991). As a result, we obtained an individual "optimal" fat and protein content range per cow. In a second step, the fat-protein ratio (**FPR**) was calculated and categorized into "normal" (FPR \leq 1.4) and "in risk of ketotic metabolic status" (**ROK**; FPR >1.4). According to Glatz-Hoppe et al. (2019b), a ketotic metabolic status can quite accurately be identified by an FPR >1.4 together with a milk protein content lower than the calculated individual lower milk protein limit or a milk fat content higher than the calculated individual higher milk fat limit. As a third step, for each cow, the 3 most recent milking data sets before the farm visit were scanned for ROK (yes or no) and the total number of ROK events (possible range 0-3) on the basis of these conditions. In a fourth step, the ECM performance was calculated with the following formula (Sjaunja et al., 1990):

$$\text{ECM} = \frac{\text{milk yield} \times \begin{bmatrix} 0.38 \times (\text{milk fat in \%}) + \\ 0.21 \times (\text{milk protein in \%}) + 1.05 \end{bmatrix}}{3.28}$$

Information of the relevant raw milking data (milk fat content, milk protein content, milk urea, and SCC) as well as ECM from the 3 most recent test-day samplings before the day of scoring was condensed to a single value using a Bayesian nonparametric bootstrap procedure with 1,000 samples with replacement. This yielded Bayesian medians for each of the variables to reflect the individual animal in a single value. The ECM bootstrap medians were categorized by breed and quantiles (<25%, 25–75%, <75%) into low (GH: <25.9 kg; SIM: <22.2 kg; other: <23.0 kg), average (between low and high) and high (GH: >36.2 kg; SIM: >31.6 kg; other: >34.6 kg) performing cows, likewise the SCC was categorized into low (<39.9 cells \times 1,000/mL), increased (between low and high), and high (>198.5 cells \times 1,000/mL). The raw values for milk fat and milk protein were categorized into low, optimum, and high using the individually calculated upper and lower limits, while for milk urea, limits were set to the established limits of 150 mg/L (lower limit) and 250 mg/L (upper limit) for categorization into low, optimum, and high.

Statistical Analyses

All statistical analyses were performed with the R software for statistical computing version 1.2.5042 (R Core Team, 2020) using the R studio interface (RStudio Team, 2020). A complete list of implemented packages can be found in the supplemental materials (Rittweg et al., 2023).

Univariable binary logistic regression was performed on animal level for each variable with regard to the binary target variable lame (1/0). No variable selection based on *P*-values was conducted during the univariable analyses and all predictors analyzed at the univariate level were entered into the multivariate process. This procedure was chosen to detect potential confounding effects of variables as the effect of single predictors may be covered up by other factors (Dohoo et al., 1997). Moreover, univariable analyses were performed to identify relevant associations between covariates and lameness that could translate into relevant associations or confounding effects during multivariate modeling. Subsequently, one multivariate logistic regression model was built on animal level for each region individually, including a farm-specific random effect to account for potential clustering at the herd level as well as for between-farm heterogeneity. A manual stepwise backward selection process was chosen and hence one variable at a time was removed from the model. After each excluded variable, candidate models were compared and ranked based on the Akaike's information criterion (**AIC**; Akaike, 1969) and the Bayesian information criterion (**BIC**; Schwarz, 1978). A decrease of the AIC or the BIC when comparing 2 candidate models indicated a better fit in the model with the lower AIC or BIC values (Ding et al., 2018). If the removal of one covariate induced a change in the estimate of another predictor by 20% or higher, the variable was regarded as a confounder and remained within the model.

Correlation among predictors was checked by assessing the correlation matrix of the models. If a correlation > |0.7| was encountered, the biologically more relevant factor was selected based on biological reasoning and the extant literature. A network structure was drawn via the free software DAGITTY (Textor et al., 2016) to visualize presumed relationships among variables and identify potential confounders (see supplemental materials, Rittweg et al., 2023). This network was used to guide the modeling procedure (i.e., after the backward selection process had identified an optimal candidate model) potential confounders were re-entered into the model one by one based on the network structure (observer, pasture access, visit year, herd size, farming type [conventional vs. organic], DIM, breed, and parity) and model fit was again evaluated based on the aforementioned criteria. As a last step during the model building process, biologically plausible 2-way interactions between the remaining covariates were tested. The constructed network structure guided this process by providing an overview of all variables and aiding at identifying potential interactions between predictors. To acknowledge potential observer effects during locomotion scoring, the variable observer was forced into the model. Throughout the analyses, statistical significance was set at a *P*-value ≤ 0.05 and the CI was set to 95%.

The R function vif() from the car package (Fox and Weisberg, 2019) was used to assess variable inflation (i.e., potential [multi-]collinearity among variables). As none of the variable inflation scores were >1.4 (N and S: max. 1.4, E: max. 1.3), we inferred that (multi-) collinearity was not a considerable issue (Hair et al., 2018; Kim, 2019).

One receiver operating curve (**ROC**) was generated per model and the area under the curve (**AUC**) was calculated, to assess the model performance after having selected a good fit model for each region. The model selection process allowed the models to differ between regions. The ROC curves were generated using the R package ROCR (Sing et al., 2005), and the AUC was calculated with the pROC package (Robin et al., 2011). The AUC value for region North was 0.75 (95% CI 0.74-0.76), while the corresponding values for East and South were 0.76 (95% CI 0.76-0.77) and 0.76 (95% CI 0.75-0.77), respectively.

RESULTS

Descriptive Results and Characterization of the 3 Regions

All descriptive results for each region can be found in the supplemental materials (Rittweg et al., 2023). The total number of farms and animals in each region included in this study is displayed in Figure 2, the number of invited farms including the response rates is displayed in Table 1.

Animal-level prevalence of lameness in the present study was 27.5% (North), 42.6% (East), and 27.5% (South).

In North and East the main breed was GH, with 94.1% (North) and 92.7% (East) of the cows; in region South, SIM was the main breed (78.9% of the cows). The percentage of first lactation cows was 31.1% (North), 33.8% (East), and 28.9% (South). The percentage of cows in second lactation was 25.1% (North), 26.0% (East), and 25.6% (South), and in third or higher lactation, 43.8% (North), 40.1% (East), and 45.5% (South).

Median ECM was 29.9 kg (North), 31.0 kg (East), and 27.0 kg (South). One ROK event during the last 3 test milkings was detected in 15.6% (North), 16.2% (East), 18.8% (South) of cows. Two ROK events were present in 3.9% (North), 3.4% (East), and 4.4% (South) of animals, with 1.8% (North), 1.0% (East), and 1.8% (South) of cows showing evidence of a ROK event in all assessments. The percentage of cows having had at least 1 ROK event within the last 3 mo was 21.3% (North), 20.6% (East), and 25.0% (South).

The mean herd size was 146.0 (North; interquartile range: 95), 458.9 (East; interquartile range: 327) and 67.4 (South; interquartile range: 35). Although parttime farming concerned only 0.4% of cows in North and 0.1% of cows in East, it was more present in South with 3.6% of animals being managed part time. Pasture management also clearly differed between regions: 71.1% (North), 45.6% (East), and 25.3% (South) of the cows had access to pasture. The percentage of cows housed under organic farming conditions was 3.9% (North), 4.0% (East), and 11.2% (South). More than half of these animals were housed on farms with less than 100 animals (50.1%), and organic farming was not present on farms with herd sizes of more than 300 animals at all. The percentage of cows housed on organic farms were 3.4%, 9.0%, and 15.9% for GH, SIM, and other breeds, respectively.

In addition, univariable analysis were performed. Results can be found in the supplemental materials (Rittweg et al., 2023).

Multiple Mixed-Logistic Regression Model

Results of the final multiple mixed-logistic regression are displayed in Table 3. Parity, BCS, and farming type were associated with lameness in all 3 regions. The BCS appeared to be an important covariate in all study regions: undercondition was associated with higher odds for lameness, whereas overcondition was associated with lower odds for lameness in comparison with a recommended BCS. A parity of either 1 or 2 was associated with lower odds of lameness compared with older cows (parity \geq 3), whereas organic farming was associated with lower odds of lameness compared with conventional farming.

The models differed in the following covariates: ECM was associated with lameness only in North and South (i.e., low ECM was associated with higher odds for lameness and high ECM with lower odds for lameness compared with average ECM). Low milk protein contents were associated with higher odds, and high milk protein content with lower odds, of lameness in North and East, but this predictor was not retained in the model for South. Early lactation stage showed an association with lower odds for lameness in East but with higher odds in South, whereas the mid-lactation stage was associated with higher odds in all regions compared with late-lactation stage. Access to pasture was associated with lower odds for lameness cases in North and South, but not in Easta. "Other" breeds appeared to be associated with lower odds for lameness in East and South compared with the main breed of the respective region (GH in East and SIM in South), while this predictor was not retained in the North model. Presence of at least one ROK event was associated with higher odds of lameness in North but not in the 2 other regions. Similarly, high SCC was associated with higher risk of lameness only in region East.

Parity, DIM, farming type (organic vs. conventional), and observer appeared as confounders in all 3 regions. In East and South also breed, in region North and South pasture access, and in region North und East visit year appeared as additional confounders.

DISCUSSION

The aim of the present study was to explore animallevel factors associated with lameness in dairy cows housed in freestall barns, and to comparatively assess these factors in 3 structurally different regions of Germany. Even though previous studies focusing on factors associated with lameness have been published in abundance (Green et al., 2002; Solano et al., 2015; Foditsch et al., 2016; Bran et al., 2019), information on factors related to milk production variables and ketosis on animal level is limited (Oehm et al., 2019). We aimed at contributing to this field of knowledge using the dynamic milking variables assessment limits suggested by Glatz-Hoppe et al. (2019a), giving an individual range of optimal milk protein and milk fat content for each cow taking into account it's breed and total milk yield, to ensure a modern, advanced approach of assessing the metabolic status of dairy cows.

The unique aspect of comparing dairies of 3 structurally diverse regions in one study allowed a deeper understanding of associations that are consistent between different types of farming and those associations that might be related to certain farming practices. In addition, as the majority of studies has presented knowledge with regard to GH cows, the composition of breeds in one of the regions (majority of German SIM cows with only few GH and other breeds, such as Brown Swiss in South) is a clear novelty of the present study.

Limitations

To correctly interpret the results of the present analyses, it is paramount to be aware of the cross-sectional design of the study, which entails certain limitations that need to be acknowledged (Setia, 2016). The target variable lameness and most predictors were recorded simultaneously during the same occasion. Therefore, no conclusions can be drawn to infer causalities, which is a general drawback of cross-sectional studies (Carlson and Morrison, 2009).

Observer effects are a common issue in locomotion assessments of dairy cows (Oehm et al., 2022). We are convinced to have minimized the observer bias by an extremely close-meshed evaluation of observers as well as by following a distinct, standardized protocol for data collection, as well as incorporating repeated training of observers. The interobserver reliability of observers was assessed several times during the study period and kappa values varied substantially, which was expected. To address this issue to a certain extent, we have included observer in the model and hence separately tested the relevance of this covariate for each region: The model performance improved clearly while interestingly estimates of the other covariates were only slightly affected. Hence, in the presence of observer, the results of the remaining covariates were adjusted for the observer effect. This appears as a clear asset of this study and corroborates our results while simultane-

			Nort $(n \text{ cows} =$	North ws = 17,248)	(8)			(n cow	$\begin{array}{c} \text{East} \\ \text{(n cows} = 34,413) \end{array}$	3)			(n co ¹	South $(n \ cows = 6,483)$	3)	
Predictor	Category	OR	CI (95%)	P-value	<i>P</i> -value Estimate	\mathbf{SE}	OR	CI (95%)	P-value	<i>P</i> -value Estimate	${ m SE}$	OR	CI (95%)	<i>P</i> -value	<i>P</i> -value Estimate	SE
BCS Parity	Undercondition Recommended Overcondition 1	2.15 Ref. 0.51 0.32 0.51	$\begin{array}{c} 1.96-2.34 \\$	<0.001 <0.001 <0.001 <0.001 	$\begin{array}{c} 0.76 \\ - \\ - 0.67 \\ - 1.15 \\ - 0.67 \end{array}$	$\begin{array}{c} 0.05 \\ - \\ 0.08 \\ 0.05 \\ 0.05 \end{array}$	$2.66 \\ \\ 0.51 \\ 0.19 \\ 0.41 \\ 0.$	$\begin{array}{c} 2.45{-}2.88\\\\ 0.48{-}0.54\\ 0.18{-}0.20\\ 0.39{-}0.44\end{array}$	<pre><0.001</pre> <pre><0.001</pre> <pre><0.001</pre> <pre><0.001</pre>	$\begin{array}{c} 0.98 \\ - \\ - 0.68 \\ - 1.68 \\ - 0.89 \end{array}$	$\begin{array}{c} 0.04 \\ \\ 0.03 \\ 0.03 \end{array}$	$2.45 \\ \\ 0.65 \\ 0.28 \\ 0.49 \\ 0.49 \\ 0.49 \\ 0.49 \\ 0.49 \\ 0.49 \\ 0.41 \\ 0.$	2.01-2.98 	<0.001 	0.90 -0.44 -1.28 -0.71	$\begin{array}{c} 0.10 \\ - \\ 0.09 \\ 0.08 \\ 0.08 \end{array}$
Stage of lactation	≥ 3 <102 DIM 102-204 DIM	Ref. 0.99 1.15	0.89-1.10 0.89-1.10 1.05-1.27	0.005	-1.15 -0.67	$0.05 \\ 0.05 \\ 0.05$	$\begin{array}{c}\\ 0.92\\ 1.24\end{array}$	0.86-0.98 1.17-1.32	0.007 0.007 0.001	$-0.09 \\ 0.22$	$0.03 \\ 0.03 \\ 0.03$	$\frac{1.28}{1.38}$	1.08-1.50 1.18-1.62	0.004 0.004 0.001	0.24 0.32	
ROK	<204 DIM No Ves	Ref. Ref. 1 11	$\frac{-}{-}$ 1 01 -1 99	0.095	0 11	0 02	1 06	1 00–1 13	0 068	0 06	0 03	1 13	 	0 088	0 13	
Farming type	Conventional Organic	Ref. 0.18	0.11-0.32		-1.69	0.28	0.39	0.28-0.56		-1.00	0.17	0.45	0.29-0.68		-0.81	
Pasture access ECM ²	No Yes Low	Ket. 0.64 1.16	${0.50-0.82} 1.05-1.27$	$< 0.001 \\ 0.002$	$-0.44 \\ 0.15$	$0.12 \\ 0.05$						$\begin{array}{c}\\ 0.65\\ 1.43\end{array}$	${0.47-0.88}$ 1.22-1.69	0.006 < 0.001	-0.43 0.36	
Milk protein content ³		Ref. 0.88 1.34 Ref.	$\begin{array}{c}$	0.014 <0.001	-0.13 0.29	$\begin{array}{c} 0.05 \\ 0.06 \\ 0.06 \end{array}$	1.17	1.08-1.28	<0.001	0.16	0.04	0.80	0.68-0.94	0.008	-0.22	
SCC^4	Low Low Increased	0.79	0.11-0.00	100.0>	62.0-	en.n	0.64 Ref.	$\frac{0.79-0.90}{-1.03-1.17}$	0.003	0.09	0.03 0.03					
Breed	High GH SIM Other						1.08 Ref. 0.83	1.01-1.16 0.43-1.63 0.42-0.53	0.031 $ 0.596$ ~ 0.001	$\begin{array}{c} 0.08 \\ -0.18 \\ -0.75 \end{array}$	$0.04 \\ \\ 0.34 \\ 0.06$	$\begin{array}{c} 0.62 \\ \mathrm{Ref} \end{array}$	0.43 - 0.90	0.012	-0.47	
Visit year	2017 2018 2019	Ref. 1.25 1.94	${0.98-1.59}_{1.49-2.54}$	-0.077 < 0.001	$0.22 \\ 0.67$	$\begin{array}{c} - \\ 0.12 \\ 0.14 \end{array}$	$0.88 \\ 0.61$	$\begin{array}{c}$	v	-0.12 -0.51	$0.09 \\ 0.13 \\ 0.13$		H 000			
Observer	− c2 co 4	Ref. 0.86 0.82 0.94	$\begin{array}{c}$	$\begin{array}{c}\\ 0.347\\ 0.218\\ 0.742\end{array}$	-0.15 -0.20 -0.06	$\begin{array}{c} - \\ 0.16 \\ 0.17 \\ 0.17 \end{array}$	$^{}_{$	$\begin{array}{c}\\ 0.83{-}1.20\\ 0.81-1.36\\ 0.70-0.96\end{array}$	${0.988} 0.732 0.016$	$\begin{array}{c} -0.001 \\ 0.04 \\ -0.19 \end{array}$	$\begin{array}{c} - \\ 0.09 \\ 0.13 \\ 0.08 \end{array}$	$0.68 \\ 0.48 \\ 1.24$	${0.45-1.03} 0.45-1.03 0.31-0.74 0.79-1.94$	$\begin{array}{c} \\ 0.068 \\ 0.001 \\ 0.354 \end{array}$	$-0.39 \\ -0.73 \\ 0.21$	
	0 -1 C a	$0.88 \\ 1.01$	0.63 - 1.22 0.72 - 1.41	0.447 0.957	-0.13 0.01	$0.17 \\ 0.17$	1.98 0.98 1.00	$\frac{1.60-2.45}{0.82-1.17}$ 0.83-1.22	$< 0.001 \\ 0.811 \\ 0.962 \\ 0.$	-0.02 0.005	$0.11 \\ 0.09 \\ 0.10 \\ $	$\begin{array}{c} 0.59 \\ 1.08 \\ 1.10 \end{array}$	$\begin{array}{c} 0.39{-}0.90\\ 0.71{-}1.66\\ 0.69{-}1.73\end{array}$		-0.52 0.08 0.09	
	8 10 11						$ \begin{array}{c} 1.04 \\ 0.78 \\ 1.48 \\ 0.80 \\ \end{array} $	$\begin{array}{c} 0.59 - 1.21 \\ 0.57 - 1.06 \\ 0.86 - 2.55 \\ 0.55 - 1.18 \end{array}$	$\begin{array}{c} 0.003\\ 0.114\\ 0.157\\ 0.266\end{array}$	-0.25 -0.25 -0.22	$\begin{array}{c} 0.08\\ 0.16\\ 0.28\\ 0.20\end{array}$					

Journal of Dairy Science Vol. 106 No. 12, 2023

Rittweg et al.: CROSS-REGIONAL RISK FACTORS FOR LAMENESS

9296

 3 Categorial limits for milk protein content: individual upper and lower limits as displayed in supplemental materials (Rittweg et al., 2023; https://b2drop.eudat.eu/s/fRjGckCjERjLfFF). 4 Categorial limits for SCC: low = <39.9 cells × 1,000/mL; increased = 39.9–198.5 cells × 1,000/mL; high = >198.5 cells × 1,000/mL.

ously acknowledging the relevance of observer effects in locomotion scoring and varying interobserver agreement measures.

The animal-level prevalence of lameness in the present study is similar to reports by other authors: Solano et al. (2015) reported a prevalence of 20.8% for Canada, Barker et al. (2010) of 36.8%, and Griffiths et al. (2018) of 28.2% for the United Kingdom. One aspect to consider in regard to the reported prevalence of the current study is the voluntary participation of farm managers which may have been a source of potential participation bias. The response rate of the first invitation round was lower than expected and varied considerably across regions. Hence, a second round of invitations had to be sent to farmers to compensate for this. It may be plausible to assume that proactive farmers are overrepresented in our study by being generally more interested in scientific progress and improvement of animal husbandry. More refined housing conditions and closer monitoring, a focus on higher longevity of cattle, and lower lameness prevalence may be present on their farms. Hence, the true lameness prevalence in the underlying dairy cow population might be higher than the value reported in the current work. On the other hand, it could also be possible that especially those farmers with lameness problems or an overall inferior health situation on their farms were interested in participating in our study. In the latter case, the presented lameness prevalence would be an overestimation. Even though we cannot entirely rule out a certain degree of bias, we are confident that based on the alignment with other published work, the results do reflect the situation within the target population.

According to extant literature (Clarkson et al., 1996; Murray et al., 1996; Becker et al., 2014a), the most common lameness-associated claw lesions are white line disease, sole ulcer, heel erosion, and digital dermatitis, which all develop rather slowly and often represent chronic processes rather than acute events. It seems plausible that a cow recorded with an LS of $\geq 3 \mod 2$ likely had developed the underlying pathology some time beforehand (with the exceptions of foot rot and trauma, which occur as an acute condition), and it is therefore likely that the evaluated DHI milking characteristics could be reflecting an emerging lameness already some weeks before the detection of the clinical lameness by the farmer. This reasoning aligns with the findings of previous studies (Green et al., 2002; Reader et al., 2011), which reported a decrease in milk yield several months to weeks before the cows became visibly lame. To address this issue, a requirement for the inclusion of cows in the study was the availability of at least 3 test-day sampling milk values within the last 4 mo before the day of the individual locomotion and body condition scoring (exception: cows in the first 3 mo of lactation). We regard this threshold as reasonable from a biological point of view, but we need to underline that animals that did not have 3 complete DHI records before the farm visit were not part of the current analysis (with the exception of cows in the first 3 mo of lactation). This might consequently have resulted in the exclusion of animals without DHI data due to disease associated with lameness or metabolic imbalances during the selected period. The population analyzed in our work might hence be subjected to a certain level of bias and represent a group of animals with a better health status. Although we have been able to find relevant associations despite this issue, it is important to acknowledge it as a possible limitation.

The fact that some predictors were not consistent across regions lends support to the idea that these factors are biologically relevant in a region-specific manner only. Nevertheless, this finding could also indicate noise in the data. Although we are aware that we cannot definitively rule this out, we regard this possibility as rather unlikely, because biologically plausible explanations can be found for all affected predictors.

Structural Differences in the 3 Regions

The apparent differences between regions result from the regionally differing farming structures (Merle et al., 2012). In North and East the main breed is H, a breed with a clear focus on high milk yield (Yan et al., 2006), while the main breed of South is SIM, a dualpurpose breed (Bundesanstalt für Landwirtschaft und Ernährung, 2016, Grimm et al., 2019). The ECM per cow was the highest in East and so dairy production in general, feeding strategies, and ketosis prevention are assumed to be more intensive in this region.

While farmers often are unaware of the actual extent of the lameness prevalence in their herds (Whay et al., 2002, Šárová et al., 2011, Jensen et al., 2022) or do not regard the detected gait disturbances as actual lameness cases (Horseman et al., 2014), monitoring of individual animals due to a larger staff per animal ratio may be enhanced in the smaller farms in North and South which could be a potential explanation for the lower lameness prevalence there.

The structure and management of farms and especially conventional versus organic farming has to be considered. Over the last decades, farming in North and East in general has developed more and more toward large farm cooperatives and company managed farms, while in South farm management is still mostly family run. In addition, half of the entire German organic milk production volume is produced in region South (Bayerisches Staatsministerium für Ernährung, 2022). Although organic farming in North and East is rare, organic farming in South has a long tradition (BMEL, 2021), and has been developed and promoted by dairies and the federal government for many years (Bayer-

and the federal government for many years (Bayerisches Staatsministerium für Ernährung, 2022) and is an established part of the Bavarian dairy industry.

Differences in pasture management in the different regions have to be considered. If pasture is available on the farm, cows in regions North and South typically all have access to pasture during all days with suitable weather conditions. However, the pasture management in East focuses on cows in the dry period, while lactating cows in East rarely have any access to pasture.

Discussion of Model Results: Animal-Level Factors

Body Condition Score. A low BCS appeared to be a key predictor in all models, associated with higher odds of lameness in all regions. The thickness of the bovine digital cushion is an important factor in the development of claw horn disruption lesions and closely connected to the BCS (Bicalho and Oikonomou, 2013; Newsome et al., 2017a; Randall et al., 2018). During periods of negative energy balance, fat is mobilized from subcutaneous and intra-abdominal body reserves as well as from the digital fat pads (McNamara et al., 1995; Iqbal et al., 2016). Cows become lame as the decreasing dimensions of the digital cushion lead to an impaired ability to dissipate pressures of weight bearing and shielding the corium and the germinal epithelium from forces of the distal phalanx exerted on the corium (Newsome et al., 2017a,b). Subsequently, claw horn lesions develop as a result from contusions within the horn capsule.

Being lame and ranking low in the social hierarchy of the herd has been proven to correlate noticeably (Galindo and Broom, 2000). Animals with a lower social rank spend less time lying and more time standing, especially on the passageways, which was found to be associated with an increased number of clinical cases of sole, interdigital, and heel lesions (Galindo and Broom, 2000). Because of their lower rank, those animals might find themselves in a situation of ongoing competition for resources such as cubicles and access to the feed bunk. This situation may then lead to a negative energy balance and body condition loss, a hypothesis supported by the reported negative correlation of association between social dominance and body condition (Hohenbrink and Meinecke-Tillmann, 2012). As previously reported, a low or decreasing body condition is considered an important driver for lameness development (Lim et al., 2015; Newsome et al., 2017a,b). In addition, Schöpke et al. (2013) found cows with a low BCS to be more susceptible to interdigital hyperplasia.

Parity. The odds for lameness were lowest in first lactation, increasing with higher lactation numbers in all regions concurring with the findings of others (Espejo et al., 2006, Bicalho et al., 2009). Potential explanations include the accumulation of calving associated metabolic stress, and prolonged standing times on hard flooring. This contributes to the development of claw horn disruption lesions. Hormonal changes associated with calving that lead to loosening of the suspensory apparatus of the claw (Tarlton et al., 2002) may accumulate in multiparous cows. Incorrect and delayed treatment of lameness may lead to permanent damage (Newsome et al., 2016; Wilson et al., 2021), resulting in chronic lameness that is often difficult or impossible to treat (Thomas et al., 2016). Prolonged exposure to harmful aspects of housing (Kerr, 1998), previous lameness events (Hirst et al., 2002), and chronicity of claw pathologies (Alban et al., 1996) as well as development of arthritis with increasing age exert an additive detrimental effect. Early detection and treatment of lameness, especially in first- and second-lactation animals, are, therefore, crucial to continuously maintain the foot health of the herd (Hirst et al., 2002; Leach et al., 2012; Thomas et al., 2016).

Stage of Lactation. Cows in mid lactation appeared to have higher odds for lameness in this study compared with those in early and late stage of lactation (with the exception of early lactation cows in S). The influence of the lactation stage on lameness is complex. The potential convalescence during the dry period, which early lactating cows still may benefit from, could be one aspect to consider; however, dry period appeared to be a period of increased risk for lameness development in a study by Daros et al. (2019). Kempson and Logue (1993) found that horn quality in heifers worsens in late gestation, and horn growth increases from dry period to about 12 wk after calving and then decreases (Livesey and Laven, 2007). Considering these findings, increased odds for lameness in mid lactation might indicate that an additional trimming in mid lactation could confer positive effects specifically for cows in mid lactation. In this very context, it is important to be aware of the fact that claw horn lesions developing earlier in lactation may not become evident as clinical lameness until mid lactation (Collick et al., 1989; Leach et al., 1997; Green et al., 2002).

Energy-Corrected Milk. The fact that high ECM was associated with lower odds for lameness similar to recent results by O'Connor et al. (2019), can be explained by the circumstance that cows with sufficient energy supply are more likely to tap their full potential in milk yield performance than those having troubles to meet their metabolic requirements. The basis for a high milk yield is a good health condition, which is as-

sociated with good housing, feeding, and management conditions and, in consequence, good animal welfare (Quality, 2009). Cows hence may be at a lower risk to develop lameness as well. Additionally, as clinically lame cows were found to show decreased milk production already long before and while lameness cases were detected (Green et al., 2002; Reader et al., 2011), the reverse direction of the causality (i.e., lameness impairing productivity) could explain the association between low ECM and higher odds for lameness events. This concurs with findings by Solano et al. (2015), who reported a decrease in the odds for severe lameness cases with increasing milk yield. Of course, feeding strategies, feed composition, quality and supply are important factors in this context and should, therefore, not go unmentioned, but are beyond the scope of this study.

Risk for Ketosis. The occurrence of ketosis events represents a metabolic status of insufficient energy supply (Glatz-Hoppe et al., 2019b). The higher odds for lameness for cows with one or more ROK events complement the hypothesis of a negative energy balance to be an important factor associated with lameness, both as a risk factor and possible consequence. The reason that this parameter was not consistently associated with lameness in this study might be related to the findings of Schöpke et al. (2013), who reported a decreased milk fat content and FPR in cows diagnosed with laminitis in the following month.

Breed. Lower odds for lameness events within the population of other breeds than GH and SIM might include genetic factors (e.g., the recently discovered polygenic background to the digital cushion thickness; Barden et al., 2022) and the heritability for digital dermatitis and sole ulcer reported by Oberbauer et al. (2013). In alignment with our findings, Barker et al. (2010) reported lower lameness prevalences in herds with few or no GH cows compared with pure GH herds. Becker et al. (2014b), who also found GH cows to be more frequently affected by certain lesions, suggested that this might be result of breed-specific differing angles of the dorsal wall of the claw. In addition, highyielding breeds are suspected to be at greater risk for metabolic imbalances which again, are associated with lameness, but it is likely that the lower odds for lameness in breeds other than GH and SIM may also be an indirect effect originally caused by other circumstances. Those could include the fact that cows summarized in the breed category "other" were more likely to be housed on organic farms than of the breed SIM and GH in this study.

Somatic Cell Count. The association between low SCC and decreased odds for lameness could be ascribed to generally healthier cows, good housing and management conditions. In addition, ascending DIM (being as-

sociated with increased odds for lameness in this study) have been associated with an increase in SCC before (King et al., 2016).

Discussion of Model Results: Management Factors

Organic Farming. Organic farming has been associated with lower lameness prevalence in previous work (Rutherford et al., 2009). In Germany, organic farming underlies strict regulations regarding animal welfare such as obligatory access to pasture and increased space in exercise pens. Management regimens are less intensive and farmers are to improve the overall housing conditions. Moreover, organic farmers have a better awareness concerning lameness (Jensen et al., 2022). Smaller herd sizes and, therefore, closer monitoring might also lower the prevalence of undetected lameness cases and speed up the detection of evolving health problems. Furthermore, organic farms often have a lower average milk production compared with conventional farms (Slagboom et al., 2016), and since high milk production is reported to be associated with increased lameness risk (Green et al., 2002; Onyiro et al., 2008; Archer et al., 2010), this might be another aspect to consider.

Access to Pasture. The negative association between access to pasture and lameness was not surprising, because the positive influence of pasturing on lameness events, lameness recovery, and general welfare is well known, even when pasture is provided for only short periods (Hernandez-Mendo et al., 2007; Rutherford et al., 2009; McLellan et al., 2022). An appropriate pasture is considered a more comfortable and secure surface to walk and stand on compared with artificial indoor flooring systems, avoiding injuries and allowing a species-appropriate locomotory behavior (van der Tol et al., 2005; Alsaaod et al., 2017). In addition, cows on pasture spend more time lying in comfortable positions, and the absence of stall compartments when transitioning between lying and standing reduces the risk of injuries and lameness (Haskell et al., 2006).

Visit Year. The influence of the visit year may be a rather indirect factor being associated with lameness. It could be attributed to differing climatic conditions, allowing more or less access to pasture, altering the quality of the forage or exposing the cows to heat stress, which is associated with lameness (e.g., the years 2018 and 2019 featured hot and dry summers, especially in North; King et al., 2016). Because lameness prevalence is associated with season and appears to be generally more pronounced in winter and spring (Clarkson et al., 1996; Cook, 2003; Rutherford et al., 2009), other factors include the uneven distribution of farm visits in the different years and seasons (e.g., the largest farms were

visited in 2017, and data collection was terminated in summer 2019, leaving 2019 with no visits in autumn and less winter).

CONCLUSIONS

This cross-regional study evaluates a large data set collected across several years and 3 structurally differing regions and has therefore the potential to be extrapolated to a wide variety of loose-housed dairy production systems. The outcomes suggest a consistent and robust association of BCS, parity, and organic farming with lameness in dairy cows. However, as other factors differed from region to region, the results also suggest that region-specific characteristics should be taken into account in comparable future projects.

ACKNOWLEDGMENTS

Farm visits and data collection of this study were initiated and financially supported by the German Federal Ministry of Food and Agriculture (BMEL; Bonn, Germany) through the Federal Office for Agriculture and Food (BLE), grant numbers 2814HS006 (University of Veterinary Medicine Hannover), 2814HS007 (Freie Universität Berlin), and 2814HS008 (Ludwig-Maximilians-Universität Munich). We extend our cordial gratitude to all participating farmers as well as to the members of the PraeRi project. Our special gratitude is given to G. Knubben-Schweizer for supporting this work. All data collected were completely anonymized and processed in strict compliance with German and European data protection legislation. Participation in the study was voluntary and only farm- and management-specific data (no personal data) were collected in interviews and questionnaires. Written consent was obtained from the farmers to retrieve the required farm-specific data from the DHI and HIT databases. The authors have not stated any conflicts of interest.

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