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Bayesian evaluation of the accuracy of a thoracic auscultation scoring system in dairy calves with bronchopneumonia using a standard lung sound nomenclature

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Abstract

Background: Although thoracic auscultation (AUSC) in calves is quick and easy to perform, the definition of lung sounds is highly variable and leads to poor to moderate accuracy in diagnosing bronchopneumonia (BP).

Hypothesis/Objectives: Evaluate the diagnostic accuracy of an AUSC scoring system based on a standard lung sound nomenclature at different cut-off values, accounting for the absence of a gold standard test for BP diagnosis.

Animals: Three hundred thirty-one calves.

Methods: We considered the following pathological lung sounds: increased breath sounds (score 1), wheezes and crackles (score 2), increased bronchial sounds (score 3), and pleural friction rubs (score 4). Thoracic auscultation was categorized as AUSC1 (positive calves for scores ≥1), AUSC2 (positive calves for scores ≥2), and AUSC3 (positive calves for scores ≥3). The accuracy of AUSC categorizations was determined using 3 imperfect diagnostic tests with a Bayesian latent class model and sensitivity analysis (informative vs weakly informative vs noninformative priors and with vs without covariance between ultrasound and clinical scoring).

Results: Based on the priors used, the sensitivity (95% Bayesian confidence interval [BCI]) of AUSC1 ranged from 0.89 (0.80-0.97) to 0.95 (0.86-0.99), with a specificity (95% BCI) of 0.54 (0.45-0.71) to 0.60 (0.47-0.94). Removing increased breath sounds from the categorizations resulted in increased specificity (ranging between 0.97 [0.93-0.99] and 0.98 [0.94-0.99] for AUSC3) at the cost of decreased sensitivity (0.66 [0.54-0.78] to 0.81 [0.65-0.97]).

Conclusions and Clinical Importance: A standardized definition of lung sounds improved AUSC accuracy for BP diagnosis in calves.

Abbreviations: AUSC, thoracic auscultation; AUSC1, data categorization thoracic auscultation 1; AUSC2, data categorization thoracic auscultation 2; AUSC3, data categorization thoracic auscultation 3; BCI, Bayesian credible interval; BP, bronchopneumonia; covDn, covariance between TUS and WCRS in truly negative animals; covDp, covariance between TUS and WCRS in truly positive animals; DIC, deviance information criterion; IQR, interquartile range; NLR, negative likelihood ratio; NPV, negative predictive value; PLR, positive likelihood ratio; PPV, positive predictive value; STARD-BLCM, standards for the reporting of diagnostic accuracy studies that use Bayesian latent class models; TUS, thoracic ultrasonography; WCRS, Wisconsin calf respiratory scoring.

KEYWORDS
calves, respiratory disease, thoracic auscultation, thoracic ultrasonography

1 | INTRODUCTION

There is great interest in studying bronchopneumonia (BP) diagnostic strategies in calves because a more accurate indication for antimicrobial treatment for sick patients is needed, and no practical and affordable gold standard test currently exists.[1] One of the main obstacles to diagnosing individual BP cases is that the clinical signs expressed by affected animals are often variable.[2-4] Thoracic ultrasonography (TUS) is an accurate confirmatory test with good interrater agreement.[5] Existing research on the accuracy of TUS assessed using a Bayesian approach indicates that this method has high specificity (Sp) but variable sensitivity (Se) values depending on the geographic area, the different populations of calves enrolled in clinical trials, and different thresholds used to define BP (Se range, 59.8%-89%).[6,7] On the other hand, the diagnostic accuracy of clinical scoring systems remains debated because of low interobserver reliability[8] and poor correlation with ultrasound-defined lung injury.[9]

Thoracic auscultation (AUSC) is considered a cornerstone of respiratory tract examination in cattle and is often the first diagnostic approach used by practitioners.[4,10] In dairy calves, the interpretation of respiratory sounds showed poor interrater reliability[4] and poor and limited accuracy when correlated with TUS.[2,10] One of the critical limitations is that the definition of lung sounds is highly variable and complex.[11] Confusion regarding the terminology and numerous problems in interpreting auscultated sounds have been reported.[12,13] Another issue is the subjectivity of auscultation sounds[10] and poor agreement on the definition of the sick calf in published studies.[2,10,14-16] These same difficulties have made it a relatively underused clinical method in respiratory research in human medicine. Different authors argue that standardizing and simplifying the description of pathological sounds in respiratory AUSC of humans could improve international communication and accuracy.[17,18] In this regard, the task force on lung sounds of the European Respiratory Society aimed to standardize the nomenclature of thoracic AUSC findings in humans.[11] From this consensus, it emerged that crepitations or crackles should be used to describe crackling sounds and that rhonchi should describe low-pitched wheezes. Similarly, another report[12] recommended the use of terms such as increased breath sounds, crackles, wheezes, and increased bronchial sounds to describe pathological sounds commonly heard in large animals.

A standard definition and classification of lung sounds based on that study[12] might increase diagnostic accuracy in affected calves. Our objective was to evaluate the diagnostic accuracy (Se and Sp) of AUSC scoring systems (our primary test of interest with different definition thresholds) at different cut-off values characterized by an objective classification and definition of lung sounds for the diagnosis of BP and to determine the robustness of the test accuracy findings based on different priors obtained from Wisconsin calf respiratory scoring (WCRS) and TUS accuracy information as a part of a Bayesian sensitivity analysis framework.

2 | MATERIALS AND METHODS

2.1 | Study design

A cross-sectional diagnostic accuracy study was performed according to appropriate standards using Bayesian latent class models (STARD-BLCM).[19] The study design (approval number 104/2020, January 15, 2021) and publication of data from our clinic activity (approval number 47/2017, November 28, 2017) were approved by the institutional animal care and use committee of the University of Milan. All calves included in the study were managed according to standard protocols for diagnosing BP in compliance with the professional ethics of veterinarians and the standards for protecting calves.[20]

2.2 | Selection of farms

A convenience sample was selected from dairy farms requesting the Clinic for Ruminants and Swine, Department of Veterinary Medicine and Animal Sciences, University of Milan respiratory disease diagnostic service from January 2020 to December 2021. Criteria for farm inclusion were a history of spontaneous cough from calves in at least 1 calf pen detected by the herd practitioner, no history of treatment for BP in calves in the 15 days before the start of the study, farm location (no more than a 1-hour drive from our clinic), and willingness to use data for scientific purposes. This specific selection of farms with a relatively high anticipated prevalence of BP was used to mimic the potential context of applying the test under investigation (AUSC) and obtain a relatively narrow AUSC Se credible interval width by having a true BP prevalence higher than observed in the general calf population. On these farms, calves usually were separated from their dams immediately after birth and received 4 L of good-quality colostrum (Brix ≥22%) within 6-8 hours after birth. Calves were housed individually for up to 15-20 days and fed 4-5 L of milk replacer twice a day from a bucket before entering multiple pens with an automated calf feeder, where they remained for up to 75-80 days. After weaning, calves were transferred to multiple pens until they were 6 months old.

2.3 | Selection of calves

On the day of the study, both pre-and-post weaned dairy calves were observed. One primary author (first or corresponding author)

observed calves for detection of spontaneous cough. This approach was used to ensure a wider spectrum of the disease (cases at various stages of BP) in the affected pen to optimize sample size for both groups of calves (calves with and without BP) because cough is associated with increased probability of lung consolidation (which also is associated with true BP status).[9] Calves that coughed were scored using the WCRS by the same author who detected the cough. If 1 coughing calf was given a total respiratory score ≥5, it was considered a positive case.[21] Consequently, all calves in the pen were considered eligible for the study unless they showed lameness, dehydration, or umbilical pathology.

The selected animals were male and female Holstein Friesian calves aged between 1 and 6 months. A maximum of 20 calves was examined per farm. If >20 eligible calves ere present, the recruited calves were randomly selected using an application that runs on an Android smartphone (Randomizer, Darshan Institute of Engineering and Technology, Rajkot, India) with the list of identification numbers (written on the ear tag of each calf) provided by the farmer. The calf with WRSC >5 was released in the pen and reevaluated if it was on the randomization list or constantly reevaluated if <20 calves were in the pen. For each enrolled animal, identification, date of birth, age at clinical examination, and sex were recorded.

Enrolled calves were assessed using the same 1-gate design protocol. Two helpers gently captured each calf. Without moving it from the capture site, 1 of the principal authors performed WCRS for each calf. At this point, the calf was submitted first to AUSC and then TUS. Calves from the same herd were subjected to the clinical protocol on the same day. Each enrolled calf's ear tag, serial number, age, WCRS, AUSC, and TUS findings were written on predetermined tables.

2.4 | Wisconsin calf respiratory scoring chart

The same postdoctorally-trained veterinarian (GS) performed the WCRS in all enrolled calves. Each calf was examined and assigned a clinical score of 0 (normal), 1 (slightly abnormal), 2 (abnormal), or 3 (severely abnormal) for temperature, nasal discharge, cough, ocular discharge, and ear position, considering the highest score of eyes and ears. The scoring system resulted in a minimum score of 0 and a maximum of 12. If 1 calf reached a total respiratory score ≥5, it was considered a positive case.[21]

2.5 | Thoracic auscultation

Enrolled calves were subjected to bilateral AUSC by the first author using a conventional stethoscope (3M Littmann Master Classic II Veterinary, 3M Italy, Milan, Italy). The AUSC area was divided topographically into the ventral, middle, and dorsal thirds for both the right and left hemithorax (Figure 1). Each field was auscultated for at least 3 respiratory cycles (inspiration and expiration phases, totaling 18 cycles per calf). The clinician observed the right costo-abdominal region during the procedure to differentiate inspiration from expiration. Auscultation then was scored according to the nomenclature proposed previously[12] and summarized in Table 1. The term normal breath sound was used to describe the sounds produced during inspiration by normally aerated lung parenchyma. Increased breath sounds were defined as a moderate increase in loudness of breath sounds audible during inspiration and expiration when the difference between inspiration and expiration was always identifiable. Signs of bronchial diseases, including wheezes and crackles, also were noted. Signs of lung consolidation as increased bronchial sounds were defined as an actual increase in expiratory sounds reaching the same inspiration tone, simulating the sounds generally audible during tracheal auscultation, and leading to clear difficulty in distinguishing between inspiration and expiration sounds.[12,22,23] Signs of pleural anomalies, including pleural friction rubs, were recorded. In the case of multiple pathological sounds in the same calf, the highest score was recorded based on auscultation at 6 sites.

2.6 | Categorization of AUSC data

Data from the clinical examination were analyzed using 3 categorizations to explore the impact of different lung sound classifications (see Table 1). The first categorization (AUSC1) assessed accuracy of the AUSC when all the pathological lung sounds (increased breath sounds, wheezes, crackles, increased bronchial sounds, pleural friction ribs) were considered to qualify the animal as a positive case (calves with AUSC score ≥1). Calves with an AUSC score of 0 (normal breath sounds) were considered negative.

A second categorization (AUSC2) assessed the accuracy of the AUSC when only wheezes, crackles, increased bronchial sounds, and pleural friction rubs, but not increased breath sounds were considered to qualify the animal as a positive case (AUSC score ≥2), and therefore calves with a score <2 (normal breath sounds or increased breath sounds) were considered negative.

A third categorization (AUSC3) assessed AUSC when only severe pathological sounds (increased bronchial sounds, pleural friction ribs) were considered to qualify the animal as a positive case (AUSC score ≥3), whereas calves with a score <3 (patients with normal breath sounds, increased breath sounds, wheezes, or crackles) were considered negative.

2.7 | Thoracic ultrasonography

Systematic TUS (intercostal spaces [ICS] 10-1 on the right and ICS 10-2 on the left) then was performed on all auscultated calves based on landmarks described previously[24] by 1 of the main authors (DP) who was blinded to the AUSC and WCRS results. Ultrasonographic examination was performed using a portable unit (Ibex Pro, EI Medical Imaging, Loveland, Colorado) with a 7.5 MHz linear transducer designed for transrectal purposes, set to a depth of 8 cm and gain of 16 dB. The thorax was not shaved, and 70% isopropyl alcohol was applied to the hair as a transducing agent. Lung lobes were examined and scored based on the mass of lung tissue involved as

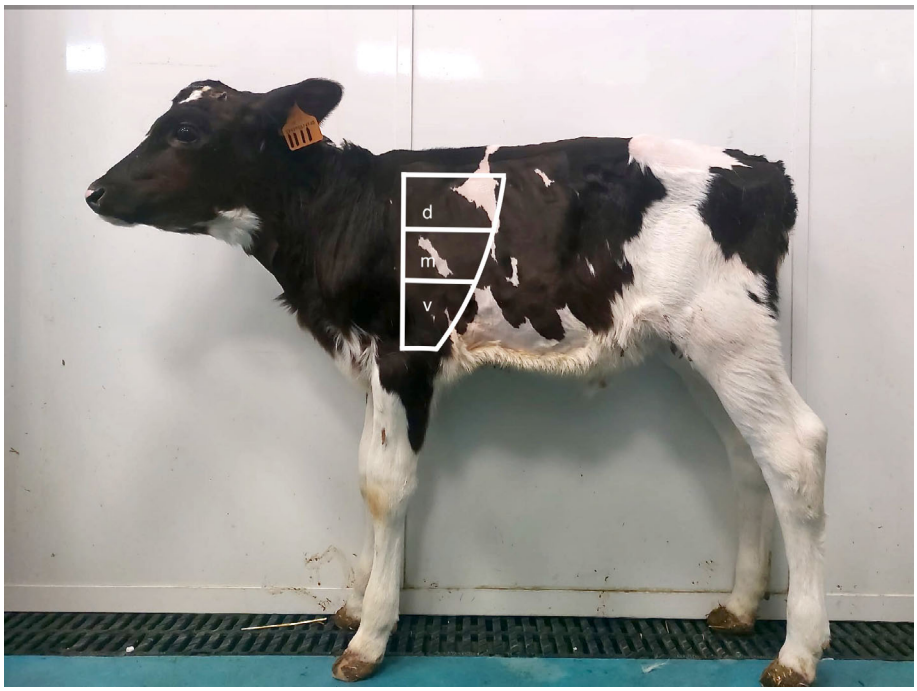

FIGURE 1 Illustration of thoracic auscultation sites. The total auscultation area ranges from the 10th intercostal space in the dorso-caudal regions of the lung projection area to approximately the third intercostal space located below the axillary region of the cranial thorax. The total area was divided into dorsal (d), median (m) to ventral (v) thirds and was auscultated for at least 3 respiratory cycles. The auscultation area was identical for both the right and left hemithorax.

TABLE 1 Thoracic auscultation (AUSC) scoring system and categorizations used to explore the impact of different lung sounds classification for distinguishing calves with and without bronchopneumonia.

Score	AUSC findings	Description	AUSC1	AUSC2	AUSC3
0	Normal breath sounds	Soft blowing sounds, longer and louder on inspiration than on expiration. In heavier subjects, the sound from expiration may be nonaudible (Lung sound file 1)	−	−	−
1	Increased breath sounds	Increase in loudness of breath sounds mainly on inspiration but also on expiration. The difference between inspiration and expiration is always identifiable (Lung sound files 2 and 3)	+	−	−
2	Signs of bronchial disease: (a) Wheezes or (b) Crackles in at least 1 auscultation site	(a) Variable-toned, intermittent, or continuous musical whistling sounds ("huin")[35] that are usually heard on expiration but can also be heard on inspiration (Lung sound file 4) (b) Crepitating nonmusical sounds ("knack")[35] (Lung sound file 5)	+	+	−
3	Signs of lung consolidation: Increased bronchial sounds in at least 1 auscultation site	High and harsh audible tone like what is usually possible to hear during trachea auscultation. Difficulty in assessing the difference in tone between expiration and inspiration (Lung sound file 6)	+	+	+
4	Signs of pleuritis: Pleural friction rubs	Grating sounds during inspiration and first phase of expiration (Lung sound file 7)	+	+	+

Note: For this study, we evaluated the accuracy performance of respiratory sounds by categorizing them into 3 different models: AUSC1 (score of 0 [negative] vs ≥ 1 [positive]), AUSC2 (score of 0 or 1 [negative] vs ≥ 2 [positive]), and AUSC3 (score ≤ 2 [negative] vs ≥ 3 [positive]). +: Sound considered in categorization of positive cases. −: Sound not considered in categorization of negative cases.

described previously,[7] which showed that ≥3 cm of depth yielded excellent accuracy for diagnosing active pneumonia in calves, and another study[25] that considered consolidation positive if ≥3 cm of consolidated lung was present. Ultrasonography scores ranged from 0 to 3 (0 = no lesions or < 1 cm consolidation; 1 = diffuse comet tails; 2 = patchy lesions, consolidation of ≥1 cm but <3 cm between normal aired lung parenchyma; 3 = lung consolidation area ≥3 cm). Consolidation was defined when the normal reverberation artifact was replaced by a hypoechoic structure similar to liver. For each calf, the maximal depth of consolidation on TUS was recorded. The maximal depth of lung consolidation (cm) was calculated by manual count using the lateral grid of the ultrasound image.

2.8 | Categorization of TUS data

Data from ultrasonography then were analyzed using the following categorization: calves with a score of 3 (consolidation depth ≥3 cm) were considered positive, and calves with a score <3 were considered negative.

2.9 | Descriptive statistics

Data storage and analyses were performed using IBM SPSS Statistics version 27.0 for Macintosh (IBM Corp., Armonk, New York). Descriptive statistics were performed, and age was reported with median, interquartile range (IQR), minimum and maximum because they were nonnormally distributed (Shapiro-Wilk test). At the same time, categorical variables (sex, AUSC score, TUS score) were expressed as frequencies and percentages.

2.10 | Contingencies tables

A first contingency table was obtained considering the 5° AUSC score combined with the 4° TUS score (Table 2). A second cross-classification table was built using data from WCRS (positive calves when WCRS scores were ≥5 and negative calves when WCRS scores were <5), TUS dichotomized as previously reported (positive calves with score = 3 and negative calves with score <3) and the 3 categorizations for AUSC (AUSC1, AUSC2, and AUSC3; Table 3).

2.11 | Bayesian latent class model

2.11.1 | Main model with informative priors on TUS and WCRS accuracy

The evaluation of AUSC at different cut-offs was determined using 1 population and 3 imperfect diagnostic tests under a Bayesian latent class analysis framework. The latent variable was the true BP status of the examined animals, which was evaluated using AUSC, WCRS, and TUS. A priori, we used a model with no covariance between tests because these tests are based on different modalities and because of the lack of identifiability of the model if allowing covariance between all tests. However, because it was difficult to rule out conditional dependence between WCRS and TUS as previously reported,[7] the possibility of positive conditional dependence between these tests in truly diseased (covDp) and truly nondiseased (covDn) calves was positively modeled using the Dendukuri and Joseph[26] parametrization as follows:

- covDp ~ Uniform (0, min [Se{WCRS}, Se{TUS}] − [Se{WCRS} × Se{TUS}]);
- covDn ~ Uniform (0, min [Sp{WCRS}, Sp{TUS}] − [Sp{WCRS} × Sp{TUS}]).

Informative priors were used to feed the model using previously obtained accuracy information from the WCRS[6] and TUS[7] at a specific threshold of positivity of ≥3 cm depth. For the WCRS, the median Se (95% credible intervals) and Sp were 62% (48%-76%) and 74% (65%-83%), respectively. Elicited beta distribution was obtained using the PriorGen R package.[27] The equivalent beta distributions were beta (21.47, 13.29) and beta (53.79, 19.11) for WCRS Se and Sp, respectively. For TUS accuracy, median Se and Sp were 89% (55-100) and 95% (92%-98%), respectively. The equivalent beta distributions were beta (4.62, 0.86) and beta (77.55, 4.4) for TUS Se and Sp, respectively.

2.11.2 | Sensitivity analysis: non informative and weakly informative models

Sensitivity analysis is an important part of Bayesian latent class models to determine the impact of priors on posterior findings.[14] Two specific scenarios were used for Se analysis. Noninformative models using uniform probabilities from 0 to 1 (beta [1]) were used for the accuracy of all the tests and the true prevalence of the disease as a default Se analysis.[28] However, using purely noninformative priors also has been criticized because a flat distribution can strongly

TABLE 2 Cross-tabulated results of thoracic auscultation (AUSC) and thoracic ultrasounds exam (TUS) on 330 dairy calves for diagnosing bronchopneumonia.

	AUSC (0-4)					
TUS (0-3)	**0**	**1**	**2**	**3**	**4**	**Total**
0	25 (7.6%)	1 (0.3%)	14 (4.2%)	0 (0%)	0 (0%)	40 (12.1%)
1	44 (13.3%)	33 (1%)	8 (2.4%)	0 (0%)	0 (0%)	85 (27.8%)
2	21 (6.4%)	19 (5.7%)	5 (1.5%)	14 (4.2%)	0 (0%)	59 (17.9%)
3	25 (7.6%)	22 (6.6%)	16 (4.9%)	79 (24%)	4 (1.2%)	146 (44.2%)
Total	115 (34.8%)	75 (22.7%)	43 (13.0%)	93 (28.1%)	4 (1.2%)	330 (100%)

Note: Systematic TUS (intercostal spaces [ICS] 10-1 on the right and ICS 10-2 on the left) score ranged between 0 to 3 (0 = no lesions; 1 = diffuse comet tails; 2 = patchy lesions; consolidation of ≥1 cm but <3 cm, between a normal aired lung parenchyma; 3 = lung consolidated area ≥ 3 cm). Thoracic auscultation scoring system ranged from 0 to 4 (0 = normal breath sounds; 1 = increased breath sounds; 2 = wheezes or crackles in at least 1 auscultation site; 3 = increased bronchial sounds; 4 = pleural friction rubs).

TABLE 3 Cross-tabulated results of Wisconsin calf respiratory scoring (WCRS), 3 different thoracic auscultation (AUSC) examinations definitions and thoracic ultrasonography (TUS) examinations with score ≥ 3 conducted on 330 dairy calves for the diagnosis of bronchopneumonia.

AUSC1					AUSC2					AUSC3				
		AUSC +	AUSC −	Tot.			AUSC +	AUSC −	Tot.			AUSC +	AUSC −	Tot.
WCRS +	TUS +	68	5	73	WCRS +	TUS +	21	35	56	WCRS +	TUS +	52	21	73
	TUS −	31	15	46		TUS −	38	117	155		TUS −	10	36	46
WCRS −	TUS +	53	20	73	WCRS −	TUS +	61	12	73	WCRS −	TUS +	31	42	73
	TUS −	63	75	138		TUS −	20	26	46		TUS −	4	134	138
		215	115	330			140	190	330			97	233	330

Note: AUSC +; calves with a AUSC score ≥ of 1, ≥2 or ≥3 for AUSC1, AUSC2 and AUSC3 respectively (pathological lung sounds); AUSC −; calves with a AUSC score < of 1, <2 or <3 for AUSC1, AUSC2 and AUSC3 respectively (physiological lung sounds). WCRS +; calves with a Wisconsin calf respiratory scoring ≥5 (considered positive). WCRS −; calves with a Wisconsin calf respiratory scoring <5 (considered negative). TUS +; calves with a TUS score of 3 (calves with lung consolidation ≥3 cm considered as positive). TUS −; calves with a TUS score < 3 (calves considered negative).

influence posterior findings and therefore could be considered informative.[28] Therefore, a third approach using a weakly informative approach was used. We used a horseshoe prior-like which allows lower probability for extremely improbable values (eg, values close to 0 or 100%). The horseshoe prior-like type used had a most probable value of 60% and 95th percentile of the distribution at 95% leading to a distribution beta (1.58, 1.16) for TUS and WCRS Se and Sp. Therefore, 6 different models were run for each auscultation definition (informative vs weakly informative vs noninformative, with or without conditional dependence). The Se analysis investigated whether posterior densities 95th Bayesian credible intervals of AUSC Se and Sp included the median estimates found from noninformative and weakly informative models.[6,19] Other model estimates also were analyzed using the same approach.

Models were run in OpenBUGS[29] using the interface of the package R2OpenBUGS in RStudio.[30,31] Three different chains were run starting from different units. Thirty thousand iterations were performed with a 5000 burn-in resulting in a total of 25 000. A specific thinning was added if needed based on autocorrelation plots.

2.12 | Evaluation of the models

Convergences of the different models first were assessed by visual inspection of the history and density plots.[29] This assessment was further formalized using Brooks-Gelman-Rubin (BGR) statistics measuring the ratio of the total variability combining multiple chains (between-chain plus within-chain) to the within-chain variability, which is close to 1 when convergence is achieved. Posterior distributions of each parameter were reported as medians and corresponding 95% Bayesian credible interval (BCI). The models were evaluated using deviance information criteria (DIC). A difference in DIC ≥5 was considered an indicator of better fit and used to assess model differences.[32]

3 | RESULTS

Fifteen males (4.5%) and 315 female calves (95.5%) belonging to 18 dairy farms were enrolled in the study (330 calves in total). On 14 farms, >20 calves were eligible, and therefore a randomization system was used. On 4 farms, 12, 9, 16, and 13 calves were eligible. The median age of the calves was 65 days (IQR, 47-85 days; range, 12-171 days). Cross-tabulated results of AUSC and TUS are shown in Table 2 and Figure 2. One-hundred and fifteen calves (34.8%) had physiological lung sounds (AUSC score = 0). Of these, 69 (60%) calves showed normal aerated parenchyma on TUS (TUS scores of 0 and 1). On the other hand, 46 (40%) calves showed lung lesions with TUS score ≥2 (21 calves with TUS score = 2, 25 with TUS score = 3). Two-hundred and fifteen calves had an AUSC score ≥1 (pathological lung sounds). Of these, 56 (26%) calves had normally ventilated parenchyma (TUS scores of 0 and 1), and 159 (74%) had a TUS score ≥ 2. Ninety-seven (45%) calves had severe pathological lung sounds related to lung consolidation or pleuritis. Of these, 14 had a TUS score of 2, and 83 had a TUS score of 3. On TUS examination, none of these calves showed normally aired parenchyma (TUS scores of 0 or 1). Of the 233 (70%) calves with lung sounds not related to consolidation or pleuritis (AUSC scores of 0, 1, 2), 63 had ≥3 cm of lung consolidation on TUS (TUS score of 3), and 45 had a patchy lesion pattern (TUS score = 2). The remaining 125 calves had normally aerated lungs (TUS scores of 0 and 1).

Figure 3 presents the posterior densities of the different AUSC Se and Sp definitions (AUSC1, AUSC2, and AUSC3) based on different modeling strategies (informative vs weakly informative vs noninformative priors and with vs without covariance between TUS and WCRS). The informative models (Table 4) had a higher deviance information criterion (DIC) than the others (noninformative or weakly informative models, see Supplementary Table 1). The DIC was relatively similar when comparing TUS and WRSC models with vs without covariance

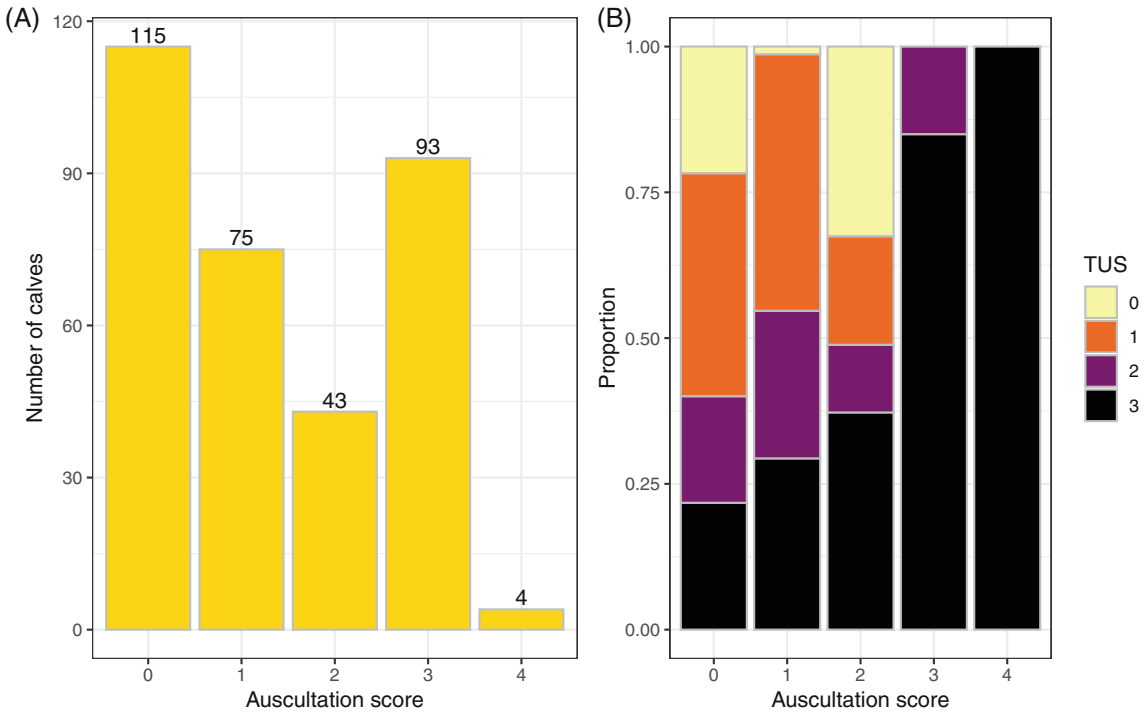

FIGURE 2 Distribution of auscultation score in 330 dairy calves (A) and correspondence between auscultation and thoracic ultrasonography (TUS) findings (B). Systematic TUS (intercostal spaces [ICS] 10-1 on the right and ICS 10-2 on the left) scores ranged from 0 to 3 (0 = no lesions or <1 cm consolidation; 1 = diffuse comet tails; 2 = patchy lesions; consolidation of ≥1 cm but <3 cm, between normally aired lung parenchyma; 3 lung consolidation = consolidation area ≥3 cm). Thoracic auscultation scoring system ranged from 0 to 4 (0 = normal breath sounds; 1 = increased breath sounds; 2 = wheezes or crackles in at least 1 auscultation site; 3 = increased bronchial sounds; 4 = pleural friction rubs or absence of breath sounds).

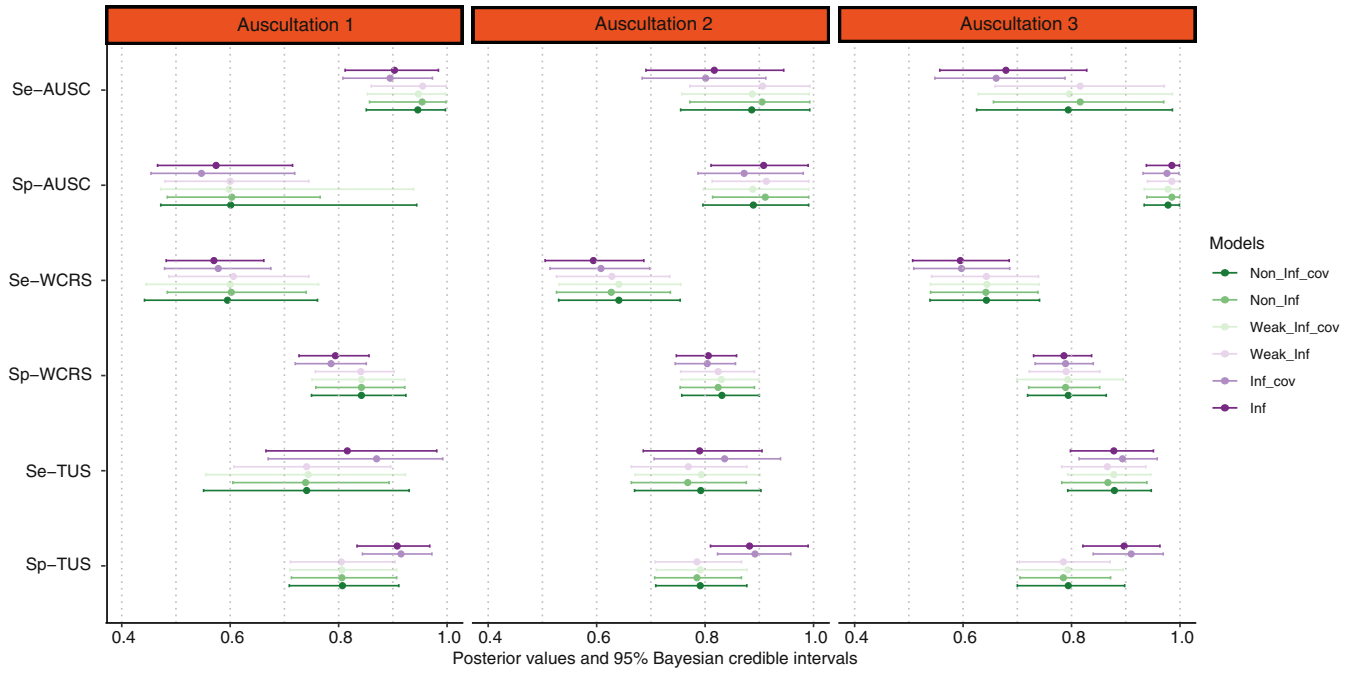

FIGURE 3 The distribution of posterior densities (median estimate and 95% Bayesian credible intervals of the accuracy [i.e., sensitivity (Se) and specificity (Sp)]) of thoracic auscultation (AUSC), thoracic ultrasonography (TUS), and Wisconsin clinical respiratory score (WCRS) based on different modeling strategies (informative vs weakly informative vs noninformative priors and with vs without covariance between TUS and WCRS) in 3 different auscultation categorizations (AUSC 1, 2, and 3).

TABLE 4 Posterior densities of Bayesian latent class main model with informative priors on thoracic ultrasound (TUS) and Wisconsin calf respiratory score (WCRS) for the diagnosis of bronchopneumonia (BP) in 330 dairy calves using 3 different auscultation (AUSC) definitions.

Informative priors for TUS and WCRS								
			Auscultation 1 (0 vs 1+)		Auscultation 2 (0, 1 vs 2+)		Auscultation 3 (0, 1, 2 vs 3+)	
Test	Parameters	Priors	No covariance Median (95% BCI)	Covariance Median (95% BCI)	No covariance Median (95% BCI)	Covariance Median (95% BCI)	No covariance Median (95% BCI)	Covariance Median (95% BCI)
AUSC	Se	beta (1,1)	0.903 (0.812-0.984)	0.895 (0.808-0.973)	0.817 (0.691-0.945)	0.801 (0.684-0.912)	0.679 (0.557-0.828)	0.661 (0.548-0.788)
	Sp	beta (1,1)	0.574 (0.466-0.715)	0.547 (0.454-0.719)	0.908 (0.811-0.990)	0.872 (0.787-0.981)	0.985 (0.938-0.999)	0.976 (0.932-0.998)
TUS	Se	beta (4.62,0.86)	0.816 (0.666-0.981)	0.870 (0.670-0.992)	0.790 (0.686-0.905)	0.836 (0.706-0.939)	0.878 (0.798-0.951)	0.894 (0.814-0.958)
	Sp	beta (77.55,4.4)	0.908 (0.834-0.0.968)	0.915 (0.844-0.972)	0.882 (0.810-0.990)	0.892 (0.823-0.958)	0.897 (0.821-0.963)	0.910 (0.840-0.969)
WCRS	Se	beta (21.47, 13.29)	0.570 (0.482-0.662)	0.578 (0.479-0.675)	0.594 (0.505-0.687)	0.608 (0.514-0.698)	0.595 (0.507-0.685)	0.597 (0.509-0.686)
	Sp	beta (53.79,19.11)	0.794 (0.727-0.856)	0.786 (0.720-0.851)	0.806 (0.747-0.858)	0.804 (0.745-0.856)	0.786 (0.730-0.837)	0.789 (0.733-0.840)
BP prevalence	P(BP)	beta (1,1)	0.469 (0.349-0.607)	0.447 (0.342-0.608)	0.459 (0.352-0.570)	0.443 (0.348-0.552)	0.420 (0.329-0.508)	0.425 (0.342-0.508)
Covariance	covDn	[a]	–	−0.006 (−0.024-0.022)	–	−0.009 (−0.025-0.017)	–	−0.007 (−0.024-0.018)
	covDp	[b]	–	−0.054 (−0.008-0.048)	–	−0.017 (−0.054-0.033)	–	−0.021 (−0.049-0.010)
DIC	DIC		46.7 (39.1-56.8)	47.6 (39.7-58.1)	47.0 (38.9-59.1)	47.5 (39.3-60.3)	46.0 (38.3-56.9)	44.8 (37.3-56.1)

Note: AUSC1 (score of 0 [negative] vs ≥ 1 [positive]), AUSC2 (score of 0 or 1 [negative] vs ≥ 2 [positive]), and AUSC3 (score ≤ 2 [negative] vs ≥ 3 [positive]) were associated with different definitions based on auscultation findings (see Table 1). Calves with a WCRS ≥5 were considered positive. Calves with a maximal consolidation depth ≥3 cm with TUS were considered positive.

Abbreviations: covDn, covariance between TUS and WCRS in truly negative animals; covDp, covariance between TUS and WCRS in truly positive animals; DIC, deviance information criterion; Se, sensitivity; Sp, specificity.

[a]min (Se-WCRS, Se-TUS) − Se-WCRS × Se-TUS.

[b]min (Sp-WCRS, Sp-TUS) − Sp-WCRS × Sp-TUS.

parameters. The main differences between informative priors and other models were that the AUSC3 Se median estimate of the noninformative and weakly informative models was not included in the 95% BCI of the informative model. However, the differences between the upper limit of BCI of the informative model and median estimates were small (<3%).

The AUSC Se and Sp (95% BCI) of informative models with no covariance were 90.3% (81.2%-98.4%) and 57.4% (46.6%-71.5%) for AUSC1, 81.7% (69.1%-94.5%) and 90.8% (81.1%-99.0%) for AUSC2, and 67.9% (55.7%-82.8%) and 98.5% (93.8%-99.9%) for AUSC3 respectively. Interestingly, the different prior scenarios only impacted posterior findings of TUS Sp which was higher in the informative prior model (i.e., approximately 90%) vs noninformative or weakly informative models (where the posterior median was approximately 80%). No meaningful impact was observed for TUS Se and WCRS Se and Sp (ie, posterior distribution medians included in the informative model 95th percent credible interval).

4 | DISCUSSION

We report the accuracy of 3 different AUSC criteria used to define BP. As expected, using all pathological sounds in AUSC1 led to high Se but low Sp. On the other hand, not considering increased breath sounds in AUSC2 or using only severe pathological sounds in AUSC3 resulted in high Sp at the cost of lower Se. The posterior findings of the different models generally were not impacted by the different prior scenarios except for AUSC3 Se and TUS Sp.

Assessment results of the accuracy of AUSC1 regarding all pathological lung sounds were consistent with a previous study.[10] Unfortunately, interpretation of all pathological sounds of AUSC1 suffered from lower Sp. These results could be related to the increased breath sounds included in the score, which occur in the early stages of BP but also in conditions unrelated to BP, such as exercise or stress.[22] On the other hand, when increased breath sounds were not considered, the Sp of both AUSC2 and AUSC3 was high (90.8% and 98.5%, respectively). The Sp of these models is higher than found in previous studies that reported an Sp of 53.3%.[10] In addition, regarding AUSC3, the high Sp was characterized by a narrow 95% BCI in all Bayesian latent class models, further strengthening the robustness of the findings. In addition to the absence of increased breath sounds in the model, another possible explanation for these results could be related to an unambiguous sound classification. According to a previous study,[12] an increased bronchial sound is the essential pathological sound in any lung disease in which ≥1 large bronchi remain open in lung tissue that has been replaced by consolidated tissue. In these conditions, forced respiratory sounds are transmitted better than in normally ventilated parenchyma, leading to a harsh sound similar to that usually heard during tracheal auscultation. Following the recommendations of previous studies,[12,22] we defined increased bronchial sounds as sounds from inspiration that were identical to those from expiration, differentiating increased bronchial sounds from increased breath sounds when the difference between inspiration and expiration was identifiable. Although, according to a previous study,[12] "there is no sharp line of demarcation between increased breath sounds and increased bronchial sounds," we believe that, in clinical practice, it is not difficult to recognize tracheal-like sounds during auscultation of the thorax when severe lung consolidation is present concurrently. Our results regarding the high Sp of AUSC3 showed that these sounds occur infrequently in healthy calves. Moreover, in our study sample, in relation to the contingency in Table 2, all calves with increased bronchial sounds had at least 1 cm of lung consolidation on TUS, whereas the increase in bronchial sounds did not occur in TUS-negative calves. The low Se of AUSC3 could be related to the fact that these sounds are not commonly produced, even in the presence of obvious lung damage. To produce increased bronchial sounds, at least 1 large bronchus must still be patent at the site of sound generation. In dairy calves, severe or chronic suppurative BP is characterized by evident intrabronchial purulent exudate, bronchiectasis, abscessation, or some combination of these which increases the risk of total obstruction of the airways.[33] Obstruction leads to a lack of ventilation of the distal airways with complete gas reabsorption, resulting in poorly-ventilated atelectatic areas.[34] Furthermore, with partial obstruction of the airways, which also may occur during a BP episode, a concomitant decrease in the strength of the sounds is also possible because a loss of intensity of the turbulent flow. This mechanism plays a pivotal role in the genesis of physiological and pathological lung sounds.[35] Airflow limitation during lung inflammation also has been reported in calves inoculated experimentally with *Pasteurella multocida*.[36]

The AUSC2 results were expected. Although wheezes and crackles generally indicate bronchial disease, both are found in cases of BP. Different mechanisms of origin have been described for these sounds, high-velocity air passage through intrathoracic airway obstructions (wheezes) and sudden opening of airways or rupture of fluid menisci in the case of moderate density exudate (crackles).[11,22] Whatever the pathogenesis, most bronchial anomalies derive from increased fluid from inflammation associated with BP. However, intrabronchial mucopurulent exudate is not always necessarily abundant. Its quantity also is related to the stage of the inflammatory process (ie, it usually increases in chronic stages of the disease).[33] In addition, a pathological process at the bronchial level is possible without injury to the alveolar tissue, especially in the case of infection by moderately virulent pathogens.[37] These factors may explain the relatively lower robustness of AUSC2 compared to AUSC3, particularly concerning the increased width of the BCI that characterizes the (albeit high) Sp of the AUSC2 model.

Our results raise intriguing questions regarding the use of AUSC for diagnosing BP in dairy calves. Severe pathological sounds (increased bronchial sounds and pleural friction rubs) were found to be highly specific pathological sounds for diagnosing BP. The low Se of these sounds results in missed BP cases because of a high false negative rate, thus highlighting the need for a composite reference to better identify positive cases. At the same time, these observations support the hypothesis that standardizing and simplifying the description of lung sounds may be helpful in the diagnosis of lung diseases in

farm animals. Because of the continual use of imperfect diagnostic tests, AUSC still should be considered a helpful tool, especially in field conditions. Thoracic ultrasonography could be beneficial in addition to AUSC to correctly identify affected animals, especially in calves in which increased bronchial sounds are not auscultated, or to better identify calves with pathological sounds less specific to lung disease. Thus, higher accuracy in diagnosing BP could be achieved, leading to more judicious use of antibiotics.

We performed various Bayesian latent class models allowing for covariance between TUS and WRSC and with various types of priors for TUS and WRSC. It is essential to point out that this Se analysis is inherent to latent class modeling.[19]

Using the different prior scenarios helped in finding consistent AUSC accuracy parameters results except for AUSC3 Se, for which the informative model gives lower Se results than the non and weakly informative models. However, the difference between the upper limits of informative models and median estimates of models used in the sensitivity analysis was small and negligible from a clinical point of view. The impact of priors therefore was limited to AUSC accuracy parameters. Other secondary test accuracies (e.g., TUS Se, WCRS Se, and Sp) were relatively stable independent of the prior information except for TUS Sp. The WCRS posterior findings were similar to those of the previous study used for informative prior specification (even in models where noninformative priors were used for WCRS accuracy). On the contrary, the TUS Sp was higher in informative models (close to 90% with BCI >80%) vs non or weakly informative models (median estimate close to 80%). The informative priors from TUS and WRSC were taken from previous dairy studies performed in North America.[6,7] Interestingly, the informative prior for TUS Sp could be considered a strong prior with low uncertainty (median, 95%; 95% BCI, 92%-98%). Therefore, it is not unexpected that it markedly impacted TUS Sp posterior, especially for total sample size including several hundred animals. However, the magnitude of the difference was limited from a clinical standpoint (from 90% to 80% Sp). Part of the variability among the different models could be partially associated with these priors not accounting for the BP pathogens, which may vary from 1 geographical area to another. The accuracy of the diagnostic tests, primarily TUS, also may depend on the specific pathogens involved in BP, which can lead to different ultrasonographic findings and more variable accuracy of lung consolidation for BP diagnosis. Unfortunately, we could not test this hypothesis, but it would be necessary in the future to determine if TUS accuracy depends on potential calf or farm characteristics. The different modeling scenarios covered a wide range of settings and could be considered relatively robust for most accuracy parameters with the limitation previously discussed.

Our study had some limitations. First, the study was performed using a convenience sample of farms and enrolled a limited number of animals with a high BP prevalence. Second, although 2 independent operators performed the AUSC and TUS to ensure blinding between examinations, only 1 operator performed the AUSC. Therefore, future studies on the interobserver agreement of AUSC to detect BP are recommended. In humans, such studies have led to improved standardization of lung sound definitions.[17] We did not control for potential conditional dependence between AUSC and TUS or WCRS because of the limitation of the degrees of freedom associated with the study design (1 population, 3 tests). As previously reported, we only include a conditional dependence between TUS and WCRS and could not rule out that AUSC accuracy is dependent of TUS or WCRS. Microbiological tests could not be performed on these farms. Therefore, we cannot determine the auscultation accuracy findings according to the specific pathogens encountered.

In conclusion, we showed that the AUSC3 described here has, on average, a high Sp for detecting BP in dairy calves. These results indicate that the definition of lung sounds used in our study could help practitioners detect BP based on inexpensive tools and easy-to-obtain clinical parameters. Therefore, AUSC with rigorous and objective definitions should be further investigated and preserved in the required skills of veterinarians. Additional studies are needed to assess the accuracy of AUSC in populations with a different prevalence of BP in dairy calves and the interrater agreement of different operators using a precise definition of lung sounds.

Supporting information: Additional supporting information for this article is audio files recorded with an electronic stethoscope (3M Littmann mod. 3200 Electronic Stethoscope, 3M Italy, Milan, Italy) in patients with various pathological conditions (Lung sound files 1-7).

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CONFLICT OF INTEREST DECLARATION

Dr. Sébastien Buczinski serves as Consulting Editor for Experimental Design and Statistics for the Journal of Veterinary Internal Medicine. He was not involved in review of this manuscript. No other authors declare a conflict of interest.

OFF-LABEL ANTIMICROBIAL DECLARATION

Authors declare no off-label use of antimicrobials.

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE (IACUC) OR OTHER APPROVAL DECLARATION

The study design (approval number 104/2020, Jan. 15, 2021) and publication of data from the ordinary activity of our clinic (approval number 47/2017, Nov. 28, 2017) were approved by the IACUC of the University of Milan.

HUMAN ETHICS APPROVAL DECLARATION

Authors declare human ethics approval was not needed for this study.

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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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