

## Long-term monitoring of environmental risk factors for bovine respiratory disease complex in different dairy calf rearing conditions

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### Abstract

Bovine respiratory disease complex (BRDC) is still one of the most challenging problems in calf rearing, therefore identification and continuous real-time monitoring of contributing environmental factors might play a role in mitigation of the damage caused by the disease. Microclimatic variables (temperature, relative humidity, air velocity, airborne particles expressed in particulate matter [PM], aerial germ load and gaseous pollutants) of a conventional calf barn and outdoor placed small-group hutches with pens were real-time monitored in a dairy herd by mounted sensors from summer to winter. Among the risk factors for BRDC, the small-group outdoor rearing units were characterized by high relative humidity, air velocity, and PM<sub>2.5</sub> particulate matter concentration. Although the conventional calf barn was poorly ventilated, most variables were more favourable than expected, in which proper farm management may have played a role. We were able to identify long term and intraday periods with limit-breaking values, daily fluctuations as well as specific patterns of individual indicators in different calf husbandry environments. Based on obtained data, contributing technological processes may be reviewed and the effect of changes can be monitored under farm conditions. In addition, assessment of prevailing microclimatic conditions is also possible before investing in modernization of calf rearing units.

*Particulate matter, ammonia, wind speed, humidity*

Bovine respiratory disease complex (BRDC) is one of the major causes of dairy industry losses (Gorden and Plummer 2010; Panciera and Confer 2010). As a multifactorial disease it is related to viral and bacterial infectious agents, individual resistance, microclimatic, husbandry and management factors (Gulliksen et al. 2009; Griffin et al. 2010; Ózsvári and Búza 2015; Buczinski et al. 2018b; Stokstad et al. 2020). The latter ones contribute to the development of BRDC by promoting the survival and spread of pathogens and weakening the overall resistance of the defence mechanisms of the calf respiratory system by increasing stress (Ackermann et al. 2010). Group housing and large group size, poor litter, direct contact with older animals, unfavourable climatic conditions (heat, cold, snow, frost) and inappropriate air quality such as high humidity, dust and harmful gases can be cited as main risk factors (Lundborg et al. 2005; Lago et al. 2006; Svensson and Liberg 2006; Taylor et al. 2010; Ózsvári and Búza 2015).

Aerosols of the livestock buildings may also play a critical role, as they are made up of feed, litter, organic substances from animals (e.g. epithelial cells, hair, urine, faeces), microorganisms and toxins. Due to their size, PM<sub>10</sub> and PM<sub>2.5</sub> particles are important: they irritate the conjunctiva and the airways when inhaled. Moreover, PM<sub>2.5</sub> particles accumulate in the lung parenchyma and can lead to serious respiratory and systemic disease (Losacco and Perillo 2018). High temperature, low humidity, air movement (especially drafts), and the increased activity of animals also help the litter to dry out, turn into dust,

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and allow particles to enter the air (Urso et al. 2021). Bioaerosols on farms can serve as vectors for microorganisms (Islam et al. 2019), and even though most of them are non-pathogenic, they can burden the respiratory defence mechanisms (Wathes et al. 1983). Therefore, the total number of aerial germs (CFU/m<sup>3</sup>) can be used as an air hygiene marker (Nordlund 2008). Among the gaseous pollutants in barn indoor air gases, ammonia may be of the greatest importance, as it directly damages the respiratory epithelium (Brscic et al. 2010) and even with exposure to a concentration of 4 ppm, tissue changes in the lungs can be detected by ultrasound examination (van Leenen et al. 2020).

In our study, we investigated whether continuous measurements could be used to monitor the long-term dynamics and diurnal patterns of critical microclimatic factors and their correlations, and thus provide a basis for the development of a BRDC predictive precision livestock farming (PLF) system.

### Materials and Methods

Air quality indicators were investigated in different calf housing conditions between 2019 July and 2020 February on a Hungarian large-scale dairy farm. Mounted climate monitoring devices (U1, U2, and U3) were used in individual calf hutches (ICH) and group calf pens (7GCP, U2), in group calf hutches (GCH, U1) and in conventional group barn (CGB, U3). Outdoor ICHs and 7GCPs were emplaced on the central farm (47°38'06.91" N; 19°54'59.72" E). As both kinds of rearing units were in the same area, we considered that their microclimates could be monitored with the same standing device (U2). The ICH consisted of a 1.2 × 1.0 m metal cage run and a 1.5 × 1.0 m fiberglass calf hutch. In each 7GCP seven calves were raised. The pen consisted of a 3 × 3 m cage run and a 3 × 3 m fiberglass hutch. After weaning, calves were transported to a replacement farm about 4 km away (47°37'23.74" N; 19°58'07.09" E), where they were raised in a CGB, which is still widely spread type of building in Hungary. The CGB was an approx. 60 × 10 m brick constructed, tent roofed building with an unroofed runway on the east side. The building and the run were divided into 3 compartments (for 3 × 40 calves). During the 2019/2020 winter two modern small outdoor GCHs with pens (Holm&Laue Multimax Duo Veranda pen, Westerrönfeld, Germany and Hampel Calf-Tel hutch, Germantown, WI, USA) were also used as experimental units for weaned calves (6 calves in each). The GCHs consisted of a 6 m<sup>2</sup> plastic calf shed and a 7 m<sup>2</sup> roofed run. The calf rearing units are referred to as U1, U2, and U3 in this article, based on the names of mounted devices.

Properties of the calf rearing units and measurement conditions are presented in Table 1; the technical data of sensors and previously reported limits and hazardous levels of measured microclimatic factors are summarized in Table 2. All three mounted devices (built by Boreas Ltd., Érd, Hungary) were continuously monitoring the temperature (T) and relative humidity (RH) with a BHP-06 sensor, the wind speed (WS) with a BWS-06/HW hot wire anemometer, and PM2.5 and PM10 with a Honeywell HPMA115S0 sensor. The device in barn U3 was also equipped with BGS-06 amperometric sensors for the monitoring of gaseous indicators: carbon monoxide (CO), carbon dioxide (CO<sub>2</sub>), hydrogen sulphide (H<sub>2</sub>S), ammonia (NH<sub>3</sub>) and methane (CH<sub>4</sub>). The devices recorded data at 10- (U1 and U2) and 5-min intervals (U3), then the data were transmitted to the operating company's server. During the study period the calves were being raised at U2 (about 1–3 month old, until weaning) and U3 (> 3 month old) locations, except U1, where calves were being kept only from December 2019 till January 2020 (3–4 month old). The U1 and U3 locations were used to investigate the climatic characteristics of two different yet widely used calf rearing units for weaned dairy calves. Due to technical reasons, continuous measurement was only available until the end of November in unit U3.

In winter months (according to the life stages of calves kept in these units), three times weekly point measurements of temperature, relative humidity, and airborne particles (PM2.5 and PM10 concentrations in µg/m<sup>3</sup> and number of 0.3–10.0 µm particles by sizes in pcs/m<sup>3</sup>) were carried out in U1, U2, and U3 units with a hand-held particle counter (HPC, Trotec PC220, Trotec GmbH, Heisenberg, Germany). Data were recorded at 3 different points of the units in a row. According to the same scheme, but once weekly air samples were also taken with portable direct-to-agar impaction sampler (MicroBio MB2, Cantium Scientific Ltd., Dartford, United Kingdom) to measure the aerial germ load. Briefly, 10-litre air sample was collected on plate count agar (PCA). Before sampling, the metal, fenestrated casing of the device was disinfected with alcohol. Inoculated plates were stored at 4–8 °C and transported to the laboratory (Eurofins Food Analytica, Gyula, Hungary) at the day after sampling and tested according to standard for microbial count of workplace air (collision method, colony counting, MSZ EN 13098:2001 and MSZ EN ISO 4833-2:2014; incubation at 30 °C, 72 h). Results were expressed in colony-forming units (CFU)/m<sup>3</sup>.

Data were processed with R-project (Posit, Boston, MA, USA) statistical software and Microsoft Excel (Microsoft Corporation, Redmond, WA, USA) spreadsheet. During the data cleaning process observations were compared with the lower and upper limits of the linearity interval. Measurements that had fallen outside of these limits were considered non-linear measurements and were not taken into account during statistical analysis. Descriptive statistics contain monthly, daily and hourly number of observations, minimum, maximum, median, mean values and standard deviations, as well as monthly means by hours and number of limit-exceeding

Table 1. Properties of calf rearing units and measurement conditions (U1, U2, and U3).

Place	Device	Location properties	Calf age groups	Timeframe	Mounting of the devices
ICH	U2	• open towards South or North	until 5 weeks	Sept 2019-	• on a stand • at ca. 1.5 m height • at the border of the hutch and the adjacent run
		• straw bedding (in- and outside)		Feb 2020	
7GCP	U2	• open towards South	5–12 weeks (until		
		• straw bedding (in- and outside)	weaning, conventional)		
GCH	U1	• drinking buckets, hay rack, roofed manger			
		• open towards South	12–18 weeks (after		
CGB	U3	• straw bedding (in- and outside)	weaning, experimental)	July 2019–	• inside the building • on the wall • at ca. 2 m height • in the central compartment
		• drinking buckets, hay rack, roofed manger		Nov 2019	
CGB	U3	• open towards South and North	> 12 weeks (after		
		• small windows and door openings on the longer sides	weaning, conventional)		
CGB	U3	• straw bedding inside, concrete floor outside			
		• unroofed mangers alongside the run, ball drinkers			

U1, U2, and U3 are names of mounted climate monitoring devices and also refer the calf rearing units; ICH = individual calf hutches (U2); 7GCP = group calf pen for 7 calves (U2); GCH = group calf hutch (U1); CGB = conventional group barn (U3).

Table 2. Technical parameters of U1, U2, and U3 mounted sensors and applied literature limits.

Unit	Sensor	Parameter	Detection range	Resolution	Limit	Reference
		T	–50 to +70 °C	0.1 °C	-	
U1	Boreas BHP-06	RH	0–100%	0.1%	80%	Webster (1984)
					85%	Webster (1984); Lorenz et al. (2011)
		WS	0.01–10 m/s	0.01 m/s	0.2 m/s	Roland et al. (2016)
U2	hot wire anemometer				0.6 m/s	Roland et al. (2016)
U3	Honeywell HPMA115S0	PM2.5	0–1000 µg/m <sup>3</sup>	1 µg/m <sup>3</sup>	25 µg/m <sup>3</sup> 15 µg/m <sup>3</sup>	WHO (2005) WHO (2021)
		PM10	0–1000 µg/m <sup>3</sup>	1 µg/m <sup>3</sup>	50 µg/m <sup>3</sup> 45 µg/m <sup>3</sup>	WHO (2005) WHO (2021)
U3	Boreas BGS-06 amperometric sensors	CO	0–200 ppm	12.5 ppb (0.0125 ppm)	10 ppm	EFSA (2009)
		CO <sub>2</sub>	200–40000 ppm	1 ppm	3000 ppm	EFSA (2009)
			200–40000 ppm	1 ppm	2000 ppm	Rafai (2003)
		H <sub>2</sub> S	0–50 ppm	1 ppb (0.001 ppm)	0.5 ppm	EFSA (2009)
					20000 ppb	EFSA (2009)
					(20 ppm)	
	NH <sub>3</sub>	0–100 ppm	1 ppb (0.001 ppm)	10000 ppb (10 ppm)	Lundborg et al. (2005), (Woolums et al. (2009)	
				4000 ppb (4 ppm)	van Leenen et al. (2020)	
	CH <sub>4</sub>	0–30 ppm	1 ppb (0.001 ppm)	-	-	

U1, U2, and U3 are names of mounted climate monitoring devices; T = temperature, RH = relative humidity, WS = wind speed; PM2.5 and PM10 = 2.5 and 10 µm particulate matter; CO = carbon monoxide; CO<sub>2</sub> = carbon dioxide; H<sub>2</sub>S = hydrogen sulphide; NH<sub>3</sub> = ammonia; CH<sub>4</sub> = methane.

days and hours for environmental indicators in every location. Inductive statistics contain Pearson correlations between microclimatic parameters (all data, as well as data by locations) and logistic models to compare locations to each other.

## Results

Temperature (T), relative humidity (RH), wind speed (WS), and particulate matter (PM 2.5 and PM10) concentration data were collected from July to November in U3, and from September to February in the small group rearing units (U1 and U2), whose monthly mean values are summarized in the tables below (Table 3), while daily means are plotted in diagrams (Fig. 1). Monthly mean values of forenoon data of mounted devices and of HPC (point measurements) are shown in Table 4. Logistic models were calculated considering previously reported limits of hazardous levels (Table 5) to individual microclimatic factors, where U2 was appointed as basis, as it was used until weaning and U1 and U3 were compared to it (all three units compared based on data from September to November, while U1 and U2 units based on data from September to February as well). Probability with 95% confidence interval show the proportion of measured data above the limit. The effect of month and location : month interactions were also investigated (data not shown, significant results are marked in text). Number and proportion of days when daily mean values of different microclimatic indicators fell above recommended limits are presented in Table 5, while the number and proportion of days and hours when average harmful gas levels in U3 exceeded limits are shown in Table 6.

### Temperature, relative humidity

The CGB (U3) barn was always warmer during autumn, while similar T characterized the GCH (U1) and 7GCP (U2) units. Monthly mean RH varied inversely with T and similar statement can be made for daily average T and RH too. The RH of each location showed a significant difference during autumn (Table 5). Depending on whether a RH value of 80 or 85% is considered as limit, about half or more of the measured values exceeded that in the small group units, where U1 was the most humid. Compared to them, values above the 80% limit rarely occurred in the U3 unit. As no measured data were > 85% in U3, the model could not compare it to U2 and could not estimate the probability and confidence level for this limit in U3. Examining the autumn and winter values of U1 and U2 together, more than half, or even two-thirds of the values breached the limit. When the month and location : month interactions were also taken into account in the model, it could be seen that the frequency of RH values above the limit was not determined by the location, but by the month ( $P < 0.0001$ ). Analyzing the pattern of monthly means by hours in the autumn months (Fig. 2), the coldest T and highest RH was observed around 6–7 in the morning at each location, then T rose and RH decreased gradually. The warmest T and lowest RH could be measured between 14:00 h and 17:00 h, after which T gradually decreased and RH increased until the morning.

### Wind speed

Wind speed at each location showed a relevant difference during autumn (Table 3). Unit U2 was significantly draughtier compared to U1 and U3. When the limit was 0.2 m/s three-quarters, while at 0.6 m/s about half of the measured values in small group units fell above the limit value, while we rarely experienced this in U3 (about 5% of the data at 0.2 m/s, and occasionally at 0.6 m/s). When the autumn and winter periods of the U1 and U2 units were analysed together, we could see limit breaks in similar proportions. When the month and location:month interactions were taken into account in the model, it could be seen that the frequency of windspeed values above the limit were determined by the location ( $P < 0.0001$ ). Analyzing the pattern of monthly averages by hours and by location,

Table 3. Average monthly values of measured microclimatic parameters in U1, U2, and U3 units.

Device (site)	T (°C)		RH (%)		WS (m/s)		PM2.5 (µg/m <sup>3</sup> )		PM10 (µg/m <sup>3</sup> )		CO <sub>2</sub> (ppm)		H <sub>2</sub> S (ppb)		NH <sub>3</sub> (ppb)		
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	
U1 (GCH)	Sept	17.18	5.62	71.31	20.01	0.63	0.77	7.64	7.83	8.69	8.01						
	Oct	12.51	5.84	77.02	16.88	0.57	0.74	22.54	24.27	23.92	24.72						
	Nov	8.69	3.44	94.27	7.95	0.75	0.68	22.94	19.13	24.31	19.51	not		not		not	
	Dec	2.89	3.96	92.19	7.65	0.67	0.65	28.84	19.08	30.41	19.47	measured		measured		measured	
U1 (GCH)	Jan	-0.66	2.59	92.68	7.73	0.61	0.69	43.06	29.40	44.82	29.79						
	Feb	4.89	4.43	73.55	13.29	1.47	1.41	16.59	21.09	17.83	21.49						
U2 (7CGP)	Sept	17.46	5.60	68.51	20.20	1.05	1.07	9.31	28.09	10.39	28.47						
	Oct	12.80	5.86	72.92	15.60	0.89	0.89	19.83	28.41	21.12	28.86						
	Nov	8.82	3.30	88.49	7.79	0.90	0.86	17.76	14.12	18.94	14.41	not		not		not	
	Dec	2.65	3.83	88.97	8.24	0.88	0.92	24.53	15.10	26.03	15.52	measured		measured		measured	
U2 (7CGP)	Jan	-0.90	2.49	91.19	8.95	0.71	0.69	43.66	53.86	45.44	54.52						
	Feb	4.85	4.22	70.30	13.51	1.45	1.56	13.78	18.07	14.98	18.42						
U3 (CGB)	July	28.01	4.52	52.66	11.16	0.07	0.16	6.26	8.44	7.18	8.63	932.92	647.31	6.29	14.52	528.04	581.78
	Aug	29.30	4.41	51.98	9.83	0.06	0.15	6.52	6.96	7.42	7.14	1075.00	807.40	7.88	15.64	284.64	351.75
	Sept	22.33	4.20	55.15	11.34	0.04	0.13	6.22	6.69	7.21	6.86	1022.73	883.71	5.47	10.94	1055.87	1073.48
	Oct	17.81	4.63	58.11	8.72	0.05	0.16	17.31	17.90	18.54	18.23	1413.05	1135.33	1.64	1.11	307.17	299.16
U3 (CGB)	Nov	14.78	2.45	73.06	4.73	0.01	0.09	11.08	9.50	11.85	9.97	1005.22	825.17	1.77	5.88	70.41	85.42

U1, U2, and U3 are names of mounted climate monitoring devices and also refer the calf rearing units; 7GCP = group calf pen for 7 calves (U2); GCH = group calf hutch (U1); CGB = conventional group barn (U3); T = temperature, RH = relative humidity, WS = wind speed; PM2.5 and PM10 = 2.5 and 10 µm particulate matter; CO<sub>2</sub> = carbon dioxide; H<sub>2</sub>S = hydrogen sulphide; NH<sub>3</sub> = ammonia.

Table 4. Monthly mean values of forenoon data on different microclimatic and aerosol parameters measured with HPC, MAS, and mounted devices between 9:00 h and 13:00 h in winter months in units U1, U2, and U3, and comparison of the units by HPC results according to measurement periods.

Device	Site	Month	T °C	RH		PM2.5 µg/m <sup>3</sup>	PM10 <sup>3</sup> µg/m <sup>3</sup>	0.3 µm		0.5 µm		1.0 µm		2.5 µm		5.0 µm		10.0 µm		Germ load CFU/m <sup>3</sup>				
				Mean	SD			Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean
HPC and MAS (9:00–12:00)	U1 (GCH)	Dec	6.33	82.05	87.03	148.12	139109.97	51132.97	8928.64	728.00	232.42	50.16	8100.00											
			2.37	8.54	99.87	153.91	80775.83	39817.37	8080.78	687.95	232.81	57.57	6318.23											
		Jan	2.32	83.80	213.10	352.18	222237.37	91697.69	18320.39	1267.82	359.83	75.43	17744.44											
			1.30	11.04	228.87	352.30	96599.08	50046.79	11736.66	897.36	257.42	65.07	16495.39											
		Feb	7.05	62.91	18.21	48.43	126698.82	25183.70	2354.68	238.21	59.68	15.18	not measured											
			1.80	18.96	33.43	79.72	114371.68	21919.90	1741.04	156.70	37.47	7.96	10700.00											
	U2 (7GCP)	Dec	6.20	78.43	279.45	314.45	175306.83	76586.18	14290.30	1169.11	292.50	63.75	10700.00											
			2.03	14.67	387.50	336.75	66889.76	44795.09	12688.60	786.49	229.40	44.45	9165.70											
		Dec	6.12	77.58	97.94	146.94	137304.42	47615.09	7214.23	727.67	166.90	43.38	6333.33											
			3.25	10.14	221.10	204.61	93518.90	42487.40	6721.49	722.66	180.56	46.76	6380.18											
		U3 (CGB)	Jan	2.50	77.24	115.02	244.29	192304.52	77676.71	13653.83	1361.76	344.21	99.74	6866.67										
				1.59	10.26	113.75	236.52	81604.98	41022.69	8592.72	669.20	202.34	85.65	4927.47										
U2 (GCP)	Feb	7.54	61.40	13.20	37.48	144436.48	21729.60	2411.13	255.10	65.65	17.83	6100.00												
		3.60	17.87	10.43	30.98	100716.29	17502.46	2283.82	190.15	47.91	13.75	4782.26												
Continuous measurements (9:00–12:00)	Dec	4.58	89.83	29.02	30.62	not measured																		
		3.45	8.64	18.00	18.41																			
	U1 (CGB)	Jan	0.18	90.93	44.85	46.55	not measured																	
			2.36	8.20	29.68	30.10																		
	U2 (GCP)	Feb	7.98	66.60	11.52	12.70																		
			2.47	14.64	15.79	16.14																		
	P values of ANOVA	Dec	4.15	87.66	22.78	24.30																		
			3.11	9.48	12.32	12.76																		
		Contrast	T	RH	PM2.5	PM10	0.3 µm	0.5 µm	1.0 µm	2.5 µm	5.0 µm	10.0 µm												
				0.3485	0.5512	0.4694	0.7166	0.6960	0.8591	0.8989	0.9812	0.7134	0.7814											
		Dec	U1-U3	0.6004	0.8409	0.7187	0.7804	1.0000	0.9851	0.8966	0.9293	0.9342	0.9986											
				0.0497*	0.8775	0.9053	0.9917	0.6958	0.9335	1.0000	0.9053	0.9917	0.7608											
Dec-Feb	U1-U3	0.6916	0.0026*	0.0168*	0.0289*	0.2952	0.0494*	0.0064*	0.9432	0.1038	0.7564													

HPC = handheld particle counter; MAS = microbial air sampler; U1, U2, and U3 are names of mounted climate monitoring devices and also refer the calf rearing units; 7GCP = group calf pen for 7 calves (U2); GCH = group calf hutch (U1); CGB = conventional group barn (U3); T = temperature, RH = relative humidity, WS = wind speed; PM2.5 and PM10 = 2.5 and 10 µm particulate matter; 0.3–10.0 µm pcs/m<sup>3</sup> are number of 0.3–10.0 µm particles by sizes in 1 m<sup>3</sup> air; CFU = colony forming unit. \* Significant result (P < 0.05)

Table 5. Logistic models considering previously reported limits of hazardous levels to individual microclimatic factors to compare microclimatic properties of units U1, U2, and U3, with U2 appointed as basis.

Season	Parameter	Limit	Site	<i>P</i> value	Probability (LCL–UCL)	LED/Total days (proportion)
Autumn (U1, U2, U3)	PM10 μg/m <sup>3</sup>	50	U2		0.04 (0.03–0.04)	1/91 (1.10%)
		50	U1	0.0007	0.05 (0.05–0.06)	4/91 (4.40%)
		50	U3	0.0013	0.02 (0.01–0.03)	1/80 (1.25%)
		45	U2		0.05 (0.04–0.06)	4/91 (4.40%)
		45	U1	0.0001	0.07 (0.06–0.09)	5/91 (5.49%)
	PM2.5 μg/m <sup>3</sup>	45	U3	< 0.0001	0.03 (0.02–0.03)	2/80 (2.50%)
		25	U2		0.16 (0.15–0.17)	18/91 (19.78%)
		25	U1	< 0.0001	0.24 (0.22–0.26)	21/91 (23.08%)
		25	U3	< 0.0001	0.11 (0.10–0.13)	6/80 (7.50%)
		15	U2		0.37 (0.35–0.38)	39/91 (42.86%)
	RH %	15	U1	< 0.0001	0.44 (0.42–0.46)	43/91 (47.25%)
		15	U3	0.0005	0.32 (0.30–0.34)	26/80 (32.50%)
		80	U2		0.55 (0.54–0.57)	36/91 (39.56%)
		80	U1	< 0.0001	0.61 (0.59–0.63)	42/91 (46.15%)
		80	U3	< 0.0001	0.001 (0.000–0.004)	0/80 (0.00%)
	WS m/s	85	U2		0.45 (0.43–0.46)	29/91 (31.87%)
		85	U1	< 0.0001	0.54 (0.52–0.56)	36/91 (39.56%)
		85	U3	Not estimable	Not estimable	0/80 (0.00%)
0.2		U2		0.85 (0.84–0.86)	90/90 (100%)	
0.2		U1	< 0.0001	0.76 (0.74–0.78)	84/90 (93.33%)	
Autumn and winter (U1, U2)	PM10 μg/m <sup>3</sup>	0.2	U3	< 0.0001	0.05 (0.04–0.06)	3/89 (3.37%)
		0.6	U2		0.57 (0.56–0.59)	66/90 (73.33%)
		0.6	U1	< 0.0001	0.49 (0.47–0.51)	40/90 (44.44%)
		0.6	U3	< 0.0001	0.01 (0.00–0.01)	0/89 (0.00%)
		45	U2		0.13 (0.12–0.13)	18/175 (10.29%)
	PM2.5 μg/m <sup>3</sup>	45	U1	< 0.0001	0.17 (0.15–0.18)	22/172 (12.79%)
		50	U2		0.09 (0.09–0.10)	12/175 (6.86%)
		50	U1	< 0.0001	0.12 (0.11–0.13)	18/172 (10.47%)
		15	U2		0.50 (0.49–0.51)	98/175 (56.00%)
		15	U1	0.0001	0.54 (0.52–0.55)	100/172 (58.14%)
	RH %	25	U2		0.32 (0.31–0.33)	61/175 (34.86%)
		25	U1	< 0.0001	0.37 (0.36–0.39)	67/172 (38.95%)
		80	U2		0.63 (0.62–0.64)	91/175 (52.00%)
		80	U1	< 0.0001	0.68 (0.66–0.69)	104/172 (60.47%)
		85	U2		0.52 (0.51–0.54)	75/175 (42.86%)
	WS m/s	85	U1	< 0.0001	0.60 (0.58–0.61)	86/172 (50.00%)
		0.2	U2		0.86 (0.85–0.87)	167/172 (97.09%)
		0.2	U1	< 0.0001	0.78 (0.76–0.79)	160/171 (93.57%)
0.6		U2		0.56 (0.55–0.57)	116/172 (67.44%)	
0.6		U1	< 0.0001	0.50 (0.48–0.51)	82/171 (47.95%)	

LCL = lower confidence level, UCL = upper confidence level, LED = limit exceeding days; U1, U2, and U3 are names of mounted climate monitoring devices and also refer to the calf rearing units; T = temperature, RH = relative humidity, WS = wind speed; PM2.5 and PM10 = 2.5 and 10 μm particulate matter.

The *P* values show significant differences ( $P < 0.05$ ). Probability (with 95% confidence interval) shows the proportion of measured data above the limit.

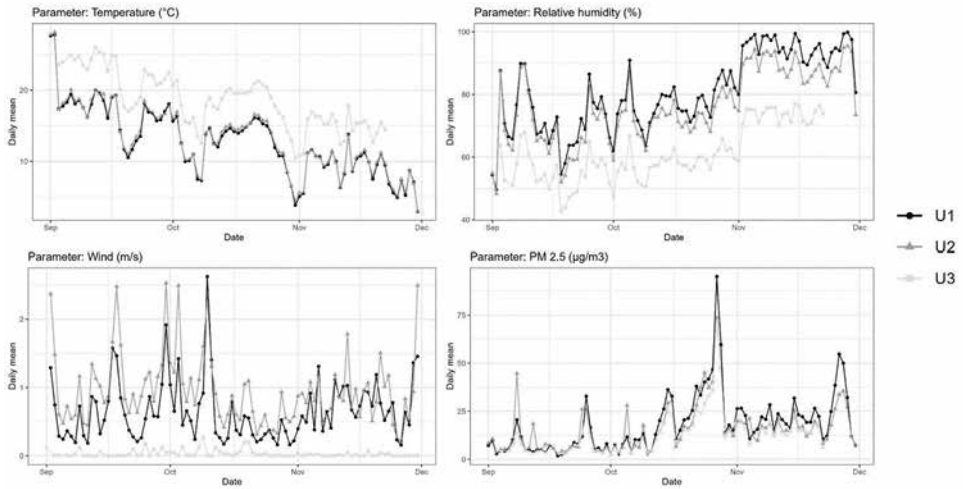


Fig. 1. Daily means of temperature, relative humidity, windspeed and PM2.5 particulate matter in U1 (group calf hutches), U2 (group calf pen for 7 calves) and U3 (conventional group barn) units from September to November 2019.

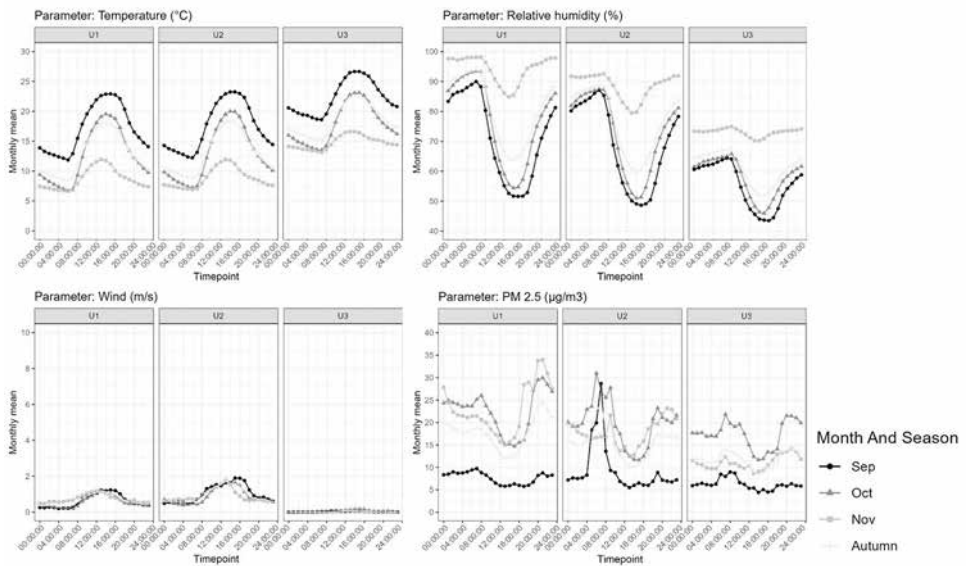


Fig. 2. Monthly means by hours of temperature, relative humidity, windspeed and PM2.5 particulate matter in U1 (group calf hutches), U2 (group calf pen for 7 calves) and U3 (conventional group barn) units from September to November 2019.



Table 6. Number and proportion of days and hours when daily and hourly mean values of different harmful gas concentrations in U3 exceeded recommended limits during summer and autumn 2019.

Indicator	Limit	Season	LED/Total days (proportion)	LEH/Total hours (proportion)
NH <sub>3</sub> ppb	4 000	Summer	0/48 (0%)	0/1144 (0%)
		Autumn	0/80 (0%)	11/1910 (0.58%)
	10 000	Summer	0/48 (0%)	0/1144 (0%)
		Autumn	0/80 (0%)	0/1910 (0%)
CO <sub>2</sub> ppm	2 000	Summer	0/41 (0%)	77/9760 (7.89%)
		Autumn	10/80 (12.5%)	294/1910 (15.39%)
	3 000	Summer	0/41 (0%)	9/9760 (0.92%)
		Autumn	3/80 (3.75%)	134/1910 (7.02%)
H <sub>2</sub> S ppb	500	Summer	0/48 (0%)	0/1144 (0%)
		Autumn	0/80 (0%)	0/1910 (0%)
CO ppb	10 000	Summer	0/48 (0%)	0/1144 (0%)
		Autumn	0/80 (0%)	0/1910 (0%)

LED = limit exceeding days, LEH = limit exceeding hours; U3 is the name of mounted climate monitoring device and also refers to the conventional group barn (CGB); NH<sub>3</sub> = ammonia; CO<sub>2</sub> = carbon dioxide; H<sub>2</sub>S = hydrogen sulphide; CO = carbon monoxide.

as temperature rose during the day, wind speed was also higher during the warmer period of the day. Wind speed was higher than average during forenoon and afternoon in the small group units, peak values were measured right after noon, and lower in the evening and night (Fig. 2). In CGB we observed the same pattern with lower values and intraday fluctuations. Based on our results, the U1 and U2 units were draughty (depending on the limit) more than half and three quarters of the days, whereas barn U3 was almost windless with practically stagnant air.

#### Aerosols (particulate matter and aerial germ load)

The monthly average PM values increased during autumn. Summer PM concentrations in U3 were generally low, and rose during autumn. The three units differed significantly in terms of the PM concentrations. Unit U1 was significantly dustier than U2 and U3, but U2 was also dustier than U3. In case of PM10, values were rarely higher than the 50 or 45 µg/m<sup>3</sup> limits. At the same time, the 25 µg/m<sup>3</sup> limit of PM2.5 particles was more often exceeded, in case of U1 we experienced this in nearly a quarter of all measurements, while at 15 µg/m<sup>3</sup> almost half of the measured data fell above the limit (Table 5). Winter values (except February) were high and fairly similar in U1 and U2 (Table 3), however, comparing U1 and U2 based on the autumn-winter period, U2 was also the dustier, and depending on the limit > 30 and > 50% of the measured data were above the PM2.5 limit at both locations (Table 5). In the two small group units, the month and the location:month interaction ( $P < 0.0001$ ) both influenced the frequency of high dust concentrations in both fractions.

Daily mean PM2.5 values fluctuated at all locations (Fig. 1). Concentrations of PM10 were usually only slightly greater than PM2.5 (a few µg/m<sup>3</sup> difference). Analyzing the pattern of monthly averages by hours, PM2.5 concentration profiles are different by location. In all places early afternoons were the least dusty periods in each month. Higher PM2.5 concentration could be observed in the morning and late afternoon/evening at all three locations, although this pattern was not typical for every month (Fig. 2).

On the contrary, monthly mean PM10 concentration measured by HPC was clearly greater than PM2.5 concentration, and both fractions were higher in all rearing units compared to forenoon mean values of mounted devices (Table 4). Comparing the locations based on HPC-data from December (when HPC-measurements were carried out at all three locations), no differences were present between the rearing units, except for the U2-U3 contrast, where only the T differed significantly. However, a relatively low number of datapoints were available. On the other hand, comparison of U1 and U3 based on data from December to February, several significant results were found: In U3 the RH, PM2.5 and PM10 concentrations, as well as 0.3 and 1.0 particle numbers were significantly lower than in U1. Monthly means of aerial germ concentrations (Table 4), were higher in U1 and in U2 compared to U3, however, comparing all three locations were not feasible due to limited number of datapoints in December. The U1 and U3 locations were compared by ANOVA based on December and January datapoints, however, the difference was non-significant ( $P = 0.0706$ ).

### Harmful gases

The monthly average values of  $\text{CO}_2$ ,  $\text{H}_2\text{S}$ , and  $\text{NH}_3$  were generally low (Table 3), however, daily means of these gases and  $\text{CH}_4$  fluctuated (Fig. 3). Analyzing the pattern of monthly averages by hours, a slight increase in concentrations of  $\text{H}_2\text{S}$ ,  $\text{NH}_3$ , and  $\text{CH}_4$  can be seen from morning to evening (Fig. 4). The  $\text{H}_2\text{S}$  and CO concentrations never exceeded limits, and hourly means of  $\text{NH}_3$  concentrations rarely rose above 4 ppm. However,  $\text{CO}_2$  concentrations were elevated in some parts of the measured days and hours, especially in autumn (Table 6).

### Correlation between microclimatic factors

Correlations between individual factors measured by mounted devices were usually significant, probably because of large number of data ( $P < 0.0001$ , Tables 7 and 8), however, the strength of correlations was variable according to the classification by Evans (1996, as cited in Papageorgiou 2022). The T and RH showed strong negative correlation in all units, while T and  $\text{CH}_4$  showed strong positive correlation in U3. Correlations were moderate

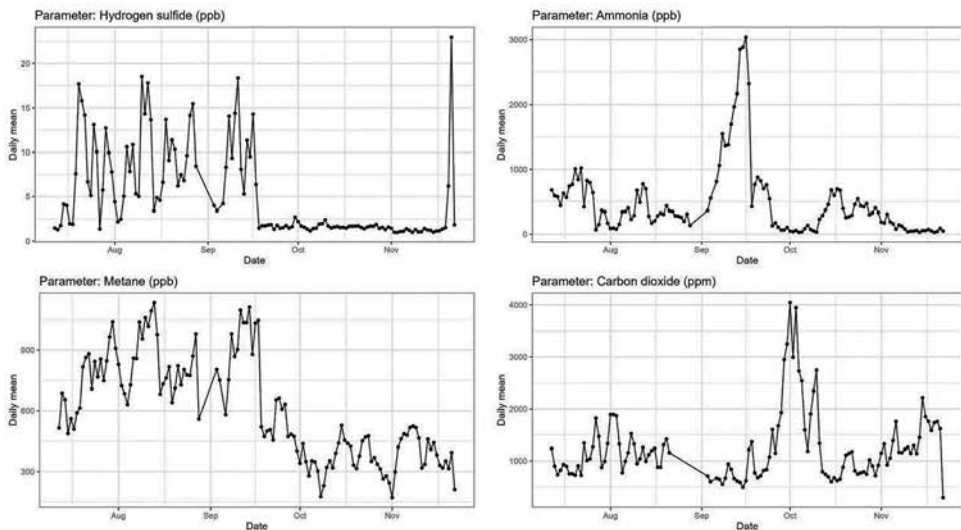


Fig. 3. Daily mean concentrations of hydrogen sulphide ( $\text{H}_2\text{S}$ ), ammonia ( $\text{NH}_3$ ), methane ( $\text{CH}_4$ ) and carbon dioxide ( $\text{CO}_2$ ) in U3 unit (CGB, conventional group barn) from July to November 2019.

among some gas parameters, and between T and PM particles, while weak correlations were observable between RH or T and some gas parameters. PM10 and PM2.5 concentrations were strongly correlated, which was the same when measured by HPC. The PM particles and RH correlated medium to strong depending on mounted and HPC device (Table 7).

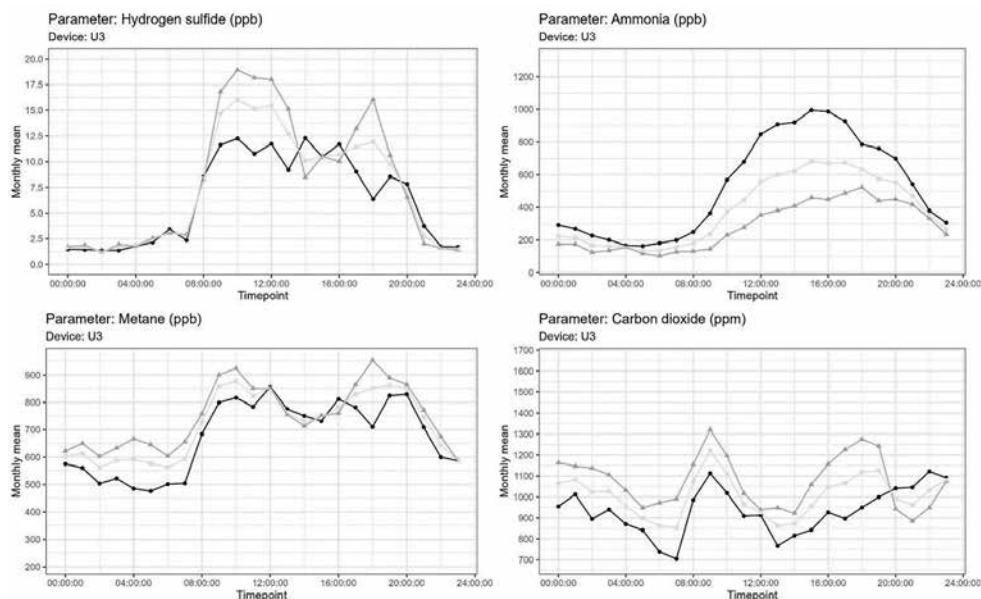


Fig. 4. Monthly mean concentrations by hours of hydrogen sulphide ( $\text{H}_2\text{S}$ ), ammonia ( $\text{NH}_3$ ), methane ( $\text{CH}_4$ ) and carbon dioxide ( $\text{CO}_2$ ) in U3 unit (CGB, conventional group barn) from July to August 2019.

Table 7. Pearson correlation coefficients ( $r$ ) between microclimatic factors measured by mounted and handheld devices in U1, U2 and U3 units.

Variable 1	Variable 2	Mounted device continuous measurement			Handheld device point measurement
		U1 $r$	U2 $r$	U3 $r$	All units $r$
PM10	PM2.5	0.99*	1.00*	1.00*	0.96**
RH	PM2.5	0.41*	0.30*	0.29*	0.65**
RH	PM10	0.40*	0.31*	0.29*	0.67**
WS	T	0.15*	0.21*	0.17*	N.m.
WS	PM10	-0.30*	-0.21*	-0.09*	N.m.
WS	PM2.5	-0.31*	-0.21*	-0.09*	N.m.
WS	RH	-0.34*	-0.38*	-0.18*	N.m.
T	PM10	-0.40*	-0.30*	-0.31*	-0.29
T	PM2.5	-0.40*	-0.30*	-0.31*	-0.21
T	RH	-0.62*	-0.63*	-0.67*	-0.31

U1, U2, and U3 are names of mounted climate monitoring devices and also refer the calf rearing units; T = temperature, RH = relative humidity, WS = wind speed; PM2.5 and PM10 = 2.5 and 10  $\mu\text{m}$  particulate matter, N.m. = not measured.

Asterisks mark significant correlations ( $*P < 0.0001$ ,  $**P < 0.01$ ). Correlation strengths can be interpreted according to Evans (1996, as cited in Papageorgiou 2022) as follows: 0.00–0.19 = very weak; 0.20–0.39 = weak; 0.40–0.59 = moderate; 0.60–0.79 = strong; 0.80–1.0 = very strong.

Table 8. Pearson correlation coefficients (r) between microclimatic factors measured by mounted device in unit U3.

Variables	CO <sub>2</sub>	NH <sub>3</sub>	H <sub>2</sub> S	CH <sub>4</sub>	CO
T	-0.07*	0.33*	0.36*	0.62*	0.08*
RH	0.02*	-0.30*	-0.20*	-0.20*	0.08*
WS	0.13*	-0.08*	-0.01	-0.10*	-0.08*
PM10	-0.09*	-0.11*	-0.06*	-0.18*	0.61*
PM2.5	-0.09*	-0.11*	-0.06*	-0.18*	0.61*
CO	-0.11*	0.15*	0.18*	0.25*	
CH <sub>4</sub>	-0.16*	0.54*	0.47*		
H <sub>2</sub> S	-0.07*	0.32*			
NH <sub>3</sub>	-0.37*				

U3 is the name of mounted climate monitoring device and also refers to the conventional group barn; T = temperature, RH = relative humidity, WS = wind speed; PM2.5 and PM10 = 2.5 and 10 µm particulate matter; CO<sub>2</sub> = carbon dioxide; NH<sub>3</sub> = ammonia; H<sub>2</sub>S = hydrogen sulphide; CH<sub>4</sub> = methane; CO = carbon monoxide.

\* Significant correlation ( $P < 0.0001$ ). Correlation strengths can be interpreted according to Evans (1996, as cited in Papageorgiou 2022) as follows: 0.00–0.19 = very weak; 0.20–0.39 = weak; 0.40–0.59 = moderate; 0.60–0.79 = strong; 0.80–1.0 = very strong.

## Discussion

In naturally ventilated barns, inside climatic conditions correlate with the outside temperature and a seasonal pattern can also be observed (Seedorf et al. 1998). High stocking density and shared airspace with older animals, RH > 75%, poor air quality (toxic gases, dust, high germ load), the type of bedding and cleanliness of the equipment have to be considered as risk factors in terms of respiratory problems (Lago et al. 2006; Woolums et al. 2007). Others claim that there is also no correlation between PM concentrations and stocking density in calf stables (Islam et al. 2020). Twenty-four-hour measurements in Belgian herds found no relationship between PM concentration and a number of environmental variables such as temperature, RH, air movement, aerial germ load, NH<sub>3</sub> and CO<sub>2</sub> (van Leenen et al. 2021). However, we found weak and moderate correlations of PM with T, RH, WS and strong correlation with CO.

### Temperature, relative humidity, and wind speed

Low temperature not alone but in combination with other adverse factors (humidity and wind) lead to increased morbidity and mortality. Optimal RH in calf barns and hutches should vary from 50 to 70–80%, while low RH (< 50%) can dry the epithelial surface of the respiratory tract, leading to infections (Malá and Novak 2021; Müller 2021). Other sources claim that problems start in naturally ventilated calf barns when the RH cannot be brought below 85% in cold and damp weather (Webster 1984; Lorenz et al. 2011), as low temperatures and high RH (< 10 °C and > 85% RH) impair the health of calves (Nonnecke et al. 2009). In U1 and U2 similar Ts were present, however, U1 had ca 2-5%pt higher RH compared to U2. Moreover, in both U1 and U2 units, RH levels exceeded the limit of 80% and 85% in half or even more of all measurements, whereas in U3 it happened only occasionally. According to our results, monthly average T below 10 °C and monthly average RH above 85% were typical in the small group units from November to January, which means that a calf born in autumn can spend the crucial stage of its life (including weaning and grouping) in risky conditions. Unit U1 was a more enclosed location and WS was also lower compared to U2 (explained below). There was a bigger difference compared to the very low velocity of U3. For this reason, we assume that

WS and the surrounding windbreaking landmarks may have played a role in the formation of the humidity of these units. From the point of view of BRDC, this can be important if the air with higher RH associated with low T cannot exchange due to low air movement, so favourable conditions for aerial germs can develop.

However, a draughty environment is not favourable either. Air movement faster than 0.3 m/s (Buczinski et al. 2018a) or 0.5 m/s (Lundborg et al. 2005) already constitutes a draught, while others claims that > 0.2 m/s velocity at low T, and > 0.6 m/s at high T should be avoided (Malá and Novak 2021). The draughty environment in our study may be explained by the location of the small group units. The U1 unit was less draughty but it had a roof extending over the runway and the measuring device and it was surrounded by nearby vegetation (bushes, trees). In contrast, U2 was located in an open field from three sides (a barn bordered the northern site of the hutch). As we found significant differences between units, placement of the hutches and surrounding landmarks is decisive in mitigation of draught. Tissue lesions detectable by ultrasound may be present in a draughty environment (Buczinski et al. 2018a). We found no correlation between windspeed and T, and between windspeed and RH, and it was the same in the study of van Leenen et al. (2020), as well. However, they assumed that presumably the draught caused first airway inflammation which developed further into pneumonia as the prevalence of lesions  $\geq 1$  cm was 81.8% in the draughty and 54.2% in the warm, dry, and NH<sub>3</sub>-rich environment, compared to 31.6% under normal environmental conditions (van Leenen et al. 2020).

#### Aerosols (particulate matter and aerial germ load)

Average PM concentrations in U1, U2, and U3 units were in accordance with other studies in dairy environments, as we reviewed earlier (Sáfár et al. 2023). In humans the 24-hour average airborne dust concentration cannot exceed 25  $\mu\text{g}/\text{m}^3$  for PM<sub>2.5</sub> and < 50  $\mu\text{g}/\text{m}^3$  for PM<sub>10</sub> taking into account the old WHO standard, while the new standard specifies < 15  $\mu\text{g}/\text{m}^3$  for PM<sub>2.5</sub> and < 45  $\mu\text{g}/\text{m}^3$  for PM<sub>10</sub> (WHO 2005, 2021). The daily and hourly mean PM<sub>10</sub> concentrations rarely exceeded this limit. In contrast, the PM<sub>2.5</sub> concentration was above the 15  $\mu\text{g}/\text{m}^3$  limit in about third to half of the time. Conventional group barn seemed more favourable from this point of view. In a Polish study carried out in a dairy barn in the winter period of 2019/20 PM<sub>2.5</sub> and PM<sub>10</sub> concentrations exceeded the limits also in a significant number of days (Nieckarz et al. 2023). Some authors report higher concentrations of aerosols in winter (Tan and Zhang 2004), and we got the same results in U1 and U2 units. Although there are no recommendations in case of respiratory diseases in animals, PM<sub>2.5</sub> and smaller particle fractions accumulate in the lung parenchyma, which can lead to serious respiratory and systemic diseases (Losacco and Perillo 2018). The source of PM particles can be the bedding or the calf itself, but it can also be influenced by air movement, litter management, animal activity and stocking density. We also observed a diurnal PM profile with higher concentration in the morning and late afternoon/evening, and lowest values in early afternoons. A diurnal profile was also observed by Joo et al. (2013) in naturally ventilated dairy barns: PM<sub>2.5</sub> and PM<sub>10</sub> concentration also peaked late in the evening, while they were lowest from night to morning. Peak values are thought to be related to bedding or feeding, as they are the most significant sources of PM, but can also be influenced by ventilation and animal movement (van Leenen et al. 2021). Higher PM concentrations can be measured during the day than at night, according to animal activity (Tan and Zhang 2004), and as the activity of cattle increases in the evening, dust formation may be reduced by the timing of feeding so that rumination is shifted to the period of increased activity (Urso et al. 2021). However, others claims that increased activity of animals had no significant effect, albeit lower morning peaks were observed in relation to feeding (Joo et al. 2013). Morning feeding can be an explanation for the observed peak values in the U2 unit in the early morning. In the

case of U3, the same is less likely, although the feeding area was located outside, away from the measuring unit. On the other hand, in U1 no calves were kept during the autumn period, but morning and afternoon peaks were characteristic at this unit as well. In our study, concentrations measured by HPC were higher compared to mounted devices, and PM10 concentration was about 1.5–2 times higher than PM2.5. Particles larger than 20  $\mu\text{m}$  settle approximately at 1.0 m/s, while particles of 5  $\mu\text{m}$  settle at 0.1 m/s (Pearson and Sharples 1995). The number of particles equal or less than 2.5  $\mu\text{m}$  measured by HPC was orders of magnitude larger compared to those of 5.0 and 10.0  $\mu\text{m}$ . This can explain that differences between PM2.5 and PM10 concentrations were larger measured by HPC, compared to mounted devices. As HPC was used at about 1.0 m height, and bedding was probably stirred by calves or our movements when we entered inside the barn or hutches, therefore larger PM particles probably had not yet settled. In contrast, mounted devices were placed higher and further from the HPC measurement points, and recorded data at 5- or 10-min interval allowing time for sedimentation of larger particles, while smaller ones remained suspended in air for longer time. From this we conclude that data from mounted devices can better represent the real characteristics of the buildings and results of HPCs can be misleading.

We found that PM emission is negatively correlated with ambient T, while positively correlated with RH, and it was claimed by Nieckarz et al. (2023) too, however, others found correlation in the opposite direction (Joo et al. 2013). Presumably, the role of surrounding, non-animal PM sources are also not negligible, and less windy environment (lower clearance by wind) may also explain it. We experienced at all locations that early afternoons were the least dusty periods in every month and at the same time, the windiest periods were also early afternoons. Moreover, February was the windiest but less dusty month in winter in U1 and U2 as well, while January was the dustiest and least windy. It is likely that in open rearing units the wind did not cause an increase in PM2.5 concentrations but on the contrary, it cleared the air from dust in the windiest early afternoon period.

Literature sources on aerial germ load in calf barns have shown large differences depending on the season and location (Sáfár et al. 2023). Aerial germ concentrations in U1, U2, and U3 units were relatively low, however, standard deviations were high and outliers also occurred and data varied sometimes even within the same unit and day. Monthly mean values are in accordance with data that outdoor air contains airborne germs of usually up to 20 000 CFU/m<sup>3</sup>, whereas in well-ventilated barns, 5 000 to 30 000 CFU/m<sup>3</sup> aerial germ loads are possible (Nordlund 2008). Sampling and evaluation techniques were basically developed to monitor clean rooms, however, bacteria outgrow the plate even under very small volume of air samples in barn conditions (Nordlund 2008), making it difficult or impossible to count the colonies. For that reason, dilution series (Islam et al. 2019) or small volume of air samples can be used, then data can be extrapolated for 1 m<sup>3</sup> but values calculated from these data may be hard to evaluate. Moreover, the stirring of bedding and manure when stepping inside the area or any movement of the calves is also likely impairing the results, as bacteria in the air come mainly from the animal's body surface, manure, and bedding (Webster 1984).

### Harmful gases

Daily dynamics of the concentrations of harmful gases can be plotted based on continuous measurements, and related technological processes can be identified (Zou et al. 2020). The U3 gas concentrations rarely exceeded the relevant limit values. CH<sub>4</sub> and CO<sub>2</sub> in dairy barns predominantly originate from cows through digestion and respiration (Jungbluth et al. 2001). CO<sub>2</sub> is suitable for monitoring the ventilation of barns: taking into account the 3000 ppm CO<sub>2</sub> limit allowed in livestock buildings (EFSA 2009), the traditional barn was poorly ventilated for part of the study period, especially in autumn, as was expected based on the low wind speed.

The concentrations of H<sub>2</sub>S and NH<sub>3</sub>, as the most significant barn gases were generally low, never reaching 0.5 ppm (500 ppb, EFSA 2009) and 20 ppm (20 000 ppb, EFSA 2009), respectively. Although no clear relationships between concentration vs. detrimental effect have been described (Seedorf and Hartung 1999; Kaufman et al. 2015), a negative effect of NH<sub>3</sub> could be significant even at low concentrations. In calves, lung lesions of  $\geq 1$  cm were detected by ultrasound at  $> 4$  ppm NH<sub>3</sub> concentrations; at increasing NH<sub>3</sub> concentration, bronchoalveolar lavage samples showed elevated nucleated cell counts and increased epithelial cell ratios (van Leenen et al. 2020). The daily mean concentration of NH<sub>3</sub> never exceeded this limit, while hourly means only rarely, which coincides with other studies in dairy environments, as reviewed earlier (Sáfár et al. 2023).

According to some research, there is no correlation between T and NH<sub>3</sub> or between RH and NH<sub>3</sub> concentration of cattle barns (Seedorf and Hartung 1999; Kaufman et al. 2015). Others claim that the NH<sub>3</sub> concentration is temperature-dependent (Jungbluth et al. 2001; Ngwabie et al. 2011), showing a diurnal pattern and being the highest in the afternoon (Teye et al. 2008), which can be explained by increasing urease activity of the manure at higher T (Zou et al. 2020). Low NH<sub>3</sub> concentration at lower T and higher RH may be due to the dissolution of NH<sub>3</sub> in water (Saha et al. 2014; Zou et al. 2020). In accordance with Zou et al. (2020), we found only a weak correlation with T and RH, and observed the diurnal changes in H<sub>2</sub>S and NH<sub>3</sub> concentrations, which were higher in the afternoon, especially in summer. The drastic decline in the NH<sub>3</sub> concentration in early September may be related to manure removal and replacement by fresh straw at that time. Low values in autumn can be explained by declining T and elevating RH, while smaller peaks and declines may be attributed to refreshed bedding which highlights the importance of litter management.

Methane is important as greenhouse gas, but has no limit value for animals and humans (EFSA 2009). Quantity of CH<sub>4</sub> produced in manure should increase by temperature (Ngwabie et al. 2011). We found a strong positive correlation between CH<sub>4</sub> and T. The CH<sub>4</sub> concentration was higher in the summer periods of the study, whereas in early September there was a sudden decrease in CH<sub>4</sub> concentration after deep litter was removed. On the contrary, in a dairy cow barn, where slurry was regularly removed by scrapers, negative correlation was found between daily CH<sub>4</sub> emissions and indoor T, as well as positive relation between CH<sub>4</sub> emissions and cow activity on both diurnal and daily basis. It was explained by increasing T having the effect of decreasing animal activity which, in turn, decreased the quantity of CH<sub>4</sub> produced (Ngwabie et al. 2011). In our study, the CH<sub>4</sub> concentration also showed diurnal changes in summer period and peak values were observable around 10 a.m. and 6 p.m., which was likely connected to feeding and animal activity too, as reported by others (Ngwabie et al. 2011). However, the diurnal pattern was less evident in autumn, when CH<sub>4</sub> concentration was lower as well. Our conclusion is that besides temperature, manure management is also an important factor in CH<sub>4</sub> production in naturally ventilated barns.

#### Practical experience in the operation of a real-time monitoring system

Summarizing our long-term experiences, we suggest the following for real-time monitoring systems for farm use:

- 1) Measurements should represent all phases of the farm work.
- 2) Measuring parts of devices should be placed at the animals' height and preferably surrounded by a large-holed mesh wire box to avoid damage by calves or husbandry processes (e.g. machine bedding), but still allow air flow around the measurement unit.
- 3) Cables of measuring devices should be covered with flexible metal tubes to avoid damage caused by calves.
- 4) Ongoing technical support and software indicating errors immediately (e.g. loss of internet connection or power supply, validity of data is doubtful) is necessary.

- 5) Farm management personnel (i.e. shift managers, electrician) should know how the system works and have basic troubleshooting skills.
- 6) Validation period or preliminary testing is useful before commissioning.
- 7) Regular inspections are necessary (security camera and on-site inspections) to check the integrity and cleanliness of the devices (e.g. covered by manure or straw).
- 8) Collected data should be checked even when notifications about unfavourable conditions are not present. Change in data trends may have a significant role as well. If unlikely data are present, always think about false measurement and ask for technical support from developer.

### Benefits of real-time monitoring of microclimatic factors in farm conditions

Our system was suitable for real-time monitoring of microclimatic factors under farm conditions. We could identify long-term and intraday periods with limit-breaking values, daily fluctuations as well as specific patterns of individual indicators in different calf husbandry environments. Considering data and operating experiences, specific software can be developed, which could monitor the different calf rearing units in real time. As there is no or only weak correlation between individual microclimatic parameters, this kind of monitoring system probably will be not able to forecast critical PM or gas values. However, it could notify users in case of unfavourable conditions, and based on the results, contributing technological processes can be reviewed and the effect of adjusted processes can be monitored. For example, in case of elevated PM concentration, dust forming processes such as manure removal, straw and forage allocation or any other activities that promote animal movements could be timed at those periods of the day when the PM concentration is lower anyway, but relocation of mangers, use of less dusty bedding material or spreading technology may also be considered. Further investigations are needed into the use of real-time microclimatic monitoring as a predictor of clinical BRDC cases or pathological slaughterhouse findings.

On the other hand, replacing traditional barns with modern structures requires large investments. Therefore, we recommend to assess the prevailing microclimatic conditions of the given building and review the connecting management factors prior to building new calf rearing units. Low-risk microclimatic environment from the point of view of BRDC might also be ensured in a less modern barn with appropriate management measures, as we experienced in case of barn U3. Although we monitored only one (yet wide-spread) building type, these results can serve as a starting point for veterinarians and farmers who would like to assess the typical climatic microclimatic conditions on their own buildings.

In conclusion, real-time monitoring PLF systems may help in early recognition of adverse environmental conditions, and on its basis, preventive measures and mitigating steps can be enforced, or clinical diseases can be diagnosed in a timely manner. Finally, they can enable early intervention, thus reducing economic damage and drug consumption. The use of such systems can be useful especially on those farms where other predisposing factors such as incomplete biosecurity measures and poor health state are also present and any preventive step can be important in mitigation of the adverse effects of BRDC.

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