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# Animal hygienic assessment of air carbon dioxide concentration in semi-open freestall barns for dairy cows

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### Abstract

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The objective of this study was to determine the  $CO_2$  concentration in the different animal service zones in semi-open freestall barns for dairy cows. The study was conducted in 3 dairy cattle farms, with production buildings with different capacities – two farms for 120 and one farm for 500 cows. The following microclimate parameters were monitored: temperature (°C), relative humidity (RH, %), temperature-humidity index (THI) and speed of airflow (SAF). The average  $CO_2$  values in the three buildings surveyed ranged from 434.85 to 635.27 ppm, with a maximum deviation up to 2130 ppm. The highest  $CO_2$  concentrations were recorded in the building with the largest capacity – 500 cows. In the building with a capacity of 120 cows, a trend for the lowest  $CO_2$  levels and the least variation during the day was reported. In the summer the lowest  $CO_2$  values in the air above the stalls – 515.9 ppm were reported and in the winter the highest – 626.2 ppm. Highest  $CO_2$  values in the morning and afternoon were recorded in all investigated barns when the animal's locomotor activity was greatest associated with eating, drinking, and moving (other activities). The correlation between the temperature-humidity index (THI) values and the  $CO_2$  values was significant and negative (-0.23). The lowest  $CO_2$  concentrations were reported at THI values 68-72.

*Keywords:* carbon dioxide; dairy cows; freestall housing; welfare *Abbreviations:* THI – temperature-humidity index,  $CO_2$  – carbon dioxide, SAF – speed of air flow

## Introduction

According to EFSA (2009) 18% of greenhouse gas emissions in the EU are due to ruminant livestock. One of these gases is carbon dioxide. Its high concentration in production buildings has a negative effect on both livestock and livestock breeders. Long-term exposure of animals to a concentration of  $CO_2$  above 1% in the air may cause chronic intoxication, lower productivity and disease resistance (Vtoryi et al., 2017). In the dairy cows' premises optimal microclimatic conditions must be provided as they have a great impact on their health, well-being and productivity, and thus on milk production efficiency. In this regard, microclimate and ventilation are important factors, which determine the air quality in the farm production buildings (Mijid, 2013). Poor ventilation can increase the relative humidity and the concentration of harmful gases such as carbon dioxide and ammonia. The concentration of carbon dioxide depends to a large extent on the type of building, the ventilation system and the density of the animals (Jovović et al., 2015). The carbon dioxide sources in cow buildings are: animal breathing, fodder emissions, manure, and a small amount of technological energy (fuel, electricity) (Schiefler, 2013).

In Bulgaria during the last 10–15 years many technological approaches were introduced referring to dairy cattle breeding – new cattle farms with larger capacity, freestall production system in semi-open barns with natural ventilation, etc. (Slavov et al., 2013). Breeding a large number of high yielding cows under one roof is a challenge for each production system to provide an optimal microclimate and especially with regard to harmful (NH<sub>3</sub>, H<sub>2</sub>S) and greenhouse (NH<sub>4</sub>, CO<sub>2</sub>) gases. Of all these gases, the CO<sub>2</sub> content in barns for dairy cows was least studied. All of this gave us basis to conduct the present study, which purpose was to determine the carbon dioxide concentration in the air of semi-open freestall barns for dairy cows and the factors influencing its variation.

# **Material and Methods**

#### Study area

The study was conducted for a one-year period (July 2014 – June 2015) in three dairy farms for Holstein cows from different districts of Central Southern Bulgaria – Stara Zagora, Haskovo and Plovdiv. Farms were labeled with numbers 1, 2 and 3. The areas of the studied farms fall under one climatic zone with transitional continental climate. The average milk yield per cow in all farms was in the range of 8000 to 9000 kg milk for 305-day lactation. The feeding was *at libitum* with total mixed ration. In all three farms the cows were housed in semi-open freestall barns with height 3.50 m at the roof overhangs (eaves). The buildings in farms differed in some technical indicators as follows:

**Farm 1:** the building, object of study was with capacity of 120 cows (60.00/18.00 m) with 43.4 m<sup>3</sup> volume per cow provided. There were two 4 m wide manure alleys in the building, located between the stall rows and one feeding alley also 4 m wide, situated at the longitudinal open side of the building. The building was equipped with 5 fans (60000 L/h air) above the manure alleys which were automatically switched on at temperatures above 28°C. The building was also equipped with curtain sidewall opening that lifts automatically at speed of airflow over 4 m/s, in rain and snow, and when the air temperature drops below 2°C. The feeding was with a feed trailer (mixer) twice at 7.00 and 16.00 h. Cows were milked twice at 7.00 and 19.00 h in a milking parlor. The manure cleaning was with a delta scraper, which moves automatically at an interval of 90 min.

**Farm 2:** the building, object of study was with capacity of 120 cows (66.00/18.00 m), with 43.98 m<sup>3</sup> volume per

cow provided. There were two manure alleys in the building, located between the stall rows and one feeding alley, situated at the longitudinal side of the building (as in the building of Farm 1). There were 8 fans (17000 L/h air) in the building above the manure alleys which were manually switched on at the discretion of the workers. The building was equipped with a polyethylene curtain, which was lowered manually in adverse weather conditions. Feeding was with mobile mixer twice at 8.00 and 16.00 h. Cows were milked three times at 5.00, 12.00 and 19.00 h in a milking parlor. The manure cleaning was with a delta scraper, which moves automatically at an interval of 60 min.

**Farm 3:** the building, object of study was with capacity of 500 cows (90.00/45.00 m), with 58.7 m<sup>3</sup> volume per cow provided. There were 5 manure alleys in the building, located between the stall rows and two feeding alleys – one located centrally of the building and the other at the outer open longitudinal wall. During the winter period, in adverse weather in front of feeding alley a polyethylene nets were descended. The building was equipped with 15 fans (17000 L/h air) above the freestalls, which were manually switched on at the discretion of the workers. The feeding was with a feed trailer (mixer) twice at 8.00 and 16.00 h. Cows were milked three times at 5.00, 12.00 and 19.00 h in a milking parlor. The manure cleaning was with a delta scraper, which moves automatically at an interval of 20 min.

#### Monitoring points

The farms were visited two times every month for the period from July 2014 till July 2015. At each visit all indicators were measured five times per day, respectively, at 10.00; 12.00; 14.00; 16.00 and 18.00.

At all visits temperature, relative humidity, speed of airflow and level of carbon dioxide were recorded. All indicators were reported both outside the premises at a distance of 10 m from the building and inside the premises in the three service areas – over stalls and manure and feed alleys.

The measuring of the data from the measuring devices was in the living area of the cows at a height of 1 m from the floor. The indicators were measured as follows: temperature, relative humidity and carbon dioxide level – with Lutron MCH-383SDB and the speed of airflow with Lutron EM-9300SD.

Temperature-humidity index (THI) was calculated by the formula proposed by Kibler (1964), quoted by Bouraoui et al. (2002) and Gantner et al. (2010):

THI = 1.8 x Ta - (1-RH) x (Ta - 14.3) + 32,

where: Ta is the air temperature,  $^{\circ}\mathrm{C};\,\mathrm{RH}$  – the relative humidity, %.

#### Statistical analysis

To obtain a better approximation for the analysis of the variance, temperature-humidity index (THI) and the speed of airflow (SAF) values were divided into the following classes:

for THI: <50; from  $\ge50$  to  $\le58$ ; from  $\ge58$  to  $\le68$ ; from  $\ge68$  to  $\le72$ ; from  $\ge72$  to  $\le74$  and >74;

for SAF:  $\leq 0.1$  m/s; from 0.2 m/s to 0.5 m/s; from 0.6 to 1.0 m/s and  $\geq 1.1$  m/s.

To assess the influence of the factors on  $CO_2$  values in the three service areas in the buildings, the following model was used:

 $Y_{ijkl} = \mu + F_i + S_j + H_k + e_{ijkl}$ 

where:  $Y_{ijkl}$  is the dependent variable (CO<sub>2</sub>);

 $\mu$  – the average for the model;

 $F_i$  – the effect of the farm;

 $\dot{S}_{i}$  – the effect of the season of reporting;

 $\dot{H}_{k}$  – the effect of the hour throughout the day of reporting;

 $e_{iikl}$  – effect of uncontrolled factors (the error).

To assess the impact of microclimate parameters on  $CO_2$  values in the buildings, the following model was used:

 $Y_{ijkl} = \mu + TVI_i + Sj + e_{ijkl},$ 

where:  $Y_{iikl}$  is the dependent variable (CO<sub>2</sub>);

 $\mu$  – the average for the model;

 $TVI_i$  – the effect of the temperature-humidity index (in classes);

 $S_i$  – the effect of the speed of airflow (in classes);

 $e_{iikl}$  – the effect of uncontrolled factors (the error).

For the basic statistical processing of the data, the MS Excel package was used, and in order to obtain the statistical indicators and the analysis of the variance, the corresponding STATISTICA modules of StatSoft (Copyright 1990–1995 Microsoft Corp.) were used.

### **Results and Discussion**

For the areas of the farms, included in the study, the main climatic indicators were reported – temperature and humidity of the air (of which the temperature-humidity index was calculated), speed of airflow and carbon dioxide level (Table 1). From the presented average daily values of the three climatic indicators, no significant differences for the areas of the three farms were reported. All three farms were located in an area with a transitional continental climate. The average January temperature was from -1.5 to +1°C, the average in July was 22-24°C and the maximum summer temperatures reach 40°C (Alexandrov, 2006).

During the summer season in the regions of the three farms, that define conditions for heat stress in dairy cows were reported – THI from 74.43 to 76.69. There was a certain risk of such conditions also in the spring – an average daily THI values above 69. Areas of the three farms were not exposed to strong air current, relatively low SAF was measured 0.1 m/s for almost all seasons. Relatively higher SAF was found in winter – 0.40 m/s.

The average daily  $CO_2$  values in the areas of the three farms were within the limits of the average for atmospheric

Table 1. Average daily temperature-humidity index (THI), speed of airflow (SAF) and carbon dioxide (CO $_2$ ) value	es and
theirs deviations outside of the production buildings by seasons	

Farm	n	TI	THI SAF, m/s CO <sub>2</sub> , ppm		SAF, m/s		ppm
		x±SE	SD	x±SE	SD	x±SE	SD
			Sum	nmer	_	_	
Farm 1	25	76.60±0.60	2.93	$0.33 {\pm} 0.09$	0.40	363.60±15.16	67.80
Farm 2	25	74.43±0.96	4.78	$0.23 \pm 0.04$	0.19	408.68±21.43	100.54
Farm 3	25	76.69±0.60	2.70	$0.29{\pm}0.06$	0.28	408.20±25.24	112.86
			Aut	umn			
Farm 1	20	61.55±2.18	9.73	$0.26 \pm 0.04$	0.17	403.45±17.22	77.00
Farm 2	20	61.44±2.32	1.37	$0.21 \pm 0.02$	0.10	376.55±6.97	31.15
Farm 3	15	66.08±2.58	9.99	$0.26 \pm 0.09$	0.33	370.73±9.37	36.29
			Wii	nter			
Farm 1	15	47.95±1.38	5.34	$0.37{\pm}0.08$	0.30	443.47±21.29	82.46
Farm 2	15	50.40±0.95	2.66	$0.40{\pm}0.11$	0.44	374.67±26.25	101.68
Farm 3	15	51.40±1.29	4.99	$0.23 \pm 0.04$	0.17	412.47±13.51	52.33
Spring							
Farm 1	10	69.22±1.38	5.17	$0.28 \pm 0.09$	0.30	356.70±16.43	51.96
Farm 2	10	70.44±0.76	2.39	$0.10 \pm 0.00$	0.00	336.50±15.48	48.94
Farm 3	10	70.46±1.18	3.74	0.18±0.03	0.10	357.00±14.25	45.06

air. For all seasons, the daily average values were in the range of 336.50 to 443.47 ppm, as slightly higher values in winter were reported. Rustad (2017) pointed out that for 2013 the average global atmospheric carbon dioxide was over 400 ppm and 402.9 ppm in 2016.

Daily THI, SAF and  $CO_2$  values were reported in the three animal service areas in the buildings – above stalls, above manure alleys and above the feeding alleys at a height of 1 m from the floor. No statistically significant differences between the daily averages values of THI in the three service areas in the three farms were reported (Table 2). It should also be noted that there were no significant differences between the average daily values for THI inside the building and outside the building as by seasons so and by farms. The buildings on the three farms were of the same type – semi-open, as the open side was next to the feeding alley and only in bad weather was closed by curtains. This type of building does not provide isolation from the external climatic conditions, especially from high temperatures.

In the buildings of all three farms high THI values during the summer season in all animal service areas were reported – THI  $\geq$  73 (Table 2). THI values are usually distributed to classes which show different heat stress levels. Definitions of these levels vary with different indexes and authors. For example, Armstrong (1994) classified an index below 71 as a comfort zone, and values ranging from 72-79 as mild stress, 80-89 moderate stress, and values over 90 as severe stress. According to recent studies, Segnalini et al. (2013) offered an even lower threshold to assess the effect of THI on dairy cows, respectively 68 $\geq$ THI<72 (mild discomfort). From these classifications, it can be seen that during the summer, in the buildings of the three surveyed farms the reported THI values during the day, which were outside the dairy cows' comfort zone, were determined by different authors from a mild stress and uncomfortable conditions to a danger signal from heat stress (Segnalini et al., 2013).

The average SAF in the buildings in all three service areas varied within very narrow limits and was around 0.10 m/s. Higher values, up to 0.34 m/s, were reported in the summer when at high daily temperatures fans on all three farms were switched on. According to the Regulation No. 44 (2006) for the warm periods with air temperatures above 15°C, SAF is recommended to be from 0.5 to 1.0 m/s, and for the cold periods – 0.3 m/s. The three buildings had no mechanical ventilation, and even cooling fans to run in the summer, SAF was under the standards set by Regulation No. 44 (2006) during all seasons.

Jovović et al. (2015) also established a low average SAF in dairy cows' buildings 0.12 m/s, which was lower than the recommended by regulatory requirements in Bosnia and Herzegovina – about 0.2 m/s in winter and 0.6 m/s in the summer.

The reporting of the values of  $CO_2$  in this study was only during the light part of the day when a higher activity of the animals was reported. Van Ouverkerk and Pedersen (1994) emphasized that animal activity was an important factor that must be taken into account when calculating the  $CO_2$  balance for periods shorter than 24 hours. The studies of Wachenfelt et al. (2001), showed that the production of  $CO_2$  during the

Farm	n	THI			SAF			
		Stalls	Manure alley	Feeding alley	Stalls	Manure alley	Feeding alley	
Summer								
Farm1	120	73.79±0.23	73.74±0.25	74.13±0.34	$0.23 \pm 0.023$	$0.34{\pm}0.035$	$0.27 \pm 0.052$	
Farm2	220	73.47±0.26	73.37±0.25	72.93±0.34	$0.16 \pm 0.015$	$0.21 \pm 0.018$	$0.15 \pm 0.009$	
Farm3	100	73.48±0.27	73.60±0.28	73.60±0.35	$0.19{\pm}0.016$	$0.13 \pm 0.010$	$0.11 \pm 0.005$	
			Aut	umn				
Farm1	120	60.07±0.64	59.76±0.65	60.33±1.03	$0.11 \pm 0.003$	0.11±0.003	$0.13 \pm 0.009$	
Farm2	200	60.21±0.59	60.16±0.58	59.64±0.79	$0.10{\pm}0.002$	$0.11 \pm 0.002$	$0.13 \pm 0.003$	
Farm3	200	63.71±0.64	63.79±0.67	63.87±0.86	$0.19{\pm}0.024$	$0.10{\pm}0.001$	$0.16{\pm}0.028$	
			Wii	nter				
Farm1	90	48.10±0.45	48.27±0.44	48.29±0.65	$0.10{\pm}0.001$	$0.10{\pm}0.001$	$0.10{\pm}0.004$	
Farm2	150	50.13±0.21	50.11±0.23	49.76±0.31	$0.11 \pm 0.003$	$0.12{\pm}0.007$	$0.13 \pm 0.011$	
Farm3	150	51.27±0.32	50.79±0.33	51.45±0.46	$0.10 \pm 0.001$	$0.10{\pm}0.001$	$0.10{\pm}0.000$	
Spring								
Farm1	90	$66.78 \pm 0.60$	66.90±0.60	66.82±0.89	$0.10{\pm}0.002$	0.11±0.006	$0.22 \pm 0.033$	
Farm2	100	68.83±0.29	68.68±0.30	68.51±0.38	$0.11 \pm 0.006$	$0.15 \pm 0.028$	$0.12 \pm 0.010$	
Farm3	100	67.35±0.41	67.40±0.42	67.83±0.49	$0.10{\pm}0.001$	$0.11 \pm 0.001$	$0.10{\pm}0.001$	

Table 2. Average daily temperature-humidity index (THI) and speed of airflow (SAF) values in three service areas of the production buildings by seasons

night hours was only 66% of that during the daytime hours.

The carbon dioxide values were different for the buildings on the three farms (Table 3). The highest values in all three cow servicing areas were recorded in the building of Farm 3, respectively from an average of 590.8 ppm above the two feeding alleys, up to 635.3 ppm (with a variation of 321 to 1550 ppm) above the free stalls. The building of Farm 3 had the largest capacity – 500 cows, which was a prerequisite for the release of a larger amount of carbon dioxide from the animals. Moreover, this building was also the widest, whereupon a large part of the stalls and the two manure alleys were inward the building, too distant from the open, longitudinal wall. Vtoryi et al. (2016) found that the level of  $CO_2$  concentration increased 2.0 to 2.5 times inward along the width of the building compared to the levels at front open wall.

The buildings of three farms had the same height but differ in provided area per cow. Farm 3 was with the largest volume provided per cow – 58.7 m<sup>3</sup>, compared to 43.4 m<sup>3</sup> and 43.98 m<sup>3</sup> in the other two farms. Although Erbez et al. (2015) pointed that more spacious animal premises can help disperse CO<sub>2</sub>, but this was not confirmed in our study. It is likely that this claim refers to differences that relate to the height of the buildings.

A trend for lowest  $CO_2$  values, especially above the stalls and manure alley, was observed in the building of Farm 1, respectively 517.4 and 518.2 ppm (with a variation of 191 to 1243 ppm). The buildings of Farm 1 and Farm 2 are the same. The main difference was the automatic turning on of almost all technological lines – for cleaning, closing the side openings and switching the fans on and off. In the building of Farm 2 only the alley scraper moved automatically, the curtain lowering and the fans switching on and off were manually, both at the discretion of the workers. Romaniuk and Mazur (2014) also found that in dairy cow buildings with robotic cleaning systems a lower level of harmful gases was maintained, including carbon dioxide, respectively 845.5 ppm and 665.5 ppm, which was below the half of the recommended levels.

Vtoryi et al. (2017) pointed that in the process of manure accumulation in manure collection channels there was a constant emission of carbon dioxide and a particular microclimate was formed depending on the number of animals in the building, the design characteristics of the ventilation systems or the external climatic conditions. According to Vtoryi et al. (2017), when chain-and-scraper conveyor begins to move the manure is shuffled, and carbon dioxide emissions concentrations in the building are increasing. During the removal of manure, which lasts 15-17 minutes, the CO<sub>2</sub> concentration increases from 1.3 to 1.5 times in different parts of the

building. In our study with the shortest interval of moving (20 min) was the scraper in Farm 3, and with the largest (90 min) the one in Farm 1.

~ .	1					
Service area	n	CO <sub>2</sub> , ppm				
		x±SE	min-max			
	Farm	1				
Above the stalls	390	517.41±7.43 <sup>a</sup>	215-1243			
Above the manure alley	390	518.15±8.19 <sup>b</sup>	191-1122			
Above the feeding alley	195	482.97±9.18 <sup>ab</sup>	255-970			
Farm 2						
Above the stalls	670	543.63±6.50ª	236-1204			
Above the manure alley	670	533.88±7.41 <sup>b</sup>	149-2130			
Above the feeding alley	335	434.85±7.16 <sup>ab</sup>	180-1704			
Farm 3						
Above the stalls	600	635.27±6.68ª	321-1550			
Above the manure alley	600	628.43±6.67 <sup>b</sup>	271-1447			
Above the feeding alley	300	590.84±8.94 <sup>ab</sup>	211-1406			

Tabl	e 3.	Mean	values	and	variatio	on of	carbon	dioxide	in
the t	hre	e anim	al servi	ice a	reas by t	farm	IS		

\*Between the values marked with identical letters for the different service areas by farms there was a statistically significant difference at P < 0.05

The lowest values of carbon dioxide in the buildings of the three farms were reported in the area above the feeding alley, the differences with the other service areas were statistically significant in all three buildings (P < 0.05). In the buildings of Farms 1 and 2 the feeding alley was located on the open, longitudinal wall of the building. This was a prerequisite for the better ventilation of this area and, accordingly, the reporting of low levels of carbon dioxide of 483 and 434.9 ppm, which was almost the background level. There were two feeding alleys in the building of Farm 3, of which only one was located along the outer open wall, the other was internal, between the stalls. Here as an average higher values of carbon dioxide were reported - 590.84 ppm, although they were lower than the average values reported above the stalls and manure alley respectively with 7 and 6%. Vtoryi et al. (2016) indicate that the concentration of carbon dioxide is highest in the central part of the building, where the level can reach 1.4 to 1.9 times higher than in other parts.

Regardless of the reported differences in the buildings of the three farms the values as the average so and the maximum deviations were below the threshold value determined by Regulation 44 (2006), respectively, up to 0.3% or 3000 ppm.

In a study conducted by Teye et al. (2007) in dairy cows' farms in Finland and Estonia the  $CO_2$  concentration was from 522 to 1678 ppm. Erbez et al. (2015) found that the

lowest carbon dioxide level reported in buildings for loosehousing production system, an average of 627.5 ppm (ranging from 390 to 890 ppm), while in the tie-stall barns it was 936,7 ppm (with a variation of 390 to 1690 ppm). Kavolelis (2006) in Lithuania reported a much higher level of carbon dioxide in tie-housing dairy cow buildings with good insulation, respectively 1520 ppm (with varying from 500 to 2450 ppm) and much lower levels in free stall barns – 765 ppm (ranging from 480 to 930 ppm).

Table 4 presents the results of analysis of variance for the influence of the environmental factors Farm, Season and Hour of reporting on the values of  $CO_2$  in the three animal service areas.

The Farm, Season and Hour of reporting had a statistically significant effect (P<0.001) on CO<sub>2</sub> values in all three service areas. Associated factors Farm\*Hour, Season\*Hour and Farm\*Season\*Hour had no significant effect on the level of carbon dioxide only above the feeding alley. This was probably due to the fact that in all three farms the feeding alley was on the open side of the building.

By seasons (Fig. 1), the highest LS-mean  $CO_2$  values in the three service areas were reported in winter – from 548.3 to 626.6 ppm in the different areas followed by these during the autumn. For the winter season, the highest maximum values for dioxide of 2130 ppm were also recorded. The lowest  $CO_2$  values were reported in the summer – from 479.4 to 517.9 ppm, followed by these during the spring.

Jovović et al. (2015), in a survey of 38 dairy cow buildings in the regions of 10 municipalities in Bosnia and Herzegovina, reported average  $CO_2$  values for the winter season of 871.57 ppm, ranging from 390 to 1690 ppm. The authors indicated that in summer, the concentration of carbon diox-



Fig. 1. LS-mean values of CO<sub>2</sub> in the three service areas by seasons

ide in cow buildings was lower than in winter due to the opening of windows and doors and more intense ventilation. When the outside air temperature increases, the carbon dioxide content of the building air decreases and vice versa (Vtoryi et al., 2016).

The lowest LS-mean values for dioxide were recorded at noon hours – 12:00-14:00 h (Fig. 2). At that time in Farms 2 and 3 was the midday milking, and most animals were outside the building, in traffic area, in holding area in the waiting room, and in the milking parlor. In the morning, around 10.00 h and after noon at 18.00 h, an increase in carbon dioxide in the premises was recorded. In the area above the stalls, the increase of  $CO_2$  compared to noon hours was 16.3% for Farm 3 and 23.9% for Farm 2. On the three farms in the morning after milking (at 8 o'clock) and after noon about 16 o'clock, the animals were fed. At these time intervals, some veterinary and zootechnical activities were also carried out.

Table 4. Analysis of variance for the influence of the controlled factors on the level of carbon dioxide in the area above the stalls, manure and feeding alleys

Sources of varia- tion	Degrees of freedom	Above t	he stalls	Above the manure alley		Above the feeding alley	
	(n – 1)	MS	F P	MS	F P	MS	F P
Total for the model	59	207171.5	9.38***	215556.9	8.22***	59999093	7.03***
Farm	2	1909496	86.45***	1771670	67.52***	2000478	118.25***
Season	3	1153834	52.24***	1079201	41.13***	310025	18.33***
Hour	4	166555	7.54***	217855	8.30***	84238	4.98***
Farm* Season	6	294030	13.31***	341549	13.02***	160542	9.49***
Farm* Hour	8	119635	5.42***	99419	3.79***	28433	1.68-
Season* Hour	12	31364	1.42-	55604	2.12*	20297	1.20-
Farm* Season* Hour	24	43540	1.97**	53472	2.04**	23165	1.37-
Error	1600(770)	22087		26238		16917	

\* significant at P < 0.05; \*\* significant at P < 0.01; \*\*\* significant at P < 0.001; ns - no significant effect



LS- means of CO<sub>2</sub> depending on the Farm and Hour of reporting above the manure alley

Fig. 2. LS-mean values of CO, depending on the farm and the hour of reporting above the stalls and manure alley

This was a prerequisite for increased animal activity and the release of more carbon dioxide. The studies of Pedersen et al. (2008) confirmed the need to take account of animal activity.

LS-means of CO<sub>2</sub> depending on the Farm and Hour

Least variation in the level of carbon dioxide at different times during the day was reported in the building of Farm 1 – within 50 ppm. This once again showed that automated technological activities were better option than to rely on workers' solutions and senses. From the analysis of the influence of the two microclimatic indicators – THI and SAF, on the carbon dioxide values in the dairy cows' buildings a statistically significant effect of both indicators was found (P < 0.001) (Table 5).

The lowest LS-means for carbon dioxide (Fig. 3) were reported at THI values from 58 to 72, respectively 519.27 and 500.78 ppm, and the highest were reported for THI values up to 58. The differences were respectively higher by 25.8% and 31.0%. A slight increase in carbon dioxide was also reported at the highest THI values - over 74 - an average of 578.6 ppm, the increase from the lowest was about 15.5. Segnalini

Table 5. Analysis of variance for the influence of temperature-humidity index (THI) and speed of airflow (SAF) on the values of carbon dioxide in the dairy cow buildings

Sources of variation	Degrees of	For all areas		
	freedom $(n-1)$	MS	F P	
Total for the model	8	1448908	52.88***	
THI	5	1980131	72.28***	
SAF	3	367695	13.42***	
Error	3311	27395		

\* significant at P < 0.05; \*\* significant at P < 0.01; \*\*\* significant at P < 0.001; ns – no significant effect

et al. (2013) determined the values of  $68 \le \text{THI} < 72$  as a prerequisite to mild discomfort in dairy cows. Armstrong (1994) classified a THI below 71 as a comfort zone, values ranging 72-79 as mild stress, 80-89 as moderate stress, and values over 90 as severe stress.

THI values above 74 were a prerequisite for varying degrees of heat stress in dairy cows. They react on it in several ways: reducing feed intake and increasing water intake, accelerating the metabolism and increase the loss of water by evaporation, increased frequency of breathing, change in the concentration of hormones in the blood and increased body temperature (Koubková et al., 2002). We believe that the slight increase in carbon dioxide values in our study was due to the increased breathing rate at these THI values.

The correlation between CO<sub>2</sub> and THI was -0.23, statistically significant at P < 0.05. The lowest carbon dioxide values



Fig. 3. LS-means of CO, depending on the THI values

are reported at the optimal values of SAF from 0.2 to 1 m/s, respectively from 495 to 502.6 ppm (Fig. 5). Compared with the lowest values, at low SAF – up to 0.2 m/s an increase in the level of dioxide emissions by 16.3% was reported. There was a slight increase in the level of dioxide and at the highest SAF values (over 1.1 m/s), by about 7%. As pointed by Vtoryi et al. (2016), carbon dioxide was heavier than air, so with a low SAF in the room, the highest concentration was at floor level and rises with the water vapors. When the SAF is more than 0.2 m/s, due to the mixing of air, the concentration of all gases at the height of the premises is equalized.



Figure 4. LS-means of CO, depending on SAF

The slight increase in the values of the carbon dioxide at a high SAF (over 1.1 m/s) was due to the fact, that such speeds were reported in the summer at high THI, whereat respectively a slight increase in CO<sub>2</sub> was reported. The correlation between carbon dioxide values and SAF was -0.08, statistically significant at P < 0.05.

## Conclusion

The larger capacity and width of the building, despite the more  $m^3$  of the building space per cow provided, were a prerequisite for higher CO<sub>2</sub> levels in the area above the stalls. A trend for the lowest levels of CO<sub>2</sub> and the least variation of its values during the day was reported in the building with the automation of all technological processes – cleaning, fans, side openings and the largest scraper movement interval.

The semi-open type of buildings for dairy cows does not provide isolation for the animals from the external climatic conditions, especially from high temperatures. Generally, they maintain good microclimate conditions, but high risk temperature, high THI values and insufficient ventilation were reported during the summer, especially in larger buildings. The  $CO_2$  levels in the studded barns were within the limits set by the Bulgarian legislation. The larger capacity and width of the building, despite the more m<sup>3</sup> of the building space per cow provided, were a prerequisite for higher  $CO_2$  levels in the area above the stalls. A trend for the lowest levels of  $CO_2$  and the least variation of its values during the day was reported in the building with the automation of all technological processes – cleaning, fans, side openings and the largest scraper movement interval.

During the summer, the lowest  $CO_2$  values in cow's buildings were reported due to increased ventilation and the highest in winter. Highest  $CO_2$  values were reported in the morning and in the afternoon when the animal's motor activity was greatest, associated with eating, drinking, and other activities. At the optimum values of THI of 58 to 74 and SAF of 0.6 to 1 m/s, the lowest  $CO_2$  values were recorded. At the lower THI and SAF values (of these parameters) the highest  $CO_2$  values were reported.

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