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Effect of light intensity, spectrum, and uniformity on the ability of dairy cows to navigate through an obstacle course

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ABSTRACT

The most suitable light intensity for cows during nighttime has not been thoroughly investigated. Recommendations on the night-time lighting regimen on dairy farms differ between countries and range from light throughout the night to darkness to allow the animals a rest from artificial light. Commercial actors recommend red light for night-time lighting in cattle barns to facilitate livestock supervision with minimum disturbance for the animals. However, little is known about how light intensity, spectrum, and uniformity affect the ability of cows to navigate their indoor environment. Thus, in a change-over study with 12 pregnant, nonlactating dairy cows, we observed how the cows walked through an obstacle course under different light treatments. Obstacles were positioned differently for every run, to present a novel challenge for each light environment. Fourteen different light treatments were tested, involving intensity ranging from <0.01 (darkness) to 4.49 μmol m^{-2} s^{-1}, high or low uniformity, and white or red color. Light was characterized in terms of illuminance, photon flux density, spectral composition, and uniformity. Additionally, assessment of the environmental light field was used to describe each lighting condition from a bovine and human perspective. Data were analyzed in a generalized mixed model to assess whether lighting conditions affected cow walking speed or stride rate. Pair-wise post hoc comparisons showed that the cows walked at a slower speed in nonuniform red light compared with uniform white light or uniform red light. Interestingly, darkness did not alter walking speed or stride rate. The odds of different behaviors occurring were not affected by lighting conditions. In conclusion, darkness did not affect the ability of cows to navigate through the obstacle course, but medium-intensity, nonuniform red light affected their speed. Hence, cows do not necessarily need night-time lighting to navigate, even in a test arena with obstacles blocking their way, but nonuniform light distribution may have an effect on their movements.

Key words: activity, night light, dim light, arena test

INTRODUCTION

In loose-housing systems, cows can move freely in the barn and access available resources throughout the 24-h period. Loose housing combined with robotic milking in automatic milking systems (**AMS**) assumes cow activity and ability to navigate around the clock. The ability of cows to navigate in dim light environments has been examined in some previous studies, with various outcomes. Hjalmarsson et al. (2014) compared the effect of 3 night light intensities (11 ± 3, 33 ± 1, and 74 ± 6 lx) on gate passages on AMS farms and found that cows passed through the gates more frequently during daytime than at night, but that the total number of gate passages per 24 h did not differ between the light intensities. Phillips and Morris (2001) investigated whether cows would choose a bright or dark passageway in a y-maze through which they were trained to walk by providing a reward at the end of the passageway, and found that nearly all cows avoided the dark passageway. Phillips et al. (2000) found that stride rates were higher and stride lengths were shorter in dim light than in bright light, but that walking speed (m/s) was similar in bright and dark light environments. Willson et al. (2021) found that light distribution can affect walking behavior, with sharp contrast shadows on the floor hindering cow traffic.

In humans, it is known that artificial lighting during night hours and shift work poses a health risk (Costa,

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2010). Research on health risks in relation to artificial lighting during night hours in cows is scarce. However, it has been shown that light at night suppresses melatonin secretion (Tähkämö et al. 2019), which could affect the circadian rhythm negatively. To maintain a circadian secretion of melatonin in cows, a dark phase at night is essential, but a threshold value for the level of darkness required is yet to be determined (Hedlund et al., 1977; Muthuramalingam et al., 2006; Bal et al., 2008; Kollmann et al., 2008; Elsabagh et al., 2020). Recommendations for night-time lighting regimens on dairy farms differ between nations. For example, recommendations in the UK state that cows kept indoors should have a period of rest from artificial light (DEFRA, 2003), whereas in Sweden, dimmed night light is mandatory (Statens Jordbruksverk, 2019). However, light intensity, uniformity, and spectral composition of the light are currently not specified in regulations or recommendations, and hence night-time lighting regimens differs in practice. For example, a study by Jakobsson (2016) investigating night light intensity (illuminance) on 15 Swedish farms found that it varied between 0.6 and 54.6 lx, while a study by Reksen et al. (1999) on 104 Norwegian farms found that supplementary light intensity ranged from 4 to 160 lx.

Red light has been promoted as suitable night-time lighting to enable farmers to work in the barn at night without disturbing the circadian rhythm of the cows, and particularly of the farmer. However, the actual benefit of red light for circadian rhythm in mice (Dauchy et al., 2019) and circadian rhythm and IGF-1 secretion in dairy cows (Lindkvist et al., 2021) has been questioned, especially when relatively bright. Red light is a weak stimulus for the melanopsin-containing intrinsically photosensitive retinal ganglion cells important for adjusting the circadian rhythm to the 24-h dark-light cycle, but also for rod photoreceptors. Hence, dim red light maintains the retina in a relatively dark-adapted state. With brighter red night lighting, the retina will become more adapted to light, and the number of photons will start to activate the long wavelength-sensitive or medium- to long-wavelength-sensitive cones (**L-cones**, **ML-cones**, respectively) under mesopic light conditions (Pokorny et al., 2006). Bright red light will eventually saturate the rod photoreceptors under photopic light conditions, and vision is then mediated through the cone photoreceptors (Ofri and Ekesten, 2021). Interestingly, at similar light intensities, red light did not cause the same level of pupil constriction as blue or white light (Lindkvist et al., 2021), which implies that a greater number of photons reaches the retina and stimulates the cone photoreceptors in bright red light.

Although there is some available evidence on the ability of cows to navigate through dim or dark alleys, the effect of light intensity, spectral properties, and uniformity has not been examined in detail. Therefore, the overall aim of this study was to investigate the ability of dairy cows to navigate in different indoor light environments, including light of high and low uniformity, and also with 2 different spectral compositions, red or white light. Specific hypotheses were that cows walk at a slower speed in a dark environment than in bright light, spend a longer time navigating an environment in red light than in white light, and stop more often in light with low uniformity.

MATERIALS AND METHODS

Animals and Housing

The study was conducted at the Swedish Livestock Research Centre, Uppsala, Sweden, in an indoor test arena with a controlled light environment without contamination from external light (hereafter called the "light lab"). All animal handling was approved by the Uppsala Ethics Committee for Animal Research, Uppsala, Sweden (reference no. 5.8.19–06780/2020).

The light lab comprised 3 individual adjacent pens (each 3.0 × 3.0 m) with solid walls (height 1.41 m) except for a headlock opening in the front, facing an open arena (19.8 × 10.8 m) where the test arena including the obstacle course was placed. In total, 12 pregnant nonlactating dairy cows of the Swedish Holstein (n = 3) and Swedish Red (n = 9) breeds were included in the study. Before enrollment, all cows were examined to ensure clinically good claw and leg health. All cows received an ad libitum mixture of grass and maize silage with 5% straw inclusion, delivered at 0700, 1200, and 1600 h, topped up in between if needed, and water from a pressure-valve water bowl. The pens were cleaned in conjunction with feeding and the bedding material, consisting of wood shavings, was replaced daily. The cows had visual, auditory, and olfactory contact between each pen and the test arena, and limited physical contact between pens.

The light lab was equipped with 18 dimmable light-emitting diode (**LED**) light fixtures (Elixia LX602G, Heliospectra AB, Gothenburg, Sweden), lighting the obstacle course. The light fixtures were placed in 2 rows at 2.9 m above the floor (see Figure 1 for more details), and were connected to a computer that controlled the intensity and spectral composition of the light provided by each light fixture. The light was measured at cow eye level, approximately 1.3 m above the floor, with a photosensor directed toward the ceiling, in a grid pattern of 1 × 1 m covering the obstacle course. A lux meter (Hagner Screenmaster, B. Hagner AB, Solna, Sweden), 2 different spectrometers (Jaz, Ocean Insight Inc., Dunedin, FL; PAR200 Quantum Spectrometer, UPRtek, Aachen, Germany), and the environmental light field (**ELF**) method (Nilsson and Smolka, 2021) were used for this purpose. Illuminance (lx), photon flux density (**PFD**; μmol m^{-2} s^{-1}), light spectrum (μmol m^{-2} s^{-1} nm^{-1}), and spectral photon radiance (lit; log_{10} photons m^{-2} s^{-1} sr^{-1} nm^{-1}) were quantified. To simplify reporting, the expression "light intensity" refers to the amount of light in the barn, instead of the 4 correct terms. Moreover, the different mixtures of wavelengths used in the experiments are called "colors" based on the hues a normal human trichromat would perceive.

Experimental Design

A change-over design was used in the study, with 4 batches of 3 cows blocked according to days since dry-off (25–46 d), days before predicted calving date (25–35 d), and parity (1–4). Cows were moved into the light lab in early morning on d 1 and were left to acclimatize to the environment for a few hours. Later that same day, the cows were individually trained to walk through the obstacle course 3 times, without video-recording. A bucket of concentrate was used to actively encourage the cows to move. The person holding the bucket walked at least 5 m in front of the cow in a straight line in the middle of the arena. If a cow stopped for more than 5 s, she was encouraged to walk by a person following behind her. As an additional incentive for the cows to continue forward in the arena, they always walked toward the pens where the remaining cows in the batch were waiting. Days 2 to 5 were test days, during which 5 different obstacle courses were tested per cow and day, giving in total 21 different obstacle courses per cow and batch. Cows were tested one at the time, while the 2 others remained in their individual pens. Between every cow and run, feces were removed from the test arena. The arena was 3.8 m wide and 14.5 m long, with dark rubber mats covering the floor (Figure 1). The obstacles were white cavalletti poles and blocks (Safety-System AB, Enköping, Sweden) commonly used in horse training. Once all 3 cows within a batch had navigated an obstacle course, the light regimen and the obstacles were changed, creating a new course for each light treatment.

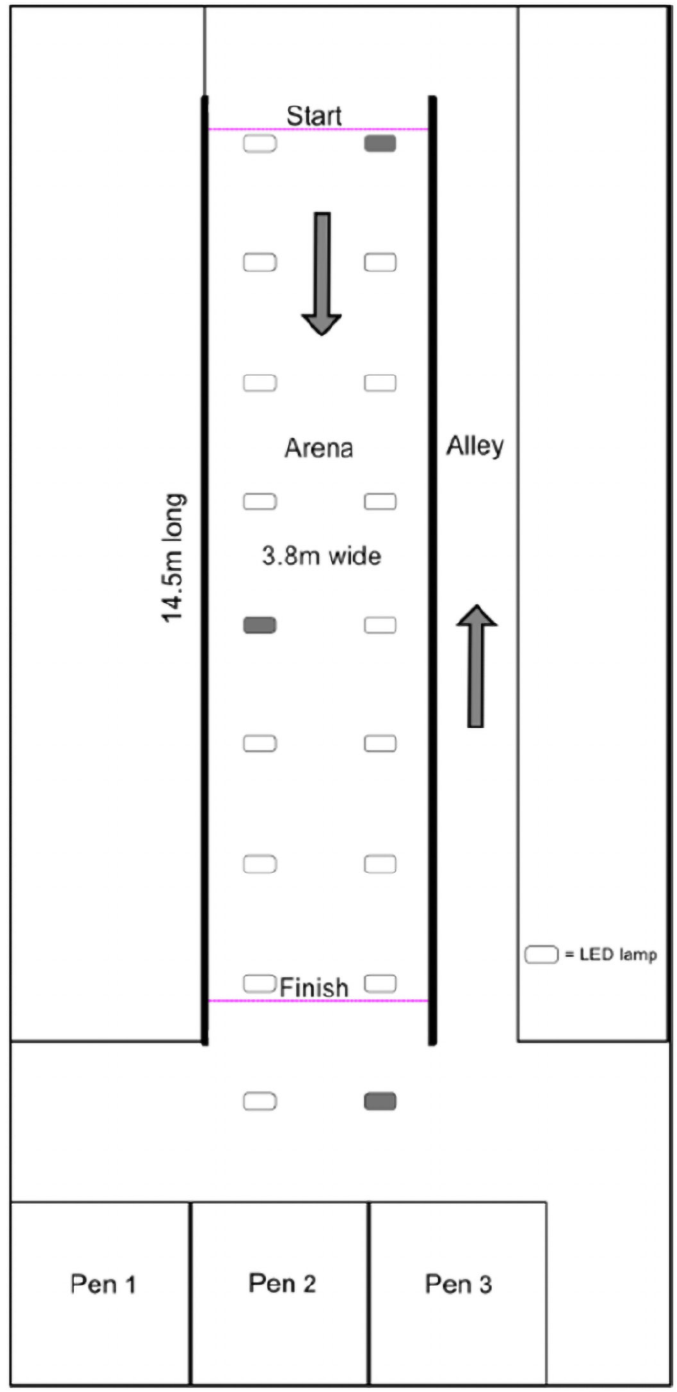

Figure 1. Schematic illustration of the room with a controlled light environment (light lab) in which nonlactating dairy cows were observed while walking through a test arena with obstacles in different light environments. The cows were housed in individual pens 1 to 3, and during the tests they walked down the alley and then through the arena (14.5 m long, 3.8 m wide). The start and the finish line are indicated by purple lines. Light was supplied by 18 light-emitting diode (LED) lamps placed 2.9 m above the floor. Filled rectangles indicate lamps used in nonuniform light treatments. In uniform light treatments, all 18 lamps were used.

Light Treatments

Fourteen different light treatments (described below) were grouped into 2 light regimens: one with uniform (**U**) light and one with nonuniform light (**N**). All 18 light fixtures were used for the uniform light scheme, while the nonuniform light was achieved by keeping only 3 fixtures switched on (as indicated in Figure 1 and described through heat maps in Figure 2). The light uniformity followed the European Standard for lighting in workplaces regarding illuminance uniformity (CEN Technical Committee, 2021), where minimum illuminance level ($\mathbf{E_{min}}$) divided by average illuminance level ($\mathbf{E_{av}}$) with high uniformity ($\geq$0.7) was considered uniform light treatment, and with low uniformity ($\leq$0.7) as nonuniform light treatment (Table 1). The color of the light was also tested, using either white (**W**) or red (**R**) light. White light contained a mixture of blue (32.5%, 400–500 nm), green (36.9%, 500–600 nm), and red (28%, 600–700 nm) light, similar to sunlight, whereas in the red-light treatments, 97.5% of photons fell within the wavelength spectrum of red light (Figure 3). Five different light levels were tested within the light scheme and identified as follows: dark, low1, low2, medium, and high. However, low1 could only be achieved in uniform white light and the intensity "dark" meant that all light fixtures were turned off. Illuminance and PFD are described in Table 1. Photon flux density, measured with a PAR200 spectrometer, was used to create similar light treatments within the light scheme and for both colors. The PAR200 device had better absolute calibration than the Jaz spectrometer, but was not sensitive enough to measure the lowest light levels. Therefore, light levels below 0.07 μmol m^{-2} s^{-1} were measured using the Jaz spectrometer and the values were multiplied by a correlation factor for PAR200/Jaz.

The ELF method was used to quantify essential biological aspects of the light environments by analyzing pictures of the light treatments, following the protocol developed by Nilsson and Smolka (2021). We used a digital camera with a 180° fisheye lens and 25 scenes with 3 exposures per scene within the arena, following the passage of cows through the obstacle course. To gain an impression of how the bovine eye (or a red-green colorblind human protanopia) perceived the arena under different light treatments, photographs from the light lab were manipulated with a digital filter (Sim Daltonism; Fortin, 2022).

Cows were exposed to 3 different light treatments per day, in addition to a dark treatment (**DU**; <0.01 μmol m^{-2} s^{-1}) that was applied as the first light intensity every morning and used to assess the possible effect of the factor "test day." For the same reason, cows also went through the arena in WU_{high} every day. All cows underwent all 14 light treatments (Table 1), over 4 test days during which the light treatments were blocked by light intensity, randomized within block, and divided by 4. This ensured 3 different light treatments per test day, always going from the dimmest light treatment to brighter light, with 10 min of light adaption time between tests.

Recordings and Data Handling

One observer manually recorded the times when the cow entered and exited the obstacle course, and the number of strides (defined as the right hind leg lifted above the floor and relocated to another spot) taken through the obstacle course, using direct observations. Time through the obstacle course, measured with a stopwatch, started and finished when the right rear leg entered and left the obstacle course, respectively. Speed was calculated by dividing the length (m) of the obstacle course by the time (s) it took for each cow to pass through it. Stride rate was calculated by dividing the number of strides by the time (s) taken to move through the obstacle course. Stride length was calculated by dividing the number of strides by the length (m) of the obstacle course. Other behaviors (see ethogram in Table 2) were indirectly mapped from video recordings, using a binary approach, and marked 1 if the event occurred and 0 if it did not. The events were then summarized per treatment, and the proportion of a specific event was calculated by dividing it by the total number of events per treatment.

A thermal camera (NightLux JSA IR-635, JSA Night-Lux, Albershausen, Germany) was used to facilitate observation of the cows' performance in light treatments below 3 μmol m^{-2} s^{-1} regardless of light color, and video recording with a mobile camera (iPhone 8, Apple Inc., Cupertino, CA) in light treatments above 200 lx. When using indirect observations, the observer was blind to the treatments <3 μmol m^{-2} s^{-1}, as those video recordings were all made by the thermal camera.

Cow heart rate (**HR**) and respiratory rate (**RR**) were measured in the pens at the start of every trial day. Heart rate was assessed using a stethoscope and RR by manually counting the number of flank movements during 1 min. Both HR and RR were then measured in the pen before and after every run by a cow through the course, and the difference between the 2 values (postrun HR − prerun HR; postrun RR − prerun RR) was used to assess the effect of light treatment ($\mathbf{HR_{diff}}$ and $\mathbf{RR_{diff}}$, respectively).

Statistical Analyses

All data were checked for normality and outliers using the univariate procedure in SAS (SAS version 9.4, SAS Institute Inc., Cary, NC). Where applicable, data were log_{10}-transformed.

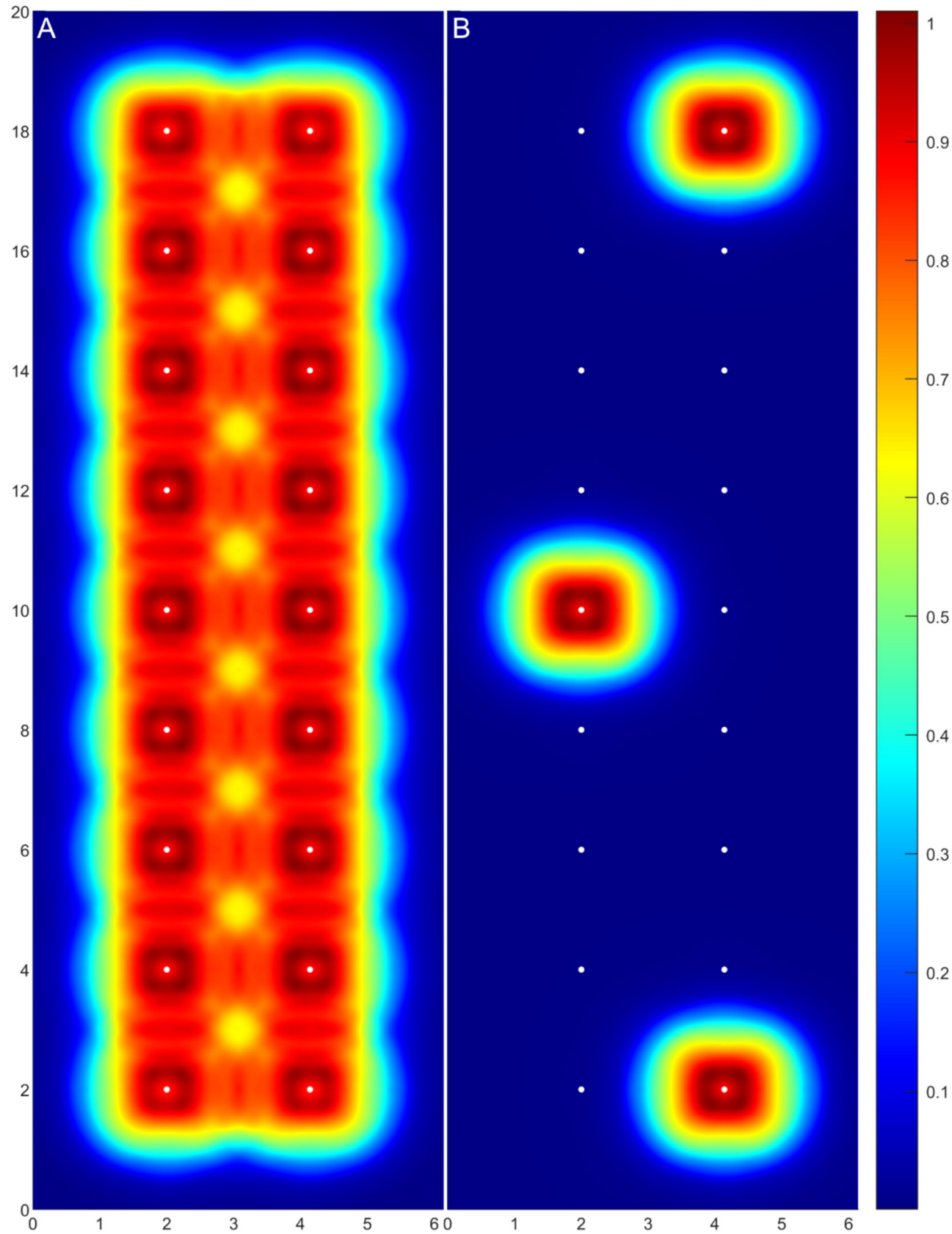

Figure 2. Heat maps of the light environment (controlled using light-emitting diode [LED] lights) in the room with a controlled light environment (light lab) in treatments with high or low light uniformity. The difference in uniformity can be illustrated by simulating light intensity distribution over the obstacle course when all lamps are lit (A), corresponding to the high-uniformity treatment, and when only 3 lamps are lit (B), corresponding to the low-uniformity treatment. The lamps are shown as white dots in the so-called heat map, and the maximum light intensity is normalized to one. This simulation does not take reflections into account.

A generalized mixed model in SAS was used to test whether time through the obstacle course (s), number of strides, stride length (m), stride rate (strides/s), and speed (m/s) were affected by light treatment (referred to using abbreviations for conditions, with light intensity level as a subscript: DU, WU_{low1}, WU_{low2}, WU_{med}, WU_{high}, WN_{low2}, WN_{med}, WN_{high}, RU_{low2}, RU_{med}, RU_{high}, RN_{low2}, RN_{med}, and RN_{high}), in addition to differences

Table 1. Light intensity and uniformity regimens tested in the study, expressed as illuminance (lx) and photon flux density (PFD; µmol m^{-2} s^{-1}) per light treatment and calculated illuminance uniformity according to European Standards (CEN Technical Committee, 2021)

	Illuminance (lx)			PFD (µmol m^{-2} s^{-1})			
Treatment[1]	Average	Minimum	Maximum	Average	Minimum	Maximum	Uniformity
DU	0	0	0	<0.01[2]	—	<0.01[2]	—
WU_{low1}	8	7	11	0.12	0.11[3]	0.15	0.9
WU_{low2}	19	17	24	0.28	0.25	0.34	0.9
WU_{med}	50	43	63	0.73	0.65	0.89	0.9
WU_{high}	209	181	265	3.6	3.19	4.48	0.89
RU_{low2}	3	3	5	0.28	0.23	0.38	0.82
RU_{med}	8	0.5	11	0.65	0.55	0.89	0.85
RU_{high}	40	33	55	3.23	2.62	4.47	0.81
WN_{low2}	8	5	15	0.04	0.01[3]	0.22	0.21
WN_{med}	19	3	39	0.09	0.01[3]	0.59	0.09
WN_{high}	35	1	162	0.46	0.02[3]	2.85	0.03
RN_{low2}	2	2	2	0.02	0.01[3]	0.2	0.32
RN_{med}	4	3	6	0.08	0.01[3]	0.46	0.1
RN_{high}	17	12	28	0.38	0.02[3]	2.28	0.04

[1]Treatment code abbreviations were as follows: D = darkness; W = white light; R = red light; U = uniform light; and N = nonuniform light. Light intensity was added as a subscript (from least to greatest intensity): dark, low1, low2, medium (med), and high. Light treatments with calculated uniformity (PFD minimum divided by PFD average) ≥0.7 are described as uniform and with calculated uniformity ≤0.7 as nonuniform.

[2]All light fixtures were turned off, but small amounts of light leaked into the room through slits around the door and from emergency exit signs.

[3]PFD was measured with a PAR200 spectrometer (UPRtek), but a Jaz spectrometer (Ocean Insight Inc.) was superior at low light intensities. Therefore, measurements below 0.07 µmol m^{-2} s^{-1} were calculated by multiplying the values obtained using the Jaz spectrometer with a correction factor for PAR200.

in physiological data (HR_{diff} and RR_{diff}). None of the behavior variables were normally distributed and they were therefore $\log_{10}$-transformed. In all models, light treatment (n = 14) and batch (1–4) were included as fixed effects, and cow nested within treatment and batch as repeated effect, with a first-order autoregressive covariate structure. In addition, the effects of test day, time of day, and obstacle course were tested. Interactions of fixed effects were excluded using stepwise backward elimination, where any interaction effect with $P > 0.10$ was excluded from the model. Only light treatment × batch remained in the final model. Both means and standard error of the mean were back-transformed, using the delta method (Onofri et al., 2010).

After reviewing the occurrence of all other behaviors, it was decided to analyze the occurrence ratio only for behaviors that all cows performed in all light treatments (i.e., standing still, interaction with an obstacle, interaction with surrounding, and interaction with the floor). The odds ratio of a cow performing one of these 4 behaviors in a light treatment was estimated with a generalized linear mixed model with a binary data distribution using Proc Glimmix in SAS. Light treatment (n = 14) and batch (1–4) were included as fixed effects, and cow nested within treatment and batch as a random effect. Proportion of behaviors occurring in each treatment was descriptively illustrated by adding up the number of events per light treatment and dividing by the total number of behaviors occurring.

Unless otherwise stated, the values presented are least squares means ± standard error of the mean. Differences were considered significant at $P \leq 0.05$, while a trend was assumed for probability values $0.1 > P > 0.05$. Post hoc means separation for significant main effects was applied using Tukey-Kramer's adjustment of probability values.

RESULTS

Descriptive Results of the Light Environment

The obstacles and the barrier tape were readily visible to humans with normal color vision, and to a human protanopia or cow under both red and white illumination (Figure 4). However, the nonuniform dim light made the extent of the obstacles appear less distinct and created shadows and fuzzy surroundings. According to the ELF method, the nonuniform light treatments were slightly less bright, closer to moonlight, whereas the uniform light treatments appeared as "mid-dusk" (Figure 5). Unsurprisingly, the relative color of red was highest in all red-light treatments, as indicated in Figure 5. The relative color of red was also highest in the white light treatments, but the difference between red, blue, and green was minor.

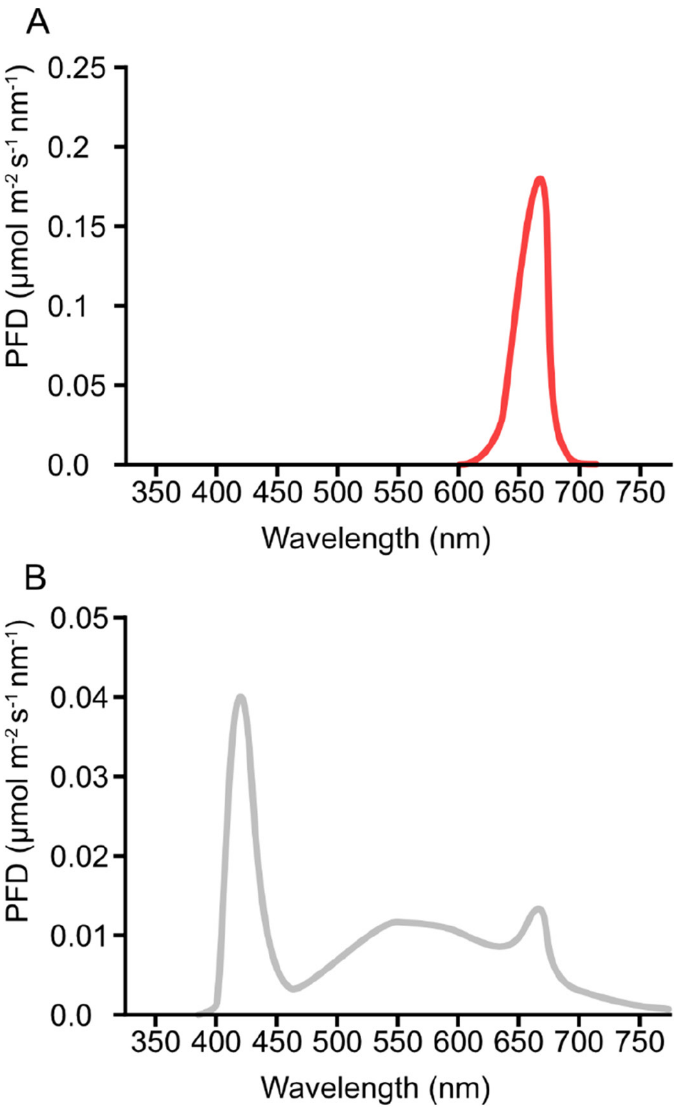

Figure 3. Spectral composition of the (A) red and (B) white light-emitting diode (LED) light environment used in the room with a controlled light environment (light lab). Light intensity and uniformity are shown in Table 1. PFD = photon flux density.

Strides and Other Behaviors Performed in the Obstacle Course

Cow speed (m/s; $P = 0.006$; Figure 6), stride rate (strides/s; $P = 0.014$), and time (s; $P = 0.006$) through the obstacle course were affected by light treatment. Interestingly, pair-wise post hoc comparisons showed that DU did not alter cow speed or stride rate. Instead, cows spent a longer time in the obstacle course and walked at a slower speed in RN_{med} compared with WU_{high} ($P = 0.01$ for both) or RU_{low2} ($P = 0.001$ and $P = 0.02$, respectively). In addition, there was a tendency for cows to walk more slowly in RN_{med} than in RU_{high} ($P = 0.08$). There were no other significant pair-wise differences in speed, stride rate, or time through the obstacle course. There was a tendency for treatment to affect the number of strides ($P = 0.08$; Figure 6) and stride length (m; $P = 0.08$), but obstacle course ($P = 0.13$), day in treatment ($P = 0.46$), and time of day ($P = 0.48$) had no effect on any of the parameters measured. As visualized in Figure 7, there was no obvious pattern in behaviors occurring in the different treatments. The odds of behaviors occurring were not affected by treatment ($P = 0.41$), obstacle course ($P = 0.65$), or batch number ($P = 0.39$).

In HR_{diff}, there was no difference between the light treatments ($P = 0.25$). Mean HR during rest and testing was 71 ± 6.8 (range 60–84) and 77 ± 8.0 (range 56–104), respectively. There was a tendency for light treatment to affect RR_{diff} ($P = 0.07$). Average RR during rest and testing was 37 ± 9.5 (range 20–64) and 39 ± 7.3 (range 20–60), respectively.

DISCUSSION

This study investigated the ability of dairy cows to navigate in different indoor light environments, including light of high and low uniformity. Tests using an obstacle course showed that cows walked slower and with shorter strides in medium nonuniform red light,

Table 2. Ethogram of behavioral observations within the obstacle course

Behavior	Definition
Standing still	The cow stands with at least 3 hooves on the floor for at least 5 s.
Interaction with obstacle	The cow interacts with obstacle by sniffing, licking, or touching it with any part of the body, without knocking the obstacle to the floor.
Interaction with surrounding	The cow interacts with any other surrounding by sniffing, licking, or touching it with any part of the body.
Interaction with floor	The cow interacts with the floor by sniffing or licking.
Knock-down	The cow touches an obstacle, and the obstacle falls to the floor.
Defecation	The cow defecates or urinates.
Self-grooming	The cow interacts with herself by licking or scratching with tongue or claw.
Jumping over obstacle	The cow steps or jumps over an obstacle with all 4 legs without knocking it down.
Slipping	The cow does a sliding movement with a leg along the floor.
Vocalization	The cow creates a sound with her vocal cords for at least 1 s.

Figure 4. Images showing light treatments of medium intensity (med; left column) used in the room with a controlled light environment (light lab), while testing the cow's ability to negotiate an obstacle course. (A) White light, uniform (WU_{med}), (C) red light, uniform (RU_{med}), (E) white light, nonuniform (WN_{med}), (G) red light, nonuniform (RN_{med}); and the corresponding images (right column) as possibly perceived by a cow (or human protanope). (B) White light, uniform with protanope filter (WU_{med}), (D) red light, uniform with protanope filter (RU_{med}), (F) white light, nonuniform with protanope filter (WN_{med}), (H) red light, nonuniform with protanope filter (RN_{med}). The spectral composition is shown in Figure 3, and light intensity and uniformity in Table 1.

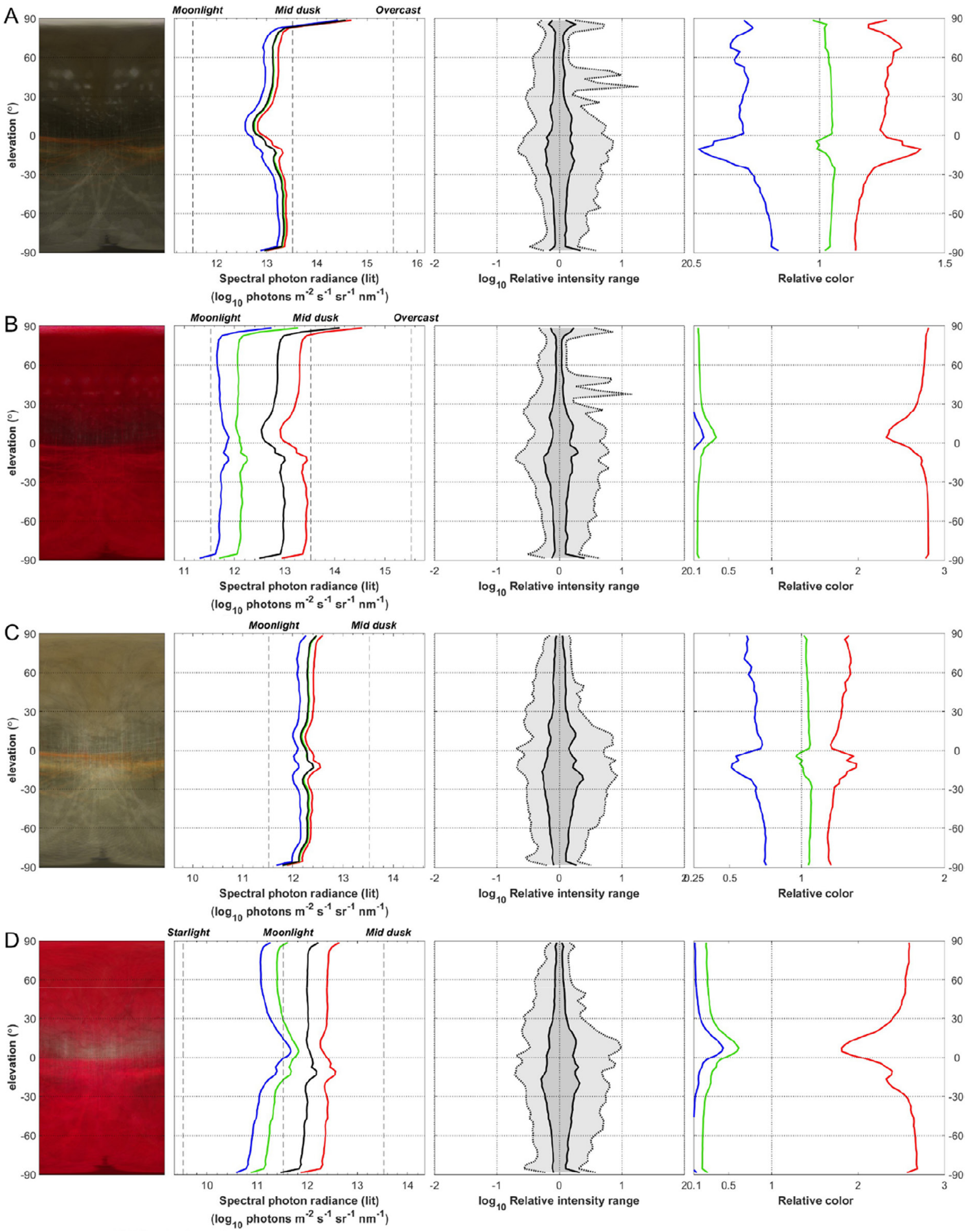

Figure 5. Environmental light field (ELF) analysis of light treatments of medium intensity (med): white (W), red (R), uniform (U), and nonuniform (N). The ELF method uses 180° high dynamic range images taken within the room with a controlled light environment (light lab), with multiple exposures of 25 photos per light treatment taken from different environmental positions, following the cows' progress within the obstacle course. An average image (compressed in azimuth) from the contributing scenes (180° by 180°) is shown to the left, followed by panels showing the intensity (radiance) on an absolute log scale, the intensity range on a relative log scale (dark gray, 50% of all intensities; light gray, 95% of all intensities) and, on the right, the contribution of red, green, and blue light plotted on a relative log scale. (A) White light, uniform (WU_{med}), (B) red light, uniform (RU_{med}), (C) white light, nonuniform (WN_{med}), (D) red light, nonuniform (RN_{med}).

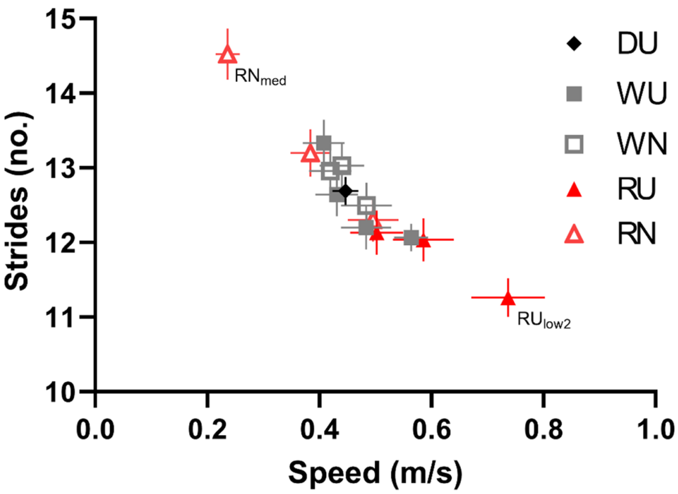

Figure 6. Speed (m/s) relative to number of strides of nonlactating dairy cows navigating through an obstacle course in different light treatments: darkness (D), white (W), or red (R) light, and uniform (U) or nonuniform (N) light.

but there was no effect of complete darkness on movement through the obstacle course, contradicting our hypothesis. None of the high-uniformity light environments affected cow behavior. The lack of difference between the darkest light environment and any of the other light treatments is interesting, because Phillips et al. (2000) found that cows walked faster in darkness than in brighter light intensity. However, the cows in our study walked slower in general than the cows in that study. This could be due to differences in study design, as Phillips et al. (2000) used a passageway and we used an obstacle course in an arena that was not part of the cows' regular housing unit. Hence, the novelty of the experience and the obstacles were likely to have prevented cows from walking at high speed in both dim and bright light intensities in our study. In one of the trials reported in Phillips et al. (2000), cows had a higher stepping rate with shorter stride length in the dark, which was suggested to be a strategy to maintain speed in the dark and at the same time manage the risk of slipping or encountering obstacles. The cows in our study did not change their stride length or rate between the brightest and lowest light treatment, indicating that they were sufficiently comfortable and aware of their surroundings to maintain normal speed and stride length regardless of the light environment. It has been argued that a dark environment can make cows more hesitant (Phillips and Morris, 2001; Stookey and Watts, 2007), where hesitation can be interpreted as cows being more fearful or cautious of their surroundings. Our results did not show any effects of light treatment or darkness on HR, RR, vocalization, or frequency of defecation, which are commonly used as stress indicators (Grandin, 2001).

According to our image analysis, the RN_{med} light treatment was close to "moonlight" and subjectively perceived as dim twilight conditions by human observers. This light level could be on the verge of bovine mesopic vision, where rod photoreceptors are close to saturation and cones are still weakly stimulated (Ofri and Ekesten, 2021). In humans, increasing light above the scotopic level, where vision is essentially rod-mediated, allows L-cones and, to a lesser extent, M-cones to contribute to vision (Pokorny et al., 2006). At higher light levels but still within the mesopic range, the rod input lessens and short-wavelength sensitive cones (**S-cones**) become active. Under even brighter light cone vision prevails, and objects reflecting longer wavelengths appear relatively brighter than those reflecting

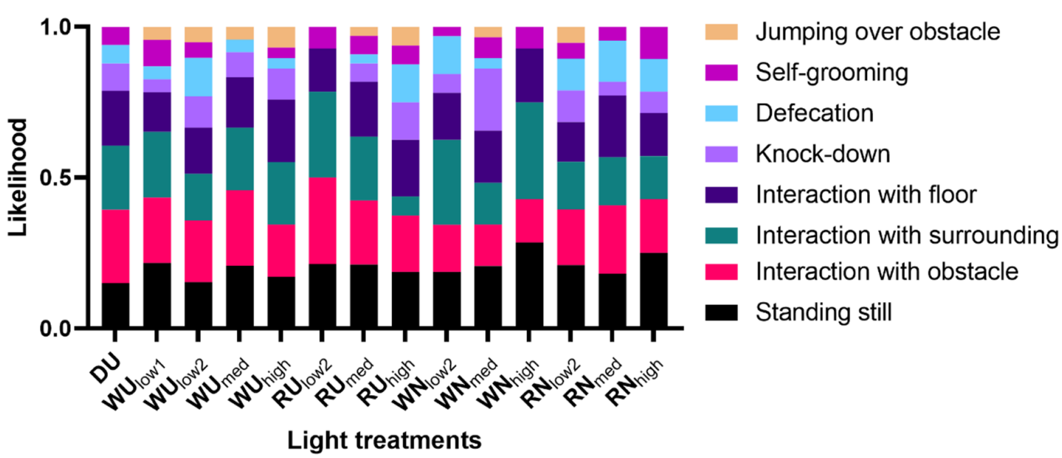

Figure 7. Likelihood of different behaviors occurring in the 14 different light treatments while nonlactating dairy cows were passing through an obstacle course (for ethogram of behaviors, see Table 2). Light treatment combinations include darkness (D), white (W), red (R), uniform (U), and nonuniform (N) light, at 5 light levels: dark, low1, low2, medium, and high.

shorter wavelengths (the Purkinje shift). Cattle are dichromats with S- and ML-cones (Jacobs et al., 1998). Although not studied in cattle, as far as we know, a similar transition from rod to cone vision over a range of twilight-like light intensities and a Purkinje-like shift in color perception are likely.

Color and reflectance of surfaces in the test arena probably affected the ability of the cows to see the obstacles. The black rubber mats on the floor had low reflectance compared with the white surface of the obstacles (Gilchrist, 1979). The cows could probably perceive the white obstacles quite easily in all light treatments due to their reflectance, whereas the interior of a cow barn usually has lower reflectance. The effect of a Purkinje-like shift on perception of the obstacles and other items in the environment is unclear. When light levels were dimmed or low in some areas due to low uniformity, the short-wavelength (blue) component of white light may have made the obstacles easier to perceive against the dark floor than with red light illumination containing a similar number of photons. Red objects appear relatively brighter than blue objects in daylight, whereas the opposite is true when the light is dimmed.

It has been found that sheep are more hesitant to move when the light comes from below the floor (Hutson, 1981). A white floor could be perceived as similar to light coming from below, whereas in nature the ground is always darker than the sky. The floors in dairy barns are not white, so this should be kept in mind when designing light environments for dairy cows. It would be interesting to investigate whether the strong contrast in color between the dark floor (black) and white obstacles was essential for the results in the present study and to determine the effect of smaller (more dairy barn-like) differences in brightness between the obstacles and floor on the performance of cows under different lighting conditions.

A strength of our study was good precision in the light treatments and detailed characterization of the treatments, with intensity, wavelength, and spectrum of light within the arena measured in more detail than in previous studies. Nevertheless, understanding cows' perception of their light environment remains challenging. Although the measured PFD was <0.01 μmol m^{-2} s^{-1} for the darkest light treatment, the human eye did not perceive the light lab as pitch black. Small amounts of light leaked into the room through slits around the door and from emergency exit signs. It is known that human rod photoreceptors can detect single photons (Hecht et al., 1941). Because the cow eye is larger and has a tapetum lucidum, the light pollution in the arena was highly likely to have been sufficient for some rod-mediated vision in the cows, which can have aided them in navigating the obstacle courses without altering their walking behavior.

Illuminance uniformity affected how the cows in this study acted within the arena. For example, the behavior of jumping over obstacles was never observed in nonuniform red light of high intensity, but did occur in nonuniform red light of low intensity and in uniform white light. Jumping over obstacles instead of knocking them over indicates that the cows were confident in distinguishing the obstacles in the dim environment and were comfortable with the floor surface. We interpreted the numerical increase in interactions with obstacles in the medium-intensity, nonuniform red light, in combination with a decrease in speed, as a possible sign of insecurity or fear, as proposed by Hughes (1997). This highlights the importance of a well-planned light environment in loose-housing systems to promote efficient cow traffic. Our findings have bearings on how night lighting is designed in cow barns. Relatively dim red lights are often used at night, and the light fixtures currently used for night-time light often create low uniformity, as they are placed far apart in most dairy barns (Jakobsson, 2016; Reksen et al., 1999). High uniformity is recommended in work environments for people (CEN Technical Committee, 2021; Reinhold and Tint, 2000). Therefore, the use of nonuniformly distributed red night-time light needs to be re-assessed, indeed our results indicate that the cow traffic at night-time could benefit from having no light rather than red light of low uniformity. Newer versions of LED light fixtures can easily be dimmed, creating a uniform light environment in low light intensities. Low uniformity can also be avoided by indirect lighting, for example light fixtures facing the ceiling (Makaremi et al., 2019).

Although an even distribution of cow activity over the 24 h is preferred in systems with automatic milking, cows naturally arrange their activities in a circadian pattern, with more active behaviors such as eating and socializing occurring during the day, and more resting and sleep at night (Munksgaard et al., 2011; Kilgour, 2012; Ternman et al., 2019). Using both daylight lighting and dimmer lights at night may disturb the circadian rhythm in cows, and negatively affect their health and welfare, similar to what has been shown in people working night shifts (Costa, 2010). Light 24 h/d has negative effect on lactogenesis and lactation persistency (Stanisiewski et al., 1988), as well as melatonin secretion (Shuboni and Yan, 2010; Russart and Nelson, 2018; Tähkämö et al., 2019). Hence, the use of night lights, as well as light intensity and distribution in cow barns warrants further investigation.

In conclusion, darkness did not affect the ability of cows to navigate through a test obstacle course, but medium, nonuniform red light affected their ability to navigate. Hence, cows do not necessarily need night-time lighting to navigate, even in a context with obstacles blocking their way. Furthermore, dim red and white lighting did not alter behavior, stress indicators, or cows' ability to navigate an obstacle course, indicating that either form of light can be used in cow barns at night. However, we did not study other physiological parameters that may be affected by night-time light regimen.

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