Bedding system influences lying behaviour in dairy cows

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Leach, K.A., Charlton, G.L., Green, M.J., Lima, E., Gibbons, J., Black, D. and Bradley, A.J. (2021) 'Bedding system influences lying behaviour in dairy cows', *Veterinary Record* (e1066).

21 November 2021

VetRecord

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Journal:	*Veterinary Record*
Manuscript ID	vetrec-2021-106573.R1
Wiley - Manuscript type:	Original research
Date Submitted by the Author:	16-Sep-2021

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Abstract:	BACKGROUND The nature and depth of bedding material have an important influence on cow lying behaviour and comfort. Increasing use of recycled manure solids (RMS) as bedding led to an investigation of the influence of this material on cow lying behaviour. METHODS Leg mounted accelerometers were used to estimate daily lying time and number and duration of lying bouts in four groups of 40 cows. Each group spent two two-week periods on each of four bedding systems: deep sand, deep RMS, sawdust on mattresses and RMS on mattresses. RESULTS Total daily lying times were significantly shorter on both RMS treatments than on sawdust. Number of lying bouts per day was greater on sawdust than any other treatment, while lying bouts were 2.6 minutes longer on deep RMS and 9.3 minutes longer on sand, than on sawdust. CONCLUSIONS Greater depth and apparent softness of bedding material does not necessarily result in longer total daily lying times. RMS may have some characteristics that reduce its attraction as a bedding material for cows. The influence of bedding system on number and duration of lying bouts and the resulting total lying time appears complex.

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Running title: Bedding systems and dairy cow lying behaviour

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Acknowledgements
The authors are grateful to collaborators at Newton Rigg College (a part of Askham Bryan College) for use of farm facilities, and particularly to farm manager Jonathan Fisher, herd manager Wayne Stead and the rest of the farm staff for day to day management of the cows. Cow monitoring equipment was lent by IceRobotics.

Funding
The study was conducted as part of a project whose funding was obtained through the 'Improving the Welsh Dairy Supply Chain' project, via the Rural Development Plan for Wales 2007-2013, which was in turn funded by the Welsh Government and the European Agricultural Fund for Rural Development.

ABSTRACT

BACKGROUND

The nature and depth of bedding material have an important influence on cow lying behaviour and comfort. Increasing use of recycled manure solids (RMS) as bedding led to an investigation of the influence of this material on cow lying behaviour.

METHODS

Leg mounted accelerometers were used to estimate daily lying time and number and duration of lying bouts in four groups of 40 cows. Each group spent two two-week periods on each of four bedding systems: deep sand, deep RMS, sawdust on mattresses and RMS on mattresses.

RESULTS

Total daily lying times were significantly shorter on both RMS treatments than on sawdust. Number of lying bouts per day was greater on sawdust than any other treatment, while lying bouts were 2.6 minutes longer on deep RMS and 9.3 minutes longer on sand, than on sawdust.

CONCLUSIONS

Greater depth and apparent softness of bedding material does not necessarily result in longer total daily lying times. RMS may have some characteristics that reduce its attraction as a bedding material for cows. The influence of bedding system on number and duration of lying bouts and the resulting total lying time appears complex.

INTRODUCTION

Cow comfort is an important element of dairy cow welfare, for which farmers have responsibility. Comfort when lying is an influential aspect, as cows typically spend 12 - 14 hours per day lying down (1,2). Lying times of 12 hours per day are widely accepted as the target for housed dairy cattle (3). Understanding the conditions that provide good cow comfort relies on interpretation of cow behaviour. Development of animal-mounted sensors (“wearable technology”) has enabled automated assessment of lying behaviour and measurement of lying time (4). Individual cow data are now readily available, but require interpretation.

Cows are highly motivated to lie down and in certain situations will prioritise lying over eating (5). While lying, cows rest and ruminate, their legs and feet are rested and blood circulation through the udder increases by up to 30 per cent, all of which are beneficial for production (6) (7). Excessive standing time is considered detrimental, being a risk factor for increased lameness (8) and time spent lying is taken as an indicator of cow comfort (9).

Within the daily time budget, many factors influence a cow’s decisions on whether to lie down, and for how long. These include physiological state (10), motivation to eat (5), social interactions within the herd (11), ambient conditions, and the physical conditions of beds and bedding (12)(13)(14). It is important to understand the effects of such factors to enable optimal design of buildings and management systems, that enhance cow comfort within the confines of productive systems.

A developing interest in using recycled manure solids (RMS) as bedding in the UK led to questions about its risks and benefits. One benefit claimed for this material has been “improved cow comfort”, based on its being soft, not abrasive, deformable, and,

not least, readily available, and therefore likely to be used in generous quantities (15). The material is commonly used in one of two bed designs: in an “enclosed” bed, which has a “kerb”, “bedding retainer”, or “heel-stone”, preventing loss of bedding material from the back of the bed, (often referred to as “deep beds”), or as a covering for mats or mattresses, with no rear retainer, from which bedding is more easily lost as cows move. This generally results in a shallower covering than in an “enclosed” bed.

In an investigation of the potential risks and benefits of RMS (16), a number of aspects of cow health, behaviour and productivity were studied, when RMS was used in “deep” beds, and on mattresses, in comparison with more conventional materials, namely “deep” sand and sawdust on mattresses. This paper explores the influence of these combinations of bedding material and bed design (referred to as “bedding systems”), on lying behaviour. The hypothesis was that, in terms of cow comfort, deep beds would be better than shallow (reflected in longer lying times), deep RMS would be comparable to deep sand (similar lying times), and RMS on mattresses would be superior to sawdust on mattresses, (longer lying times). The effects on the number and duration of lying bouts were also investigated.

MATERIALS AND METHODS

Experimental herd and site

The study was conducted at Sewborwens Farm, Newton Rigg, Penrith, Cumbria, UK (Grid Reference NY 492303). The herd comprised 221 Holstein-Friesian cows with an annual mean milk yield of 9,379 litres/cow/year, and weight approximately 700 - 750 kg. The mean herd somatic cell count at the start of the study was 115,000 cells/ml. The study was entirely observational and did not involve any departure from treatment or management of cows found on commercial farms, therefore no ethical issues arose.

The study was conducted in a building approximately 6 m high (at the eaves), with doors and space boarding at both gable ends. The sides of the building were solid to a height of approximately 2 m, above which were automatically operated curtains. Cubicles with cantilever divisions were arranged in head-to-head rows, or facing a wall; all allowed forward lunge space. The base of each cubicle measured 130 cm x 250 cm with a brisket board 210 cm from the kerb. Diagonal length from neck-rail to kerb was 200 cm and the neck-rail was approximately 125 cm above the surface of the bedding. Kerb height was 15cm. Passages were 5m wide between the feed face and the cubicles and 4.5m wide between cubicle rows, with a surface of rubberised textured concrete (Flexscreed; Quattro , Penrith, UK). The study ran from 28th January to 20th May 2015.

Experimental design and cow allocation

A modified crossover design used four groups of 40 cubicles within one shed. Each group of cubicles had a different bed type: deep sand (Sand), deep RMS (Deep RMS), RMS on mattresses (RMS mat) and sawdust on mattresses (Sawdust mat). Four cow groups, each of 40 cows, were rotated around the four cubicle areas, spending two weeks on each bedding system, and cycling twice around four treatments (16 weeks

in total). On each cycle, all groups moved around the treatments in the same order, although starting on a different treatment (Table 1). Apart from the bedding system, all groups were under the same management regime, and received the same diet.

Prior to the study all cows had been kept in the building used for the research, in cubicles with deep RMS beds, of the style used in the trial. Cows were initially blocked, by parity and days in milk, prior to being randomly allocated, within blocks, to one of four groups (Red, Green, Blue, Yellow), using random number generation (Microsoft Excel). There were no significant differences between the allocated groups in yield, days in milk or parity at the start of the study (Table 2). On the fortnightly "changeover days", when the group moved to the next treatment, any cows due for drying off were removed and replaced with freshly calved cows entering the next parity. The four cows that were withdrawn during a treatment period, due to injury or illness, were replaced with cows of equal parity.

Bed design and bedding materials

In deep beds, a layer of sand or RMS covered a hardcore base. The top of the rear retaining kerb of each cubicle was typically between 1 and 4 cm above the surface of the bedding material. There were no dividing structures on the base or near the bed surface between cubicles; the surface was continuous across a row of cubicles. The depth of bedding was approximately 8-10 cm at the rear of the cubicles and greater at the front. The mattress beds were created from existing deep beds, which were filled with hardcore and concrete to recreate conventional concrete cubicles to which the mattresses (35 mm thick, Luxury Mattress; Quattro, Penrith, UK) were fitted during the week prior to the trial. The deep sand beds were created by replacing existing RMS with sand two days before the start of the study.

Quarry sand obtained locally was stored outdoors, uncovered. Fine powdered kiln dried sawdust was obtained in sealed bags which were stored under cover.

RMS was produced from the slurry scraped from the study area, excluding the sand cubicles, and an adjacent shed housing approximately 30 additional milking cows. This slurry, along with water from washing the parlour and milking plant (but no whole milk) was collected in a reception pit (volume 100 cubic metres). To make the bedding material, following agitation, slurry was pumped from this pit through a FAN Press Screw Separator F10113782 (PSS 3.3-780) with standard 1mm screens. Bedding was prepared on two days per week. Dry matter (DM) content of the material was monitored subjectively, by feel, aiming for a DM content of at least 35 per cent, based on the operator's previous experience of regular assessment by oven drying. Any material considered too wet was discarded. The material was collected under cover, beneath the separator, and applied to beds within four hours of separation.

Bed and passageway management

Sand cubicles received fresh bedding every two weeks, RMS beds twice a week and sawdust beds twice daily, representing typical UK farm practice. More RMS was delivered to the front of the cubicles to allow for movement back over the following

days. Twice a day, when cows were absent for milking, all beds were inspected, any dung removed, and bedding raked from front to back of cubicles. The deep RMS beds were levelled twice a week using a tractor-mounted rake. All passageways were scraped twice daily, by a tractor-mounted scraper.

Data and sample collection

Cow milking and individual cow yields

Cows were milked twice daily, preparations beginning at 4.30 am and 1.30 pm. The order in which cow groups were milked altered daily, following a predetermined order generated using random number tables. Individual cow yields were measured at each milking using Fullwood meters and collated into Fullwood Crystal software. On the day prior to the end of each two-week period, the average yield for the previous ten days was calculated for each cow.

Cow behaviour data collection

On entering the study, cows were fitted with an IceQube™ sensor (IceRobotics, Edinburgh, UK) on the left hind leg. Three dimensional accelerations were collected at 4Hz and summarised in 15 min intervals on the sensor. Data were downloaded via IceReader™ and reported via IceManager™ software (IceRobotics, Edinburgh, UK). For each cow and for each 15 minute interval, time lying, time standing, number of lying bouts, and number of changes of position from standing to lying were reported. Prior to analysis, data were cleaned by removing potentially spurious lying bouts lasting less than 33 seconds, using a threshold established for IceQubes on cows (4). The cleaned data were used to calculate a daily summary of total time spent lying, and the number and mean duration of lying bouts for each cow on each calendar day. To allow a period of adaptation to each treatment, only data from days seven to 13 of each treatment period were analysed. For each cow group, on each day, the time when no cows in the group were lying, around milking time, was calculated and designated “time standing for milking”.

Environmental monitoring

Temperature and relative humidity were monitored hourly inside the shed using wall mounted Digitron Monolog2® data loggers.

Dry matter and depth of bedding material

Once during each treatment period a sample of fresh and used bedding of each material and from each bed type was taken for determination of DM content by oven drying. “Used” bedding samples comprised a pooled sample from ten separate cubicles, taken immediately prior to replenishment of bedding, following a standard protocol for position and depth of the sample. At the same time, the depth of bedding was measured, at the sampling site, which was towards the rear of each cubicle, in the region of contact with the udder. These samples and measurements were taken on Day 6 of each treatment period. Time from previous replenishment was between 6 and 12 days for sand, 3 days for RMS and 4 hours for sawdust.

Statistical power

Power calculations were conducted to provide an estimate of the required sample size. Calculations were conducted without the inclusion of repeated measures in the study design to minimise the assumptions needed and to provide a conservative (safe) estimate of cow numbers required. Based on assumptions of 20 minutes' difference between groups in daily lying time and a standard deviation in lying time of 30 mins per day, the sample size needed with a power of 0.8 and significance probability of 0.05 was a minimum of 36 cows per group. This was rounded to 40 cows per group to provide a safety margin.

Statistical analysis

Data were collated in Excel and descriptive statistics produced in Minitab 13. Data were analysed with 'cow-day' as the baseline unit of observation with repeated measures nested within cows. The outcome variables of interest were "total lying time per 24 hours", "number of lying bouts per 24 hours" and "mean length of lying bout per 24 hours". Mixed effect modelling was conducted using the statistical package R (v 3.6, http://www.R-project.org, R Development Core Team, 2009). Models were built using the lme4 package of R v3.6 (17)

Models took the form:

$$y_{ij} = \beta_0 + \sum_1^p \beta_j X_j + \sum_1^m \beta_{ij} X_{ij} + u_j + e_{ij}$$

where y_{ij} was the outcome for the ith reading from the jth cow, β_0 the model intercept, X_j represented one of p cow level covariates (e.g., parity) with estimated coefficients β_j, X_{ij} represented one of m cow-day level covariates (e.g., yield, time standing milking) with estimated coefficients β_{ij}, u_j was a random effect for cow, assumed to be normally distributed with mean = 0 and variance = σ_u and e_{ij} represented the residual model error, assumed to be normally distributed with mean = 0 and variance = σ_e.

The key explanatory variable of interest was bedding system. Other potentially confounding fixed effects tested in the models were: cow group, treatment period (a dummy variable representing the first or second rotation round treatments), time standing for milking, parity and mean yield over the previous 10 days. Significance probability was set at 0.05 and confounders were retained in the model when they resulted in a biologically meaningful change in interpretation of the coefficient for bedding system.

Model fit was assessed using conventional residual analysis including quantile-quantile plots of standardised residuals, evaluation of homoscedasticity and assessment of the influence and leverage of outlying data points.

RESULTS

Cow numbers and balance between experimental groups of cows

A total of 203 cows were included in the trial. Forty-three replacements were made, 39 of these on changeover days as cows became due for drying off. Four cows were replaced during a treatment period due to illness (one case of pneumonia, one of acidosis) or leg and back injuries, unrelated to the bedding treatments. Among cows that completed the trial, ten cows were treated for mastitis (one in the blue group, two in yellow, three in red and three in green). Two cows were treated for lameness but remained in the study, one in the yellow group with foul in the foot, and one in the green group with undiagnosed front foot lameness.

There were no significant differences between the allocated groups with respect to yield, individual cow somatic cell count, days in milk or parity at the start of the study (Table 2) or during subsequent experimental periods. During the trial, ten-day average yields, collated for each group at the end of each two-week treatment block, did not vary significantly between the treatment groups (P=0.714). Mean yields were 34.0, 34.4, 34.4 and 33.5 litres per day for cows bedded on deep RMS, sand, sawdust on mats and RMS on mats respectively.

Descriptive statistics of main outcome variables

Individual observations of lying time, number of lying bouts and bout duration were available for 8783 cow days. The distributions of the three outcome variables - lying time, number of lying bouts and bout duration - are illustrated in Figures 1 - 3. Across all treatments, daily total lying time ranged from 125 to 1182 minutes per day (2.1 to 19.7 hours per day), mean 686 minutes (11.5 hours), median 693 minutes (11.6 hours). Number of lying bouts per day ranged from 2 to 34 (mean 10.1, median 10.0). Daily mean bout length ranged from 10.8 minutes to 295 minutes, mean 72.5 minutes, median 69.8 minutes.

Ambient conditions

Daily mean temperature in the shed ranged between 2.6 ^{o}C and 9.8 ^{o}C in Period 1 and 5.6 ^{o}C and 13.3 ^{o}C in Period 2. (Period 1 absolute min -3.6 ^{o}C and max 17.5 ^{o}C; Period 2 absolute min 1.5 ^{o}C and max 19.1 ^{o}C). The mean temperature was lower in Period 1 (6.4 ^{o}C) than in Period 2 (9.9 ^{o}C) (P< 0.001). Mean Relative Humidity was higher in Period 1 (83.0%) than in Period 2 (76.0%) (P<0.0001).

Bedding Dry Matter content & bedding depth

Results for DM content of fresh & used bedding samples, and depth measured at the time of sampling, are reported in Table 3.

Statistical models

The number of observations available for the final models was restricted to 8677 by the absence of one two-week treatment period's yield data for eight cows, and two two-week treatment periods' yield data for one cow. These omissions appeared to occur at random. The mean and standard error for each of the outcome variables, by treatment, are illustrated in Figures 4 - 6. The full model results are shown in Table 4 (total lying time), Table 5 (number of lying bouts) and Table 6 (lying bout duration). Cow group was not significant and did not influence the outcome of the models, so was not included.

Total lying times were shorter on Deep RMS (by 23.6 min, P<0.001)) and on RMS mat (by 13.1 min, P<0.001) than on Sawdust mat. Lying times on Sand and Sawdust mat did not differ significantly (Figure 4, Table 4).

The number of lying bouts was significantly less on all other treatments than on Sawdust mat. The difference between Sawdust mat and both RMS treatments was less than one bout but cows on sand had 1.36 fewer lying bouts per day than those on sawdust (Figure 5, Table 5). The number of lying bouts across all bedding systems was increased by 0.29 in Period 2 compared with Period 1. The effects of yield and time standing milking were statistically significant but numerically very small (Table 5).

Lying bouts were longer on Deep RMS (by 2.6 min, P<0.001) and on Sand (by 9.3 min, P<0.001) than on Sawdust mat (Figure 6, Table 6). There was a very small but statistically significant negative effect of increasing yield on length of lying bouts (P<0.01) (Table 6).

DISCUSSION

The study aimed to compare lying behaviour of dairy cows on a novel bedding material (RMS), used in two different bed designs, with more commonly used materials, to investigate whether RMS could be considered a "comfortable" bedding material. It was hypothesised that lying times would be longer with deeper and softer bedding, as these features were assumed to provide superior cow comfort, reflected in longer lying times.

Surprisingly, lying times were longest with the shallowest bedding treatment (sawdust) and shortest on a deeper, enclosed bed treatment (Deep RMS), and there was no significant difference in lying times between sawdust and deep sand. Deep sand beds are generally considered to provide the ultimate in comfort, and encourage cows to lie down. Current American recommendations for sand depth are 6-8 cm (18). Comparisons between farms have demonstrated longer lying times on sand than on sawdust (19)(20) and on "deep bedding" (not defined), versus "concrete, mattress, rubber mat, and so on" (21). However, in Ito's study (21), "deep bedding" was associated with other management factors, including newer barns, which might also

have promoted longer lying times. On the other hand, there is evidence of cows lying for shorter times on sand beds, compared with deep straw on concrete, or straw on mattresses (Manninen et al 2002). In the current study, it was also surprising that longer total lying times were identified for cows on RMS on mattresses, compared with deeper RMS, in view of reported preferences for deep beds of sawdust compared with sawdust on mattresses (22). It is possible that the drier nature of the thinner layer of RMS made it more attractive to cows than the damper conditions of the deep RMS (23).

The pattern was reversed in terms of lying bouts, with significantly fewer lying bouts on sand and Deep RMS than on sawdust, which might indicate better comfort (24). These bouts were also longer on the deeper beds. Some authors would consider this to indicate greater comfort (12) (19). Despite this, total lying time was shorter on these bedding systems.

Comparing the two designs of RMS beds, lying bouts were 2.5 minutes longer on deep RMS than on RMS mat. Longer bouts might suggest either that cows became more settled, or they found it harder to change position, on the deeper RMS, contributing to the longer lying time recorded here. The current finding agrees with previous research that reported longer lying bouts on deep beds than shallow (19), and on sand compared with mattresses covered with 5mm sawdust (25). Our results fall within the spectrum of mean bout lengths reported by others, e.g., 1.3 hours on deep sand, 1 hour on shallow sand (14), 1.5 hours on sand, 47 minutes on mattresses with sawdust (19).

The relative benefits of short and long lying bouts, number of bouts, and the interactions between these is difficult to unravel, and much is open to interpretation, particularly whether longer bouts indicate greater comfort (24) or reluctance to get up and change position, which might indicate discomfort (26) (27). Deeper analysis of the duration and time sequence of consecutive standing and lying bouts, and the relationship between these, will be needed for a better understanding.

Some findings were surprising and deserve further consideration. It is possible that the relatively short lying times on RMS compared to sawdust arise from characteristics that make RMS less attractive for cows to lie on, even when provided in a deeper layer. Such characteristics might include dampness (23), smell, ammonia released, pH, heat caused by microbial activity or transferred from the cow's body and stored, or even recognition of faecal material. Cows have been shown to prefer lying on drier bedding (28) (13) and clean surfaces (14). Sawdust was the material replenished most frequently in our comparison of "bedding systems" (representing common practice with the various materials in the UK), so may have been perceived by cows as the "cleanest" bedding. Sawdust also had a higher DM content than RMS; deep RMS remained at lower DM content than RMS on mattresses.

The mattresses in this study were new, and likely to have been softer, more deformable and potentially more comfortable than older mattresses or mats, which might partially explain the lack of advantage of the deeper beds here. In a recent choice experiment, cows spent more of their time lying on new mattresses than on sand, in cubicles (29). Other possibilities for limited lying time in the sand cubicles are the nature of the sand, exposure of the kerb at the rear of the cubicle (30), and cows'

unfamiliarity with sand. Cubicles were only replenished with sand every 14 days, and lowering of the bedding surface level to expose 5cm height of kerb prior to re-bedding may have reduced comfort. Lying time has been shown to decline by 10 min for every 1 cm drop in sand level below the kerb (30). The enclosed beds contained less bedding than in some reports describing “deep” beds (e.g., Tucker et al, 2003 compared 30 - 40 cm bedding with 2-3 cm on mats (14)) However, the depth of bedding in enclosed beds and on mattresses was statistically and, we believe, biologically, different.

It is possible that there could have been some interaction between air temperature, relative humidity and the thermoregulatory properties of the surfaces, for example with the sand (if it was colder) or the RMS (being wetter). Cows might have found some surfaces more challenging than others for maintenance of thermoregulation in certain conditions. Longer lying bouts could perhaps be explained by cows lying for longer once the surface had been warmed up. However, this is speculation in the absence of data on surface temperature of bedding.

Although total lying time is widely considered to reflect comfort, the combination of bout number and bout length is also likely to be important (31). In other studies, fewer bouts of longer duration have combined to give greater total lying time on beds that would appear to be more “comfortable” (21), however, it was not the case here. This may be a result of the within-farm comparison, in contrast to a between-farm comparison (21). Treatments within a farm are more likely to represent bed features only, while at farm level other factors will introduce influence and potentially create confounding. Alternatively, the human view of apparently comfortable conditions may not coincide with the cows’ experience. Also, other intrinsic and extrinsic factors must be considered when interpreting measures of lying and standing times and bouts in the assessment of cow comfort and animal welfare.

A limitation of the current study was that, for practical reasons, each treatment (bedding system) remained in the same position within the shed. It is possible that localised influences of environmental conditions, such as draughts, or disturbance due to daily routines, confounded any bedding treatment effect but local environmental conditions were not recorded. Also, for 14 months prior to the study, cows were bedded on deep RMS, so initially unfamiliarity with sand and mattresses might have made them reluctant to lie down on these. However, a one-week adaptation period on each treatment was given, to reduce any such effect, and the results did not reflect a preference for lying on a familiar material. Cows did not have any apparent difficulty in adapting to lying on sawdust on mats, as this was where they lay longest. The new experience of moving on shifting sand might have affected their behaviour on this treatment (32). Because of the timing of the trial, ambient temperatures increased between period 1 and period 2, so it would be difficult to separate any effects of familiarity and environmental influences.

In conclusion, equal or higher daily lying times were achieved on new mattresses frequently replenished with sawdust, compared with enclosed, relatively deep beds, of either sand or RMS. However, the length of each lying bout was longer on RMS and sand compared to sawdust. Greater understanding of the interaction between number and duration of bouts of lying and standing, and interpretation of these in terms of cow comfort and welfare, is needed. RMS may have some characteristics that reduce

cows' lying times, and relationships between properties of bedding materials, environmental conditions and cow behaviour should be further investigated.

Acknowledgements
The authors are grateful to Newton Rigg College (a part of Askham Bryan College) for use of farm facilities, particularly to farm manager Jonathan Fisher, herd manager Wayne Stead and other farm staff for management of the cows. Cow monitoring equipment was lent by IceRobotics.

Funding
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Conflict of Interest
The authors declare there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

The author Eliana Lima is employed with the European Food Safety Authority (EFSA) in the Unit ALPHA that provides scientific and administrative support to EFSA's Scientific Activities in the area of animal health and welfare. However, the present article is published under the sole responsibility of the author Eliana Lima and may not be considered as an EFSA scientific output. The positions and opinions presented in this article are those of the author alone and are not intended to represent any views/any official position or scientific work of EFSA. To know about the views or scientific outputs of EFSA, please consult its website under http://www.efsa.europa.eu.

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Table 1 Experimental design - rotation of four experimental groups of cows around four bedding system treatments over two experimental periods

Cow group		Green	Blue	Yellow	Red
Period	**Start date**				
1	25/01/2015	Deep RMS*	RMS mat	Sawdust mat	Sand
1	11/02/2015	Sand†	Deep RMS	RMS mat	Sawdust mat
1	25/02/2015	Sawdust mat‡	Sand	Deep RMS	RMS mat
1	12/03/2015	RMS mat§	Sawdust mat	Sand	Deep RMS
2	25/03/2015	Sawdust mat	Sand	Deep RMS	RMS mat
2	08/04/2015	RMS mat	Sawdust mat	Sand	Deep RMS
2	22/04/2015	Deep RMS	RMS mat	Sawdust mat	Sand
2	06/05/2015	Sand	Deep RMS	RMS mat	Sawdust mat

* Recycled manure solids in an enclosed deep bed

† Sand in an enclosed deep bed

‡ Sawdust on mattress

§ Recycled manure solids on mattress

Table 2 Mean (and SD) of parameters by cow group at the start of the trial

Cow group	Green	Blue	Yellow	Red	P value for ANOVA by group
Parity	2.31 (1.326)	2.25 (1.345)	2.46 (1.349)	2.41 (1.574)	0.994
Days in milk	165.2 (104.7)	159.4 (109.4)	180.7 (105.4)	181.7 (109.8)	0.989
Yield (l/day - 10 day average prior to group allocation)	35.0 (10.54)	34.6 (10.69)	31.6 (10.00)	32.7 (11.00)	0.435
Individual cow somatic cell count Jan 2015	108.9 (128.8)	93.2 (166.6)	102.9 (195.2)	108.9 (128.8)	0.593

Table 3. Bedding dry matter and minimum bedding depth for the four bedding systems: mean (and SD) of fortnightly measurements during the sixteen week trial*

Bedding system	Dry Matter (%)		Bedding depth (cm)†
	Fresh	Used‡	
Recycled Manure Solids (RMS)	33.5 (4.37)		
Deep RMS bed		50.5 (5.13)	6.8 (1.29)
RMS on mattress		55.7 (8.25)	1.36 (0.48)
Sawdust	92.2 (1.08)	78.9 (4.93)	0.89 (0.29)
Sand	91.8 (2.09)	94.4 (2.35)	5.5 (1.15)

* Measurements made / samples taken in ten cubicles per treatment once every two weeks

† Depth measured immediately prior to replenishment, in the region of the cubicle in contact with the udder

‡ Samples taken immediately prior to replenishment from the top 2.5 cm, in the region of the cubicle in contact with the udder

Table 4 Final multivariable model for factors associated with mean total daily lying time in minutes per day, for cows on four different bedding systems, over two time periods. n observations = 8677

RANDOM EFFECTS

		Variance	SD
Cow (n = 194)	(Intercept)	8957	94.64
Residual		8371	91.42

FIXED EFFECTS

Variable	Level	ß	SE	t-value	P
Intercept		696	15.3	45.61	
Bedding system	Sawdust on mattress	Reference			
	Deep RMS*	-23.6	2.82	-8.213	<0.001
	Sand	-1.38	2.79	-0.49	
	RMS* on mattress	-13.12	2.82	-4.60	<0.001
Period	1	Reference			
	2	15.8	2.40	6.57	<0.001
Time standing for milking (minutes)		-0.08	0.02	-3.60	<0.001
Yield (l)		-0.12	0.380	-0.316	

* Recycled Manure Solids

Table 5 Final multivariable model for factors associated with number of lying bouts per day for cows on four different bedding systems, over two time periods. n observations = 8677

RANDOM EFFECTS

		Variance	SD
Cow (n = 194)	(Intercept)	5.268	2.295
Residual		4.015	2.148

FIXED EFFECTS

Variable		ß	SE	t-value	P
Intercept		9.93	0.360	27.27	<0.01
Bedding system	Sawdust on mattress	Reference			
	Deep RMS*	-0.66	-0.066	-9.924	<0.001
	Sand	-1.36	0.066	-20.37	<0.001
	RMS* on mattress	-0.15	0.066	-2.304	<0.05
Period	1	Reference			
	2	0.29	0.057	-5.093	<0.001
Time standing for milking (minutes)		-0.002	0.0005	-4.846	<0.001
Yield (l)		0.022	0.009	2.441	<0.05

* Recycled Manure Solids

Table 6 Final multivariable model for factors associated with daily mean duration of lying bouts in minutes for cows on four different bedding systems, over two time periods. n observations = 8677

RANDOM EFFECTS

Groups			**Variance**	**SD**
	Cow (n = 194)	(Intercept)	215.4	14.68
	Residual		286.4	16.92

FIXED EFFECTS

Variable		**ß**	**SE**	**t-value**	**P**
Intercept		75.60	2.57	29.79	<0.001
Bedding system	Sawdust on mattress	Reference			
	Deep RMS*	2.56	0.522	5.283	<0.001
	Sand	9.321	0.522	18.004	<0.001
	RMS* on mattress	-0.421	0.521	-0.806	0.29
Period	1	Reference			
	2	-0.265	0.439	-0.605	0.44
Time standing for milking (minutes)		0.0046	0.0041	1.115	0.14
Yield (l)		-0.191	0.065	-2.934	<0.01

* Recycled Manure Solids

Figure legends

Figure 1: Distribution of total lying time per day in minutes for 194 individual cows on four different bedding systems over sixteen weeks

Figure 2: Distribution of number of lying bouts per day for 194 individual cows on four different bedding systems over sixteen weeks

Figure 3: Distribution of daily mean duration of lying bouts in minutes for 194 individual cows on four different bedding systems over sixteen weeks

Figure 4: Mean total daily lying time in minutes by treatment for 194 cows, each spending two two-week periods on each of four different bedding systems - mean and SEM. Deep RMS- recycled manure solids in an enclosed deep bed; RMS mat - recycled manure solids on a mattress; Sand - sand in an enclosed deep bed; Sawdust mat - sawdust on a mattress.

Figure 5: Mean number of lying bouts per day by treatment for 194 cows, each spending two two-week periods on each of four different bedding systems - mean and SEM. Deep RMS- recycled manure solids in an enclosed deep bed; RMS mat - recycled manure solids on a mattress; Sand - sand in an enclosed deep bed; Sawdust mat - sawdust on a mattress.

Figure 6: Daily mean duration of lying bouts by treatment for 194 cows, each spending two two-week periods on each of four different bedding systems - mean and SEM. Deep RMS- recycled manure solids in an enclosed deep bed; RMS mat - recycled manure solids on a mattress; Sand - sand in an enclosed deep bed; Sawdust mat - sawdust on a mattress.

Figure 1: Distribution of total lying time per day in minutes for 194 individual cows on four different bedding systems over sixteen weeks

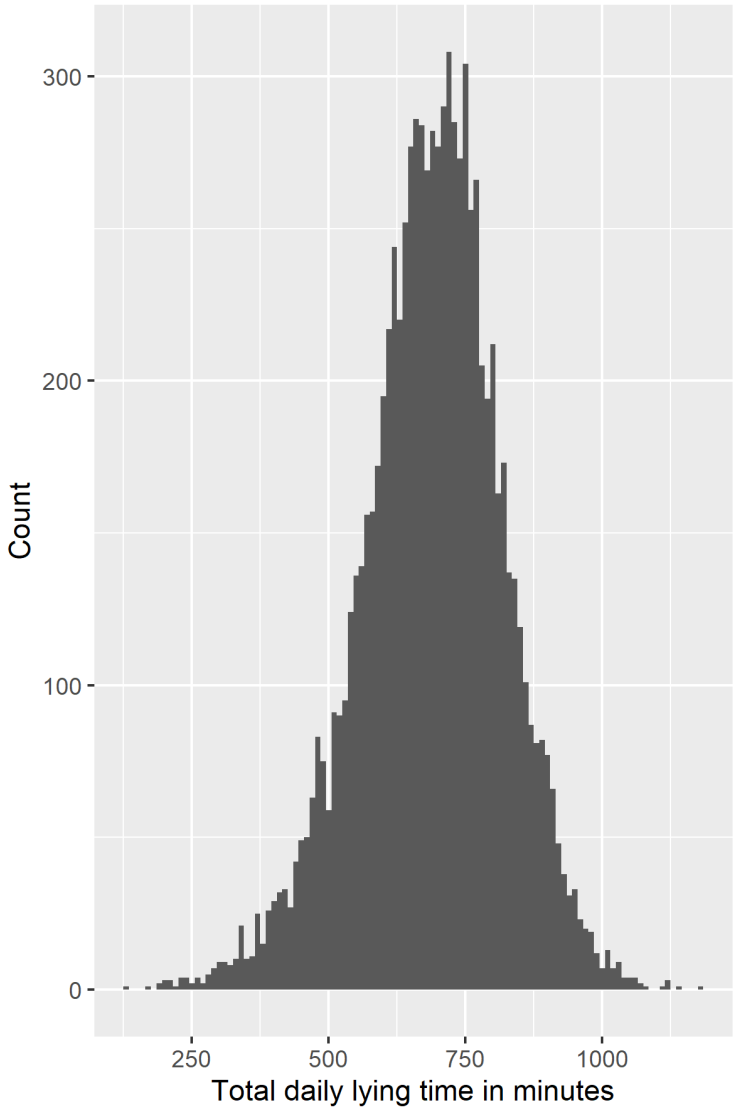


Figure 2: Distribution of number of lying bouts per day for 194 individual cows on four different bedding systems over sixteen weeks

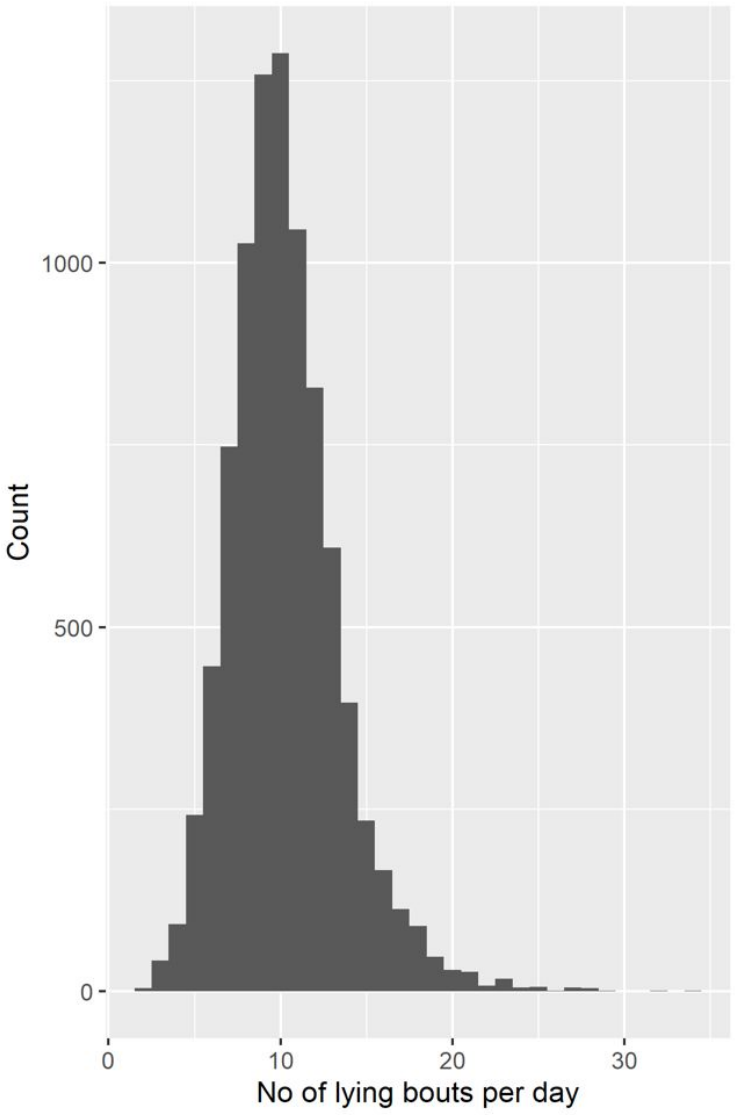


Figure 3: Distribution of daily mean duration of lying bouts in minutes for 194 individual cows on four different bedding systems over sixteen weeks

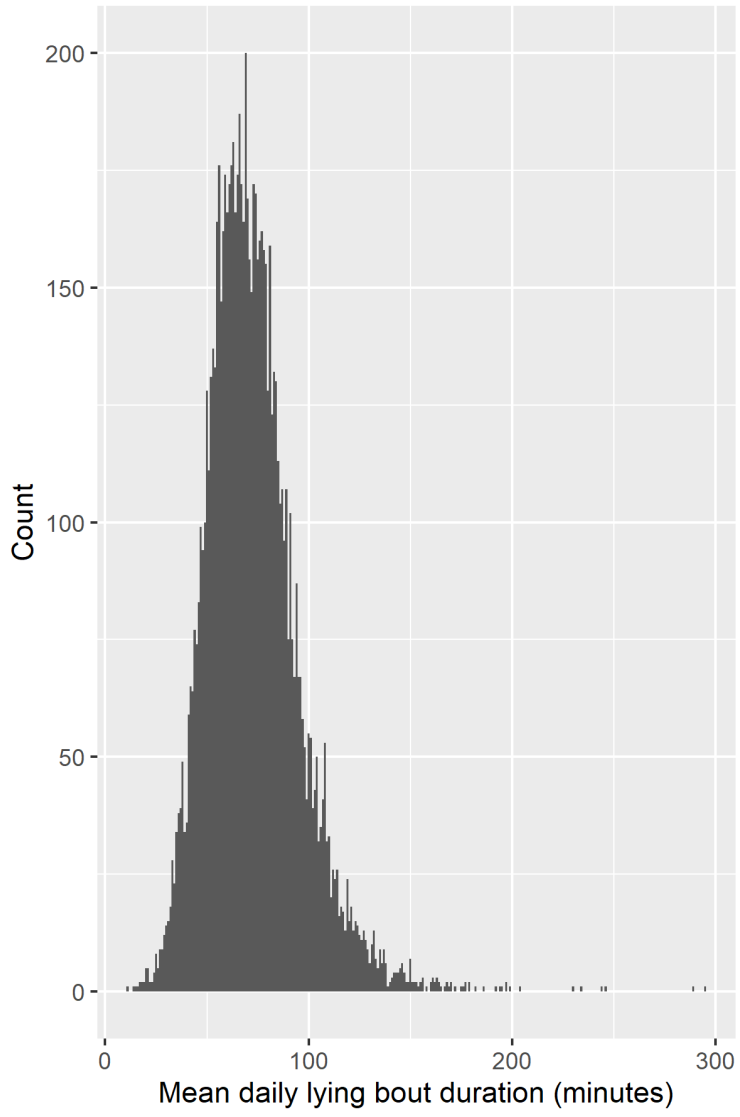
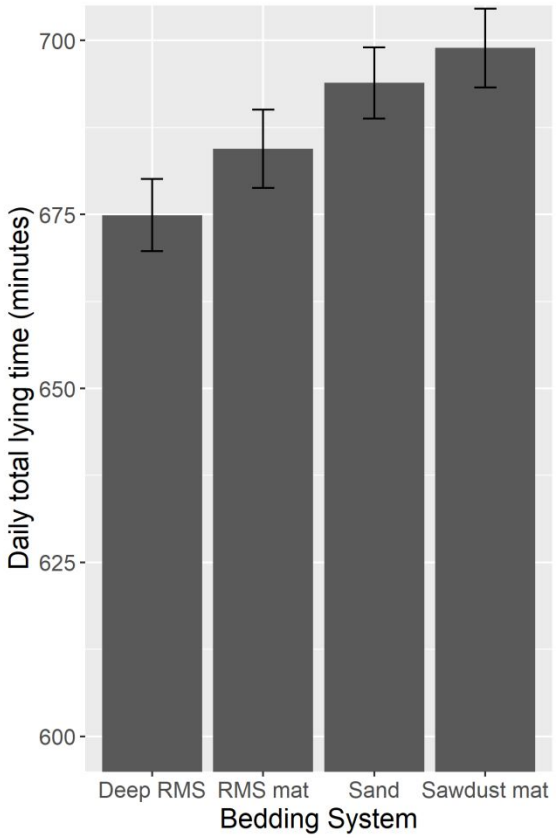


Figure 4: Mean total daily lying time in minutes by treatment for 194 cows, each spending two two-week periods on each of four different bedding systems - mean and SEM. Deep RMS- recycled manure solids in an enclosed deep bed; RMS mat - recycled manure solids on a mattress; Sand - sand in an enclosed deep bed; Sawdust mat - sawdust on a mattress.



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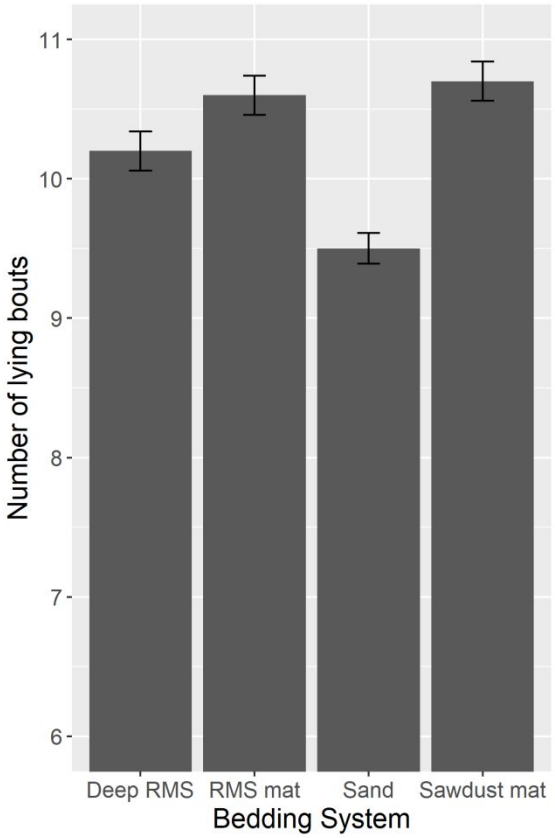
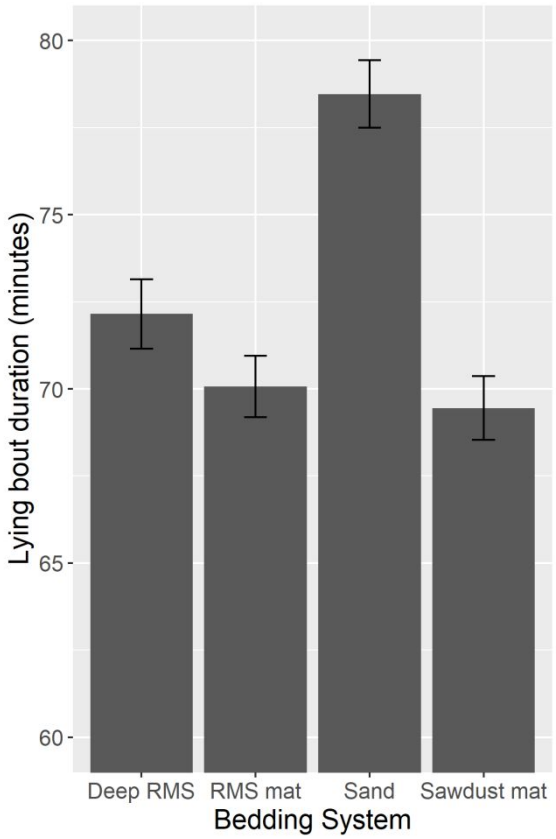


Figure 6: Daily mean duration of lying bouts by treatment for 194 cows, each spending two two-week periods on each of four different bedding systems - mean and SEM. Deep RMS- recycled manure solids in an enclosed deep bed; RMS mat - recycled manure solids on a mattress; Sand - sand in an enclosed deep bed; Sawdust mat - sawdust on a mattress.



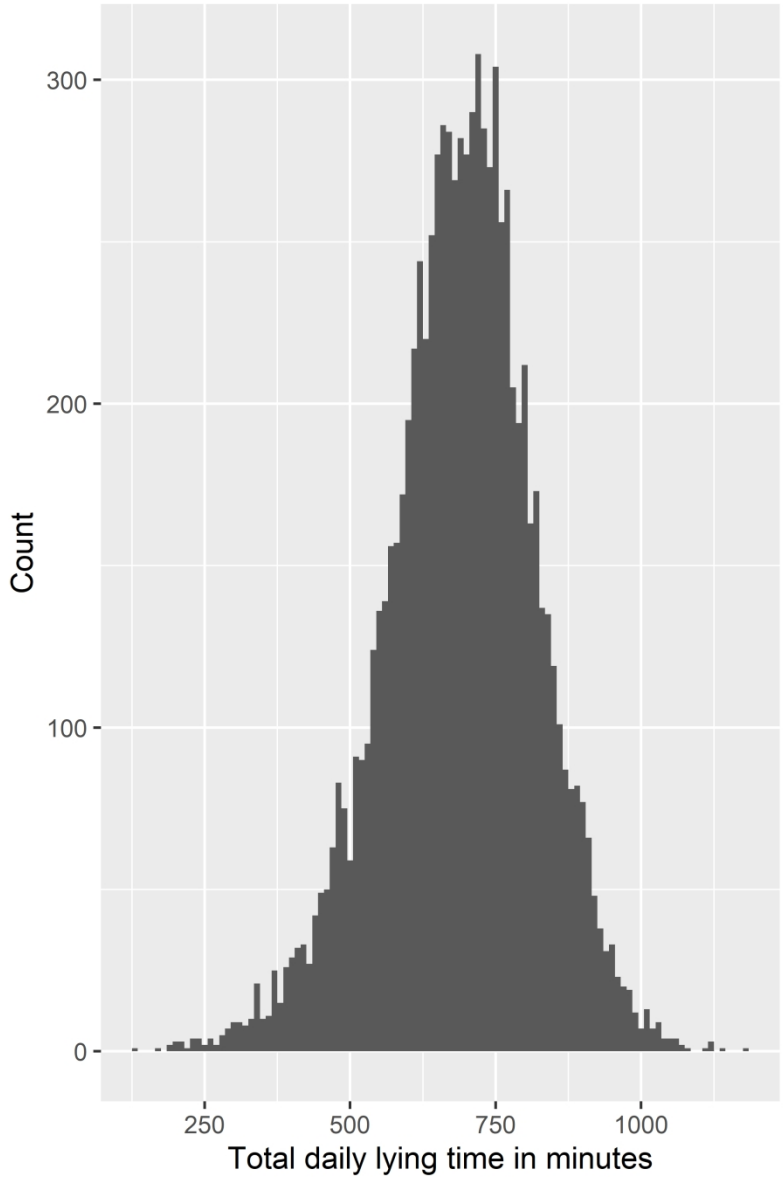
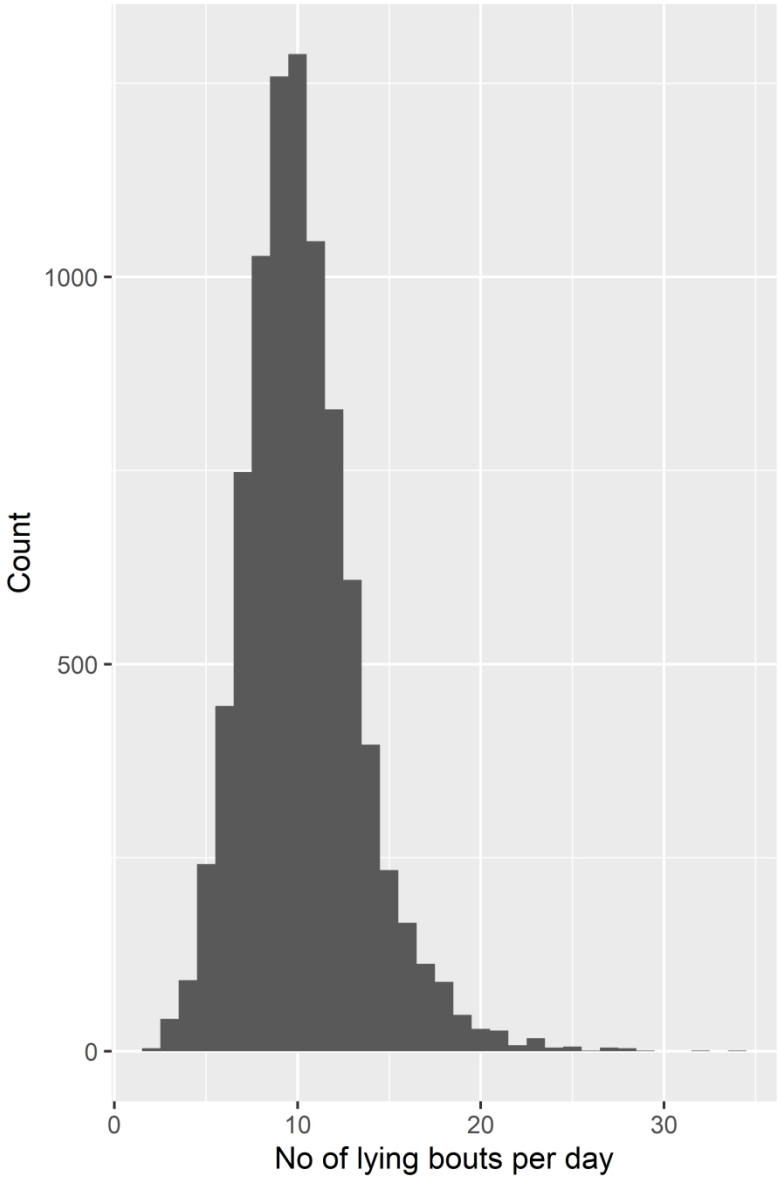


Figure 1: Distribution of total lying time per day in minutes for 194 individual cows on four different bedding systems over sixteen weeks



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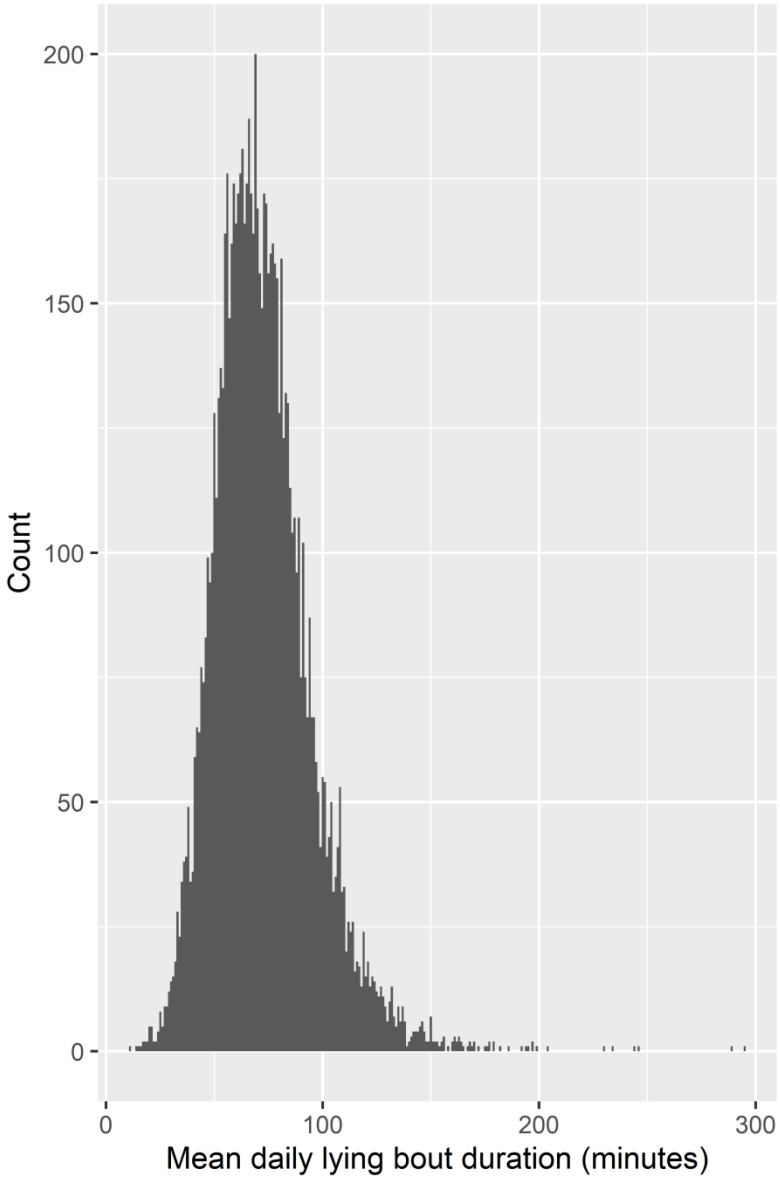

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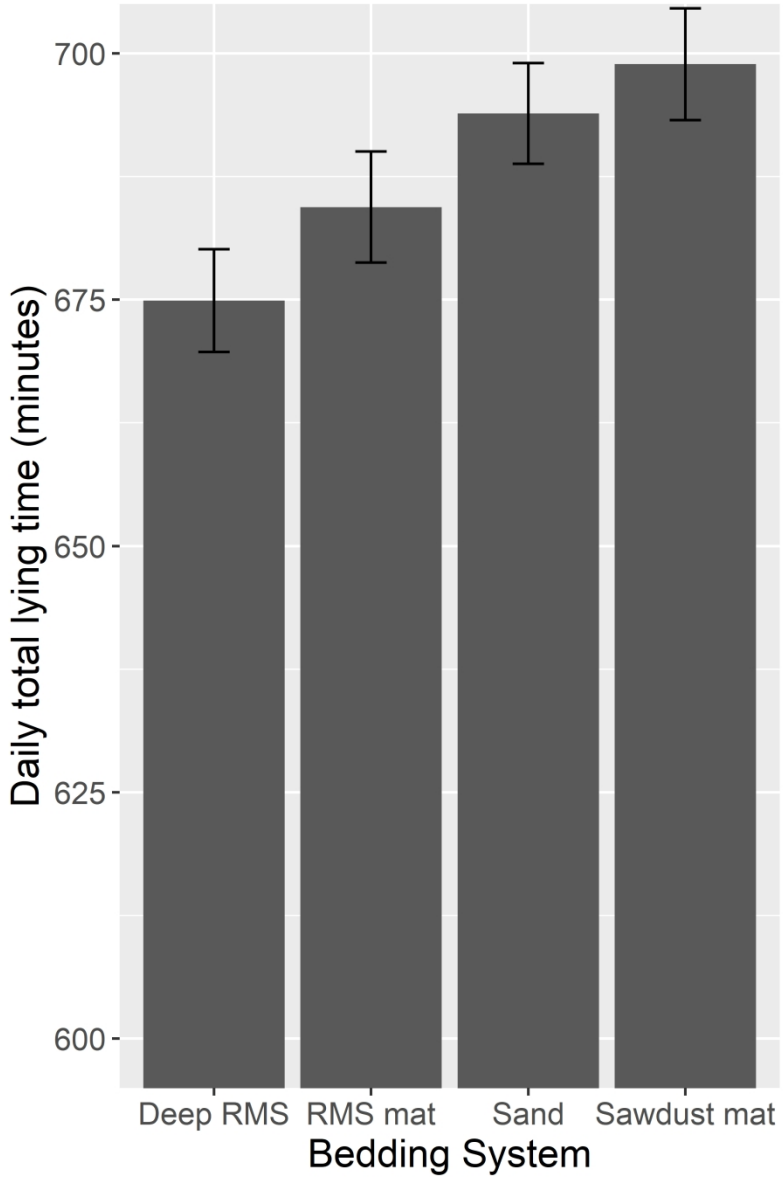
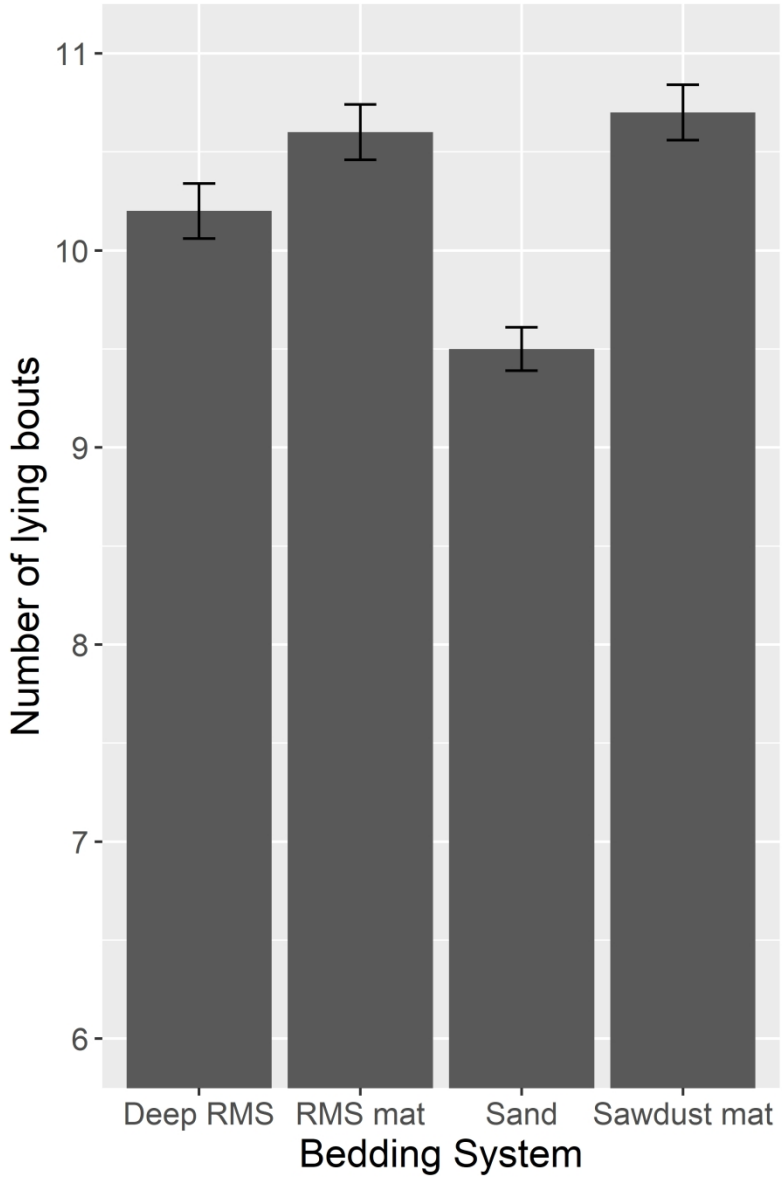


Figure 4: Mean total daily lying time in minutes by treatment for 194 cows, each spending two two-week periods on each of four different bedding systems - mean and SEM. Deep RMS- recycled manure solids in an enclosed deep bed; RMS mat - recycled manure solids on a mattress; Sand - sand in an enclosed deep bed; Sawdust mat - sawdust on a mattress.



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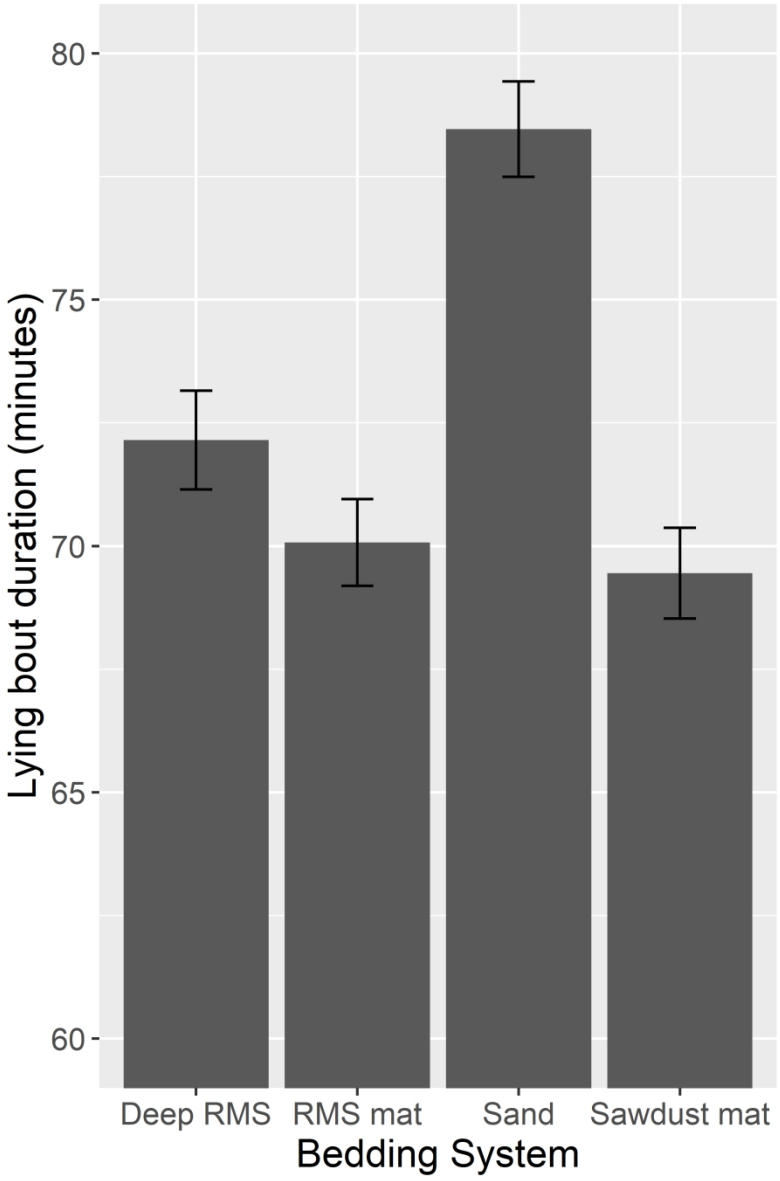

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