#### **Original Paper**

# New trends in the use of recycled manure solids in dairy housing

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The aim of the study was to compare improved bedding composition with conventional straw bedding under farm conditions, regarding its effects on the indicator microorganisms influencing hygiene levels. Dairy cows were housed in newly-built stables divided into two parts each with four sections and bedded cubicles arranged in three rows. In the first part, the bedded cubicle floors were layered with straw up to a height of 20 cm. In the second part, the cubicles were layered to a height of 20 cm with improved bedding composition consisting of recycled manure solids (RMS; 10 kg), ground limestone (200 kg), straw (40 kg) and water (110 l). After layering the bedding was treated with a concrete selector to provide strength and sufficient resistance. Samples for microbiological examination were taken from four sections according to the time interval of production and bedding. From three sections were taken bedding samples according to a new recipe in intervals of 1–3 months after its use. A control sample of bedding, consisting of straw, was taken from the last, fourth section. Comparing classical straw bedding with the improved recipe bedding, the total viable count (TVC) and coliforms bacteria (CB) in a first and second month after laying were found to be reduced. In addition to TVC and CB, decreased numbers of faecal coliforms (FC) and faecal streptococci (FS) were observed in the first, second and third months after layering. In addition to reducing the number of microorganisms, the improved bedding showed a stabilizing effect by keeping the litter thickness up to the bedding threshold (200 mm), which has a beneficial effect for increased purity of the body and udder.

Keywords: dairy cows, housing, hygiene, improved bedding, microorganisms

## 1 Introduction

Housing of cows and ensuring of optimum conditions for achieving their production potential belongs among the basic zootechnical factors. For individual categories of cattle, various housing systems have been introduced in practice, such as free housing in cubicles or housing in group pens. Loose housing in cubicles is used mainly in dairy farming. This housing can also be used successfully for other age categories of cattle. It keeps the animals clean, which is especially important for dairy cows and also addictive to heifers. It provides sufficient comfort for resting and minimizes the risk of development of dominant behaviour as well as mutual interference between animals (Mariano, 2014).

These advantages of lying boxes are achieved only with the right choice of their dimensions according to the body frame of the cattle and sufficient hygiene quality of the litter (Popescu et al., 2014). In the lying cubicles there must

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be sufficient space not only for comfortable resting but also for getting up and lying down. Otherwise, if the cows do not feel comfortable in the boxes they are looking for another place to lie down, e.g. in the areas of the feeding and manure removal passages or by the drinkers. If this is the case, the cows become excessively dirty and the risk of mammary gland inflammation and hoof disease increases. Boxes with recessed floors are best suited for underlayment (Brouček et al., 2015).

Straw is the most widely used bedding material in boxes, but its durability and hygiene effect are often insufficient. In addition to the use of straw, breeders search for alternative materials that will ensure the comfort of dairy cows and sufficient hygiene. Alternative materials suitable for undercoating include a separate, sand, limestone or zeolite (Bradley et al., 2018).

The aim of this study was to investigate the effects of improved composition of bedding used in dairy farm conditions on the indicator microorganisms influencing the level of hygiene, and compare these to the properties of conventional straw bedding.

# 2 Material and methods

## 2.1 Cows and housing

The practical part of the study was carried out in the breeding of 300 dairy cows of the Slovak Spotted Cattle breed in district Stará Ľubovňa. The dairy cows are housed in a newly built high-air stall divided into two sections, between which there is a feeding corridor. Each section is divided into 4 sections. In each section there are 42 cubicles, which are arranged in three rows. Between the rows there are movement areas with automated excrement cleaning, into which dairy cows can be moved as needed to perform technological tasks. The boxes are 2.500 mm long with an active length of the bearing area of 1.700 mm, which is defined by a 200 mm raised concrete threshold above the floor level. The removal of excrements is ensured by automatic collection with hydraulic shovels 24 hours a day.

## 2.2 Production and layering of bedding

Limestone, straw, water and separation (RMS) is used to produce a new type of bedding, which is obtained directly on the farm. The production of RMS is ensured by pumping the collected manure from the homogenization channel to a cylindrical separator where its liquid fraction is separated. Subsequently, the separate is allowed to lie under a shelter for two weeks (Figure 1). For the production of bedding in one bed box the following is required: 200 kg of ground limestone, 40 kg of straw, 10 kg of separate and 110 l of water. The production itself as well as the technological modification of the bedding in the cubicles takes place in three basic steps:

- **The first step:** Approximately half of each component (except water) is taken and placed for mixing in an old feed wagon discarded for this purpose. Straw (20 kg) is added to the mixing space of the car first, which is mixed for 10–15 minutes. Subsequently, limestone (100 kg) with separate (5 kg) is added. After another 15 min, 80 l of water are added to the resulting mixture by stirring. After the addition of water, the whole mixture is mixed for 15–20 minutes to completely mix all components and achieve a slurry (Figure 2). Subsequently, the resulting slurry is dosed from the feed wagon to half the height (10 cm) of the cleaned bed and is evenly distributed. After spreading the mixture on the bed, the mixture is compacted using a vibrating plate.
- **The second step:** In the same way as in the first step, the second half of the straw, limestone and the rest of the separate (5 kg) are mixed in the feed wagon, but only 30 l of water are added to the mixture. The smaller addition of water in the second step will ensure higher elasticity and lower hardness compared to the first layer. After spreading the second layer on the first to a height of 20 cm of the concrete barrier of the bed, this layer is again compacted by means of a vibrating plate.
- **The third step:** In the last phase, the remainder of the prepared mixture (2–4 cm) is poured onto the compacted second layer, which is no longer treated with a vibrating plate but only gently spread with rakes (Figure 3). The cubicles thus formed are mechanically held once a day by collecting faeces into the manure passage and, if necessary, a thin layer of limestone is added.



Figure 1Separate production by pumping and pressing manure



Figure 2 Production of bedding for dairy cows by mixing components



Figure 3Dosing of bedding mixture and its adjustment in the cubicle

## 2.3 Sampling of cubicles bedded

Samples for microbiological examination were taken from 4 sections according to the time delay of production and layering of bedding. Bedding samples were taken from three sections according to a new recipe with a monthly intervals of 1–3 months after use of improved bedding. A control sample of bedding, consisting of straw, was taken from the last, fourth section. Two cubicles were selected from each section, from which a four samples of bedding to a depth of 20 cm was taken. The samples of bedding were mixed and ground to a particle size of 2–4 mm prior to analysis.

## 2.4 Microbiological examination

From the bedding sample was taken 10 g and 90 ml of saline was added. Following subsequent homogenization, individual ten-fold dilutions in physiological saline were then prepared from the treated samples. Dilutions from 10<sup>-4</sup> to 10<sup>-8</sup> according to the method by Fournel et al. (2018) were used for seeding. Various culture media were used to ensure appropriate growth conditions for the individual microorganism strains tested. For total count of bacteria (total viable count, TVC) at 37 °C, Plate Count Agar (Oxoid, UK) was used. Fecal coliform bacteria (FCB) were cultivated on a petri dish containing McConkey agar (Oxoid, UK) at 43 °C. For coliform bacteria (CB) at 37 °C, Endo agar (Hi-Media, India) was used. For fecal streptococci (FS) at 37 °C, M17 agar (Oxoid, UK) was used. The culture media were prepared according to the manufacturer's instructions and poured into 90 mm diameter Petri dishes in parallel. After a specified incubation time for each microorganism, the colony forming units (CFUs) were calculated according to the appropriate formula.

## 2.5 Statistical analyses

For statistical comparison, bacterial counts were also log-transformed and expressed in log CFU/ml. The differences between tested indicator microorganisms of improved bedding and control bedding with straw were analyzed by using analysis of variance (ANOVA) followed by Dunnett's multiple range test. The minimum criteria for statistical significance was set at  $p \le 0.05$  for all.

# 3 Results and discussion

Proper selection of bedding and application of appropriate components to achieve a disinfection effect are the key factors in maintaining the cow's health. The effectiveness of disinfection is affected by the resistance of microorganisms, the selection and use of disinfectant components and the external environment in which the disinfection takes place (Ismaïl et al., 2013). Under dairy farm conditions, straw is the predominant litter, although other bedding materials may also be used (Wolf et al., 2018).

According to Tančin et al. (2013), it is ideal when cubicles filled with straw are re-bedded twice a day, always after the manure is cleared out from the end of the bed and from the corridors. In some farms the re-bedding procedure is performed only once a day with higher amount of fresh bedding, but the corridors themselves are cleared out twice a day.

On farms with a lack of straw, the separated solid fraction of the recycled manure sludge (RMS) has been applied in recent years to bedding in the cubicles (Leach et al., 2015). With a higher dry matter content, it is a suitable material for lining deep bedded cubicles. However, freshly separated sludge has a dry matter content of only about 30% and it is desirable to allowed it to dry before use. The use of RMS as an alternative source for bedding provides a sufficient effect of maintaining a dry and hygienic bedding after application to boxes. Due to its high dry matter content, the required bedding consistency and hygienic effect are achieved only for a short period of time (Fournel et al., 2018).

One of the ways how to achieve the desired consistency of bedding in the long run and ensuring its optimal height and hygienic effect is to mix RMS with straw, limestone and a sufficient amount of water and to carry out the technological treatment directly on the bed. In our study we used separated RMS which was left standing for two weeks before the production of bedding. The sludge was mixed with limestone at a ratio of 1 : 4 to increase the proportion of limestone and the accompanying disinfectant effect of the bedding formed. The increased disinfection effect in the bedding we produced was confirmed from the samples taken. A reduced total count of microorganisms (TVC) and coliform bacteria (CB) was noted in the improved litter for two months after its loading by the dairy cows compared to straw-filled bed boxes. Comparison of the counts of faecal bacteria (FB) and faecal streptococci (FS) showed that their

counts were reduced during the first, second and third month after using the bedding with an improved composition compared to the control group housed on straw (Table 1).

Bedding	Total viable count	Coliform bacteria	Fecal coliform bacteria	Fecal streptococci
Control – straw	8.6ª	6.6ª	6.5ª	9.2ª
IB 1month	6.9 <sup>b</sup>	5.6 <sup>b</sup>	4.9 <sup>b</sup>	6.2 <sup>b</sup>
IB 2 months	7.3 <sup>b</sup>	6.0 <sup>b</sup>	5.3 <sup>b</sup>	6.5 <sup>b</sup>
IB 3 months	8.4ª	6.5ª	5.7 <sup>b</sup>	7.1 <sup>b</sup>

 Table 1
 Effect of bedding on the level of indicator bacteria (log CFU/ml)

IB 1 month – one month after use of improved bedding; IB 2 months – two months after use of improved bedding; IB 3 months – three months after use of improved bedding; a, b – values in column with different superscript letters differ significantly at *p* <0.05

In addition to the reduction of faecal contamination, a stabilization of the consistency of the litter with an improved composition in the laid beds was observed, which was demonstrated by maintenance of its thickness up to 20 cm.

#### 4 Conclusions

Our results show that in the case of classical straw bedding, its height in the bedded cubicles tends to fall below the litter threshold, and the breeders have to resolve this by clearing out and adding new straw. Especially in strawbedded cubicles, at the back (level with the cow's rear) the thickness of the bedding layer is often reduced, with an accumulation of dung, urine and an abundance of bacteria, which in favourable conditions rapidly multiply, as demonstrated by their increased numbers in the first two months of our study. Conversely, we found that improved bedding maintained a stable level as well as reduced numbers of TVC, CB, FS and FCB for the same two months. The positive effect of reducing the level of faecal contamination was also reflected in the dairy cows themselves, as they kept a cleaner body and udder.

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