Risks, benefits and optimal management of recycled manure solids for use as bedding for dairy cattle

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Disclaimer

This report is the output of a study, the aim of which was to extend knowledge of the risks, benefits and optimal management of recycled manure solids for use as bedding for dairy cattle. This study is inevitably constrained by its short duration and the fact that sampling only occurred in the months of January to May. This report does NOT constitute a full risk assessment or "claim to be the definitive document of RMS use". Suggestions for modifications of existing guidance on use are based on current knowledge but cannot be expected to provide "fool proof advice". All users of RMS have to accept responsibility for their own decisions with respect to its use. The authors of this report cannot be held responsible for decisions made on the basis of the information contained herein.

Executive Summary

Background

The use of Recycled Manure Solids (RMS) as bedding for dairy cattle constitutes a "technical use" of a Category 2 Animal By-Product under the EU Animal By-Products (ABP) Regulation (EU Reg. 1069/2009). The regulation has provisions which permit "technical use" of animal by-products and derived products, provided these do not pose an unacceptable risk to public or animal health. At the time when UK farmers began to adopt the practice (attracted by perceptions that there would be benefits to farm economics and cow comfort), data on which to base an assessment of this risk under UK conditions was unavailable.

In 2013, AHDB Dairy (formerly DairyCo) commissioned a desk top scoping study to collate the available evidence on the use of RMS. An executive summary of the study can be found at http://www.dairy.ahdb.org.uk/resources-library/technical-information/buildings/rms-bedding/ The study concluded that there were significant knowledge gaps limiting the ability to assess the risks to public and animal health in UK conditions.

In June 2014, Defra agreed to allow continued use of RMS in England under prescribed management conditions, while further data were collected. For cross-reference, these conditions can be downloaded from http://www.dairy.ahdb.org.uk/rmsbedding. In a similar manner, the Scottish Government also permitted use, whilst the Welsh Government held the opinion that the information to assess the risks to animal and human health was insufficient, and therefore the use of manure as bedding was not sanctioned in Wales at that point.

To address this gap in knowledge, research was commissioned to gather data on RMS use under UK conditions. The overall objective was to provide greater technical understanding to help inform the legal position with regard to the safe use of recycled manure solids as bedding, and in particular to investigate management and husbandry options to safely mitigate any potential risks to animal or human health.

Aims and Structure of the Project

The project aimed to:

- Assess the presence (and in some cases, levels) of selected pathogens and milk spoilage bacteria in cubicles bedded with RMS in comparison with other bedding materials.
- Assess the transfer of pathogens and milk spoilage bacteria from different bedding types to bulk milk and potential mitigating factors.
- Provide robust information on the relationships between bedding (including RMS) and udder health in dairy cows under UK conditions.
- Increase our understanding of factors influencing the success of use of RMS as a bedding material for UK dairy cows.
- Assess the potential to mitigate possible adverse impacts of the use of RMS as bedding.
- Assess specific aspects of welfare and comfort of cows on RMS and other bedding.

- Predict the likely levels of MAP and *Salmonella* spp in slurry and RMS bedding over time, in farms with different disease levels, by modelling literature based values of cow excretion patterns alongside the dynamics of slurry storage and removal.
- Provide information on antimicrobial resistance patterns in organisms isolated from farms using RMS and other bedding materials.
- Contribute to information on best practice for building and managing beds using RMS.
- Provide a cost calculator enabling farmers to evaluate a variety of bedding options.
- Enable exchange of information and experiences on RMS use between the UK and the Netherlands.

The work involved collection and analysis of data from:

- An epidemiological survey of 125 farms using RMS, sawdust and sand as bedding.
- A controlled study of different bedding materials and depths.
- An observational study of different methods of initial build-up of deep RMS beds.
- *In silico* modelling to predict the likely levels of MAP and *Salmonella* spp in slurry and RMS bedding over time, in farms with different disease levels.

The project also incorporated:

- Development of an economic cost calculator to allow evaluation of the cost of converting to, and subsequent use of RMS bedding.
- Analysis of bacterial isolates collected on the survey to assess implications for antimicrobial resistance.

1. Epidemiological Farm Survey

The survey included 40 farms using RMS bedding, 41 farms using sand and 44 farms using sawdust.

Bacterial counts in "used bedding"

- Across all the species and groups enumerated, with the exception of *Streptococcus* spp, bacterial counts in "used" bedding were significantly higher in RMS than either sand or sawdust. Numerically, mean and median counts were typically lowest on sand farms. However, it is important to note that there was often as much variation within bedding type as between bedding type.

Bacterial counts in bulk milk

- Despite the high levels of bacteria in "used" RMS bedding, bacterial counts in bulk milk did not differ between the groups of farms with different bedding types, and there was no association between bacterial count in "used" bedding and in bulk milk sampled on the same day, across all bedding types.

- Across all bedding types, fore-milking was associated with a lower total bacterial count (TBC) in bulk milk (2,503 *vs* 4,800 cfu/ml; p=0.047), but not with any other bacterial species/grouping.
- However, within the population of RMS farms, higher total bacterial count in "used" bedding was associated with higher total bacterial count in bulk milk.

Somatic cell counts in bulk milk

- Somatic cell counts were not significantly different between farms bedded on the different materials, though there was a trend for SCCs to be lower on the sawdust farms compared to the RMS farms (134 *vs* 171 $x10^3$ cells/ml; p=0.06).
- Within RMS farms pre-dipping was associated with a lower bulk milk SCC (137 *vs* 206 $x10^3$ cells/ml; p=0.037).

Specific zoonotic pathogens in bedding

- *Yersinia enterocolitica* was identified in the bedding on between 4.9% and 9.8% of farms, but the prevalence did not vary between bedding types.
- *Salmonella* spp were identified in used bedding on four farms (two sand and two RMS).
- *Listeria monocytogenes* was isolated from a significantly higher proportion of bedding from sand farms (58.5%) than RMS (15.0%) or sawdust farms (31.7%) (p<0.01), which did not differ.

Zoonotic pathogens in milk

- *Yersinia enterocolitica* was identified in the bulk milk on between 0% and 12.2% of farms, but the prevalence did not vary between bedding types.
- A *Salmonella* spp was identified in the bulk milk of one sawdust farm and was subsequently identified as *S. montivideo* (APHA).
- *Listeria monocytogenes* was isolated least frequently from bulk milk from sand farms and was isolated from between 2.4% and 12.5% of farms across the bedding groups. However, the prevalence in milk did not vary between bedding types.

Udder health

- No significant differences were identified between farms utilising the different bedding materials, in any of the measures of udder heath analysed, based on either SCC or clinical mastitis cases. No significant effects of changing to RMS from a different bedding material were identified.
- The ability to identify differences in clinical mastitis rates between cows housed on different bedding types was hampered by a lack of robust data.

Culling

- No significant difference in reasons for culling cows was identified between herds using different types of bedding.

Cow comfort and welfare

- RMS beds would appear to offer some advantages with respect to cow comfort and cleanliness:

- based on measures of cleanliness and hock condition, deep RMS beds typically performed as well as sand beds.
- when used on mats, RMS demonstrated clear advantages over sawdust.

The findings of this survey need to be interpreted in the light of the fact that the use of RMS as a bedding material is still in its infancy in the UK and Europe. Whilst early indications are that there need not be an adverse effect on udder and animal health, this will need to be monitored, as and if more herds adopt the practice.

Mitigation of risk through management practices to reduce bacterial levels in used RMS bedding

- With the exception of the use of RMS as deep or shallow beds, management had no consistent effect on bacterial levels in RMS bedding.
- TBCs, *Streptococcus* spp and psychrotrophic counts were higher in RMS managed in shallow beds. *Bacillus cereus* counts were higher in RMS managed as deep beds.
- There was no detectable impact of using conditioner on bacterial counts in used RMS bedding.
- No significant relationship between the frequency of bedding and bacterial counts in bedding was identified. There was a trend for *Streptococcus* spp counts to be lower in beds to which fresh RMS was applied daily (1.08×10^8 *vs* 2.80×10^8 cfu/g; p=0.057).
- No effect of separating RMS bedding under cover was found on i)dry matter of the fresh bedding on the day of production, ii) dry matter of used RMS or iii) bacterial counts in RMS. However, only a single sample of bedding from the day of the visit was analysed and farmers without 'cover' avoided separating RMS in wet weather.

Mitigation of risk of transfer of bacteria to bulk milk through milking and bed management practices for RMS, sand and sawdust bedding

- Across all bedding types (RMS, sand, and sawdust):
  - Fore-milking was associated with a lower TBC in bulk milk.
  - Pre-milking teat preparation that involved a pre-dip followed by wiping dry was associated with a lower *Streptococcus* spp count in bulk milk (and with a lower psychrotrophic count in the subpopulation of RMS farms).
  - Cluster disinfection was not found to be associated with lower bacterial counts in milk, with the exception of thermophilic spore counts and psychrotrophic counts.
  - No difference was detected between manual and automated cluster disinfection systems.
- Within RMS farms, there were no significant differences in any of the bulk milk bacterial counts or milk constituents between farms with deep and shallow beds.

2. **Controlled Trial**

This study represents one of the most comprehensive investigations of the impact of different bedding materials and managements on udder health and milk quality conducted to date. In a modified crossover design trial, four groups of 40 cows rotated twice around the four bed types (deep sand, deep RMS, shallow RMS and shallow sawdust). Bacterial loads on "unused" and "used" bedding, and in bulk milk, were assessed. Impact on udder health was evaluated by assessment of individual quarter somatic cell counts, specifically, the acquisition of new infections at the quarter level.

Bacterial counts in "unused" bedding

- Bacterial counts varied significantly between the three "unused" bedding materials, being highest in the RMS and lowest in sawdust.

Bacterial counts in "used" bedding

- RMS beds were replenished twice weekly, sawdust beds twice daily and sand once every two weeks. Counts in used bedding need to be interpreted in this context.
- TBCs of "used" bedding varied significantly between the four bedding materials, being highest in shallow RMS and lowest in sawdust.
- Coliform counts in RMS were higher than in sawdust, but not significantly different from sand.
- *Streptococcus* spp counts were highest in shallow beds, with shallow RMS and sawdust showing no significant difference.
- *Staphylocccus* spp counts were significantly higher in shallow RMS than in other used bedding materials.
- The laboratory pasteurised counts were highest in deep RMS and lowest in sand, whilst thermophilic spore counts were high in both deep and shallow RMS beds.
- Psychrotrophic counts were significantly lower in sawdust than in other bedding materials.
- *Bacillus cereus* counts were significantly higher in deep RMS beds, being 3 logs higher than in sand or shallow RMS beds; very little *Bacillus cereus* was identified in sawdust beds.

Bacterial counts in bulk milk

- With the exception of *Streptococcus* spp and *Staphylococcus* spp, there was no effect of bedding treatment group on the bacterial count of bulk milk. *Streptococcus* spp counts were significantly lower ($p<0.05$) in milk from cows on deep beds (regardless of bedding material) whilst variation was less predictable in *Staphylocccus* spp counts.
- With the exception of *Streptococcus* spp and *Listeria* spp there was no clear relationship between bacterial numbers in bedding and in bulk milk, although this may, in part, reflect the hygiene practices during milking.

Udder health

- In this study, sawdust, applied to mats twice daily, appeared to offer the best protection against new intramammary infection (as measured by SCC). New Quarter intramammary Infections were significantly less likely to occur in cows on sawdust beds

than on deep RMS (47/961 *vs* 84/965; p=0.012) or sand beds (47/961 *vs* 78/965; p=0.04). No impact of bedding material on the likelihood of a quarter curing could be identified.

- Unlike the assessment of udder health using SCCs, the analysis of clinical mastitis suggests that RMS as a bedding material may increase the risk of clinical mastitis; there was a trend for cows bedded on RMS to be at higher risk of developing clinical mastitis than cows not bedded on RMS (7/73 *vs* 2/78; p=0.086). This is an area that warrants further research.
- No consistent, biologically plausible, repeatable correlations were found between bedding bacterial counts and udder health.

Cow comfort

- Deep beds offered the highest of cow comfort, regardless of bedding material. RMS was relatively protective when used on shallow beds, in comparison with sawdust.

3. Bed building study

Farmers have reported that RMS may not dry out optimally, and heating may occur if a large amount is initially applied during the process of establishing deep beds. Such conditions might be expected to encourage microbial growth and could have an effect on udder health. This farmer-inspired experiment was designed to test whether building beds gradually was associated with an increase in the dry matter content of the bedding and less heating. In addition the impact of the presence of cows during the bed building phase was assessed. The main findings were as follows:

- Rapid building of beds elevated temperatures of bedding in the building phase, and the effect appeared to persist to some extent for four weeks. Temperature was affected more by speed of fill than by the presence of cows.
- The effect of building speed on dry matter content at the surface during the first week and four weeks later was inconsistent.
- Dry matter content of the surface material appears to be influenced by factors in addition to the speed of bed building. These may include the presence of cows, but also environmental conditions. There appears to be a complex interaction between environmental conditions, including temperature and relative humidity, and the temperature and dry matter of RMS bedding.
- Rapid building of beds and the consequent increases in temperature and decreases in dry matter may be associated with a higher coliform count in the bedding material in the early stages of bed establishment.
- Slow building will by definition limit the depth of beds and thus the comfort provided if cows are present during the building phase.
- Further research is required to understand the behaviour of this material in different environmental conditions, as this may be a key to its optimal use.

4. In silico modelling of Levels of *Mycobacterium avium* subspecies *paratuberculosis* (MAP) and *Salmonella* spp in Cattle Slurry and RMS

Individual cow excretion patterns of MAP and *Salmonella* spp were obtained from peer reviewed literature and the potential load in slurry was modelled, using assumptions with respect to number of cows affected within a herd and factors relating to slurry storage and removal. Different scenarios of herd disease prevalence and slurry handling methods were evaluated. The estimated levels of organisms present in RMS were considered alongside potential infective doses to assess the degree of risk posed by each pathogen and scenario.

- The infective dose of MAP suggests that fairly large quantities of RMS (of the order of 100-1000 g) would need to be ingested by cattle to reach the published values for the infective dose. Furthermore, this figure is probably representative of an infective dose for calves, and for adult cows may be substantially higher. Therefore in terms of MAP, the models constructed in this research suggest that whilst bedding of youngstock using RMS should be avoided, the risk to adults may be minimal.
- The infective dose of *Salmonella* spp suggests that in a very severe outbreak, when levels in RMS may become high, only small quantities (of the order of <1g) of RMS would need to be ingested to cause disease in cattle. Clearly this depends on many factors and there will be other transmission routes, which in fact may be more significant, including, but not limited to, feed and water, wildlife (especially birds), and fomites.

5. Economics and bedding cost calculator

A Cost Calculator spreadsheet has been created which can be used to cost individual scenarios of conversion from an existing bedding material to RMS.

6. Antimicrobial resistance

Coliform organisms and *Enterococcus* spp were isolated from bedding and milk collected at the time of the visit to farms participating in the epidemiological farm survey. In addition data was collated on antibiotic use on farm. An in-depth analysis of the impact of bedding type and management on antimicrobial resistance was not envisaged as part of the research outlined in the original tender prior to this study and for that reason any findings need to be interpreted with care.

- Differences in the MICs for antimicrobials were identified between the different bedding types.
- No one bedding type was associated with higher MICs overall, with each bedding type being associated with the highest MICs for at least one antibiotic class.
- No clear evidence was identified to suggest that the short term use of recycled manure solids as bedding, as compared to sawdust and sand, is associated with a general increase in MICs of the major classes of antibiotics when considering coliforms and *Enterococcus* spp.

- Further research, using the dataset generated by this study and elsewhere, is needed to further our understanding of any potential interactions between bedding type and management and antimicrobial resistance in the environment.

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