Evaluation of Corncob as Bedding for Rodents

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Summary
As an alternative to aspen bedding, corncob may be used for rodents. Previous studies have shown that the ammonia level in cages using corncob bedding is reduced compared to cages with aspen bedding. The reduced level of ammonia prolongs the interval between cage changing, and it may therefore be beneficial for the facility to use corncob.
The aim of the present study was to measure and evaluate the animal preferences for corncob compared to aspen bedding and also analyse the properties of corncob compared to aspen bedding.
When analysing the bedding's ability to absorb water, corncob showed lower water absorption compared to aspen bedding; the more corncob in the mixture, the less water is absorbed.
Both mice and rats rejected cages with pure corncob during the day (sleeping time), and none of the animals preferred corncob mixed with aspen, only equally accepted it.
In conclusion, neither rats nor mice prefer corncob, even not in mixtures with aspen bedding and enrichment. In the light of the common standard for bedding being wood chips, and the lack of preference for corncob mixture, corncob seems to be a poor alternative to wood based bedding.

Introduction
According to European legislation (*Council of Europe, 1986*; *Council of Europe, 1997*) laboratory rodents should have access to bedding, and various kinds of materials have been applied for this purpose in different housing systems (*Perkins & Lipman, 1995*; *Blom et al., 1996*; *Potgieter & Wilke, 1996*; *Potgieter & Wilke, 1997*; *Ago et al., 2002*). In European animal facilities wood chips made of hardwood, e.g. aspen or birch, are common. The bedding serves several purposes, but the primary one is absorption of urine and eventually leaking water from water bottles. Furthermore, bedding enriches the environment as it can be used for digging and for creating a nest. Alternatively to wood chips, another kind of bedding material, which can be used, is corncob made from the granulated cob from corn after the corns have been removed mechanically. Then the granulate is dried and used for bedding. It is available in two sizes, a standard in which the granulates are 3-5 mm and a fine in which the granulates are 1-2 mm.
The use of Individually Ventilated Cage-systems (IVC-systems) is increasing and in US and Europe they are common in many rodent facilities. One of the advantages of using IVC's, is the ability to dry out the bedding material (*Krohn et al., 2006a*), due to the forced ventilation in the cages. A previous study has shown that, when corncob is used in IVCs, the ammonia level in the cages is lower compared to cages with aspen bedding (*Perkins & Lipman, 1995*), and the use of corncob may therefore be beneficial, as it may reduce the number of cage changes from twice a week to once a week. A reduction in the number of cage changes has great advantages for the budget as cage changing

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Figure 1. Aspen (left) and corncob bedding (right). The aspen is BeeKay bedding and the corncob is corncob bedding (Both from Scanbur A/S, Karlslunde, Denmark)

is very time consuming and expensive in bedding and washing of the cages. Therefore there may be good reasons for changing from aspen bedding to corncob, but there is a need to know, whether this would affect animal welfare or well-being. Another advantage of corncob is a reduced spread of allergens, as it appears that the amount of allergens spread from cages with corncob is between 57% and 77% lower compared to those spread from wood-based bedding (*Sakaguchi et al., 1990*), as corncob contains no dust particles (*Wirth, 1983*). There is increased focus on laboratory animal allergy, and any reduction in the amount of allergens spread from animals is beneficial. However, it is possible to reduce the amount of allergens by other means than using another kind of bedding (*Gordon et al., 1997*; *Gordon et al., 2001*; *Krohn & Hansen, 2004*; *Krohn et al., 2006b*).

The aim of the present study was to measure and evaluate animal preferences for corncob compared to aspen bedding and also analyse the properties of corncob compared to aspen bedding in mice and rats.

Materials and Methods

The bedding

Pure aspen, approx. 5x5 mm (BeeKay Bedding, Scanbur A/S, Karlslunde, Denmark), pure standard corncob, approx. 3-5 mm (Scanbur A/S, Karlslunde, Denmark), pure fine corncob, approx. 1-2 mm (Scanbur A/S, Karlslunde, Denmark), 50% v/v standard corncob, and 25% v/v standard corncob, were used, the balance in the mixtures being pure aspen. The aspen bedding and the standard corncob bedding are shown in Figure 1. For the rat preference cages were un-enriched, whereas for the mice one session was performed with and one session was performed without enrichment as defined below.

The animals

Ten female BkI:SD rats (Scanbur A/S, Sollentuna, Sweden) with initial weight of 200 gram and age of 12 weeks, and ten female C57BL/6JBomTac mice (Taconic, Ejby, Denmark) with initial weight of 25 gram and age of 10 weeks were used. When not in the preference-setup the rats were housed pair-wise in

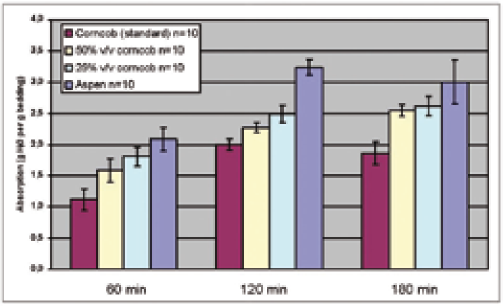

Figure 2. The absorption of water for the different bedding mixtures. There were significant differences between the different bedding mixtures ($p < 0.005$), except for between 50% v/v corncob and 25% v/v corncob after 180 min. The results are given as mean ± SD.

macrolon Type U1500 cages, 1500 cm^2 (Tecniplast, Buguggiate, Italy) with aspen bedding (BeeKay Bedding, Scanbur A/S, Karlslunde, Denmark), and with wood wool and woodblocks (Tapvei, Finland), while the mice were housed in groups of five in macrolon Type III cages, 800 cm^2 (Tecniplast, Buguggiate, Italy) with aspen bedding (BeeKay Bedding, Scanbur A/S, Karlslunde, Denmark), and with wood wool, wood sticks (Tapvei, Finland) and paper houses (Des.Res.™, Lillico, Surrey, UK). All animals were fed Altromin 1324 (Brogården, Gentofte, Denmark) and water ad libitum. All cages were changed twice a week. All animals were ear marked using ear notching with numbers from 1-10.

The absorption study

The absorption abilities of the different mixtures were tested by adding 1.0 litre of water to 1.0 litre of bedding. After 60, 120 or 180 minutes, respectively, excess water was removed by sieving the water and bedding mixture using a metal sieve, letting the water drip out for 10 minutes and measuring the amount of water dripped from the bedding. Hereafter the amount of water absorbed by the bedding was calculated. Each test was performed 10 times and means and standard deviations were calculated.

The preference studies

The preference test was set up as previously described using two type III cages (Tecniplast, Buguggiate, Italy), interconnected with a PVC-tube, placed on a computer-logged digital weight (*Krohn & Hansen, 2001*). Two set-ups were placed simultaneously in a Scantainer (Scanbur A/S, Karlslunde, Denmark) in a separate room with no other animals present, and with automatic day/light shift (06:00-18:00 h), room temperature at 23±1 °C and relative humidity at 50±10 %. The room was ventilated 10-15 times per hour and the Scantainer 70 times per hour. Two night-time (18:00-06:00 h) and two day-time (06:00-18:00 h) periods were analysed for each animal in each study. Before testing the preference for the different bedding mixtures, the set-up was calibrated. First the animals were placed individually in the set-up, and were given a choice between two identical Type III cages with aspen bedding to ascertain any possible side (left versus right) bias in the choice apparatus. The animals were then placed individually in the preference set-up, given the choice between aspen bedding and one of the mixtures described above while registering the preferences for each cage. Between tests the animals were housed socially for at least two weeks as described above.

Statistical analysis

Results from the absorption study were, after having been shown to be normally distributed (Anderson-Darling), analysed by ANOVA and subsequently differences between groups were analysed with a t-test (Minitab version 14.1, Minitab Inc., US). Results from the preference studies were analysed by the use of the Wilcoxon signed rank test, as the data are not normally distributed (Anderson-Darling) (Minitab version 14.1, Minitab Inc., US). The null hypothesis was set as there was no effect of housing on the preference (i.e. expectation of a 50/50 distribution between the two cages).

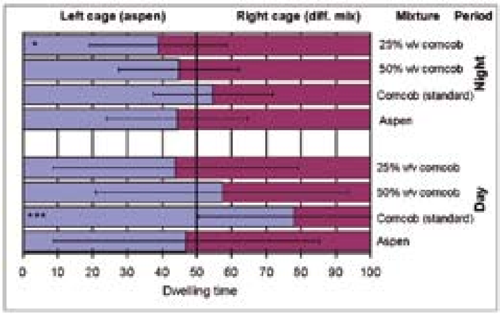

Figure 3. Preferences of female rats given a choice between aspen bedding and different mixture of corncob and aspen. The figure shows the distribution of dwelling time between left and right cage for day and night. The 50% distribution is marked with a bold line, and for each result the standard deviation is marked. Any statistical significance is marked (* $p < 0.05$, *** $p < 0.001$). N=10 for each mixture.

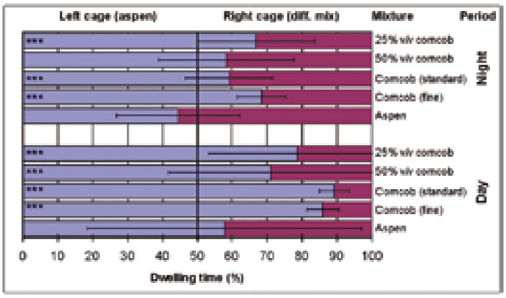

Figure 4. Preferences of female mice given a choice between aspen bedding and different mixture of corncob and aspen in an un-enriched cage environment. The figure shows the distribution of dwelling time between left and right cage for day and night. The 50% distribution is marked with a bold line, and for each result the standard deviation is marked. Any statistical significance is marked (*** $p < 0.001$). N=10 for each mixture.

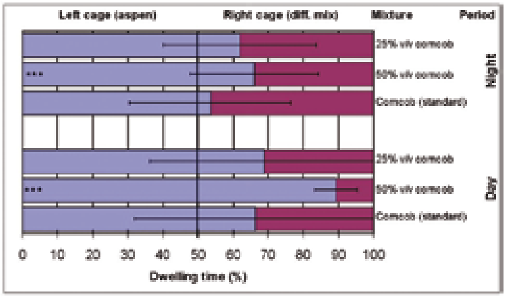

Figure 5. Preferences of female mice given a choice between aspen bedding and different mixture of corncob and aspen in an enriched cage environment. The figure shows the distribution of dwelling time between left and right cage for day and night. The 50% distribution is marked with a bold line, and for each result the standard deviation is marked. Any statistical significance is marked (*** $p < 0.001$). N=10 for each mixture.

Results

Figure 2 shows the amount of absorbed water in grams for each gram of bedding mixture for the three absorption times 60, 120 and 180 minutes. When comparing the different mixtures within the same time, there were significant differences between the different bedding mixtures ($p < 0.01$), except for between 50% v/v corncob and 25% v/v corncob after 180 minutes. The more corncob in the mixture, the less water was absorbed.

During the day (the sleeping time), the rats rejected pure corncob ($p < 0.001$), while they did not seem to prefer or reject either of the mixtures compared to aspen. Neither did the rats show any preferences at night, except for a minor preference for the 25% v/v corncob mixture ($p < 0.05$) (Figure 3).

Also the mice rejected all mixtures with corncob during the day (the sleeping time) ($p < 0.001$). During the night-time, the mice rejected any mixture with corncob ($p < 0.001$) except the 50% v/v corncob, which were neither rejected nor preferred (Figure 4).

During the day (the sleeping time) the mice neither rejected nor preferred cages with corncob although the cages were enriched (Figure 5), but the cage with 50% v/v corncob were rejected ($p < 0.001$). The results were the same during the night with the enriched cages and with rejection of the 50% v/v corncob ($p < 0.001$).

Discussion

The absorption of water by corncob was notably lower compared to aspen bedding and absorption increases with increasing amounts of aspen, as also shown by others (*Wirth, 1983*; *Burn & Mason, 2005*). A lower absorption may be advantageous in IVC-systems, in which the drying caused by the ventilation is high. It has been shown, that the use of corncob reduces the amount of NH_3 (*Perkins & Lipman, 1995*), as the amount of urine absorbed by corncob is lower and therefore the evaporation of urine from the bedding is higher, leading to a drier environment.

None of the animals tested preferred corncob, which also is the conclusion from other studies (*Mulder, 1974*; *Ras et al., 2002*; *Lanteigne & Reebs, 2006*). The reason for this might be that corncob is spherical, and thereby unpleasant to rest on (*Ras et al., 2002*), but it may also be difficult to manipulate into a nest as it cannot be shredded into small pieces, which is also the conclusion from a study in hamsters (*Lanteigne & Reebs, 2006*). When given nesting material the need for making a nest out of the bedding material becomes less important, but the animals still do not positively prefer the corncob in any way, only equally accept it. In general, rats are known to prefer the type of bedding with which they are familiar (*Manser et al., 1995*). As most breeders of laboratory rodents house the animals on aspen or other kinds of wood-based bedding, corncob means a change in bedding condition compared normal. However, even if raised on corncob rats, do not prefer corncob as bedding material (*Ras et al., 2002*).

In conclusion, neither the tested rats nor mice prefer corncob, even not in mixture with aspen bedding and with enrichment during sleeping time. As the common standard for bedding is wood chips, and as there dislike for corncob mixtures, corncob may not be the ideal alternative to wood based bedding, even though the lower water absorption capability of corncob compared to aspen makes it an option for prolonging cage changing intervals in IVC-systems, and the spread of allergens is reduced is with corncob.

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