An assessment of paper pulp dishes for safe and enriching diet delivery

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Abstract

It is common practice for laboratory animals to be provided with diet in the form of wet mash or gel for various situations such as post-weaning and post-operative recovery. This study tested the use of paper pulp dishes as a sustainable alternative to plastic dishes. Rodents often begin to chew and shred these plastic dishes as part of their normal behaviour, often incorporating the pieces into their nest. This shredded plastic can potentially pose a risk to health if ingested. In Phase 1, the paper pulp dishes successfully held the diet gel for up to three days without becoming soggy or fostering mould. Phase 2 explored how rodents interacted with the dishes as potential enrichment items, noting variations in how different species, strains and age groups used the dishes. The findings suggest that paper pulp dishes are effective for diet delivery and provide additional enrichment opportunities, such as gnawing and nesting making them a viable alternative to plastic dishes.

Introduction

The aim of this study was twofold: first, to assess the structural integrity of a sustainable dish and second, to evaluate its potential as an enrichment tool. Wet diets are commonly provided to laboratory animals for various purposes such as supporting recovery after surgery or aiding underdeveloped pups during postweaning. This is currently provided in plastic dishes, which have minimal enrichment potential and may pose a risk to the animal if ingested. As gnawing and nest building are natural behaviours seen in mice, providing a dish that is safe, shreddable and able to be utilised into a nest, can be a valuable enrichment tool for laboratory animals. Here, we analyse the behaviours observed from two different species (mice and rats) from 6 different UCL units, across a span of 2 weeks.

Methodology

Phase 1

To test the structural integrity of the dishes over time, gel was placed and cohorts photographed each day for 4 days to record the development and progression of any dampness developing around the gel.

Phase 2

At each participating facility, 5 cages of the same strain and similar stocking density were selected for each cohort. A total of 8 cohorts (7 mouse and 1 rat) were used across 6 UCL facilities. To conduct the trial in a manner consistent with the 3Rs we only used animals currently housed in the facility for existing studies, therefore ages and densities varied according to availability.

Paper pulp dishes were added to clean cages on a Monday. Recording began the following day for 4 days at daily health checks, during which observations on the animals' interactions with the dishes were recorded as 10 predefined primary behaviours and a further 12 behaviours which comprised of observed combinations of these first 10. No scoring was done over the weekend and then on the following Monday, cages were cleaned and provided with a new dish before repeating the 4 days of scoring.

Data transformation

To interpret these behaviours in terms of animal interaction with the dishes, the 22 behavioural categories were assigned to corresponding interaction categories:

- no interaction
- light interaction
- moderate interaction
- substantial interaction
- heavy interaction

Observable Primary Behaviours	
Dish completely untouched	Dish completely gone
Signs of nibbling on dish	Shredded and incorprated into nest
Up to dish ¼ gone	Whole dish used as nest
Up to dish ½ gone	Whole dish used as shelter
Up to dish ¾ gone	Dish flipped over

Table 1. Observable primary behaviours.

Results and discussion

Phase 1: Structural integrity test

The structural integrity test demonstrated that the dishes maintained their stability for a full 3-day period. Only a slight spread of moisture was observed from the diet gel and no signs of disintegration were noted. These findings suggest that the dishes can safely hold diet gel for the recommended 3-day shelf life with minimal risk of mould formation. Additionally, they allow for the advance preparation of diet cups prior to use.

Phase 2: Primary observed behaviours

Mouse

Within all cohorts of mice 22 behaviours were recorded in total. The 10 most frequently observed behaviours are displayed here in the graph. Signs of nibbling on the dish were recorded 106 times over the 2 weeks. Although some

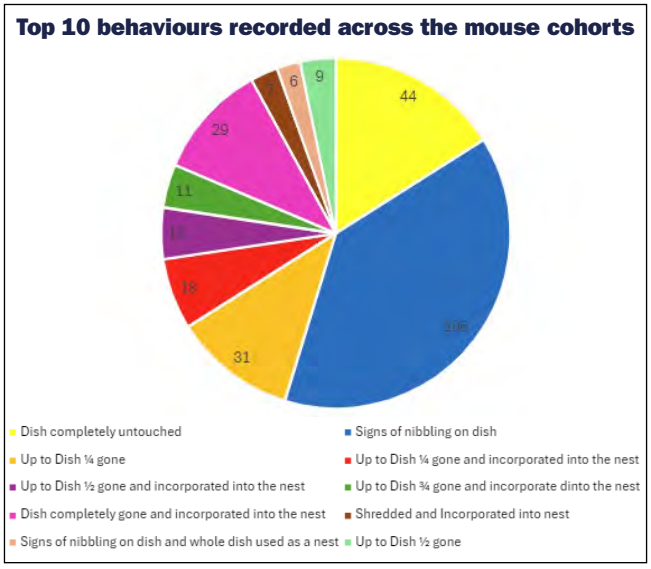

Figure 2. Ten most common behaviours recorded across the mouse population across the 2-week observation period.

left the dish completely untouched, these observations were outweighed by recordings of the dish being nibbled to varying extents and incorporated into the nest. Whilst non-interaction accounts for 15% of the total behaviours recorded, the data indicates that this was common within the first few days of the dish being placed in the cage. This could be down to neophobia or the novelty of the enrichment.[1]

Figure 3. Mouse behaviour.

Figure 1. Observation of a paper pulp dish filled with diet gel across a 4-day period.

Rat

Within the cohort of rats 7 behaviours were recorded, all rats used or nibbled the dish during the 2 weeks they had them. Rats are inquisitive by nature and will investigate any new enrichment given to them.[2]

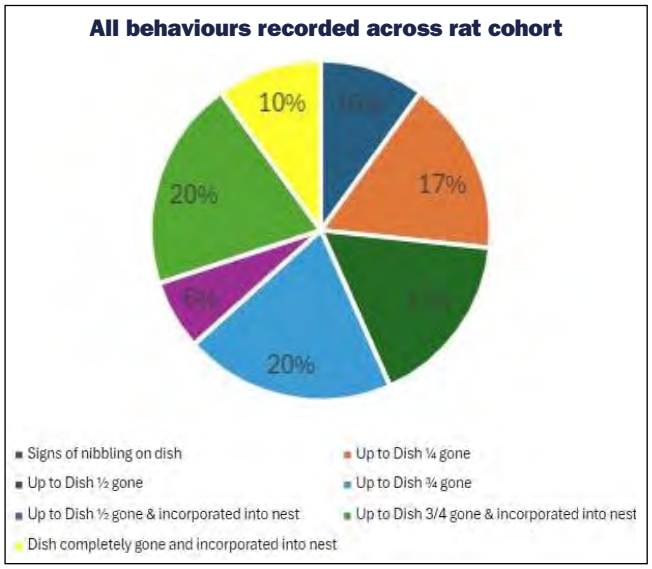

Figure 4. All behaviours recorded across the rat population across the 2-week observation period.

Average interaction scores

All facilities

There was a significant difference (p=0.00000000004175845677954l1) between the average dish interaction across all 7 mouse cohorts. This difference could be attributed to varying factors. Firstly, the ages range from 3 to 48 weeks across the facilities, which will heavily impact the usage. Our findings are also in line with the current literature, as there were observed behavioural differences between the different genetic strains, indicating that strain affects response to enrichment.[3]

Interaction between weeks 1 and 2

In 3 mouse cohorts (IoN x 2, P-Block & Prion) on average, the mice interacted with the dishes more in week 1 than in week 2, whereas the remaining 3 mouse cohorts (ICH, RF, Cruciform) all interacted with the dishes more in week 2 than week 1.

The effects of habituation are clear in this data set. In the majority of the mouse cohorts, overall. there was an upward trend of interaction in both weeks indicating that the mice interacted more with the dishes as time went on, exhibiting natural gnawing and nesting behaviours as they became habituated to them. The rat cohort interacted with the dishes more in week 2 than in week one and displayed an upward average trend.

C57s

Overall there was a significant difference (p= 0.000494) between the average interactions recorded between the 3 C57Bl/6J cohorts. This difference could be attributed to different ages within the cohorts (a range of 12 to 48 weeks old) and behaviours associated with their ages such as nest building.

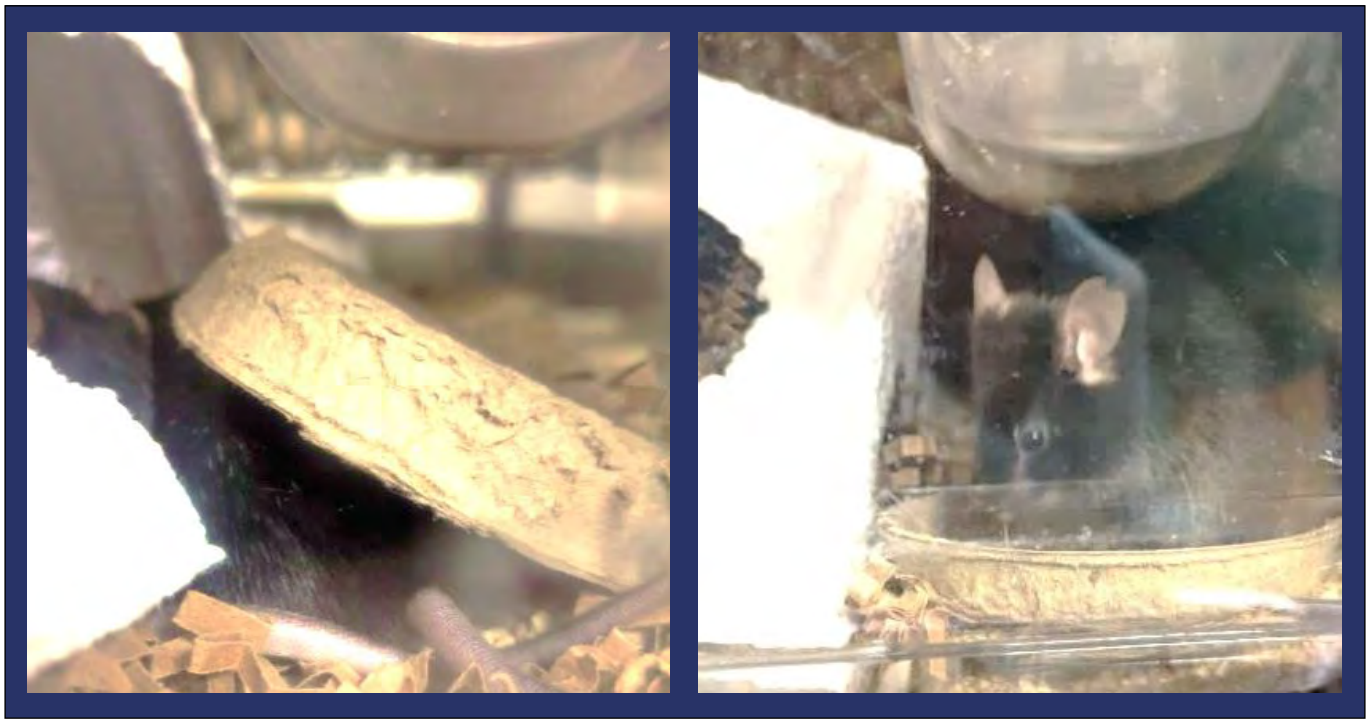

Figure 5. Rat behaviour.

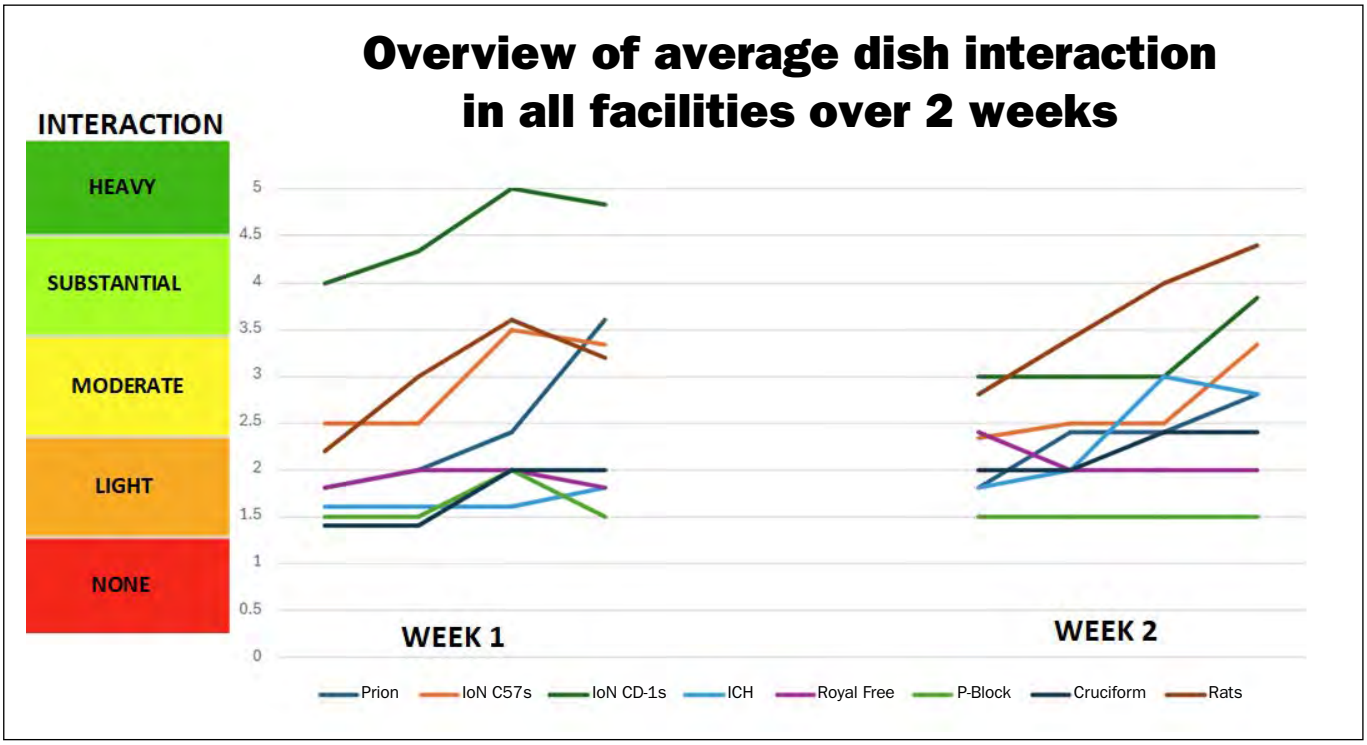

Figure 6. Graph showing average dish interaction in all facilities across the two-week observation period.

Non-C57Bl/6J strains

Overall, there was a significant difference (p= 0.00000439226) between the average interactions recorded between the remaining non- C57Bl/6J cohorts. More activity was seen in the first week, in comparison to the second week, indicating an apparent initial lack of neophobia in these cohorts and a subsequent reduction in interaction corresponding to the decrease in novelty. Mice have been noted to adapt quickly to new conditions, lessening their beneficial impact.[4] Only the P-Block cohort consistently did not interact with the dishes. These mice have a cranial phenotype and need their teeth trimmed twice a week, therefore affecting their ability to chew/gnaw.

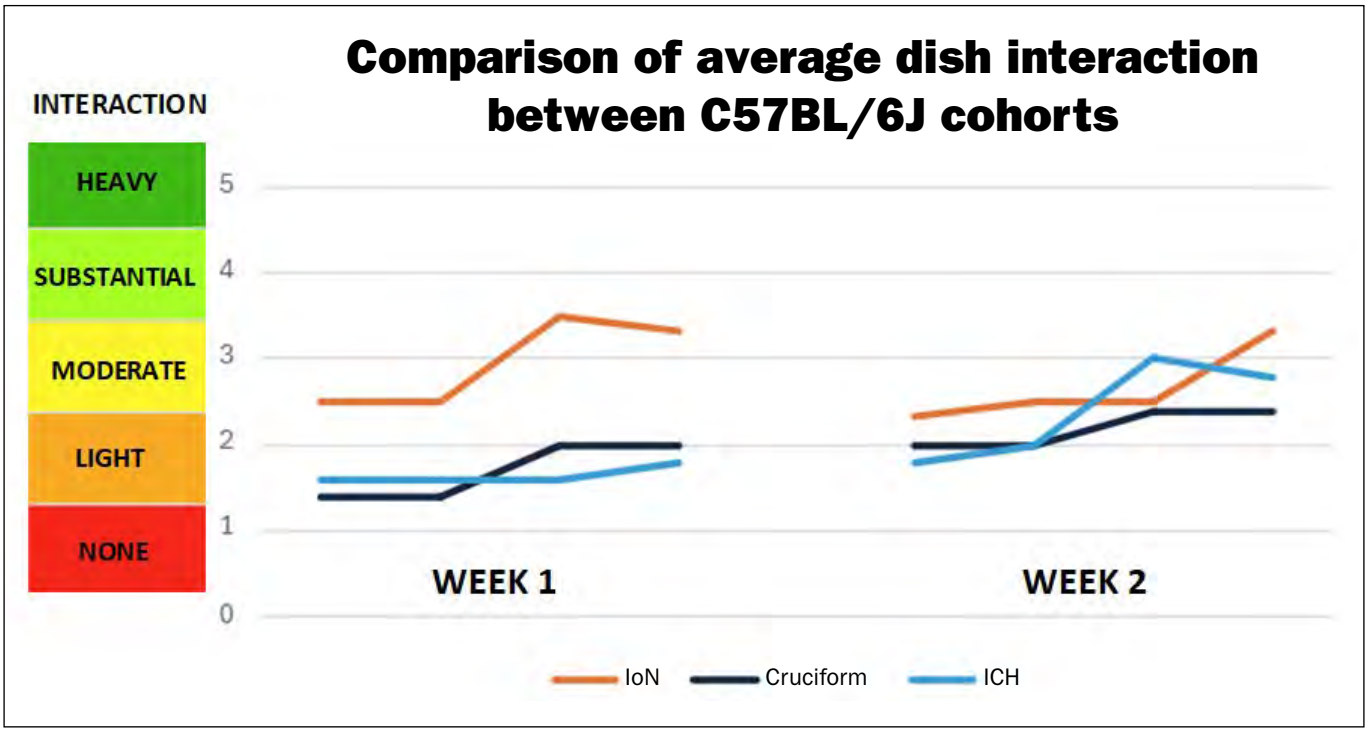

Figure 7. Graph comparing average dish interaction for C57BL/6J cohorts across the two-week observation period.

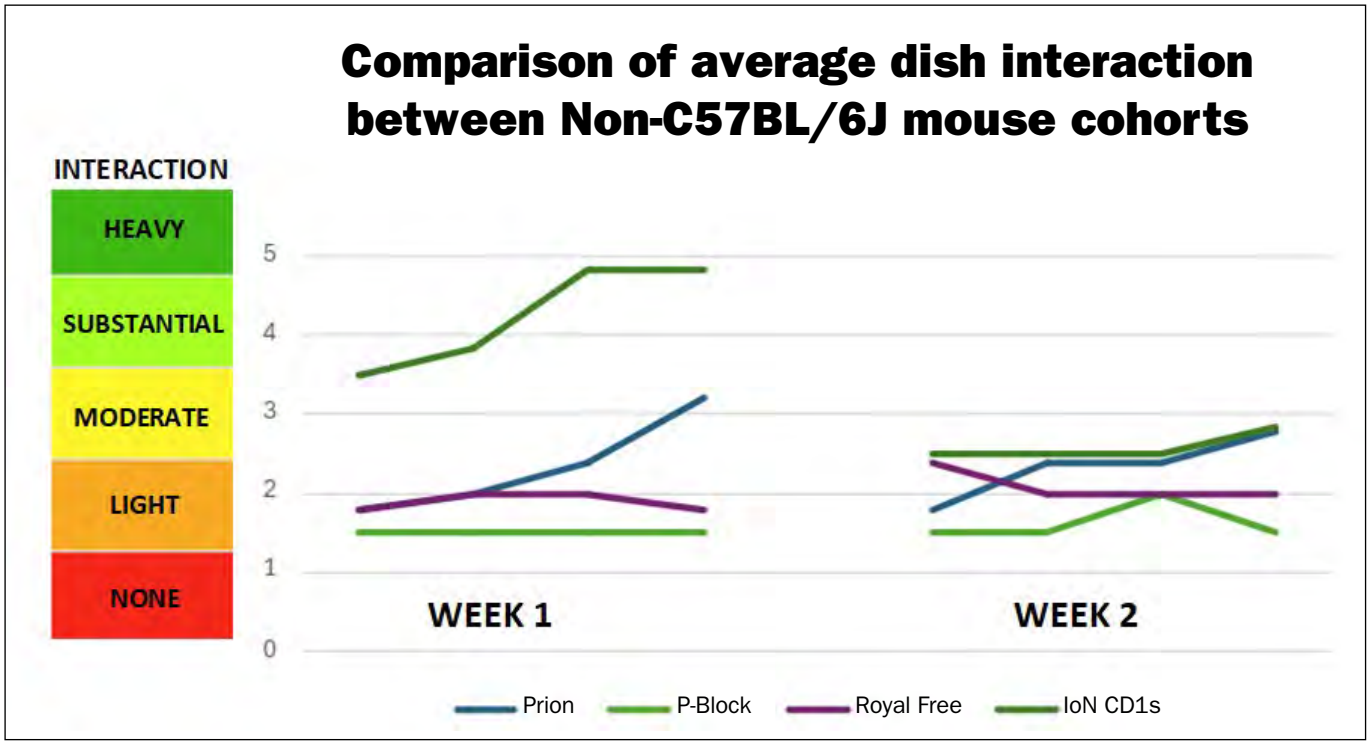

Figure 8. Graph comparing average dish interaction for non-C57BL/6J cohorts across the two-week observation period.

Conclusions

1. The dishes hold diet gel for 3 days with minimal risk of mould formation.
2. Across most cohorts, the data indicates habituation effects, as interaction with the dishes increased with exposure.
3. Within the C57BL/6J strain age likely impacts interaction levels, similarly, strain itself is also a major contributor to engagement differences with the dishes in the remaining cohorts.
4. While some mice ignored the dish, this was outweighed by gnawing and nest incorporation indicating that the dishes provide enriching opportunities for expression of natural behaviours in both rats and mice.

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