DoubleCage – an automatic system for preference testing: validation and first application

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Introduction
For social species such as mice and rats, preference and consumer demand studies should ideally be performed in groups. For this purpose, a new automatic system „DoubleCage“ was developed, validated and used in an initial preference test.

Animals, material and methods
The system consists of two Makrolon® cages connected by a Perspex tunnel (Figure 1). Via subcutaneous transponders and sensors at both ends of the tunnel, transitions of each individual mouse can be detected automatically for further analysis (crossings and dwelling times). For validation, system-recorded data were compared to corresponding videotape data.

In preference test BALB/cOlaHsd and HsdWin:NMRI female mice (16/strain) were tested in groups of four in four different designs (Table 1). All animal were tested for four weeks (five days/week/design, crossover design).

Table 1: Preference test: designs A-D

<table>
<thead>
<tr>
<th>Design</th>
<th>Cage 1</th>
<th>Cage 2</th>
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<tbody>
<tr>
<td>A</td>
<td>Standard (water, food and bedding)</td>
<td>Empty Cage</td>
</tr>
<tr>
<td>B</td>
<td>Standard</td>
<td>Standard + Mousehouse*</td>
</tr>
<tr>
<td>C</td>
<td>Cage with food and water</td>
<td>Cage with bedding</td>
</tr>
<tr>
<td>D</td>
<td>Standard</td>
<td>Cage with food, water and Mousehouse*</td>
</tr>
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*Tecniplast®, Milan, Italy

Results
No statistical difference could be detected between the data collected with the DoubleCage program and video analysis.

A highly significant correlation (p<0.0001) between DoubleCage and videotape data was found in both crossing number ($R^2=1.000$) and dwelling times in section 1 and 2 ($R^2=0.999 & 0.997$), proving the reliability of the system (Figure 2).

Figure 2: (a) Crossing frequency per day between the two cages (section 1 and section 2); (b) Dwelling times in cage 1; (c) Dwelling times in cage 2 (n=39)

In preference testing both strains strongly preferred the standard to the empty cage (A) and the cage containing only food and water to the bedding cage (C) (p<0.0001 for all, Figure 3).

Figure 3: Dwelling times [%] on section 1 and 2, design A-D (BALB/c: P<0.0001 for design A, C & D, p=0.0739 for B; NMRI: P<0.0001 for design A & C, p=0.0003 for B, p=0.0432 for D)

Strain differences were found in both preference behaviour and crossing activity. BALB/c showed a preference for the Mousehouse in B (p=0.0739), but not when the bedding was missing in D. In contrast NMRI significantly favoured the standard cage (B: p=0.0033; D: p=0.0432) (Figure 3).

For all designs crossing activity was higher in BALB/c than in NMRI mice (p<0.0001, Figure 4).

Figure 4: Strain differences in crossing activity [number] ; ***=p<0.0001

Conclusions
According to the results the DoubleCage system can be used in preference test for registering the location of animals. Both strains clearly discriminated between different housing options. A strain-specific validation of the refinement of housing conditions need to be concerned.

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