

# **TECNIPLAST**

*"Performance Standard" assessment for individually ventilated rat cage EMERAT – ER1050* 

The aim of the "Performance Standard" Assessment Test is to monitor the EMERAT ER1050 performance and its impact on animals' growth and behavior. The intra-cage micro-environment gas parameters (NH<sub>3</sub>, CO<sub>2</sub> levels and O<sub>2</sub> stability) were assessed for up to 8 days post bedding and cage change. Intra -cage and room parameters, namely temperature and relative humidity (rH%) were also measured. The potential effects on fundamental physiological parameters were evaluated, i.e., body weight, water and diet intake. Importantly, any signs of distress or stereotypical behaviors were also studied.



# **1. MATERIALS AND METHODS**

**a) Equipment.** Rack IVC ER1040X – Single-sided Ergo Rack (5W x 8H) equipped with 40 ER1050 cages and connected to a SmartFlow ventilation unit. Ventilation setting: 75ACH in Positive pressure mode and -25% differential on negative. Each cage was equipped with 2 bottles (ACBT0312SU, 312 ml/unit).



#### b) Rats.

Fifteen male Hsd:Sprague Dawley<sup>®</sup> SD<sup>®</sup> rats, of body weight between 225 -250 g at arrival. Rats were distributed 4 to a cage (3 cages) and 3 to a cage (1 cage). Cages were randomly



positioned on the rack. Rats were redistributed during the study (3 weeks) taking into account their body weight and the cage densities permitted by the current legislation.



**c) Diet, Water and Bedding.** 800 g Envigo Global Diet 2018 per feeder was distributed at the beginning of the study and at each cage change. The remaining feed was weighed at every cage change. Tap water was provided in bottles, approx. 312 ml in each bottle and two bottles per cage. Bedding: <sup>3</sup>/<sub>4</sub> Fasern, Spruce Wood, (Rettenmaier, D) 250 g per cage. Environmental enrichment in the form of a squared plastic tunnel was provided in each cage.

**d) Gas Measurement.** Drager X-am 7000. CO<sub>2</sub>, NH<sub>3</sub>, O<sub>2</sub>, sensors. The sensor was inserted through the flap of one of the two bottle hollows. When necessary, the sampling was recorded from both positions. The sensor was positioned

at different heights (min 2 cm from the bedding surface and horizontally up to the area below the feeder). Three measurements were performed for each cage: week 1 at day 8 post cage change; week 2 at day 6 and week 3 at day 7.

#### e) Relative Humidity and Temperature.

rH% and temperature were measured inside each cage by placing a 174H Data Logger (DL) Testo inside the feeder (above feed level). A further DL was placed on the rack upper floor for environmental monitoring. Recording length and frequency: reading every 30 minutes throughout the trial. Three replicates were carried out.

## 2. RESULTS

## 2.1 RELATIVE HUMIDITY AND TEMPERATURE IN ROOM (Amb) AND CAGE.

	Mean	Std.Dev
Temp A8 °C	23.65	1.5
Temp B3 °C	23.19	1.38
Temp C6 °C	23.92	1.54
Temp E2 °C	22.36	1.19
Temp Amb °C	21.54	1.35

TABLE 1. Temperature in room and cages. Mean value is representative of N=3 replicates. Range of acceptance: 20-24°C

	Mean	Std.Dev
%rH A8	48.48	7.33
%rH B3	49.99	7.64
%rH C6	53.4	7.23
%rH E2	53.54	7.56
%rH Amb	48.74	9.08

TABLE 2. rH % in room and cages. Mean value is representative of N=3 replicates. Range of acceptance 40-70%.

#### 2.2 FEED, WATER CONSUMPTION AND BODY WEIGHT.

	Weight g					
Age Weeks	7	8	9	10		
Ν	15	15	15	15		
Min	205	278	306	326.8		
Max	230	304	349.6	376.8		
Mean	219.8	292.9	329	351.9		
SD	7.32	8.47	12.24	14.77		

FEED			WATER			
Mean Intake (g rat/day)		Mean Intake (g rat/day)				
Week 1	Week 2	Week 3	Week 1	Week 2	Week 3	
20.3	19.7	21.1	24.7	28.2	23.8	

**TABLE 3.** Body weight of rats at arrival (Week 7) and during the 3 weeks of the study. N = total number of animals.

FIGURE 4. Mean feed and water intake after 1,2 and 3 weeks.

### 2.3 INTRA-CAGE GAS MEASUREMENT (NH<sub>3</sub>, CO<sub>2</sub> AND O<sub>2</sub>).

Intra-cage ammonia was measured in each cage at 8-6-7 days after cage change: the maximum concentration was 6 ppm in cages with 4 rats and a body weight of approx. 300 g and 2 rats and a body weight of 350 g.

Among all measurements, a single detection of 32 ppm of ammonia was registered after 8 days in a cage with 4 rats

and approx. 300 g of body weight. This is explained with the positioning of the probe in proximity of a latrine area, which supports rats' natural behavior and animal "zoning".

In all cages oxygen concentration was always at 20.9% throughout the study and  $CO_2$  was constantly below 0.5% (on average 0.1%).

## **3. COMMENTS AND CONCLUSIONS**



The aim of this study was to evaluate the EMERAT ER1050 performance and its impact on intra-cage environmental condition, animals' growth, physiological conditions in male rats for a period of 3 weeks.

To this aim, intra-cage gas levels (NH<sub>3</sub>,  $CO_2$  and  $O_2$ ), intra-cage and room temperature and rH% were measured. Moreover, ER1050 impact on animal growth, feed and water intake were evaluated.

NH<sub>3</sub> levels were tested at different animal density in relation to the body weight. At all densities, the identification of individual pockets of ammonia was sporadic and occurred in proximity of latrine area, which highlights the natural "zoning" of the rats when housed in the EMERAT ER1050. NH<sub>3</sub> values displayed negligible concentrations that might impact the wellbeing of the animals, indicating that ER1050 can ensure a highly efficient NH<sub>3</sub> clearance in a set-up that is typically used for large size animals. The good ventilation efficiency is confirmed by the stability of the oxygen at 20.9% and by the very low concentration of CO<sub>2</sub> in the cage.

Intra-cage temperature and rH% monitoring showed mean values ranging between 22.36 to 23.92 °C (temperature) and between 48.48 and 53.54 (rH%). The intra-cage temperature

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average standard deviation was 1.4, comparable to that from the room (1.35). This data indicates that ER1050 can properly maintain the intra-cage temperature and relative humidity within the acceptable range of respectively 20 to 24°C and 40 to 70% rH. Additionally, the intra-cage rH average standard deviation was 7.44, lower than 9.08 registered in the room. This suggests that ER1050 can smooth at cage level sudden fluctuations of extra-cage environmental parameters.

Feed and water consumption (including spillage) correlates positively with body weight indicating a physiological feeding behavior typical of that species/strain. Similarly for water consumption (including spillage), the percentage on body weight ranged between 10 and 13%, as expected and within physiological limits.

In conclusion, the Emerald ER1050 allowed rats to maintain excellent physiological conditions without impacting their behavior or being detrimental to their physical condition. These results indicate that ER1050 is the first rat cage of this size housing up to 4 experimental rats of the most used weight, while ensuring optimal animal wellbeing and technical performance.

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