An Opportunity for Refinement: Continuous In-Cage Temperature and Activity Monitoring Using the UID Matrix in Rats

Author(s): Juan Antonio Garcia Thomas[1], Kalena L. Gottwald[4], Fabrizio Scorrano[2], Carine E. Elkhoraibi[3], Colin S. Osborne[1]

Affiliations: [1]Novartis Institutes for BioMedical Research Global Health Emeryville, CA, United States; [2]Novartis Institutes for BioMedical Research Comparative Medicine, Basel, Switzerland, [3]Novartis Institutes for BioMedical Research Animal Welfare Compliance Emeryville, CA, United States, [4]Novartis Institutes for BioMedical Research Comparative Medicine Emeryville, CA, United States

P #4

Introduction

Common confounding factors in *in vivo* studies are stress related changes due to disturbances to animals routinely removed from their cage for health assessment, husbandry, or experimental procedures[1]. The Unified Information Devices, Inc. (UID) rat caging matrix is a radio-frequency identification (RFID)-enabled system that provides remote and continuous monitoring of locomotor activity and body temperature for multiple rats in their home cage environment. This technology offers an opportunity to minimize disturbances to improve data reliability and quality while expanding the parameters of routine health assessment. This may allow the refinement of rat experimental procedures by providing improved supportive care and/or introducing earlier humane endpoints.

How does the UID rat caging matrix work?

The animals have a temperature sensing microchip with a unique ID code implanted subcutaneously. They are then housed in individually ventilated cage (IVC), which are each located directly above a reader plate.

Reader plates are divided into 12 zones that continuously collect activity and temperature information from each animal's unique RFID microchip. The reader plates are connected to a matrix controller and laptop compatible with UID software, which allows the user to input study details, customize the sample frequency, and export data throughout the study (**Figure 1**).

Figure 1: UID Rat Caging Matrix System Set-up and Overview

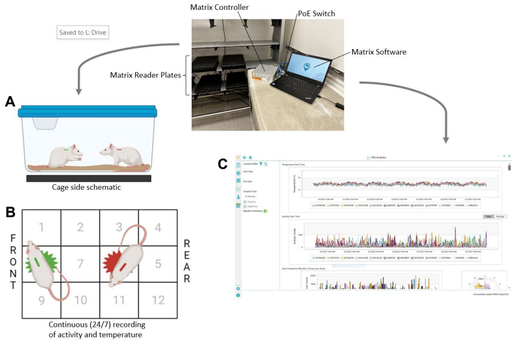

Figure 1: An overview of the UID rat caging matrix system set-up. The cage side schematic of the cage directly above the reader plate (1A). The reader plate is divided into 12 zones that communicate with each unique RFID microchip (1B). Data analytics from the Matrix software system for temperature and activity over time (1C). Activity Index is calculated based on movement between zones.

Figure 2: Manipulation sequence for each experimental group

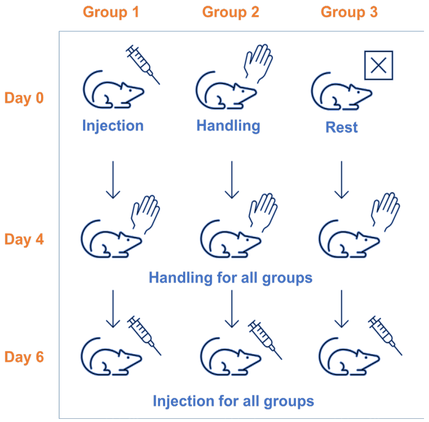

Figure 2: An overview of the manipulation sequence of each group on day 0, day 4, and day 6. Group 1 received injection, group 2 receiving handling, and group 3 received no manipulation on day 0. All groups received handling on day 4 and injection on day 6.

Objective

The objective of this study was to evaluate the effects of different manipulation techniques, such as handling and intraperitoneal (i.p.) injection, and whether the sequence of these manipulations over time impacts each animal's activity and temperature. It also aimed to examine the potential impacts of a routine cage change on temperature and activity.

Materials & Methods

In this study, 9 male Sprague Dawley rats (250-300g, n=3/group) received either handling, i.p. injection (0.9% NaCL @ 5mL/kg), or no manipulation (rest) on day 0, day 4, and day 6. Each group received a different sequence of manipulations (**Figure 2**). Group 1 underwent injection (I) on day 0, handling (H) on day 4, and injection on day 6 (I-H-I). Group 2 underwent handling on day 0, handling on day 4, and injection on day 6 (H-H-I). Group 3 underwent rest (R) on day 0, handling on day 4, and injection on day 6 (R-H-I). On day 2, all animals underwent a routine cage change that included changing the cage bottom, the lid, the wire bar feeder, and enrichment. Animals were housed in standard disposable Innocage™ IVC rat cages on an Innorack™ IVC rat rack with the UID reader plates directly beneath the cages. All cages were continuously monitored using the UID caging matrix system for 14 days with 1440 samples collected per hour. The length of the period considered for each day extended from the time the procedure was conducted until the dark cycle began at 6pm.

Results

As shown in **Figure 3**, group 1 (I-H-I) showed significant higher activity index on day 0 when injection was performed compared to day 6 when injection was repeated (**p= 0.008) (**3A**). Body temperature decreased over time with significant differences between day 0 and day 4 (**p=0.003), day 4 and day 6 (**p=0.004), and day 0 and day 6 (****p<0.0001) (**3B**). For group 2 (H-H-I), there was no difference in activity index and body temperature with each manipulation (**3A, 3B**). Group 3 (R-H-I) showed significant higher activity index on day 4 when handled compared to day 6 when injected (****p<0.0001) (**3A**).

As shown in **Figure 4,** total activity index and average body temperature on the day of cage change was significantly different than the day before (***p<0.0001) and the day after (***p<0.0001) (**4C,4D**). There was not a significant difference between total activity index and average body temperature for the day before and the day after cage change (**4C,4D**).

Figure 3: Impact of sequence of procedures on total activity index (3A) and body temperature (3B) within each cage for each day .

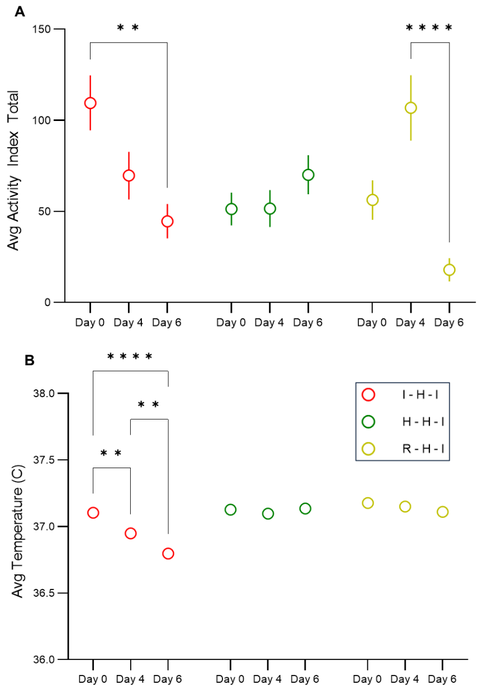

Figure 3: The average activity index total (3A) and average body temperature (3B) for group 1 (I-H-I), group 2 (H-H-I), and group 3 (R-H-I) on day 0, day 4, and day 6. Data was analyzed using mixed 2-ways Anova.

Figure 4: Impact of cage change on activity index (4A) and body temperature (4B).

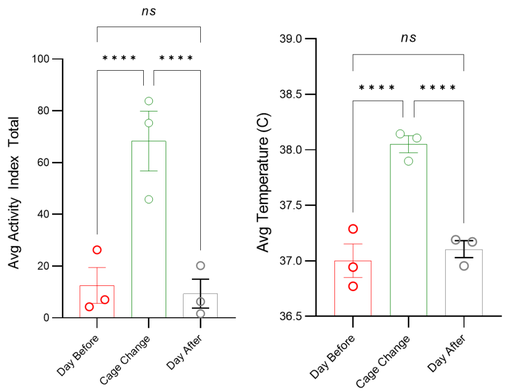

Figure 4: The average activity index total (4A) and average body temperature (4B) of all animals on the day before, the day of, and the day after a routine cage change on day 2. Data analyzed using mixed 2-way ANOVA with post-hoc comparisons via Tukey's test.

Discussion

After a prior injection or handling, the activity and temperature levels decrease or maintain for the repeated injection. This suggests that repeated animal handling and/or training before injections can help the animal habituate to the procedure. This may help reduce stress for the animal and therefore, reduce outcome variability.

Cage change resulted in increased activity and body temperature, but within 24hrs, these values returned to levels similar to those the day before cage change. This indicates that the day of cage change may be stressful for animals and is an important consideration when planning experimental procedures to avoid stress-related cofounders.

Conclusions

The UID rat matrix system is a valuable tool to noninvasively measure changes in activity and temperature in rats. The ability to evaluate the effects of different techniques can help inform husbandry, experimental procedures, and health assessments to ultimately refine the animal experience.

References

1 Balcombe, J. P., Barnard, N. D., & Sandusky, C. (2004). Laboratory routines cause animal stress. Journal of the American Association for Laboratory Animal Science, 43(6), 42-51.

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Kalena.Gottwald@novartis.com