Multi-Company Validation of the ActualHCA™ Home Cage Monitoring System for Rodent CNS Safety Pharmacology Studies

Chiara Giuliano[1], Ian Ragan[2], Aileen Milne[3], Joanne Sutherland[3], Mike Rolf[1], Greet Teuns[4], Eric I Rossman[5], Michelle McClafferty[5], Ajeesh Koshy Cherian[5], J. Douglas Armstrong[6], Rowland Silito[6], Anthony Holmes[2]

1. AstraZeneca, Cambridge, UK and Gothenburg, Sweden; 2. NC3Rs, London, UK; 3. Charles River, Edinburgh, UK; 4. Janssen, Beerse, Belgium; 5. GSK, Collegeville, PA, USA; 6. Actual Analytics, Edinburgh, UK

Introduction

In drug discovery, monitoring behavioural changes is key to many animal studies including safety pharmacology tests focusing on central nervous system (CNS) endpoints. In most cases, recording the behaviour of laboratory rats requires them to be removed from their home cage environment and cage mates and placed in novel and often solitary environments where they undergo a variety of tests for various lengths of time, e.g. the Modified Irwin test or Functional Observational Battery (FOB) [1]. The tests are often subjective, there can be significant data variability due to factors such as the presence of the experimenter and different laboratory conditions, and manual observations are usually limited to 'snapshots' during the light phase when rodents are naturally less active.

In 2011 AstraZeneca set the Rodent Big Brother CRACK IT Challenge [2] to develop a novel technology to record activity, behaviour and temperature of individual rats continuously when group-housed in conventional individually ventilated home cages (IVC) in a portable cage rack. The Home Cage Analyser (ActualHCA™) system was developed to meet the challenge and is capable of longitudinally and non-invasively tracking individual rats (using video and RFID dual identification and temperature transponders) in group-housed environments in an adapted IVC cage rack (Figure 1) [3].

Figure 1. The Home Cage Analyser (ActualHCA™) system. The system is compatible with IVC home caging and for the current study four systems were installed in a Techniplast blue-line rack. An RFID baseplate array sits under the cage with 12 antennae scanning at ~1Hz provides monitoring of location and temperature of each individual tagged animal. This is then used to generate the temperature, locomotor activity and social proximity data. Top mounted infrared lights and a side mounted infrared camera continuously records 25Hz HD video footage for on-demand review of any significant events and permits automated behaviour recognition using computer vision and machine learning algorithms. The system is currently able to identify temperature, a range of ambulatory and vertical activities, eating, drinking, rearing and social interactions.

Here we describe a multi-company validation study to build further confidence in the ActualHCA™ for CNS safety pharmacology studies. The collaboration, led by the UK National Centre for the 3Rs (NC3Rs), includes Actual Analytics (developers of the ActualHCA™), Charles River, AstraZeneca, GSK and Janssen. The study will assess the effects of three test compounds (with different pharmacology and mechanism of action) provided by the pharmaceutical company partners in group-housed rats using the ActualHCA™. Results generated in the ActualHCA™ for each of the test compounds will be compared with existing data to assess the utility of the system in identifying behaviour changes indicative of adverse effects observed in the clinic that were not identified using FOB/Irwin assessments. We present (i) baseline data confirming the consistent detection of a transient increase in temperature and activity in animals in response to dosing, and (ii) available data relating to the Janssen test compound.

Materials, methods and study design

The study is being conducted in two stages as described below.

Stage 1: to collect baseline behaviour, activity and temperature data in group-housed rats dosed daily for 28 days.

- N=12 male Han Wistar rats (sourced from Charles River UK Limited, Margate).
- Rats were implanted with temperature sensitive passive radiofrequency identification (RFID) transponders and housed three per cage.
- All rats received 0.9% saline, at a dose volume of 10 mL/kg, daily for 28 days.
- Locomotor activity, social activity and behaviour (climbing, rearing, eating drinking, social distance, seizure, tremor fighting) and body temperature were monitored continuously over 28 days.
- Cages were changed on the same day of each week, as close to the same time as possible, to ensure any disturbance was consistent.

Stage 2: To assess the effects of each test compound on group-housed rats. To date we have conducted these studies only on the Janssen compound.

- N=24 (6 per group) male Han Wistar rats (sourced from Charles River UK Ltd, Margate).
- Rats were implanted with temperature sensitive passive radiofrequency identification (RFID) transponders and housed three per cage.
- Rats received either control item (Hydroxypropyl-β-cyclodextrin) or Janssen test compound (10, 40 or 300 mg/kg), at a dose volume of 10 mL/kg, as a single dose.
- Drinking, rearing, distance moved, mobile/immobile time, transitions, separation and isolated times, zone occupancy and body temperature were monitored continuously for seven days after dosing.
- Cages were changed on the same day of each week, as close to the same time as possible, to ensure any disturbance was consistent.
- Compound classes and mode of action for each of the test compounds were blinded to Actual Analytics and Charles River to ensure this information does not influence analysis and interpretation of data generated in the ActualHCA™ system.

Results

- Data have so far been collected and analysed for the Janssen test compound (Figures 2 and 3). Studies to assess the effect of GSK and AstraZeneca test compounds will be completed by the end of October 2021.
- The highest dose of Janssen test compound (300 mg/kg) induced an increase in locomotor activity (Figure 3) and body temperature that persisted throughout the light phase the following day. This was not observed in control animals (data not shown).
- Until the data generated in the ActualHCA™ have been fully analysed it is not possible to share detailed information about how predictive the data are compared with the existing preclinical and clinical data for each test compound.

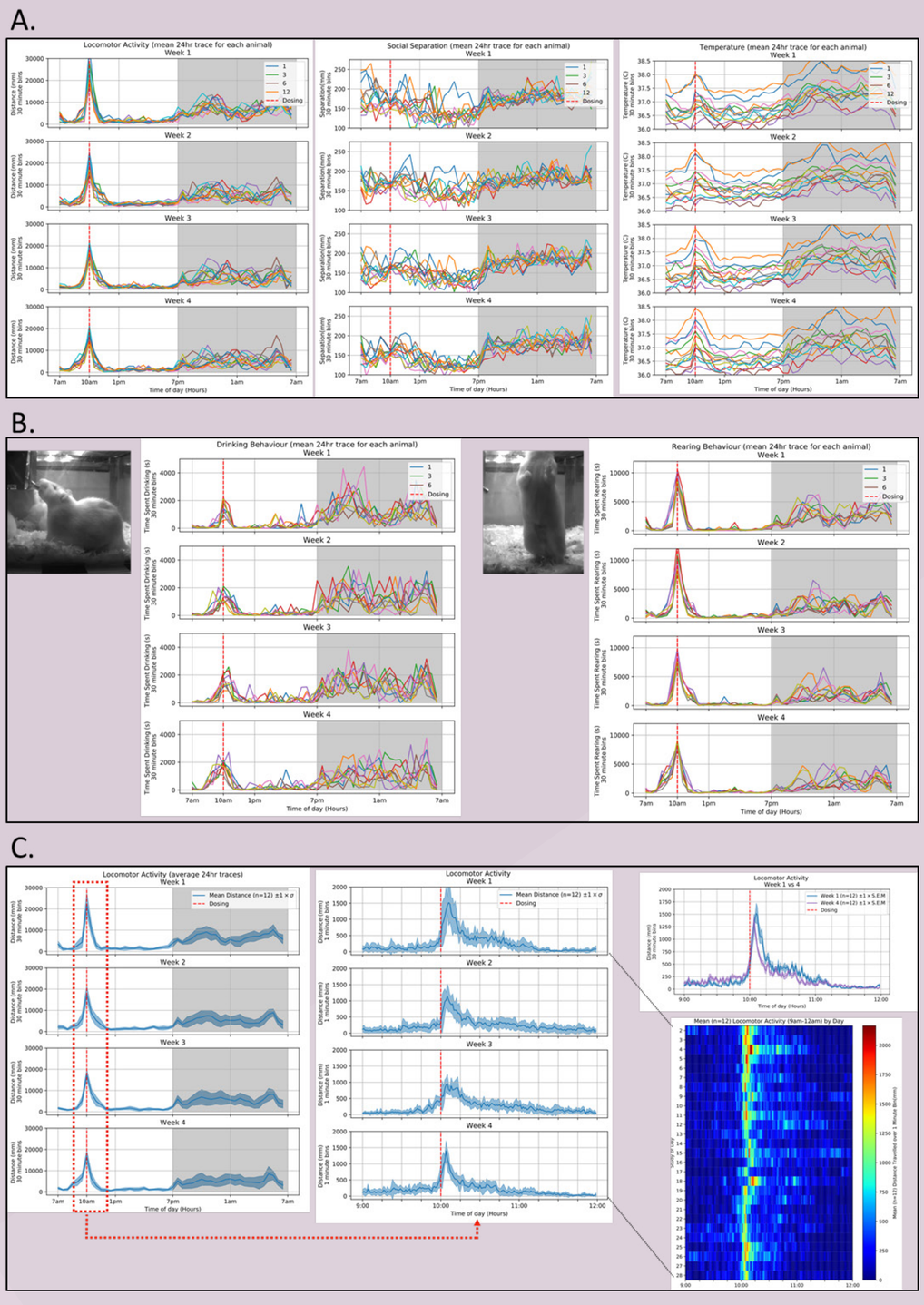

Figure 2. Stage 1 locomotor and behaviour baseline recordings over four weeks. Baseline recordings clearly demonstrate the ActualHCA™ can detect changes in locomotor activity, social separation and temperature recordings (A) and drinking and rearing behaviour (B) in group-housed individual animals. All data shown are mean 24 hour traces for individual animals. It is also possible to detect changes in locomotor activity immediately following dosing (saline) (C). Data represented as average locomotor activity (n=12).

Figure 3. Daily effect of Janssen test compound (stage 2) on locomotor activity in group-housed rats three days post-dose compared with baseline. Data generated in the ActualHCA™ clearly demonstrate a substantial increase in locomotor activity following all dose levels which continues during the light phase. This is not observed in control animals.

Conclusions

- We have confirmed that the ActualHCA™ system can consistently detect transient increases in temperature and activity (including locomotion, rearing and drinking behaviours) in control animals post-dose (Figure 2), corroborating data generated during the original validation of the system [3].
- The ActualHCA™ was able to detect increases in locomotor activity and temperature in group-housed rats in response to the Janssen test compound (Figure 3).
- Furthermore, these effects are consistent with those observed during the original CNS safety pharmacology studies conducted by Janssen using the FOB/Irwin battery of assays (personal communication). However using the ActualHCA™ this could be continuously evaluated.
- Further studies assessing the effects of GSK and AstraZeneca test compounds will hopefully continue to confirm the utility of the ActualHCA™ in identifying behavioural, locomotor and temperature changes in animals indicative of adverse effects seen in humans.
- Home cage monitoring of group-housed rats during CNS safety pharmacology studies has the potential to deliver significant animal welfare and scientific benefits over traditional approaches, e.g. FOB/Modified Irwin studies.

References

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