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HOW TECHNOLOGY CAN SUPPORT BETTER SCIENCE

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ABSTRACTS

TECNIPLAST

Vivarium of the future - the role of technologies in animal welfare, facility management and better (reproducible) science

Alon Harmelin - President's consultant, Weizmann Institute of Science, Israel

The use of animals for research and teaching began hundreds of years ago. Animal facilities have dramatically changed over the years, through an evolutionary process based on the understanding of both animal needs and science. Significant landmarks, such as the 3 Rs of Russell and Burch published in 1959 in 'The Principles of Humane Experimental Technique" and the 'Brambell Report' in 1964, which outlined the five freedoms (freedom from hunger or thirst; discomfort; pain, injury; restrictions on expressing normal behavior; and fear and distress), along with the 'Guide for Laboratory Animal Facilities and Care' and the EU Directive, have been pillars of progress in animal husbandry and care. These are supported by many national and international laws and laboratory animal organizations. The training and experience of animal care staff and veterinary specialists supporting research have grown dramatically, alongside breakthroughs in facility equipment.The next vivarium upgrade will likely combine current practices and advanced technologies, such as the DVC®, with new technologies, specifically artificial intelligence (AI). AI is expected to impact animal welfare, management, and scientific outcomes in numerous ways, including enhancing data analysis and improving decision-making, among others.

Improving Animal Welfare and Operational Efficiency using Digital Ventilated Cages (DVC®)

Jeetendra Eswaraka - Vice-President University-wide core services, Rutgers University, USA

Operational inefficiencies in daily checks and bedding change intervals is commonly accepted problems in the management of animal facilities due to the lack of appropriate tools to improve them. These inefficiencies also inadvertently have an impact on rigor and reproducibility of data from animal models of human disease. Home caging monitoring tools such as the DVC caging system have the ability to continuously monitor the animals 24 x7 presenting an opportunity to overcome some of the deficiencies in current practices. A series of studies were conducted to develop performance criteria for animal health checks and bedding change intervals. Our results indicate that the DVC caging system is able to accurately predict clinical signs of disease much earlier and with more accuracy than daily visual observations. We validated the use of bedding status index (BSI) as the primary method for bedding changes. Our results indicate that BSI method of cage change compared to a scheduled 2 week change out results in operational savings of over 70% and improves efficiencies in husbandry operations.

Transforming Animal Care and Efficiency: The Adoption of Digital Ventilated Cages in Research Settings

Anne-Lise Huot - Zootechnician, Université de Poitiers - Isabelle Petit Paris - Responsible PREBIOS, Université de Poitiers; Prebios, France

The Université de Poitiers research facility initiated the use of Digital Ventilated Cages (DVC) with a strong commitment to enhancing the well-being of laboratory animals and humans, increasing cost efficiency in animal care, and augmenting the value of scientific research. This presentation primarily focuses on exploring the introduction of DVC into the facility, the challenges encountered during implementation, and the strategies for utilizing DVC to enhance animal welfare.

Introducing DVC® Housing in a fully operational animal facility

Ronald van Os - Scientific breeding coordinator/Member Animal Welfare Body/Member Management Team UMCG, Groeningen, The Netherlands

The Central Animal Facility (CDP) of the University Medical Center Groningen (UMCG), the Netherlands, started using DVC® two years ago as a replacement for both IVC and conventional cages. The motivation for this decision was based on three fundaments: laboratory animal and human health and welfare, cost efficiency in animal caretaking and added value to scientific research. The focus of the presentation will be on: How was DVC® introduced into the facility? Which challenges came along? How do we use DVC® in order to optimize animal welfare? How can the DVC® system help us to monitor animal welfare? How are animal caretakers involved in the transition to DVC®? How are researchers using DVC®, in particular with the DVC®Analytics software? These question will be addressed using our own experience with the system.

Assessing mice activity in breeding cages using digital technology

Jason Villano - Associate Professor of Molecular and Comparative Pathobiology, John Hopkins University, USA

Discovery and innovation drive science and technology. In the age of globalization and digital media, the whole world is performing and participating on one platform. Utilizing and applying these innovations in research animal facilities modernizes the workplace, enhances work efficiency and animal welfare, and increases scientific output. Here, we use ventilated cages specifically digitally designed to investigate the activity and the use of cage floor space of breeder mice (trios and pairs) and their litters. We followed daily mouse activity through breeding cycles using several parameters like heat map, diurnality, activity pattern irregularity, and spatial activity. Our preliminary findings indicated that mice had increased activity periparturient during the light phase of the cycle, that mice had higher activity during the dark phase within 1 wk of breeding cage setup, that activity increased over time especially in the central hours of the day, that animals had increased irregularity pattern during light and dark periods around delivery date, and that there was an increased spatial activity including the use of the front sides of the cage periparturient. In conclusion, mice were more active periparturient. Additionally, the digital capability of cages used in this study provides an enhancement of animal welfare and presents a future and modern path for animal research programs.

Automated Animal Welfare Assessment: Tecniplast's Digital Ventilated Cage System and its predictive monitoring capabilities

Raphael Doenlen - Scientist, Head of the Phenotyping Unit, EPFL, Switzerland

The introduction of automated detection technologies holds promise for enhancing animal welfare within laboratory environments. In this project, we examined the Digital Ventilated Cage (DVC®) system developed by Tecniplast and designed to automatically monitor the animal activity in the home-cage. Researchers at the École Polytechnique Fédérale de Lausanne (EPFL) conducted a thorough assessment over a period of time and meticulously observed various indicators of animal welfare in a controlled environment. They also evaluated a significant aspect of the DVC® system, the Welfare Check algorithm. The results will show if this new algorithm used in the DVC technology improve the detection of potential welfare issues and if it could improve the monitoring of animal welfare performed in experimental animal facility.

Transitioning to digital housing technology within a high containment (ABSL3) biosafety facility – lessons learned, benefits gained

Betty Theriault - Director, Gnotobiotic Research Animal Facility/Biocontainment Facilities; Professor, Department of Surgery, University of Chicago; The University of Chicago, USA

Animal health and home environment monitoring is fundamental to all animal care and use programs. The ability to provide adequate animal health monitoring and home cage care is dependent upon the availability of trained animal care professionals to provide this service. The need for increased monitoring and animal care is inherent in infectious disease studies where observations may be required at more frequent intervals by personnel specially trained to work in high containment environments. The COVID-19 pandemic brought into focus the challenges of facilitating the use of animal models in infectious disease research while at the same time experiencing personnel shortages. Combined with the opportunity to renovate an aging biocontainment facility as the height of the pandemic waned was the opportunity to transition to digital housing technology. Coordinating a team comprised of digital technology experts, facility engineers, operation specialists, biosafety officers, animal care technologists, veterinarians and industry partners proved essential in installation and start-up. Training of all stake-holders inclusive of research personnel is essential in the adaptation of digital housing technology within infectious disease programs. Harnessing the potential of remote cage monitoring can enhance animal welfare and optimize husbandry scheduling and labor resources. Additionally, incorporating Bio-Exclusion technologies within a Bio-Containment environment strategically prepares for the merging of these overlapping areas of research interest. Taken together, the adaptation of bio-exclusion and bio-containment digital housing technology can enhance animal welfare, disaster preparedness, and optimize facility utilization for bio-exclusion/bio-containment research.

Evaluation of the DVC® - Digital Ventilated Cage system for facility management, assessment of animal welfare, and potential research use in preclinical drug development

Thomas Reich, Laura Breidenbach, Michaela Socher - Study Director, Abbvie, Ludwigshafen, Germany

In recent years, technologies for rodent caging emerged that promise to enhance animal welfare by allowing 24/7 home cage monitoring of selected physiological and environmental data. At AbbVie we evaluated the Tecniplast Digital Ventilated Cage (DVC®) system for its sensitivity to detect changes in activity levels and sleep patterns as well as for its suitability to guide on-demand cage changes (bedding tool) in mice. A proof-of-concept study was conducted to analyze the changes in locomotor activity patterns upon administration of well characterized test compounds for increasing locomotor activity (caffeine) and reducing activity (LPS) to mimic effects during safety testing of drug candidates. The system was sensitive to detect an increase and decrease in activity and seems thus valuable to generate endpoints in preclinical studies. In an additional study we aimed to utilize DVC®-guided cage changes to increase a fixed cage change interval (weekly). Upon implementation of the DVC® bedding tool, we could increase the mean cage change interval to 11 days (average) for 4 mice per cage (males and females) and 26 days (average) for pair-house females, while maintaining animal health as determined by ammonia measurements. Thereby, we could greatly reduce cage change associated stress for the animals and increase efficiency in terms of used personnel and material resources.

Automated detection of behavioral stereotypies using the DVC® system to enhance animal welfare and improve research outputs

Katharina Tillmann - Department of Neurophysiology and -pharmacology, Center for Physiology and Pharmacology, Medical University of Vienna, Austria

Standard housing conditions of experimental animals in biomedical research, the large majority of which are small rodents, are often poorly aligned with the physiological needs of the animals. As a consequence, animals may experience stress resulting from a disruption of their evolutionary determined homeostatic demands leading to the development of different types of behavioral abnormalities. Abnormal repetitive behaviors are classified as behaviors that are inappropriate, repetitive and unvarying in either goal or motor pattern and most frequently present as stereotypies, defined as invariantly and inappropriately repeated set of movements and/or body postures without apparent goal or function. Indeed, behavioral stereotypies are a common and significant problem in standard mouse husbandries which not only pose a substantial burden for the affected individuals, but also compromise the validity, reliability and replicability of the experimental data obtained from using these animals. Thus, the occurrence of behavioral streotypies is not only of pivotal animal welfare concern but also relevant to research outcomes. Yet, until now the abilities for a routine, large-scale surveillance of stereotypies within an experimental rodent facility have been hampered by the lack of suitable monitoring system. Here, we demonstrate the application of the DVC® home cage observation system as a non-invasive, low labor intense and easy to use system for the reliable tracking and identification of welfare-related behavioral stereotypies in a routinely used mouse caging system. The development of a dedicated algorithm for the analysis of DVC® generated data allows for the unsupervised, automated detection of stereotypies from continuously acquired data which constitutes a prerequisite for the application in routine animal care and veterinary supervision with the ultimate goal to improve the well-being of the experimental animals and to lower variability of research outcomes, enhance reliability and replicability of data obtained and thus reduce numbers of animals used in experimental research.

Influence of transfer method into new cages on mouse locomotion measured in the DVC® system

Markus Brielmaier - Head of Core Facility Laboratory Animal Science (CF-LAS) at Helmholtz Zentrum Munich, Germany

Transferring mice in a plastic tube (tunnel handling) offers many advantages over tail restraint. It has been shown in numerous publications that these animals exhibit less stress. The aim of this study is to investigate the effect of tunnel handling vs tail restraint with tweezers on the locomotion of mice. In a DVC® rack (Tecniplast), 3-week-old C57BL/6J male and female mice bought from a breeder were kept for 16 weeks in cages of 3 animals each. Eight cages of male mice and eight cages of female mice were transferred weekly to fresh cages using either tweezers or a tunnel (4 groups, 32 cages, n=96 mice). The transfer process was performed by two animal handlers simultaneously for one cage from each group to avoid any time effect. For analysis, a distinction was made between weeks one and two (after arrival of the animals) and weeks 9 and 10 (after acclimatization to the respective transfer method). The duration and locomotion activity immediately after transfer to new bedding did not differ between groups at any time. There was no difference in daily and nocturnal activity in weeks one and two. However, after week two, total locomotion was significantly reduced in in the tunnel-transferred mice compared to the mice transferred with tweezers.

A multicenter study of slow oscillations in spontaneous activity in two commonly used mouse strains.

Brun Ulfhake - Professor/Guest Researcher, Stockholm University, Sweden

Many mammalian species, including rodents, not only have a circadian rhythm in the activities of daily life, but also additional, slower recurring rhythms, the so-called infradian rhythms. These are often related to seasonal variations in climate and conditions that favour reproduction, and are thought to be triggered by environmental stimuli such as the length of the light and dark phases of the day. Recently, we have shown that laboratory mice (C57BL/6) kept under constant conditions exhibit highly significant infradian oscillations of activity with a cycle duration of 2 to 4 months. This suggests that such rhythms also exist under standardised laboratory conditions, raising the question of whether these rhythms are generated at least in part by an intrinsic oscillator and not exclusively by environmental stimuli. We have now extended these observations in a multicenter study (Basel, Toulouse, and Stockholm) in which we used undisrupted DVC recordings over a period of 1.5 years or more in the outbred Swiss and the inbred C57BL/6J mouse strains.
The preliminary results show that male and female mice of both strains, housed three or four to a cage, oscillate with peaks and lows in activity at all three sites. The oscillations are not synchronised between cages and the period ranged from 1 to 5 months with an amplitude in the range of 0.3-2 standard deviations of the mean activity. In addition, the recordings from the home cages showed that the recurrent oscillations usually occurred only after 2 and up to 5 months of environmental adaptation. In the period preceding the slow recurrent oscillations, there were often quite dramatic changes in in-cage activity. Furthermore, the slow oscillations in activity were also observed in individually housed mice, suggesting that group housing is not a prerequisite for recurrent slow oscillations in activity.
The implications of the results for the use of laboratory mice in experimental studies and the cause(s) of these oscillations are discussed.

Revolutionizing Laboratory Pup Birth Detection: an innovative acoustic monitoring approach

Jan-Bas Prins - Professor of Laboratory Animal Science, Leiden University Medical Centre, The Netherlands, Marcello Raspa - Technological Director CNR-IBBC; EMMA/INFRAFRONTIER, Monterotondo Scalo, Italy

Accurate and timely determination of pup birth in biomedical research and breeding settings remains an ongoing challenge, with often delayed observation of pups born. Historically, methods based on visual cage inspection, controlled mating and of pre-partum symptomatology are inadequate to meet precise research timelines. An innovative approach with automated detection methods was deemed essential to fill the gap. This study introduces a novel technique using mini-microphones placed in the cage to capture specific acoustic signals. We focused on the detection of low-frequency wriggling calls emitted by mouse pups and mothers' vocalisations (to verify) starting from a week before the anticipated delivery onward. Utilizing Digital Ventilated Cages (DVC®) from Tecniplast, 10 cages at CNR-IBBC in Rome and 10 cages at The Francis Crick Institute in London were each equipped with a microphone to capture these wriggling calls and vocalisations. A pregnant C57BL/6 female - with or without male - was transferred to each cage at least one week before the calculated parturition date. The capturing of sound started immediately after transfer and continued till a week after parturition. Initial results from 30 deliveries indicate that the detection of wriggling calls is sensitive and accurate to detect pup delivery. Trials are expected to be finalised by the time of DVF-3 where all results will be presented.

Revealing aggression in male mice through DVC®

Fabrizio Scorrano - Head Emerging Technologies, Novartis AG, Basel, Switzerland

The DVC® system offers significant advantages in the continuous, 24/7 monitoring of mice, providing complementary support to animal caretakers and veterinarians in their daily routines. A prevalent challenge in managing rodent facilities is the effective identification and monitoring of aggressive interactions among male mice. This presentation will delve into the experiments conducted at Novartis, focusing on the enhancement and refinement of algorithms aimed at improving the detection and monitoring of such aggressive behaviors. Through these advancements, we aim to bolster the scientific validity and ethical integrity of our research experiments.

Video-monitoring and AI solutions push animal monitoring to the next level with a version 2.0 of the DVC® rack

Pierre Lainee - Head of In Vivo Research Center, Sanofi, France

Digital biomarkers enable the collection of objectives, quantifiable and continuous data. The DVC® technology has already generated successful indexes to monitor animal locomotion, bedding status and the regulatory disruption mobility.
It is now expanding to the fight count and stereotypic detection. But we are limited by the data collected by the sensor plate, preventing from detecting more subtle or complex behaviors.
As video-monitoring is now accessible using AI platforms, the DVC® 2.0 comes as an integrated solution, opening the door to more ambitious applications. The presentation will demonstrate the detection of behaviors such as drinking, eating, rearing, climbing or grooming.
Considering the increasing performances of the algorithms, a wide range of applications can now be considered to automatize or facilitate phenotyping studies, improving capacities in domains such as preclinical pharmacology or safety. This successful evolution implanted in mice could soon be expandable to rats or event non-rodent species.

Voluntary aerobic running activity improves pathologic outcomes in a mouse model of acquired epilepsy

Valentina Kebede - Istituto di Ricerche Farmacologiche Mario Negri IRCCS, Milan, Italy

Healthy life-style was reported to improve neurological outcomes after acute brain injuries. In particular, physical activity was suggested to improve neurological deficits and reduce seizures in epilepsy. Our study investigates whether aerobic and regular physical exercise reduces the risk of developing epilepsy, the burden of the disease and neuropathology following an epileptogenic brain injury. C57BL6/N adult male mice had free access to running wheels (RW) in their home cage according two protocols: 1) mice were exposed to RW for 5 weeks before intra-amygdala kainate to induce a status epilepticus (SE) that leads to epilepsy development. Then, injured mice were allowed to run for 6 additional weeks; 2) mice were exposed to RW 24 hours after SE induction and for 10 weeks, a time required for chronic epilepsy development. Control mice were similarly exposed to SE but left in their home cage in the absence of running wheels (sedentary mice). Data show that RW activity reduced SE duration and severity ($p<0.05$ vs sedentary mice). Moreover, RW reduced epilepsy incidence independently on the running protocol and it reduced seizure burden and disease progression ($p<0.01$ vs sedentary mice). Running activity in protocol 1 reduced seizures duration ($p<0.01$) and exerted neuroprotective action on GluR 2/3-positive hilar mossy cells ($p<0.01$). Notably, seizure duration negatively correlated with the number of hilar mossy cells ($p<0.01$). Moreover, RW activity corrected aberrant neurogenesis by decreasing the number of DCX-positive neurons in the sub- and intra-granular zones of the dentate gyrus ($p<0.05$) and the hilar region ($p<0.01$ vs sedentary epileptic mice). DCX-positive cells in the dentate gyrus positively correlated with the number of seizures ($p<0.05$). RW activity in both protocols significantly reduced MMP-9 blood level, an index of blood brain barrier leakage. Data support the beneficial effects of physical activity to improve pathologic outcomes after an epileptogenic brain insult.

Urination in the Home Cage: detection of polyuria in mouse models of metabolic dysregulation

Thomas Svava Nielsen - TS Nielsen Scientific Consult, Copenhagen, Denmark.

Blood glucose is arguably one of the most common and most important in vivo parameters in metabolic research. Given the pivotal role of dysregulated glucose homeostasis in metabolic disease, particularly in diabetes, numerous pharmacological and genetic animal models of hyperglycemia are available, and widely used. However, in most models there is a large individual variation in the incidence, severity, and time course of diabetes development. This necessitates frequent measurements of blood glucose to monitor disease progression, which typically involves cutting or puncturing the tail vein of the mice.

A hallmark feature of hyperglycemia is increased urination (polyuria), and by leveraging the continuous monitoring of bedding status by the DVC system, we have developed an algorithm that can reliably detect the appearance of diabetic symptoms from the increase in bedding moisture.

Thus, we present a novel sample-free and non-invasive approach to identify the onset of diabetes in individual animals while they are ambulatory in their home cage. We believe that his technique represents a significant refinement and improvement of animal welfare as it holds the potential to drastically reduce the requirement for manual measurement of blood glucose in diabetes research

Use and validation of DVC® in research on mouse models for vascular risk factors like obesity and stroke

Amanda Kiliaan - Dept. Medical Imaging, Anatomy Donders Institute for Brain, Cognition and Behavior; Chair Preclinical Imaging, Center PRIME Radboud University Medical Center, The Netherlands

Maximilian Wiesmann and Amanda Kiliaan, Dept Medical Imaging, Anatomy Donders Institute for Brain, Cognition, and Behavior, Njimegen, The Netherlands, Chair Preclinical Imaging Center PRIME Radboud University medical center Geert Grooteplein 21N 6525 EZ Nijmegen, The Netherlands. Vascular risk factors hypertension, obesity and stroke are risk factors for cognitive decline and dementia. Brain network disruption due to reduced brain perfusion may play an important role in these neurovascular degenerative processes. In this talk we will present an overview of our research on the temporal relation between impaired brain circulation and network disruption via state-of-the-art ultra-high field imaging in mice models for obesity and stroke and the use of Digitally Ventilated Cages therein. Moreover, we will present whether exercise (running wheels in Digitally Ventilated Cages) will inhibit deterioration of brain function in an obese mouse model. This provides unique information regarding the temporal relation between vascular pathological changes, MRI features and cognitive impairment.

Sleep and circadian rhythm disruption: from mechanisms to behaviour

Stuart Peirson - Sleep and Circadian Neuroscience Institute, Nuffield Department of Clinical Neurosciences, University of Oxford, UK

Life on Earth has evolved under a predictably changing cycle of light and darkness. As a result, virtually all organisms possess a circadian clock, enabling them to anticipate rhythmic changes in the environment. The primary circadian pacemaker is located in the hypothalamic suprachiasmatic nuclei (SCN) and regulates clocks found in cells and tissues throughout the body. This SCN clock is entrained (synchronised) to the external light/dark cycle via light detected by the retina. Work on circadian entrainment has led to the identification of a new photoreceptor system in the eye, comprised of a subset of photosensitive retinal ganglion cells expressing the blue-light sensitive protein melanopsin. As well as regulating circadian rhythms, light also exerts acute effects on physiology and behaviour, including hormone production, sleep/arousal, mood, learning/memory and even pain. Sleep and circadian rhythm disruption (SCRD) is a common comorbidity in many different disorders, in both humans and mouse models. Rather than a change in circadian period or a loss of rhythms, fragmented and weaker rhythms are often observed. In some cases, altered responses to light are also seen. The circadian community has developed many tools measure these changes. In this talk I will describe the regulation of circadian rhythms and sleep, the wider consequences of SCRD, and how disrupted rhythms can be measured in the home cage.

Deciphering the Multifaceted Aging Code: gender-based insights from natural and accelerated aging in outbred mice

Angelo Parini - Institute of Metabolic and Cardiovascular Diseases, Toulouse, France

Background: Aging is a multidimensional process that constitutes the major risk factor for declining health and chronic diseases, with high variability among individuals. This variability in the aging process highlights the need to delve deeper into the intricate mechanisms related to advancing age. This lies in the notion of biological aging, which encompasses the simultaneous decline of multiple organ systems, progressing gradually and persistently. However, consensus on defining biological age remains elusive. Moreover, there is a scarcity of information concerning gender-related aging patterns.
Methods: In this context, we designed a large cross-sectional cohort of outbred Swiss mice (1576 male and female mice) in which spontaneous and voluntary physical activities were monitored from 6 to 24 months of age under either normal or high fat/high sucrose (HFHS) diet-induced accelerated aging. At different ages (6, 12, 18, and 24 months), multiorgan functional phenotyping has been carried out to identify early signs of organ dysfunction. In addition, a large biological fluids/feces/organs biobank has been generated.
Results: DVC® system-based assessment uncovered significant disparities between genders in the aging-related decline of longitudinal spontaneous and voluntary physical activities. These differences were even more pronounced in HFHS-induced accelerated aging. In addition, the analysis of urinary profile in DVC® cages indicated changes in urination linked to aging, with distinct patterns observed in males and females. Furthermore, a comprehensive assessment of evidence-based markers of physiological function identified three populations of mice at all ages: low-, intermediate-, and high-performance mice. More interestingly, without a priori Topological Data Analysis further highlighted clusters of mice closely associated with functional status, emphasizing the biological heterogeneity of the aging process. Conclusion: Overall, these findings emphasize gender-specific aging patterns, highlighting the importance of including both male and female subjects in preclinical studies. Furthermore, they contribute to understand the heterogeneity of biological aging and propose functional criteria to define biological age. These insights provide a foundation for the development of effective preclinical preventive and therapeutic strategies.

Impact of Sleep Disturbances in Adolescence on Depression Risk: insights from mouse studies

Birgitte Rahbek Kornum - Associate Professor, Visiting researcher. Neuronal Signalling, Copenhagen University, Denmark

Sleep disturbances during adolescence have been linked to an increased risk for depression and anxiety in humans. To investigate the mechanisms underlying this relationship, we conducted research using mouse models. This study aimed to understand the consequences of sleep disturbances during adolescence and its impact on depressive-like behaviors in young adult mice. Additionally, we explored how the mice would respond to a second trigger of depression. In this study, c57BL/6 mice were subjected to controlled sleep deprivation for 7 days during peak adolescence, and their behaviors were assessed using tail suspension test, open field behavior and home cage activity monitoring into adulthood. We further examined whether a second trigger could induce depressive-like behaviors in young adult mice. Our findings revealed significant behavioral changes in mice subjected to sleep deprivation during adolescence. Importantly, we observed distinct responses to this manipulation depending on whether the mice were isolated or group-housed. Furthermore, the introduction of a second trigger in young adulthood induced depressive-like behaviors in the mice. This study provides valuable insights into the relationship between sleep disturbances in adolescence and the risk of depression in young adult mice. The altered responses to depressive triggers after sleep deprivation during adolescence suggest that sleep disruptions during this critical developmental period can have long-lasting consequences on mental health. These findings underscore the importance of understanding the mechanisms involved in the link between sleep disturbances and depression and may inform future studies aimed at mitigating this risk in both animal models and humans.

A non-invasive automated home cage digital system detects the variable phenotype of two SOD1G93A mouse models of amyotrophic lateral sclerosis

Caterina Bendotti - Laboratory of Neurobiology and Preclinical Therapeutics, Research Center for ALS, Department of Neuroscience, Mario Negri Institute for Pharmacological Research IRCCS, Milano, Italy

Amyotrophic Lateral Sclerosis (ALS) is a variable disease in terms of onset and progression rate. Similar variability was found in two SOD1^{G93A} mouse models with different genetic backgrounds although their grip-strength impairment did not relate to the extent of atrophy of their hindlimb muscle, especially at early disease stage. To further characterise their distinct neuromuscular phenotype, we used an automated home-cage monitoring system, with or without voluntary running wheel (VRW) activity, to assess whether the daily home-cage and VRW activity highlight the different phenotype and influences disease progression in both models. Female SOD1^{G93A} mice on C57BL6Ola or 129sv strain and non-transgenic (Ntg) littermates, were monitored from the early pre-symptomatic stage of the disease (6 weeks age) until the end stage using a digital ventilated cage (DVC®) system to record their daily activity and the Regularity Disruption Index (RDI), a marker of irregular animal activity. An estimate of the mice's wheel running (distance, time, speed) was recorded daily. The mice were also weighed, and the hind limb extension reflex (HLER) was monitored. DVC® with the VRW allowed us to observe a remarkable difference in activity between the two SOD1G93A mouse strains. C57-SOD1^{G93A} mice showed significantly lower running wheel activity than their Ntg counterpart, even in the early disease stage when HLER was not yet affected. Conversely, 129Sv-SOD1^{G93A} mice showed unexpected running wheel activity compared to their sedentary Ntg littermates, which last for about 8 weeks before decreasing along with the rapid loss of HLER. A similar trend occurred during the light-on period when the mice should be sleeping/resting. While both ALS models exhibited irregular RDI compared to their Ntg littermates, particularly during the light-on period, the effect was more pronounced in 129Sv than in the C57BL/6J models. The peak of irregular RDI was earlier in 129Sv^{G93A} running compared to wheel-locked mice.
In conclusion, these data suggest that physical hyperactivity and sleep disturbances are premorbid signs of an earlier and faster disease in 129Sv-SOD1^{G93A} mice, as reported in some ALS patients.

Coordinating research and building capacity for advanced home-cage monitoring – international networks and institutional core-facilities

Vootele Voikar - Helsinki Institute of Life Science, University of Helsinki, Finland

In 2021, European network of researchers started the COST Action TEATIME (https://www.cost-teatime.org/), with aim at investigating and promoting the potential of advanced technology for automated continuous monitoring of animal behavior in the home-cage environment. The "holistic" view on animal behavior was suggested to contribute beneficially to the amount and quality of research data, but also to improved welfare assessment. Two years of intensive collaborative work has already resulted in several tangible outcomes. In my presentation, I will briefly review the progress of the Action, highlighting the discussion forum (www.TheBehaviourForum.org) and training program (eg webinars, schools, short-term scientific missions). I also reflect on the current perceptions on implementing home-cage monitoring, based on our community survey (conducted in 2022) and personal communications. Finally, I will discuss and provide examples on how home-cage monitoring can be incorporated in the workflow of the behavioral phenotyping core facilities or individual research projects.

Measuring behavior with machine learning

Alexander Mathis - Assistant Professor, EPFL, Switzerland

Quantifying behavior is crucial for many applications across biology. Videography provides excellent methods for the observation and recording of animal behavior in diverse settings, yet extracting particular aspects of a behavior for further analysis can be highly time consuming and computationally challenging. I will discuss the latest developments for DeepLabCut, an efficient method for markerless pose estimation based on transfer learning with deep neural networks that achieves excellent results with minimal training data (Mathis et al., Nature Neuroscience 2018). For multiple animals, I will discuss animal-agnostic assembly and tracking methods as well as the ability to predict an animal's identity from the same backbone to assist tracking and perform Re-Identification (Lauer et al. Nature Methods, 2022 and Zhou* & Stoffl* et al. ICCV 2023). Furthermore, I will discuss state of the art methods for action segmentation to predict what beahvior animals are carrying out from video. I will illustrate the versatility of these tools for multiple species across a broad collection of behaviors from egg-laying flies, via rodents to 3D pose estimation on hunting cheetahs.

Scientific Advancements with DVC® Cages at Novartis

Fabrizio Scorrano - Head Emerging Technologies, Novartis AG, Basel, Switzerland

Novartis has recently integrated the utilization of DVC® in its research facilities located in Basel (CH) and Cambridge (USA), aiming to bolster scientific discovery and digital biomarker collection. This talk offers an insight into the ongoing applications of this system, elaborating on its advantages and identifying areas requiring enhancement. Emphasis will be laid on optimizing the system for a more comprehensive exploration of preclinical digital biomarkers and their effective translation into clinical research contexts.

Home cage measurement of food intake, operant behaviour and spontaneous activity

Joel Eliades - Monash Biomedicine Discovery Institute, Monash University, Clayton, Victoria, Australia

The homeostatic control of feeding is of fundamental importance for the maintenance of energy balance, with perturbations in feeding and motivation contributing to varied human diseases including obesity, type 2 diabetes and feeding disorders such as anorexia nervosa and binge eating. The accurate and undisturbed measure of feeding, feeding behaviours (meal size, bout number), operant behaviours (motivation and reward) and locomotor activity can provide important insight into the underlying causes of metabolic diseases. To this we have paired the FED3 open-source feeding devices with the Tecniplast Digitally Ventilated Cage (DVC) System to analyse spontaneous home-cage activity, feeding and operant behaviours longitudinally. This approach affords unrivalled opportunities to integrate feeding, locomotor activity and entrained behaviours while minimising stress factors that otherwise affect experiments using existing commercial systems.

From Data Collection to Insight Generation in Core facilities: the role of metadata in home cage monitoring technologies

Leonardo Restivo - Department of Fundamental Neurosciences, Faculty of Biology & Medicine, University of Lausanne, Switzerland

Home Cage Monitoring technology (HCMt) rests upon three interconnected themes: Behavior discovery, Animal welfare, and Good research practice. While traditional out-of-cage tests rely on affordances for eliciting specific behaviors, HCMt offers an unprecedented opportunity to delve into unbiased discovery of rodent behaviors occurring in the home cage. HCMt solutions enable earlier detection of potential welfare issues, thereby improving animal welfare and refining the analysis of rodent behavior from the home environment. In addition, developing robust guidelines for HCMt data collection and annotation will enable researchers to combine data from multiple sources, leading to the effective repurposing of data from animal experiments. However, HCMt requires a substantial foundation of data to deliver on the promises to significantly improve the quality and translational power of animal research. Here we present a roadmap for developing metadata that could leverage the vast amount of data collected from HCMt solutions and dispersed among various core facilities. Finally, we highlight a few examples hinting at the full potential unlocked by this metadata approach in data repurposing, behavior finger-printing, and covariate adjustment for animal experiments.

Sensitive detection of early neurological phenotypes in Mecp2 mice using digital cage monitoring

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Rett syndrome (RTT) is a rare neurometabolic disorder caused by mutations in X-linked methyl-CpG-binding protein 2 (Mecp2). Female RTT patients develop normally for 6-18 months, but gradually lose speech and motor skills, as stereotypic hand movements, movement disorders and sleep disturbances develop. Despite heterozygous (Mecp2/+) female mice being clinically relevant, hemizygous male mice (Mecp2/Y) are the preferred model due to their penetrant phenotype The phenotype in mice can be first detected at 4 – 5 weeks of age in males, yet molecular perturbances occur earlier. To determine if a non-subjective measure could be used to detect early neurological phenotypes, Mecp2 and control mice were housed in groups of two in Digital Ventilated Cages (DVCs) from the age of three weeks onward. Strikingly, phenotypes including movement variations over light and dark cycles can be detected very early in life; these phenotypes vary across different genetic backgrounds. Our results suggest that DVCs can be used to distinguish phenotypes in Mecp2 mice at an early age, allowing for determining the effects of a variety of treatments. This work may reduce the number of mice needed for phenotyping, and reduce the number of behavioural tests needed to use Mecp2 mice as a pre-clinical model.

A Novel Murine Model To Study Potassium Channel-Related Autism Spectrum Disorder

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Voltage-gated potassium (Kv) channels play a crucial role in controlling the excitability of neurons and can trigger a cascade of signals within the central nervous system. One specific Kv channel, potassium channel subfamily B member 1 (KCNB1), is associated with integrins (Integrin_K+ channel_Complex or IKC) and is important for converting its electrical properties into signals promoting cell proliferation and migration. Mutations in the KCNB1 gene are linked to developmental and epileptic encephalopathy (DEE). Children affected by this disorder experience severe developmental delays as well as a broad range of behavioral and neurological deficits including social interaction, anxiety, disrupted sleep, hyperactivity, and restricted and repetitive behaviors. To study this neurological condition, we engineered a Knock-In (KI) mouse model harboring the Kcnb1R312H gene variant that has been identified in children affected by DEE. In this study, we observed aberrant neurodevelopment and decreased synaptic connectivity. We assessed the behavior of the Kcnb1R312H mouse, focusing on anxiety, motor functions, activity patterns, and complex tasks, to establish a reliable model for studying ion channel-related ASD. Our results indicate the Kcnb1R312H mouse model offers a valuable tool for gaining a deeper understanding of the role of Kv channels in ASD.

Using the DVC®- LEDDY system to better understand how circadian rhythms affect behavioural outcomes in neuroinflammatory disease

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Neuroinflammatory diseases, impacting millions worldwide, have intricate interactions with circadian rhythms, influencing both disease progression and symptom manifestation. Underpinning this relationship are observations like daily fluctuations in pain intensity and increased susceptibility linked to irregular circadian patterns. In the laboratory, animal models of central nervous system injuries and peripheral nerve injuries offer insights into the mechanistic aspects of such diseases. However, investigating circadian rhythmicity in animals with locomotor challenges necessitates specialized equipment. The Digital Ventilated Cage (DVC®) system, coupled with the LEDDY integration, emerges as a quintessential tool in this regard. Our research aimed to delineate the interplay between circadian rhythms and neuroinflammation in these animal models and gauge the time-dependent variations in functional outcomes such as pain response. Furthermore, we evaluated whether manipulating circadian patterns, using the LEDDY system, could modulate neuroinflammatory responses and related outcomes. Cumulatively, our findings underscore the pivotal role of circadian rhythms in modulating the intricacies of neuroinflammatory diseases. This suggests the promising potential of chronotherapeutics in managing symptoms. Moreover, the combined utility of the DVC® and LEDDY platforms provides a robust avenue for refining our understanding of neuroinflammatory disease outcomes.

DVC® activity as a digital biomarker in an inhalation study

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Cabin air quality has been a at the core of a continuous debate for the past 60 years from a health and safety point of view. To investigate possible health effects of cabin/cockpit air contamination (CAC) events we conducted a nose-only inhalation study and included a battery of neurobehavioural tests. CAC events can be cause by different substances, for example engine-oil and de-icing fluid. This contamination is thought to occur via the bleed air system and different symptoms of crew members have been reported. Still little is known of the relation between symptoms and cause of CAC event. We are the first to comprehensively analyse possible effects of oil-based CAC events in an animal, by investigating behavioural, organ- and blood-based endpoints. Crew members have reported (amongst others) loss of consciousness and fatigue symptoms after an event and thus we used the DVC® system to monitor activity as a digital biomarker throughout the experiment. We saw that cage changes had a higher impact on activity levels compared to restraint in inhalation tubes, and that even the method of cage change (how much litter is changed) matters. We saw that the housing conditions impacts on activity. We believe that using DVC® activity as a digital biomarker has an impact on animal welfare, experimental planning and monitoring as well as reproducibility and translatability. The DVC® home cage monitoring system is a valuable complementation of the phenotyping toolbox.

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