

Animals and Society:

Striking the right balance through Ethology

ISAE 2024

Proceedings of the 57th Congress of the International Society for Applied Ethology

22-26th July 2024, Curitiba, Brazil

Animals and Society:

Striking the right balance through Ethology

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Proceedings of the 57th Congress of the International Society for Applied Ethology 22–26th July 2024, Curitiba, Brazil

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Welcome to the 57th Congress of the ISAE

Bem-vindos a Curitiba!

Ethologists the world over have been called to answer a wide range of questions about the inner lives of animals. As society expands its understanding of human-animal interactions, ethologists play a vital role in ensuring that animals also benefit from those interactions. The research that you do is integral to striking the right balance between what matters to animals and important human priorities.

Ethologists continue to play an integral role in shaping the trajectory of animal welfare science. Past research has dealt with pressing issues regarding farm animal practices and brought us a range of tools to study animals from their point of view. Today, those tools have influenced the work that ethologists do across a number of animal domains, including research involving farm, lab and companion animals, wildlife, captive wild animals, insects and other invertebrates.

At ISAE 2024 we hope to showcase the variety of methods that are applied in the study of animal behavior and seek to engage in conversations that matter to us and animals. We will also explore how ethologists are using novel technologies in their research to better understand animals and the impact humans have on them.

We hope the 57th Congress of the International Conference in Brazil brings you wonderful memories along with new collaborations and friendships. Let's work together to strike the right balance for mutually rewarding human-animal interactions through ethology!

Desejamos a todos um excelente Congresso! Best wishes!

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Brazil

Welcome! We hope you will have many fun and unforgettable experiences during your stay in Brazil! With its unique cuisine, rich culture, some of the most beautiful beaches in the world, breathtaking waterfalls, and adventure for everyone's liking, all you have to do is choose the perfect type of tourism for you and be amazed by the wonders of Brazil.

Curitiba

Curitiba is the capital of Paraná. This state capital was founded on March 29th of 1693. A "model city," this ecological capital of Brazil is currently considered the best capital to live in Brazil. Its public transport system has served as a model for several countries.

Pontifical Catholic University of Paraná (PUCPR)

Educational excellence, innovation-driven entrepreneurship, and multi and inter-cultural learning shape PUCPR's primary purposes. As a result PUCPR has earned national and international recognition from the world's most important higher education ranking: The Times Higher Education, which places PUCPR among the top universities in Brazil. With more than 60 years of dedication to higher education and research, PUCPR is the largest private higher education university in Paraná and one of the most reputable universities in Brazil.

The ISAE 2024 congress will take place on PUCPR's Curitiba campus in partnership with the Graduate Program in Animal Science (PPGCA).

Address: Imaculada Conceição, 1155 - Prado Velho, Curitiba, Parana. CEP: 80215-901

Attractive tourism

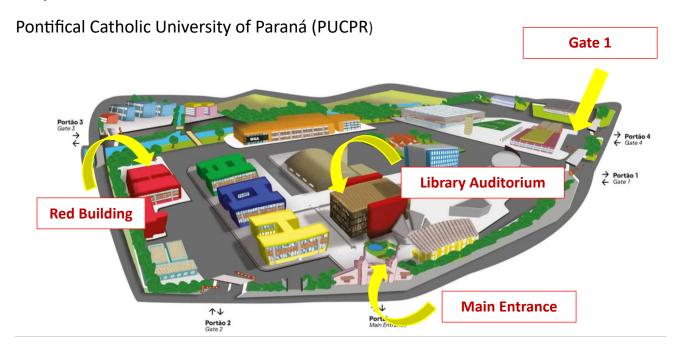
The city of Curitiba has several attractions. Below are a few listed, which we encourage you to visit while here for the Congress!

- Botanical Garden Most famous park located in the city. Once by the portal of entry, one may see extensive gardens in the French style amidst fountains, waterfalls and lakes, and the main greenhouse of 458 square meters, which shelters in its interior, specimens of plants characteristic of tropical regions.
- Tourism Line "Double-Decker Bus" The "Linha Turismo" is a special city tour that visits the main tourist attractions in Curitiba. It is possible to visit the parks, squares and the rest of the city's tourist attractions. Considered one of the best in the country, the Linha Turismo is available every thirty minutes and has a two and a half-hour tour, which travels around forty-four kilometers. The tour starts at Praça Tiradentes, but it is possible to join the tour at any moment. To go on the tour you must buy a voucher pad with five vouchers that give you the right to get on and off bus four times.
- **Municipal Market** Founded on August 2, 1958, the Municipal Market is the main and most traditional shopping destination in Curitiba. At the produce stalls,

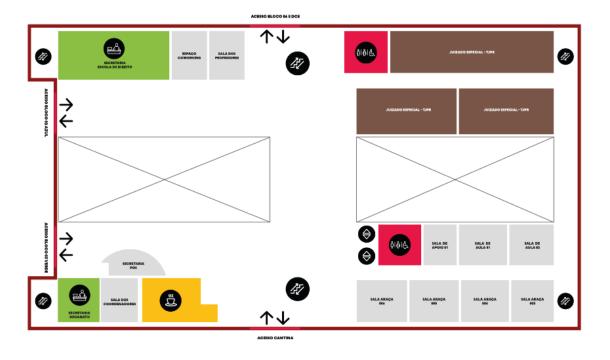
delicatessen shops, and organic section, consumers can find products such as beverages, cheeses and wines from various origins, medicinal herbs, spices and seasonings, delicacies, preserves, fish, sausages, exotic meats, and special cuts of meat. There are also decorative items, household utensils, cleaning products, haberdashery, gifts, and more. One can enjoy pleasant moments at the ethnic restaurants in the food court, which is a meeting point for Curitibanos of various generations, and also participate in events regularly held within the Market.

- Oscar Niemayer Museum This museum, named after a famous Brazilian architect, focuses on the visual arts, architecture and design. For its magnificence, beauty and for the importance of the collection, it represents a cultural institution of international significance. The complex of two buildings, installed in an area of 35 thousand square meters (of which 19 thousand are dedicated to exhibition space), it is a true example of architecture allied with art. The first building was designed by Oscar Niemeyer in 1967, faithful to the style of the time, and conceived as an educational institute, which was opened in 1978.
- Barigui Park The most frequented green area by habitants for sports or leisure activities. It offers a large number of public facilities (including a gym, sports courts, an inclusive playground, and a library) and private ones (such as an Exhibition Pavilion, a restaurant, snack bars, and an amusement park). The park is home to friendly capybaras. It is best not to approach them; observe or photograph from a distance.
- Tanguá Park Considered the best park to see the sunset, Tanguá has two lakes and an artificial tunnel. It also features a dock, a bike path, a running track, a snack bar, and two car parks (in the lower and upper areas). In the upper area is the Poty Lazzarotto Garden. Adjacent, there is a lookout point 65 meters from the lower area lake. The fauna consists of ducks, bats, opossums, armadillos, teal ducks, and other birds, water snakes, and a variety of other animals.
- Panoramic Tower This is in the highest point in Curitiba (1,050 meters above sea level), where you can have a 360-degree view of the city. Opened on December 17th, 1991, it is administered jointly by Brazil Telecom and the Municipal Institute of tourism that promotes sessions, video and provides tourism orientation. It also houses the Telephone Museum.

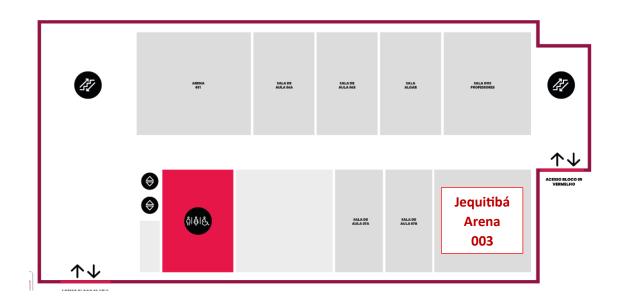
Graphical visualization



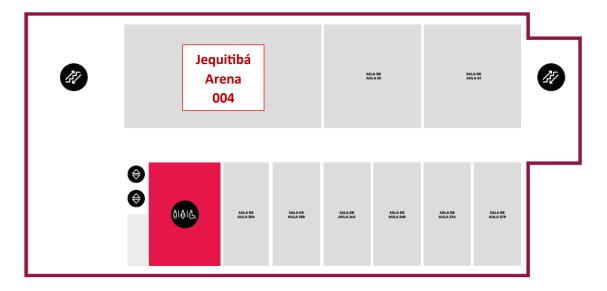
Red Building (Ground Floor)



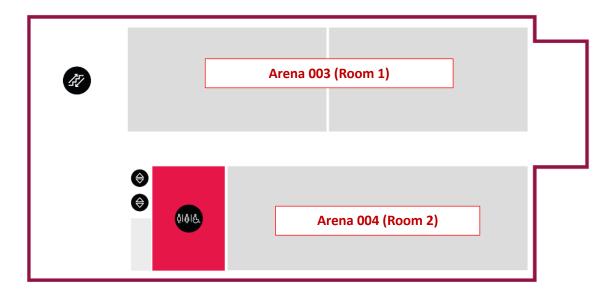
Red Building (Adjacent Building – Ground Floor)



Red Building (Adjacent Building – 2st Floor)



Red Building (Adjacent Building – 3rdFloor)



General Information

Registration and Information Desk

The registration desk is located in the Library Auditorium, which is in the Library Building. Registration will start on Monday (July 22nd) at 16:00 (4pm). The Information desk is located in the Red Building Hall. Staff members, those wearing a specific *Staff* shirt, are ready to respond to your questions.

Name badges

Name badges are required for admittance to the Congress Sessions, coffee breaks, lunches and events. Badges will be issued when you register for the conference.

Poster exhibition area

Posters will be exhibited at the Main Hall of the Red Building during coffee breaks. For program and more information, please visit our website (https://www.isae2024brazil.com/programa%C3%A7%C3%A3o-oficial).

Internet access

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For use of 5G data try electronic SIM cards through the many apps available like Airelo and HolaFly. Curitiba has 5G coverage across all areas of the city.

Parking

PUCPR offers paid parking. The main entrance is through gate 1 ("Portão 1"). You can park anywhere you'd like, but we recommend parking near the Library Building or the Red Building. Please, mind the gate directions:



Moving around

If you prefer a more agile locomotion to come and go to the conference venue, we highly recommend UBER. Curitiba is well served by this modality and cost is relatively low. Using the conference transportation service (bus) requires that you're present at the meeting spots at the set time. Note that to use the bus service you must be always wearing your conference badge.

Social program

- Welcome Cocktail (Meet & Greet): Monday July 22nd 19:00 to 22:00 Imaculada Conceição Street, 1430 Prado Velho, Curitiba Paraná.
 - o This cocktail is included in the Congress program, there are no extras fees.
- National Dinner: Tuesday July 23rd 20:00 Dona Madalena Bar, Chile Street, 2111 Rebouças, Curitiba Paraná. Bus leaves at 19:30 from PUCPR.
- **Gala Dinner:** Thursday July 25th 18:30 to 23:00 Ópera Arte Restaurant, João Gava Street, 920 Abranches, Curitiba Paraná. Bus leaves at 18:10 from PUCPR

Scientific and Social Programme

Time	Monday July 22 nd
09:00-16:45	DCCAF + ISAE Council Meetings
12:15	Lunch (only for DCAAF and Council meeting attendees)
16:00	Start of Registration
17:30	Opening Ceremony
19:00-22:00	Welcome Cocktail (Meet & Greet)

Time	Tuesday July 23 rd
09:00	Wood-Gush Memorial Lecture
10:00	Coffee Break
10:45-12:15	Oral Sessions 1 and 2
12:15	Lunch with Ethologists
13:45-14:45	Plenary
15:00-16:30	Oral Sessions 3 and 4
16:30	Coffee Break with Posters
17:15-18:45	Oral Sessions 5 and 6
20:00	National Dinner

Time	Wednesday July 24 th
09:00	Plenary
10:00	Coffee Break with Posters
10:45-12:15	Oral Sessions 7 and 8
12:15	Lunch
13:45-15:30	Oral Sessions 9 and 10
15:30	Coffee Break with Posters
16:15-18:00	Workshops
18:00	Mini Coffee Break – for AGM attendees only
18:15	AGM - Annual General Meeting

19:00 Free Night	
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Time	Thursday July 25 th
09:00	Plenary
10:00	Coffee Break with Posters
10:45-12:15	Oral Sessions 11 and 12
12:15	Lunch
13:45-14:45	Plenary
15:00-16:15	Oral Sessions 13 and 14
16:15	Coffee Break
16:45-18:00	Ending Ceremony
18:30-23:00	Gala Dinner

Time	Friday July 26 th
10:00-15:00	Farm Visit
12:15	Brunch

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Plenaries

Wood-Gush Memorial Lecture: Tuesday July 23, 09:00-10:00

The role of local communities in the co-production of knowledge for primate

conservation

Patricia Izar¹

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Within the social sciences and humanities, there is an increasing recognition that science is predominantly guided by European and US research agendas. This issue is less frequently discussed in the biological sciences, particularly in behavioral ecology, ecology, and conservation—fields that heavily influence primatology. However, there is a growing concern about diversity, equity, and inclusion in field practices. Primatologists often replicate other biologists by considering high-quality research to stem mainly from "objective" analyses conducted at universities, using data merely extracted from nature at remote field sites. This perspective assumes that scientific knowledge arises solely from academic intellectual efforts, neglecting the historically refined expertise of local individuals who assist researchers. In this contribution, I discuss the essential contributions of local community members employed as field assistants in shaping findings on the behavior and ecology of robust capuchin monkeys at three long-term Brazilian research sites. I highlight how scientists' focus on specific capuchin behaviors was influenced by field assistants' observations and how traditional knowledge informed scientific data interpretation. Thus, the activities of the local community are more accurately described as knowledge co-production rather than field assistance.

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ISAE Creativity Award Lecture: recent highlights of ethological, conceptual and human dimensions Scholarship from UBC

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In this lecture I provide a brief overview of some of our team's research contributions that I consider to be especially creative, along with some very recent contributions that I hope will inspire new work and further insights. My lecture focuses on three different but interrelated scholarly approaches: ethological, conceptual and societal. I begin by focusing on ethological approaches, with a special interest in methods that can used to draw progressively stronger inferences regarding affective states in animals, including work examining acute behavioural responses, effects of pharmacological interventions, and the use of more sophisticated learning and motivational experiments to investigate affective memories and how much it matters to animals to avoid or access certain experiences. I then cover some key examples of what I consider to be especially important conceptual contributions from our group. One key idea is that affective experiences matter not just in the moment, but also as part of a narrative; animals develop expectations about their experiences (on the basis of what they have experienced in the past), and new experiences then further shape these expectations. I argue that this context can have profound implications for animal welfare, and can also inform new approaches to improve welfare, including attempting to shape expectations in a positive manner. This conceptual work also informs the need to develop new methods for assessing expectations. Finally, I review a few examples of our group's work examining the human dimensions of animal welfare that I consider to be a special interest and relevance. If we are to be effective as applied scientists, we must have the buy in and engagement of the end users - often farmers or others responsible for decisions about animal care and use. This deeply collaborative approach with end users informs how we now do our research. Thanks largely to the insights and efforts of our students and post-docs, our more recent work in this area is now more innovative and sophisticated. A key example is in recognizing the limits of binary options often used to frame ethical issues within a narrow, status quo context; in newer work we open the

discussion to include a rejection of both options and allowing participants to reframe issue and discuss system change. I also provide examples of new work illustrating more sophisticated approaches to understanding participants responses, both quantitatively and qualitatively. I end with a discussion on whose perspectives we consider, including new conceptual and experimental work on directly considering the animal's perspective in decisions that concern them, an idea that is at once old and very new within our field.

What can elevated waking inactivity in the home environment tell about animal affective states?

Carole Fureix¹

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Captive/domestic animals are often described as inactive, with the implicit or explicit implication that this high level of inactivity is a welfare problem. Conversely, not being inactive enough can have dramatic adverse consequences for organisms, and may also indicate or cause poor welfare. Can inactivity level therefore be used as trustworthy indicators of affective states (and thus welfare) in animals? I argue they can, if we change the way we often view inactivity in behavioural and welfare studies, and acknowledge it is not a homogeneous category. There are indeed many different forms of inactivity, expressed in different contexts, and each likely associated with very different affective states. Merging these different forms into a single broad category certainly can yield erroneous interpretations with regard to the associated welfare states. In its various forms, inactivity nevertheless can be used as an indicator of affective states in animals, provided that precise descriptions of specific forms of inactivity relevant to hypotheses under test are included in ethograms prior to data collection; that clear descriptions of the contexts that trigger (or conversely decrease) the specific forms of inactivity are provided; and that additional justifications (i.e. beyond the context) are provided before inferring putative affective states associated with the specific forms of inactivity. I will illustrate this rationale by presenting results from investigations testing the hypothesis that greater time spent displaying waking inactivity in the home environment represent a marker of depression-like conditions in some animals. To test this hypothesis, I adopt a translational 'from humans to animals' approach, using depressed people as a model for inactive animals. As such, potential markers of animal depression-like conditions are expected to co-vary with a range of symptoms in animals similar to those described in human depression. Moreover, if waking inactivity reflects depression-like states in animals, its risk and curative factors should be similar to those of the human disease. I will present results in a range of mammal species: horses, mice and dogs, and discuss

further research suggestions to achieve a better understanding of the relationship between elevated waking inactivity and affective states.

Precision Livestock Management to individualize management: the relationship between individual variability and personality traits in dairy cattle for tailored management and improving welfare level

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Animal personality traits, individual behavioral differences across populations that are consistent over time and across contexts, relate to how animals respond to their environments. Personality traits therefore play a crucial role in understanding and managing dairy cattle behavior. Understanding individual variations and the expression patterns of personality traits can assist farmers in making more informed and individualized tailored decisions. Precision Dairy Technologies that monitor behavioral and physiological parameters in dairy cows enhances on-farm decision-making and enables the continuous monitoring of a large number of animals can increase individualized measures that ultimately promote greater individual cattle health and welfare. Several studies in our laboratory across environments, ages, and breeds of dairy cattle found behavioral variables measured by precision technologies were associated with traditional measures of animal personality traits, physiology, and performance. Thus, these behavioral patterns measured automatically reflected personality traits and are associated with feeding behavior patterns and performance in dairy calves and cows. Additionally, we found that personality traits to explain some of the variability in the magnitude of response of animals to stressful events (e.g. weaning, dehorning, and disease events), and social challenges and reproductive performance in dairy calves. These findings have implications for how individual calves navigate each stressor, ultimately influencing individual animal welfare levels. Another one of our studies in dairy cows investigated individual variation in rumination, locomotion, feeding activity, inactivity, and panting time (hours per day) using a comprehensive dataset from thousands of dairy cows equipped with behavior monitoring collars. The study demonstrated that the variability in daily behavioral patterns of dairy cows is highly associated with performance and health events (i.e. services per pregnancy, days open and disease bouts) during lactation. Therefore, the identification of individual variations of behavior patterns can potentially support individually tailored interventions and management for improved animal health and management. We tested this possibility of proposing a tailored intervention, a voluntary heat abatement system, which was found to have great potential and cows using this resource with great variability. Demonstrating the potential to be implemented in commercial settings. Collectively, these studies provide evidence that personality traits are associated with behaviors measured by precision livestock technologies, influencing calf development of solid feed intake, calf and cow performance and health events, and their responses to stressors. These findings underscore the importance of considering the individual in dairy management in order to achieve optimal welfare and performance of dairy cattle.

Laboratory mouse pre-weaning mortality

<u>Gabriela Munhoz Morello</u>¹, Sophie Brajon¹, Sara Capas-Peneda¹, Aurelie Thomas², Sofia Lamas¹, Hannah Wardle-Jones³, Marc Wiltshire³, Jan Hultgren⁴, Colin Gilbert³, I. Anna S. Olsson¹

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Mice are the most used mammalian species for research, but high and variable preweaning mortality hampers breeding efficiency of the most used wildtype strain, C57BL/6J. Considering a conservative estimate of 20% pre-weaning mortality, up to 1.1M mice may die annually just in the EU before becoming available for research, a major welfare and sustainability problem. A common misconception is that stressed females often commit infanticide; based on continuous video recordings, we have demonstrated that infanticide accounts for <15% of mortality. Adults do, however, often eat pup cadavers (cannibalism), leaving no trace of them and confounding estimates of neonatal mortality, which mainly happens within 24h-48h post-partum. Our research demonstrated that pup mortality can be underestimated by over 30% even if pups are counted daily. Housing laboratory mice in trios (2F+1M) exploits natural breeding behaviour of shared parental responsibility mostly between familiar sibling females, but does not improve overall pup survivability in the laboratory. In an experimental study, we found substantially higher mortality in breeding trios when pups were born into cages with existing older litters (litter overlap). A subsequent observational study with >3300 C57BL/6 pups revealed that >60% and >10% of litters from trio- and pair-housed mice, respectively, are born in overlapped litters, which indeed appears to be one of the main risk factors for pup mortality. A study performed with historical breeding records of >219 000 C57BL/6 pups confirmed litter overlap as being a risk to the survival of new-born mice and revealed other equally relevant risk factors, including advanced dam age, and increased number and age of older pups in the cage. More recently, a data-mining approach revealed that the negative social impacts can be, to some extent, compensated by small changes in the cage microenvironment. While housing laboratory mice singly is not an option, due to welfare issues, further investigation is needed to understand the

mechanisms underlying the reasons for pup death in situations of litter overlap and advanced dam age. Our group has started a task-force with partner facilities to perform pup necropsies when pups are found dead and not cannibalized. Early results indicate that most of these pups were either stillborn or died shortly after birth with no milk in their stomachs. Future studies should address litter competition for nutrition (teat access), new-born trauma/crushing by older pups, gestation disturbance, cage air and bedding quality, parental behaviour, and milk production by the dam and mate female.

Workshops

Workshop 1: Wednesday, Month 24th, 16:15 – 18:00

Using virtual farm tours as a method to teach farm animal welfare

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Virtual farm tours provide students with an entry-level way to experience and learn about farm animal welfare. Further, it provides access for students who cannot visit a farm and highlights regional practices. Of 71 students who have experienced the virtual farm tours, 43 recommended that it be used to teach students and 41 could suggest specific improvements to animal welfare. This workshop aims to model ways a virtual farm tour can be implemented in the classroom. The model provides a 360-degree interactive view of a working farm, includes hotspots highlighting text and video about animal health and best practices, and features interviews with farm managers and staff. First, we will present student perspectives on using an interactive farm tour as a way to think about dairy and swine welfare. Then, workshop participants will be asked to become students as they walk through the farm with an activity to guide their experience. This tour works on a computer, smartphone, tablet, and virtual reality goggles. The tours have been used for general introductory animal science education, biosecurity practice for veterinary learning, and welfare evaluation focused on facility design. Virtual reality goggles will be available for attendees interested in viewing the farm tours in the virtual reality format. By the end of the workshop, attendees will have an understanding of how to navigate the virtual farm tours and have a guide for helping students utilize the farm tour. Additionally, attendees will gain knowledge in previous student experience, including student learning and student perception of the value in the experience. Lastly, attendees will leave with access to the established farm tours. This workshop is in accordance with a USDA Higher Education Grant project targeting livestock animal welfare education.

The current status of science-based animal welfare evaluation protocols

Adroaldo José Zanella⁴, George Stilwell¹, Domenico Vecchio², Andreia De Paula Vieira³

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Our goal is to discuss the impact of science-based animal welfare assessment protocols, and to explore opportunities to foster collaborative work. The pioneer EU Funded Welfare Quality® (WQ) project published welfare assessment protocols for cattle, pigs and poultry. The project finished in 2009. After the Welfare Quality® (WQ) project finished it was considered suitable to maintain collaborative contact between former partners to provide relevant knowledge and services to those using the WQ® protocols. Thus, the Welfare Quality Network (WQN) was founded. The EU Funded Animal Welfare Indicators – AWIN project published welfare assessment protocols, for donkeys, goats, horses and turkeys. The AWIN project developed animal-based welfare indicators with emphasis on pain assessment and pain recognition, early development and dissemination. The project finished in 2015. AWIN translated the welfare assessment protocols into interactive apps to make data collection easy, reliable and available immediately as a management tool to help with decisions at farm level, promoting mechanisms for data storage and data-analysis. The project created the Animal Welfare Science Hub (www.animalwelfarehub.org) to disseminate scientific findings, promoting transparency and synergies among stakeholders and interested parties. The Italian Classyfarm System was created by the Directorate General of Animal Health and Veterinary Drugs (DGSA) of the Italian Ministry of Health (MINSAL) for the purpose of classifying farms according to their risk to public veterinary health. ClassyFarm is an IT platform within the national veterinary portal (www.vetinfo.it), which processes a large amount of data from different sources using business intelligence processes. Data are collected on: field surveys of animal welfare and farm biosecurity, antimicrobial use and resistance, herd data, slaughterhouse assessments on animal health (e.g. lung scores) and welfare (e.g. tail lesions in pigs). More recently partners from the AWIN were invited to join the WQN network. The WQN aims to keep or update the existing protocols, to maintain the support instruments, to regulate the training of auditors and to promote the production of further protocols

addressing other production systems or food-producing species. The WQN also aims to attract scientists from other parts of the world to work with them in adapting the protocols to local farming conditions. Currently, the WQN is solely funded from by the members, stakeholders or research funding agencies.

Research Ethics and Diversity in the ISAE

<u>Fernanda M. Tahamtani</u>¹, Katarina Buckova², Bianca Vandresen³, Mostafa Farghal⁴, Ines de Freslon⁵, Beth Ventura⁶, Ellen Williams⁷

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Adhering to ethical guidelines during research is an essential cornerstone of any work. Therefore, the ISAE Ethics Committee invites all interested members and congress participants to this workshop to deliberate on how to best meet our objectives, to maintain a high ethical standard of research across the diversity of contexts in which our members work. In this participatory workshop, attendees will be divided into moderated discussion groups to review case studies of fictional scientific abstracts brought to the ISAE Ethics Committee. These fictional abstracts, written for the purpose of this workshop, exemplify common challenges that require review by the ethics committee, such as lack of prior ethical approval, studies whose design do not follow the Ethical Guidelines, etc. After discussing their assigned abstract, all breakaway groups will regather to discuss the abstracts at plenum. Discussions will centre around the theme: How can we best ensure the welfare of our research animals, while promoting inclusivity of members and research topics in our society? We will be considering some of the various aspects required when designing ethically appropriate research projects. You will also have an opportunity to discuss some of the barriers to ethics which you have personally encountered. Come prepared to share your views and hear from others on these topics, as the ISAE ethics committee seeks to promote improved transparency and democratization of committee policies. The workshop will also have the participation of members of the EDIA committee to ensure inclusivity for all members. The goals of the workshop are (1) to have an open communication about the challenging issues the ISAE ethics committee faces when reviewing abstracts, (2) to get a democratic input from ISAE members on these issues, (3) give advice to researchers and students that are planning their studies, and (4) increase the visibility and accessibility of the ethics committee to ISAE members.

Integrating wild animal welfare science into ethology

Janire Castellano Bueno^{1,2}, Vittoria Elliott^{1,3}, Anne Clay^{1,4}, Grey Fernandez¹

¹ Wild Animal Initiative, ² Newcastle University, ³ Smithsonian Institution, ⁴ George Mason University

Welfare considerations have been progressively integrated into behavioural research and management of farm and companion animals. However, the application of such principles in wild animal research and management remains underexplored. This workshop aims to bridge this gap by offering a dynamic exploration of the intersection between wildlife and welfare through the application of ethological methods. The workshop will start with an interactive discussion session to explore participants' familiarity with wild animal welfare science. Then, we will examine the challenges that behavioural researchers have faced or anticipate facing when considering animal welfare in behavioural research on wild animals, as well as discussing the barriers to implementing animal welfare approaches in the wild. Following this, we will have a 15-minute introduction to the field of wild animal welfare and how its integration can increase reproducibility and repeatability. To delve deeper into the subject, participants will be divided into groups for a 20-minute session to discuss the unique questions they might associate with welfare assessment in wild animals. This group activity serves as a platform for participants to share insights and perspectives, enhancing their understanding of the complexities involved in studying welfare in the wild. The focus of the workshop will be to explore case studies. We will introduce participants to studies currently being conducted in wild animal welfare research and facilitate participant discussion. This part will provide participants with insights into the challenges and opportunities of studying free-ranging animals and the necessary adaptations in methodologies. These include the challenges associated with studying animals in the wild, adapting welfare science methods for freeranging individuals, and the validation of such methodologies for novel species. This approach offers participants practical experience of incorporating welfare considerations into behavioural research, contributing to a more holistic understanding of animal behaviour, and encouraging participants to consider the application of welfare methods and results to the study of wild animal welfare. The workshop will emphasize the significance of welfare in wild animal behaviour research. By addressing critical questions, the main learning outcome for participants will be gaining practical skills and a deeper understanding of wild animal welfare

and behaviour interconnectedness. This workshop serves as a valuable platform for researchers to enhance their methodological approaches, increase the rigor with which they undertake behavioural research, and foster a more ethical and comprehensive understanding of animal behaviour across diverse environments and species.

Session 1: Farm Animals Behavior and Welfare (Cattle)

Tuesday, July 23rd, 10:45 – 11:00

Do dairy cow personality traits influence their response to extended milking intervals and removal of supplemental feed in an automated milking system?

<u>Anna Schwanke</u>¹, Jessica Brasier¹, Gregory Penner², Renee Bergeron¹, Trevor DeVries¹

The motivation to visit an automated milking system (AMS) maybe be more influenced by factors such as udder pressure, for certain cow personality types, than by supplemental feed at the AMS. The study objective was to determine the influence of dairy cow personality traits on motivation to voluntarily visit an AMS in the absence of concentrate feed or under extended milking intervals. Thirty-one primiparous Holstein dairy cows (92.2±18.6 DIM), that had been milked on a free-traffic AMS for at least 14 d, were enrolled. Cow personality was assessed by observing behaviours in response to a novel environment, novel object, and novel human. Cows were exposed to each of 2 treatments in a crossover design (with 6-d periods; no washout), with exposure order balanced for DIM and milk production: 1) minimum milking interval increased from 6 to 9 h (INT), and 2) AMS concentrate allowance decreased from 6 to 0 kg/d DM (CONC). Cows were fetched to the AMS (non-voluntary visit) if milking interval exceeded 10.5 h. Principal components analysis (PCA) of personality assessments revealed 3 factors interpreted as personality traits; social- explorative, activeness, and boldness, which together accounted for 81% of the observed variance. PCA factor scores ranged from -2.3 to +1.8 for social-explorative, -1.8 to 2.3 for active, and -1.2 to 2.1 for bold. Data were analyzed using mixed-effect linear regression models. On INT, as compared to CONC, cows tended to make more total AMS visits (7.1 vs 5.0 visits/d; SE=0.81; P=0.06), more voluntary AMS visits (7.0 vs 4.8 visits/d; SE=0.84; P=0.07), and had more AMS rejections (5.0 vs 2.5 rejections/d; SE=0.73; P=0.02). There was no interaction between personality and treatment. Overall, more social- explorative cows tended to make less AMS visits (1.14 less visits/d per 1-unit increase in social-explorative score; P=0.07), but had greater milk yield (2.08 more kg/d per 1-unit increase in social-explorative score; P<0.01) compared to less social-explorative cows. More active cows made less AMS visits (1.95 less visits/d per 1-unit increase in activeness score; P<0.01) and tended to have lesser milk yield (1.2 less kg/d per every 1-unit increase in activeness score; P=0.09) compared to less active cows. There were no detected associations

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between boldness and milking behaviour or production. These data demonstrate that cow personality influenced overall behaviour and production in an AMS, while cow personality did not affect their response to extended milking intervals or removal of supplemental feed.

Investigating cross-suckling behaviour of dairy calves within a cow-driven cowcalf contact system with automatic milking

Claire S. Wegner¹, Hanna Eriksson¹, Cady W. Chan¹, Sigrid Agenäs¹, Lars Rönnegård²

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Dairy management systems where calves are reared by their dams - referred to as cow-calf contact (CCC) systems - are currently increasing as criticism of early cow-calf separation continues to grow. A prolonged suckling period is suggested to be beneficial, yet little is known about the suckling behaviours – including cross-suckling – of calves within indoor CCC systems. The aim of this research is to explore potential factors associated with cross-suckling in dairy calves reared with CCC. Dam-calf pairs (Swedish Holstein: n = 7; Swedish Red: n = 12) were housed together in a free stall barn with automatic milking and a CCC area, wherein cows and calves could access shared lying stalls and unrestricted physical contact with one another. Cows were directed to the CCC area after milking and could decide when to leave, making the CCC system cow-driven. Suckling events were registered using continuous sampling of video recordings on three 24-h periods, corresponding to average (SD) calf ages of 3 (1.8), 9 (1.8) and 15(1.8) weeks. Successful suckling bouts (calf touches udder ≥1 min and is visibly sucking) were classified as suckling on dam (0) or cross-suckling (1) and analysed using logistic regression. Model predictors included calf age, calf sex, calf birth weight (kg), and presence of other calves suckling on the focal cow at the start of event (1/0). In total, 234 suckling bouts were recorded, of which 85 were cross-suckling events. Preliminary analyses suggest that the odds of cross-suckling increased with increasing calf age (P=0.01; odds at 3 weeks: 0.05, 15 weeks: 0.23) and when other calves were already suckling (P<0.001). Female calves had 3.6 times greater odds of cross-suckling than males (P=0.06), but birth weight had no effect (P=0.61). Calves suckled other cows even when the dam was present, and this increased as the calves got older (18% of all cross-suckling events at 3 weeks, 36% at 15 weeks). We observed great individual variation between calves, with cross-suckling events constituting 10-71% of all suckling events per calf. The high frequency of cross-suckling observed in our study is – to some extent – attributable to calf-related factors, but may also have been influenced by the fact that CCC in this system was cow-driven. Further work is required to

evaluate if cross-suckling is as common in calf-driven CCC systems, where the calves have larger control in initiating contact with their dams.

Personality traits are associated with dairy calves' magnitude responses to routine stressors

Megan M. Woodrum Setser¹, Heather W. Neave², Joao H.C. Costa³

Personality traits—dimensions of behavioral traits that make up the whole animal personality—relate to how animals respond to their environment including stressors. Dairy calves routinely face stressors in early life including diarrhea, disbudding, and weaning. We aimed to determine if personality traits of dairy calves were associated with the magnitude of behavioral responses to these stressors. At 23±3d of age, Holstein dairy calves (n=49) were subjected to a series of standardized personality tests that exposed the calf to novelty and fear stimuli. A principal component analysis performed on behaviors measured from these tests revealed three factors interpreted as personality traits: 'fearful', 'active', and 'explorative'. Calves were scored daily for fecal consistency and signs of respiratory disease daily and behavior in the home environment was recorded from 4-81d of age with two precision livestock technologies (PLF): automatic feeding systems and accelerometers. Subsets of these calves were utilized for the diarrhea (n=18), dehorning (n=19), and weaning (n=30) stressor periods. A stressor period refers to the days immediately before and following the day the stressor was initiated (Day0: diagnosed with diarrhea, dehorned, no milk provided). To be enrolled calves were required to have complete PLF data on DayO, not be treated for other disease during the stressor period, and not have overlap between stressor events. For the days surrounding Day0, magnitude change in behaviors from PLF were calculated relative to the behavior performed on DayO. Regression models identified if calf scores for each factor were associated with magnitude change in behavior for each stressor period; day relative to stressor was included as a repeated measure. All personality traits were associated with the magnitude and direction of behavior change for each of these stressors, but differed by stressor type. For instance, calves that were more 'active' had a greater decrease in lying time (P=0.02) with the onset of diarrhea compared to less 'active' calves, who increased in lying time. Calves that were more 'explorative' had a greater increase in lying time (P=0.007) than less 'explorative' calves following disbudding. Calves that were more 'fearful' had a smaller magnitude increase in starter intake leading up to being weaned (P=0.02), but had a greater

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magnitude increase in starter intake following weaning (P=0.01), compared to less 'fearful' calves. Our results suggest personality traits can explain individual variation in change in and strength of behavioral responses to stressors, which has implications for individual welfare of dairy calves.

Rearing calves with full-time, part-time or no dam contact influences longerterm development of exploratory, agonistic and human-interested personality traits

<u>Heather W. Neave</u>^{1,2}, Emma Hvidtfeldt Jensen ¹, Margit Bak Jensen ¹

Dairy cattle are known to have individual personalities that are consistent over time, but little is known about how the early-life rearing environment of calves may affect the development of personality traits into later life. To this end, calves were reared in groups of 4 with either full-time (24 h/d) or part-time (10 h/d) dam contact, or were permanently separated from the dam after birth. After 10 wk, all calves were mixed and managed similarly, without dams, until personality testing at 19.3±0.7 mo of age (n= 11, 8 and 9 female calves in full-time, part-time, and separated dam-contact treatments, respectively). Each animal was individually tested in 5 consecutive tests (total 19 min): novel environment (new pen), novel object (traffic cone), novel human (standing stationary in high-visibility clothing), unfamiliar cow behind a barrier, followed by free access to the same unfamiliar cow. Animals were assigned to one of three unfamiliar, lactating cows. Each novelty test was 3 min, and each unfamiliar cow test was 5 min; the test animal was removed briefly from the test pen in between tests. A principal components analysis with varimax rotation identified 5 factors, interpreted as personality traits, that explained 79% of the total variation (TV) in behavioral responses to the tests. The effect of dam-contact treatment on each trait score was tested in a linear mixed model, with covariates (age at testing; dam parity; unfamiliar cow ID) and random effects (block; animal ID). Separated calves scored higher on 'Human-interested' trait (26% TV; quick to approach and more time spent close to and touching human) than full-time (P=0.03) and part-time calves (P=0.01). Full-time calves scored higher on 'Agonistic' trait (18% TV; more bouts of fighting and pushing unfamiliar cow), and lower on the 'Exploratory/Inspective' trait (8% TV; less time exploring novel environment; less time inspecting anogenital area of unfamiliar cow) than part-time calves (P<0.03). Dam-contact treatment did not affect 'Curious' (17% TV; quick to approach object; more time spent close to and touching object) or 'Social' (10% TV; more time spent head rubbing and touching unfamiliar cow) trait scores. These results suggest that

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the rearing environment of dairy calves, such as duration of dam-contact in early life, can influence the development of personality traits measured prior to first calving; this may have implications for how adult cows interact with farm staff and cope with introduction to new social partners or environments.

Transitioning dairy cattle from traditional parlour milking to automatic milking systems

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Technology can reduce the workload of farmers but may also affect animal welfare positively or negatively. Automatic milking systems (AMS) can benefit farmers by increasing milk yield and may also benefit cows by allowing for increased cow agency concerning when and how often they are milked. However, implementing AMS requires that the animals learn how to use an AMS unit, a transition that can induce stress responses that impacts milk production and participation in milking. Some farmers attempt to train cattle, but this training varies and there is little published research to help inform training practices. The objective of this study was to assess the effects of one training method on how often cows entered the AMS units and milk production when lactating cows transitioned from parlor milking to an AMS. We observed 45 lactating cows of varying age (55 ± 22.9 mo) before and after this transition. Half of the cows were randomly assigned to a training treatment. These animals were habituated to the AMS units during the two weeks before the switch. Every day these cows individually moved to a waiting area in front of the AMS, and for 5 min a trainer used luring techniques with grain to gradually encourage the cow to approach and then to fully enter the unit. By the end of the training period 14 of 22 treatment cows were entering on their own without trainer luring. Beginning 3 days before the switch from parlour to AMS milking, all cows (i.e. those trained and control cows) were manually moved into the AMS unit once a day by the farm staff. We found no effect of training on milk production (30.99 ± 1.53 L/day; F=0.06, P>0.05) or milking interval (8.53 \pm 0.27 h; F 0.85 = P>0.05) over the first 3 days after transitioning to the AMS. We conclude that the specific training method we applied was not effective for aiding this transition. Further work is required to assess the effects of other training methods and outcome measures.

Session 2: Wildlife Behaviour, Management and Welfare

Tuesday, July 23rd, 10:45 – 11:00

Application of the Ackonc-AWA welfare assessment protocol in ex-situ wildlife conservation: Identification of principal components

<u>Débora S. Racciatti</u>¹, Laura A. Rial¹, Alejandra Feld¹, Carlos Blanco¹

Ensuring individual and species welfare are critical obligations of ex-situ conservation. Achieving this goal necessitates the implementation of comprehensive and robust assessment tools. Our study aimed to explore three categories of animal welfare indicators (health, behavioural, and environmental) for ten individuals residing in an Argentinean zoo. The assessment was conducted using the Ackonc-AWA protocol. The studied group comprised two Loxodonta africana, one Elephas maximus, three Pan troglodytes, three Giraffa camelopardalis, and one Pongo spp. Total direct observation time was 71 hours. Statistical analyses were conducted using Infostat software. Principal components (PC) analysis was employed to reduce the dimensionality of our dataset and identify underlying patterns. Dendrograms were constructed to visualize hierarchical clustering among our variables. The significance level was 5% in all analyses. The first two dimensions of the PC analysis explained 77% of the variation (PC1 53%; PC2 24%). It revealed that agonistic behaviours, use of environmental enrichment (EE), and provision of environmental choice and control opportunities (ECCO) significantly contributed to the construction of CP1, while factors such as food presentation, water availability, stool score, and conditions of claws and teeth (CT) showed contrasting effects. Additionally, CP2 was primarily influenced by stereotypic behaviours, with use of space and group composition exerting opposing influences on its formation. Variables positively correlated were agonistic behaviours and EE; coat and CT; food presentation and stool score; water availability and CT; group composition and shelter availability. Those negatively correlated were stool score and EE; food presentation and EE; ECCO and food presentation, ECCO and water availability. The cluster analysis, represented as a dendrogram, revealed the presence of three distinct groups. One group comprised individuals of Pan troglodytes and Pongo spp., another consisted of individuals of Elephas maximus and Loxodonta africana, along with two Giraffa camelopardalis specimens. A third

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group comprised a single individual of Giraffa camelopardalis. Through the implementation of the Ackonc-AWA protocol encompassing health, behavioural, and environmental indicators, we gained valuable insights into the welfare status of ten individuals residing in an Argentinean zoo. Our analysis identified crucial factors influencing animal welfare, including agonistic behaviours, use of EE, and the provision of ECCO. These results underscore the necessity for adopting holistic approaches to welfare assessment and management within captive environments. Furthermore, our cluster analysis unveiled distinct groupings among the studied species, which highlights the importance of tailoring care strategies to the specific needs of different taxa and individuals.

Assessing behavioral and welfare outcomes of environmental enrichment in head-starting loggerhead sea turtles (*Caretta carreta*)

Raúl David Guevara^{2,3}, Maëlina Gomez¹, Clara Agusti^{2,3}, Jose Luis Crespo¹, Xavier Manteca3, Oriol Talló³

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Head-starting strategies in captivity aim to enhance the survival of endangered species during early development before releasing them back into the wild once they reach a sufficient size. However, concerns exist about whether these animals can acquire the necessary behavioral skills for survival and reproduction in their natural habitat. Strategic environmental enrichment programs, which trigger positive emotional responses and cognitive functions, have proven effective in developing crucial behavioral skills in other species. While existing studies on sea turtles in captivity often focus on optimal feeding or treatments, limited information is available regarding enrichment effects, and current head-starting guidelines do not include environmental enrichment strategies. This study aimed to evaluate the impact of environmental enrichment on the behavior and welfare of head-starting loggerhead sea turtles (Caretta caretta). The study was carried out over 6 weeks in the summer of 2020 at the Oceanogràfic of València's rehabilitation center. At the age of 43 weeks, twenty head-starting sea turtles were randomly sorted either into a control group (CON) or an enriched group (ENR). CON was housed in a circular empty tank for baseline conditions, while the ENR benefited from permanent environmental enrichments, including artificial algae and floating PVC squares. Additionally, ENR received two daily hours of extra nutritional enrichments, which varied and comprised live invertebrates, jelly, ice, and food hidden within items. Moreover, they were provided with daily supplemental environmental stimuli encompassing both floating and submersible items, and dynamic water stimulation like currents and bubbles. Behavioral observations, totaling 58 hours of recordings per individual, focused on general movements, interactions with enrichment items, and social behaviors. The findings showed that the most common behavior in both groups was "Boundary Interaction", defined as actively swimming against the tank's vertical boundary, a stress indicator due to captivity. ENR displayed less of this behavior than CON (72.42 $\% \pm 3.05$ vs. 80.69 $\% \pm 2.27$, respectively;

P<0.05), and exhibited significantly increased Active swim relative to CON ($11.60\% \pm 1.61$ vs. $6.31\% \pm 0.60$, respectively; P<0.05). Besides, ENR engaged with enrichment devices, displaying behaviors such as nosing objects, picking at rings, or biting (1.70, 0.31, 0.007 events per hour, respectively), and showed refuge-seeking behaviors (Hiding 0.25%, Resting inside floating items 0.34% mean percentage of observations with enrichment devices); opportunities not provided to CON. These results suggest an improvement in ENR welfare and the potential for an environmental enrichment plan in head-starting programs.

Why did the capybara cross the road? A case study of the Fernando Costa Campus of the University of São Paulo

André Luiz Gama Nogueira¹, Germana Vizzotto Osowski¹, Adroaldo José Zanella¹

Capybaras (Hydrochoerus hydrochaeris), the world's largest rodents, face a significant threat in Brazil due to their frequent presence along roadsides, resulting in a high incidence of vehicular accidents and making it the wild mammal species most frequently involved in collisions in São Paulo State, and being on the top of the list of most runover species on the country. This research aimed to uncover the factors contributing to the attraction of capybaras to roadsides. The study site selected was a road and its immediate surrounding, within the Campus Fernando Costa, at the University of São Paulo, in Pirassununga, known for hosting an approximately stable population of about 40 capybaras, roaming free, continuously monitored by a video camera system. The analysis of video recordings employed a scan sampling method at 10-minute intervals, systematically documenting the number of individuals involved in various behaviours. The ethogram categorized behaviours into three main groups: Social (encompassing agonistic, affiliative, and nursing behaviours), Feeding (including grass eating and ecotone eating), and Movement (encompassing walking on the road, grass, or ecotone, as well as lying down or sitting). The investigation spanned trough 30 days during autumn and 30 days in winter seasons. Statistical comparisons between seasons utilized the paired Student's t-test, with data processed using Excel and Python v.3.10.2. Results revealed a higher frequency of eating behaviour during winter compared to autumn (p = 0.001). Furthermore, capybaras were observed resting more frequently on the road in winter months than on the grass or ecotone (p < 0.001). Overall, a statistically significant preference for staying on the grass was evident, with grass-eating behaviour observed 95.5% of the time when capybaras were on that area. Although no statistical differences were found in capybara presence on different days of the week (p = 0.080), the afternoon emerged as the most active period for the animals (p < 0.001). Addressing the primary question, capybaras were observed crossing the road primarily to access and consume Zoysia japonica grass planted in one of the roadsides. As a recommended solution for managing the capybara presence in this specific area, the removal of the grass emerged as a viable strategy. The

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observed attraction of capybaras to roadsides, akin to their behaviour on highways in Brazil, may be linked to the cultivation of Zoysia japonica as an ornamental species by highway concessionaires.

Towards assessing boredom-like state in farmed spotted pacas Cuniculus paca. A preliminary study

Lucas Rossini², Carole Fureix¹, Sergio L. G. Nogueira-Filho², Selene Nogueira²

Pacas (Cuniculus paca) are large neotropical rodents farmed in South and Central America for their meat. Paca farming involves long-term confinement in an extremely barren and monotonous environment that strongly restrict behavioural opportunities, and can induce boredom-like states in captive/domestic animals. We ultimately aim to investigate boredom and its dysfunctional correlates in farmed pacas, focusing (as per existing literature on other species) on evidencing enhanced motivation for general stimulation in animals exposed to barren conditions. This preliminary study aimed to test the validity of a first set of stimuli; a priori categorised as positive, neutral, and aversive; to be used to assess motivation for general stimulation later. We predicted that pacas will attend quicker a priori rewarding than a priori aversive stimuli, with intermediate results for a priori neutral stimuli. We also explored whether greater daytime activity (when pacas are usually inactive) reflect enhanced motivation for general stimulation, predicting shorter latencies to approach the stimuli in the most active pacas. Subjects were 12 males and 4 four females (37.94±16.11 months-old), housed in severely barren pens of either single-sex male or mix-sex groups of 2-4 pacas. To measure motivation for general stimulation, pacas were exposed to three novel stimuli (testing one stimulus daily over three consecutive days). Stimuli were categorised as positive (spontaneously consumed fresh melon), neutral (metal bar lacking clear biological relevance to pacas), or aversive (broom, inducing fear responses during pen cleaning). Daytime activity (pooling all behaviours displayed outside of the shelter) was determined from 202min of video per paca recorded over three sessions. Data were analysed with GLMM. As predicted, latency to approach broom was significantly higher than melon (Chi2LRT=1.625, p=0.202, z=-6.391, p<0.001) and bar (z=-4.106, p<0.001) latencies. Latency to approach the bar also tended to be higher than for melon (z=2.275, p=0.06). Results therefore support the validity of our stimuli categorization post hoc. Latencies to approach melon and broom visually show little

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interindividual variation; daytime activity was thus compared against the latency to approach bar only, with no significant relationship between the two measures (Chi2LRT=1.625, p=0.202). Visual inspection of data, and a significant non-parametric negative correlation (rs=0.645, p2-tailed=0.007), nevertheless cautiously suggest that daytime activity may link with motivation for stimulation, supporting further testing of the hypothesis. We discuss limitations of the study and future research directions, e.g. increasing and diversifying test stimuli, informing power analyses for future studies, controlling for potential confounds, and use of enriched housing.

Managing animal welfare change: two years of the Primate Welfare Assessment Tool

Carly I. O'Malley¹, Emilie A. Paterson¹, Dawn M. Abney¹, William J. Archibald¹, Patricia V. Turner¹

Within global organizations, harmonization of animal management practices across facilities can be challenging due to different cultures, resource availability, and regional oversight. In 2022, the Primate Welfare Assessment Tool (PWAT) was implemented across 13 sites at Charles River, spanning 4 countries. The objective of the tool was to assess primate welfare at the site and company-level, outline quantitative measures of welfare, create common criteria and language for discussing welfare, and refine primate management through a culture of continuous improvement. The aim of this work was to assess the effectiveness of the tool in the first 2 years of implementation through data analysis and a user-experience survey. The assessment occurred every 6 months in 2022 and 2023. The tool assesses 6 categories: physical, behavioral, environmental, training, procedural, culture of care (employee satisfaction and training). To manage welfare change, year-end site-level reports were provided with data trends for each category and recommendations for improvement. Personnel reviewed their results and outlined 1-3 goals. The overall scores ranged from 63-90% in 2022, and from 64-90% in 2023. Between 2022 and 2023, 7 sites increased their scores, 2 sites remained the same, and 3 sites had a decrease in score. Of the 8 sites that set goals for 2023, 7 sites increased their score in the relevant categories. At the end of 2023, a userexperience survey was sent out. Tool users (n=15) rated the tool a 3.5/5 for overall satisfaction and 73% agreed that the PWAT was valuable for assessing their programs. The culture of care assessment was considered the most valuable part of the PWAT by 73% (11/15) of respondents, followed by global discussions on data trends at primate behavioral management calls (47%, 7/15). The key element of improving their programs included sitelevel discussions (82%, 9/11) and receiving management support (45%, 5/11). For sites that had a decrease in score, the factors that contributed to the decline were workload (60%, 6/10), staffing issues (50%, 5/10), and training personnel (50%, 5/10). Additional feedback from open ended responses indicate that the reports help sites prioritize projects (6 responses) but that some clarifications are needed to improve the user experience. After two

¹ Charles River

years of using the PWAT, positive changes in primate management programs are being observed at the site-level and global challenges are being addressed. Due to the feedback and results of the PWAT, additional species-specific welfare assessment tools are being developed.

Session 3: Farm Animals Behaviour and Welfare (Pigs)

Tuesday, July 23rd, 15:00 – 15:15

Response of pigs to different types and numbers of environmental enrichment: behavioural and growth performance

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Rearing pigs in confinements has altered their behavioural repertoire, leading to agonistic behaviours due to lack of stimulus to perform inherent activities. These can be curtailed by the provision of enrichment devices that provide stimuli essential for pigs to attain physiological and psychological well-being. Hence, this study was conducted to determine the effect of different numbers of soft wood-plank and polyester rope on the performance and behaviour of weaned pigs. Sixty-three (Largewhite X Landrace) weaned pigs were randomly allotted into seven treatments (3pigs/replicates) for eight weeks. Two types of objects were presented; polyester rope (PR; 12mm, blue, knotted at intervals) and soft wood-plank (SW; 40 x 4.0 x 4.5cm suspended on each side by blue 6mm PR). These objects were hung. The treatments were: T1 (without enrichment), T2 (one PR), T3 (two PRs), T4 (three PRs), T5 (one SW), T6 (two SWs), and T7 (three SWs) arranged in a 2 x 3+1 augmented factorial design. Behaviour was monitored using video cameras, Pen-Mate Manipulation (PMM), Pen Component Manipulation (PCM), and Enrichment Use (EU) were extracted in one minute of every 10 minutes for 6 hours. Growth performance data: feed intake (FI), weight gain (WG), final weight were measured, while Feed Conversion Ratio (FCR) was calculated. Data were analysed using ANOVA. The highest PMM (30.01%) and PCM (40.28%) were observed among pigs in T1, while the least PMM (12.02%) and PCM (14.05%) were in T7. However, highest EU (48.98%) was in T7 and the least (12.12%) in T2. The effect of object type (PR and SW) indicated that PMM was significantly higher (P<0.05) in T1 (30.01%) than PR (21.74%) and SW (16.89%). PCM followed the same trend. On the contrary, the EU for SW (35.05%) was significantly (P<0.05) higher compared with PR (21.87%). The effect of numbers of objects (1,2,3) showed that as the number of enrichment objects increased, the PMM and PCM decreased with significant variation (P<0.05) across the treatments, while EU increased as the number of enrichment devices increased. Highest average WG (10.39Kg) and FI (32.58kg)

were observed with pigs in T6, while the least WG (8.86Kg) and FI (29.16Kg) is with pigs in T2. Conversely, there was no significant difference in FCR. In conclusion, the use of SW and PR in this study reduces PMM and PCM. Likewise, as EU increased, appropriate stimuli that diverted the attention of pigs from agonistic behaviour were elicited, and SW was preferred.

Social dynamics and aggression in growing pigs inferred from automatic feeder data

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Aggression impacts pig welfare in modern production systems, causing low productivity due to skin lesions, increased risk of lameness, lower immunity and feed efficiency. Intensively housed pigs endure limited space, barren environments, and impossibility to perform intrinsically motivated behaviours. These conditions, along with a limited number of feeding stations, are likely to trigger competitive aggression surrounding the feeder. The objectives of this project are to assess the performance of an aggression-detection algorithm, using data collected from automatic feeders, and to use its results to characterize social dynamics in growing pigs. We used data coming from 649 groups from five farms in pens equipped with single automatic feeder stations, totalling 7772 pigs, in groups consisting of either Landrace, Duroc or Yorkshire, and either all males or all females. Each feeder records the time and date of visits, as well as duration and feed consumption. We defined feeder-detected aggressive displacements (FDAD) based on the temporal proximity between one animal exiting and the next one entering the feeder, assuming that very short time gaps between visits (< 2 seconds) reflect likely aggressive displacements. A subset of five groups in one of the five farms was also used to validate the aggression detection algorithm, by comparing FDADs detected by the algorithm with behaviour observed on video at the same timestamp, to check for aggression (e.g. pushes, bites, mounts). We observed that 96% of FDADs (n = 63) were aggressive in nature. We constructed daily social networks for each pen from FDAD events for the 50-88 days of available data during the growing period. Network density (proportion of individuals interacting on a given day) decreased over time in 97.81 % of the groups (even after correcting for the total daily visits to the feeder), reflecting that over time, certain displacer-displaced dyads no longer occur. This may be due to individuals learning their resource-holding potential during contests at the feeder, as well as that of other individual pigs they may compete with. By understanding the progression of aggression over time, we can gain insights into pig behaviour and inform strategies for improving farm management

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practices. The success of the algorithm allows constructing models that correlate group-level social properties with individual-level performance parameters. This capability could aid phenotyping of social behaviour, leading to selection strategies to improve sociability. Such advancements hold promise for enhancing pig welfare and bolstering the sustainability and efficiency of commercial pig farming operations.

Social network analysis of pregnant gilts: unraveling dynamics of social hierarchy during feeding and its impact on offspring

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Social relationships are part of the behavioral biology of pigs and can be affected by the type of animal housing. Resources available during development can affect adult pigs and their offspring. This research aimed to investigate social relations among pregnant gilts during feeding in an outdoor group-housing system and its effects on their offspring. Thus, 15 gilts were estrus synchronized and artificially inseminated with three different semen pools. The gilts were housed in a paddock with a mud pool, natural tree shade, and two nipple drinkers. Two daily meals (~2.5kg/day/gilt) were provided to the group on the ground. Their feeding behavior was assessed for three continuous days, 10 minutes in the morning and 10 minutes in the afternoon, for every third of gestation (42 hours of video). Saliva samples were collected at 6:00 a.m. and 6:00 p.m. on these same days. A trained observer evaluated all the videos using the Boris software to determine agonistic and affiliative behaviors. After farrowing, piglet data collected included sex, mortality, and weight. Elo scores were calculated to quantify the relative hierarchy levels among the gilts based on the behavior assessment. Generalized linear mixed models were used for computing the data and the significance of fixed effects was determined at p < 0.05. The higher hierarchical status of gilts was related with heavier piglets. The behavior of feeding together decreases as pregnancy progresses, but with little change in hierarchical stability. Further investigations are recommended to evaluate maternal modulation on offspring performance in pigs. The studied system allowed the observation of gilts' social feeding behaviors, evidencing the necessity for considering pigs' behavior for

genetic selection and the improvement of commercial facilities and management practices to enhance their welfare.

The impact of odourised straw on welfare of growing pigs

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The importance for pigs to explore their surroundings is widely acknowledged. In commercial housing, lack of an outlet for the motivation to explore, such as no suitable enrichment material, may result in redirection of exploration towards pen mates leading to e.g. tail biting. While studies have shown that straw provision can reduce such damaging behaviour, practical constraints limit its effectiveness, prompting a need for alternative solutions. The aim of this study was to investigate if adding odour to straw would improve the enriching qualities and thereby the welfare of growing pigs in commercial housing. The study was conducted at a commercial Swedish pig farm over 12 months. We included 1600 growing pigs housed in pens of approx. 10 littermate pigs each. Of the 160 pig groups, 80 were control groups with no treatments added (normal straw on floor), and the other 80 groups were assigned one of the four treatments: 1) Odourised straw in rack with odour changing every week, 2) Odourised straw in rack with odour changing every week-day, 3) Non-odourised straw in rack, 4) Nonodourised straw on floor (empty rack). The odours lavender, aniseed, pine, thyme and ginger were sprayed onto the straw. Welfare of the pigs was scored once a week by four trained observers. Welfare scores were recorded per individual at pen level for ear and tail damage, snout irritation and body cleanliness (scores ranging from 0=no damage/irritation/dirt to 4/3/2=high damage/irritation/dirt). The average scores per pen per week were analysed using mixed-effects models with a negative binomial distribution (due to zero-inflated data) including treatment, week and their interaction as fixed effects, with batch and pen nested within pig house as random effects. Preliminary analysis show a significant effect of week in the experiment with all welfare scores significantly increasing over the experiment (all Pvalues < 0.001). The treatments did not significantly affect the tail, ear, or body welfare scores

(P-values > 0.05) but did affect snout irritation. Posthoc pairwise comparisons show that pigs having a new odour every weekday had significantly lower snout irritation than control pigs (P-value < 0.05). These results suggest that adding odours onto existing enrichment materials such as straw did not impact tail and ear damage or body cleanliness. Further investigation of pigs' behaviour when provided odourised straw, such as play behaviour and straw manipulation will be conducted on the video material from the same experiment to elucidate the full impact of odourised enrichment.

Does sow enrichment type influence farrowing duration and piglet behaviour?

Melissa Cupido^{1, 2}, Siobhan Mullan², Laura Boyle¹, Keelin O'Driscoll¹

Traditional farrowing crates (FC) pose concerns for the welfare of sows and their offspring. FC offer some protection to piglets from being crushed by the sow while lying down but they restrict the sow's movement, are barren and uncomfortable, prevent the sow and piglets from expressing natural behaviours such as nesting, rooting and exploring, and limit social interactions. This study investigated the influence of three manipulable loose materials; Straw (n = 13), Haylage (n = 14), and Hay (n = 14), and a control (Rope; n = 14) on the duration of farrowing, and piglet behaviour in FC. There was also one rubber chew toy for piglets in each pen. Fifty-five healthy sows, balanced by weight, backfat and parity, were assigned to treatment one day prior to entering the FC. In Rope pens, a hessian rope was hung on the side of the FC near the sow's head. For the other treatments, loose material was provided via a metal box attached to the side of the trough. The farrowing duration as well as the inter birth interval (IBI) was calculated from video recordings. One day per week, pens were directly observed continuously for 5 min, 4 times/day and all occurrences of piglets performing locomotory play (scamper, chase, pivot, hop), social (piglet nudge, sow nudge, piglet interaction, sow interaction, sow climb), damaging (piglet tail and/or ear bite, or sow body and/or ear bite), fighting, and exploratory (material, toy, pen directed) behaviours were recorded. Data were analysed using SAS v9.4. There was no treatment effect on farrowing duration or IBI. However, Rope piglets tended to perform more damaging behaviour directed towards other piglets than Haylage piglets (0.08±0.01 v's 0.06±0.01 instances/pig/5min; P=0.06). Piglets engaged more with the Haylage and Straw (0.58±0.05, 0.45±0.05 instances/pig/5min respectively) than with Hay and Rope (0.17±0.05, 0.08±0.05 instances/pig/5min, respectively; P<0.01). There was no effect of treatment on pen and toy directed behaviour. The choice of enrichment material provided to sows did not influence farrowing duration under the closely confined conditions of the FC. However, it did influence the amount of engagement shown by the piglets which could influence the development of damaging behaviour.

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Session 4: Companion Animals Behaviour and Welfare

Tuesday, July 23rd, 15:00 – 15:15

Assessing shelter dog behavioral responses to various handling techniques and tools used during routine examinations

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Routine veterinary examinations are vital for monitoring health in dogs; however, unfamiliar handling can elicit fear responses in dogs, and risk handler safety. Restrictive restraints (e.g., full-body, muzzle-hold) and tools (e.g., muzzles) are commonly used in clinic settings and though minimal restraint is encouraged to mitigate fear in dogs, little to no empirical evidence supports recommendations. Thus, to support evidence-based recommendations for dogs in clinic settings, we assessed the influence of common handling techniques on behavioral and physiological indicators of welfare during routine examinations. Dogs (n=97) sourced from a local municipal shelter were randomly allocated to one of five treatments: 1) minimal restraint, 2) muzzle-hold, 3) basket muzzle, 4) soft muzzle, and 5) fullbody restraint. Age, sex, and size were balanced between treatments. Prior to restraint, an approach test was conducted to account for initial responses towards the unfamiliar handler. Dogs were restrained according to their treatment, and video-recorded behavioral (e.g., lowered posture, vocalizations) and physiological parameters (temperature, heart and respiratory rates) were assessed during 2-minute examinations (with handling applied) and 30-seconds post-handling. During handling, most lowered posture was observed during muzzle-hold (17/19, 89%), soft muzzles (13/19, 68%), basket muzzles (11/20, 55%). Posthandling, only five dogs had lowered posture (basket muzzle (n=1), soft muzzle (n=2), and fullbody restraint (n=3)). Regression results indicated increased escape attempts associated with muzzle holds (β =1.43, p < 0.001) and increased vocalizations (e.g., whining) associated with full-body restraints (β =3.7, p = 0.009) compared to minimal restraint. Time to place dogs in respective restraints was greater for basket muzzle (β =58.8, p > 0.001) and soft muzzle (β =46.7, p = 0.001) compared to minimal restraint. During post-handling, increased body shaking was associated with small dogs (<35 lbs) that received muzzle hold (β =4.21, p = 0.012), and basket muzzle (β =5.01, p = 0.002) compared to large dogs (\geq 35lbs) in a soft muzzle.

Further, post-handling, older dogs (5-8 years) that received a basket muzzle had shorter latency to escape from the mat where the examination took place compared to younger dogs (1-4 years) that received full-body restraint (β =-7.5, p = 0.028). Evidence suggests that more negative responses occurred during application of muzzle hold and full-body restraint compared to minimal restraint, and that dog-specific factors (e.g., age and sex) mediate the level of fear-related behaviors displayed. Results highlight the influence of commonly used handling techniques and tools on dog welfare and can inform current recommendations on dog handling practices.

Do Cognitive Traits Associate with Everyday Behaviour in the Domestic Dog (*Canis familiaris*)?

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Even though a large amount of research has focused on testing cognitive traits of dogs, we still know almost nothing about whether these traits associate with dogs' broader behavioural traits outside the test situation. Our aim was to deepen our understanding of what cognitive traits can tell us about dogs' behaviour in their daily lives with their owners. We used two validated owner-filled questionnaires: the Canine Behavioural Assessment and Research Questionnaire (C-BARQ) and the Dog Impulsivity Assessment Scale (DIAS) to determine daily behavioural traits of dogs. A cognitive test battery lasting approximately 1.5 hours was used to measure cognitive traits. The test battery included 7 cognitive tests based on previous research. We used linear mixed models for analysis (N = 987) and accounted for control variables such as sex, breed, age, and training level of dogs. We found that high inhibitory control in the cylinder test was positively associated with trainability (b = 0.07, p = 0.04) and negatively associated with impulsivity (DIAS) (b = -0.02, p < 0.001). A shorter latency to solve the V-detour task was positively associated with management problems (b = -0.26, p = 0.01). Human-directed behaviour during the unsolvable task was associated positively with trainability (b = 0.07, p = 0.03) and stranger-directed fear (b = 0.02, p = 0.01), and negatively with impulsivity (DIAS) (b = -0.01, p = 0.01) and management problems (b = -0.28, p = 0.01). Our findings suggest that cognitive tests could potentially help us to predict and explain dogs' behaviour, personality, and behaviour problems. These results provide important information on dogs' behaviour both in cognitive tests and in their daily lives with their owners.

Effects of white noise, cat music, and exposure time on stress levels of hospitalized cats

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Sporotrichosis is a zoonosis that raises concern in South America. Cats are the most affected species, and treatment requires long-term isolation. The Veterinary clinic environment exposes cats to a range of stressors, loud noises, and sounds unknown by the cats and at unpredictable times. Considering this, auditory enrichment can be applied to help cats to cope. This study aims to evaluate the differences on levels of stress of cats hospitalized for treatment when exposed to cat music, white noise, and no sound periods during different time periods. Cats (n = 11) were housed simultaneously at the university veterinary clinic's infectious diseases isolation ward. Cats were kept individually in double cages under the same environmental conditions and treatment routine. The experiment of 18 days was composed of 3 auditory stimuli and 2 periods without stimuli: a baseline period of no sound before cats were exposed to any auditory stimuli (BS), cat music for 30 minutes (CM), music for 12 hours (CMT), white noise for 30 minutes (WN, positive control), and no sound (NS, control) with sound averaging 55 db. The distribution of treatments was pseudorandomized, and all cats were simultaneously exposed to each treatment for two consecutive days, twice, totaling four days per treatment in. CM and WN treatments were played from 7:00 to 7:30 and CMT was played from 19:30 to 7:30 and BS and NS no sound stimuli were played. The stress levels assessment was performed through direct behavioural observation by the researchers every day between 7:30 and 8:40 (after the end of the experimental sound stimuli). Using the cat stress score (CSS) that ranged from 1 (totally relaxed) to 7 (terrified) and considered the cat's body posture, facial expression, vocalizations, and activity. Statistical analysis was made in R using mixed linear regression using cat as random effect. The CSS was affected by treatment (p < 0.05) though this effect was driven by the difference between the BS and NS (2.20 \pm 0.12; p < 0.01 vs 1.81 \pm 0.09; p < 0.01). Whereas other treatments did not differ from NS. Cats presented the same CSS means when exposed to the treatment music for 30 minutes and white noise $(1.85 \pm 0.09 \text{ vs } 1.85 \ 0.09; \text{ p} > 0.1)$.

The impact of odourised straw on welfare of growing pigs

Jacquelyn Jacobs 1, Anna Breithaupt 1, Marie Hopfensperger 1

Dogs that enter animal shelters are exposed to environmental stressors such as restricted space, sensory overload, and limited social interaction. Clinical kennel stress occurs when the dog's inability to effectively cope with environmental stress leads to chronic excessive hyperactivity or severe despondency. Behavioral and pharmaceutical intervention is often necessary to prevent further behavioral decline, however, shelters vary considerably in available resources to accurately identify these dogs. Therefore, a practical, reliable, and valid tool that measures coping behavior while in shelter would be particularly meaningful to the sheltering community, providing a means to better support shelter animal welfare. Through review of extant literature and in consultation with a veterinary behaviorist, we developed a coping assessment tool consisting of 8 human-animal interactions (HAI), including presentation at kennel door and return to kennel, social play behavior, ability to settle with a treat, and acoustic and visual distractions. Responses to the HAIs were scored and summed and fell within a range of -45 indicating severe anxious avoidance (AA) to +45 indicating severe excessive hyperactivity (EH), with near 0 scores indicating adaptive coping (AC). Ninety-one shelter dogs were enrolled in the study from June-October 2022, and each assessment was recorded to measure validity and reliability of the tool. A blinded board-certified veterinary behaviorist diagnosed participating dogs as either AC, AA, or EH through video observation, with a distribution of diagnoses as 37% AC, 41% AA, and 22% EH in our population. Quadratic weighted kappa, Cohen's kappa, and percent agreement was used to calculate inter- and intrarater reliability of each HAI. Stepwise cumulative regression and linear mixed models were utilized to assess validity and consider HAI candidates for removal. Consideration of statistical significance, reliability and clinical relevance reduced the final assessment tool to 5 HAIs, with acoustic startle tests being least meaningful. Estimated marginal means revealed the reduced assessment to be accurate at identifying AA (-3.34 \pm 0.71) from AC (0.73 \pm 0.73; p<0.01)) and EH $(2.5 \pm 0.99; p<0.001)$), but while numerically distinct, AC and EH were not statistically different (p=0.325). The coping assessment tool is currently being modified to improve

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identification and differentiation of AC and EH dogs. In sum, we expect the finalized tool to have a significant impact on the sheltering community by improving communication between staff members and providing a valid means of measuring and monitoring the effects of chronic stress and behavioral concerns in shelter dogs, thereby improving their welfare.

Scented Toy Enrichment: Exploring the influence of scented toys on owned dog activity levels

Rituparna Sonowal^{1,2}, Alissa Cisneros^{1,2}, Nathaniel J. Hall^{1,2}, Anastasia C. Stellato^{1,2}

Environmental enrichment through the provision of toys can improve the welfare of owned dogs by increasing physical and cognitive activity. Despite dog's olfactory acuity, it is unknown how providing enrichment via scented plush toys influences dog behavior and welfare. Experiment 1 assessed owned dog preference between Playology® scented (beef, chicken, peanut butter, sausage), and non-scented plush toys using multiple stimulus without replacement (MSWO) preference assessment test and level of engagement using ActiGraph accelerometers. Experiment 2 assessed the influence of toys (No-Scent, Scent-Familiar, and Scent-Novel) on dog's physical activity levels using accelerometer data. Owned dogs were recruited and all testing took place in the dog's home. For Experiment 1, dogs (n=30; aged 1-9 years), completed: 1) preference testing using MSWO, and 2) a 72-hour offsite observation period of dog, toy, and play activity with the toys in the dog's home. An array of 5 toys (4 scented and 1 non-scented) were presented to each dog in an MSWO, where the 'preferred' toy from that trial was removed for each subsequent trial; this was repeated over four days. Accelerometers were attached to dog collars and inserted in each toy to measure dog activity and toy activity, respectively. Play activity was assessed by matching the 1-min time-stamped integrated axes of the accelerometer on the collar and on the toy, to capture periods of simultaneous movement. Wilcoxon test showed that the play activity levels with preferred scented toys was higher compared to non-scented (p < 0.001). Results suggested that individual variation in dogs indicates individual preference for specific scents of toys. For Experiment 2, dogs (n=33, aged 1-7 years) were allocated to one of three treatment groups: 1) Scent-Familiar (11 dogs from Experiment 1 were provided with a scented toy that had the highest activity levels), 2) No-Scent (n=11, provided with a non-scented toy), and 3) Scent-Novel (n=11, provided with the same peanut butter scented toy from Experiment 1, as it had the highest toy activity across dogs). Linear regression results suggest that older dogs showed

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reduced activity (toy activity: p = 0.006, play activity: p = 0.001, physical activity: p = 0.0005). Toy activity levels in Scent-Familiar were lower compared to No-Scent (p = 0.005), reflecting a habituation effect where toy activity decreased with time (p = 0.01). Results highlight the importance of tailoring enrichment practices to individual dog preferences to optimize enrichment strategies to promote dog behavior and welfare.

Session 5: Farm Animals Behaviour and Welfare (Poultry)

Tuesday, July 23rd, 17:15 – 17:30

Associating maternal keel fracture severities with hatch quality and welfare of chicks

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Maternal conditions have been associated with offspring behaviour and performance in laying hens. Evidence suggests that keel fractures cause pain, result in poor egg quality and could increase stress and fear responses in affected hens. However, the impact of maternal keel fracture on chick quality and welfare remains unclear, which is highly relevant for breeder hens. This study hypothesized decreased hatch rate, chick weight and increased fear responses in chicks from hens with severe keel fractures. 120 Bovans brown breeder layers aged 61 weeks with different keel fracture severities were tagged and co-housed with 12 cocks at 20 hens+2 cocks/room. Hens were randomly assigned one of six rooms and managed under similar conditions. Egg laying was directly observed for individual collection, so eggs were labelled after each hen. Collection was done four days/week over three weeks, resulting in three batches (1 batch/week). At the end of egg collection, 111 hens were euthanized and keel bones were visually scored (0:no fracture, n=21; 1: slightly fractured, n=25; 2: severely fractured, n=65). During incubation, eggs were assigned random positions and candled on day 18. Fertile eggs were transferred into hatcher ensuring random mixing of egg positions between hens to avoid hatcher effect. Between days 21-22, number of chicks hatched/hen was recorded. 353 pullets were generated across three batches (0=69; 1=88; 2=196). Hatch rate was calculated as the number of hatched eggs relative to fertile eggs. Chicks were weighed, tagged and housed in the same room according to their mother's fracture degree. Body weight of chicks was measured weekly for five weeks. At week four, chicks' feeding behaviour within room was recorded in the morning (9-11am) and evening (4-5pm). All chicks were subjected to Tonic immobility (week four), novel arena and novel object tests (week five). Data on hatch rate and chick weight were subjected to One-way ANOVA. Tonic immobility duration was analysed using Kruskal-Wallis test. Analyses of feeding behaviour, novel arena and object tests, are currently underway. Hatch rate reduced as keel fracture

degree increased (F2,149=4.950, P=0.008) while chick weight at hatch and week 5 were similar (p>0.05) for all fracture degrees. Tonic immobility duration showed no difference (p>0.05) between fracture degrees. Conclusively, breeder layers with severe fractures had lower egg hatchability which has implications for the managerial practice of the poultry breeder industry. Maternal fracture did not affect chick growth. Results of feeding behaviour, novel arena and objects will be presented at the conference.

Domestic chicken breeds are more important in maternal reactivity and chick behaviour than whether chicks were in- or cross-fostered

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Chicks brooded by hens are less fearful, less aggressive, and exhibit reduced feather pecking later in life. To differentiate genetic and environmental effects, a comparison of chicks reared by hens of the same (in-fostering) or different breeds (cross-fostering) is of interest in terms of maternal-chick bond strength, degree of exploration and level of fear. Results from this research will inform poultry farmers on the choice of broody hens for extensive rearing or corresponding stimuli to provide maternal care for intensive housing, optimizing chick welfare, and productivity. This study investigated the effects of two phenotypically, genotypically and behaviourally divergent hen breeds (silkie and bush fowl) that both display high levels of brooding and maternal care, chick breeds (silkie and bush fowl), and maternal care duration (three or six weeks). The hens and chicks' responses to a separation test and the chicks' behaviour in a novel arena and a simulated predator test were observed. We used a total of 31 hen-chick groups. At the end of either three or six weeks of maternal care, four behavioural tests were conducted: a physical and visual separation for 10 minutes (in balanced order across hens), an exposure to a novel arena for 5 minutes, followed immediately by a simulated predator for 2 minutes. The principal components extracted from the behavioural data of each test were analysed as outcome variables using linear mixed-effects models. During the separation tests, bush fowl hens displayed increased (p = 0.0002) floor pecking, walking, maintaining high posture and head alert movement, and approached chicks by staying closer to the partition than the silkie hens. However, the behaviour of both chick breeds was similar in the separation tests. For the exploratory and simulated predator tests, bush fowl chicks displayed a shorter (p < 0.0001) latency to respond, crossed more squares in the arena, and vocalized earlier and for a longer duration (p = 0.0044) than the silkie chicks. There was no hen breed-by-chick breed interaction on hen and chick behaviour during the tests. In conclusion, the maternal response of bush fowl hens was not influenced by the breed

of chick they reared. Bush fowl chicks cared for by either hen breed were faster to explore a novel arena and respond to a predator, indicating that fostering type and maternal duration had no influence on bush fowl chicks' behaviour, but silkie chicks reared with mothers for 3 weeks were more reactive than those reared for 6 weeks.

Effects of feeding a fiber-rich diet and roughage during rearing of broiler breeders on behavior and welfare

Kaitlin Wurtz^{1,2}, Karen Thodberg¹, Anja Riber¹

Alternative feeding methods for broiler breeders have been proposed, such as qualitative feed restriction in the form of diluting the diet with a fiber source, to help alleviate some of the negative consequences resulting from feed restriction such as chronic hunger. The aim of this study was to investigate the effect of a fibre-rich diet containing oat-hulls and a daily allocation of roughage (EXP) versus a standard commercial diet (CON) on broiler breeder welfare and feeding motivation, measured using behavioural, stress physiological, and clinical welfare indicators. Nutrient content was similar between the two diets. 600 day-old female Ross 308 breeder chicks were randomly allocated to 12 pens, each receiving one of the dietary treatments. Motivation for feeding was assessed via feeding rate and frustration tests (thwarted feeding) performed at 8 and 17 wks of age. At the end of the study (18 wks), plumage condition, contact dermatitis, and number of fault bars in the feathers were assessed. Dry matter content of the litter was determined at 6, 12, and 18 wks. In wk 8, there was a tendency for CON birds to consume feed faster than EXP birds (p=0.0672), possibly indicating increased feeding motivation. During the frustration test in wk 8, EXP birds tended to perform more behavioural transitions than CON birds (p = 0.0659) possibly indicating increased frustration at not being able to access the feed. No treatment effects were observed during wk 17 for either the feed intake or frustration tests. Birds in the CON treatment had poorer plumage condition at the end of the study than the EXP birds (p < 0.0001). There was a significant difference in the odds of receiving lower scores (healthier condition) for hock burns for birds in the EXP compared to the CON treatment (OR: 8.3, CL: 1.64-41.68). CON birds were more likely to have many fault bars in their scapula feathers compared to EXP birds (p < 0.0001) possibly indicating increased stress. Litter quality deteriorated over time across both treatments, with dry matter being lower in wk 18 than both wks 6 and 12 (p < 0.0001). There was a tendency for higher dry matter content in pens fed the EXP diet (p = 0.0728). Results suggest that the EXP diet led to improved welfare indicators and litter condition and possibly

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reduced hunger, however behaviors indicative of frustration suggest that the EXP diet did not eliminate the birds' motivation to feed.

Feathered Friendships: The development of a social test paradigm to investigate prosocial behaviour in laying hens housed in commercial conditions

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Prosociality is defined as behaviour that benefits other individuals. Test paradigms to assess prosocial behaviour in different animal species have been developed in recent years but none have tested laying hens. Our study aimed to develop an apparatus to test for prosocial behaviour in laying hens. Specifically, we tested if the apparatus was biologically relevant to hens within relatively large groups (N=225), if hens were consistent in their roles (initiator/receiver), and if initiators were facing the receivers when using the apparatus. The apparatus was developed to minimize disruptions by not removing hens from their everyday environment and based on the animals' own motivation and learning. The test apparatus was designed as a seesaw with a perch platform on one side and a reward concealed under a lightweight lid on the other. When a hen was perching, the weight of the hen caused the lid to rise on the other side and reveal the reward (Test). An identical apparatus, which worked in the same way but without any reward, was used as a control (Control). Both apparatuses were additionally able to detect the presence of individual hens via leg-mounted transponders that interacted with RFID system under the apparatuses. We tested all hens (N=214) in their home pen (N=1) where they had access to both apparatuses simultaneously in an outside winter garden for 3 hours per day over 14 days during a three-week period. A total of 200 hens were registered, 67 hens as initiators on the test apparatus and 38 on the control. The frequencies of perching events and mean duration per perching event were analysed using Wilcoxon signed-rank tests, and the preliminary results showed that 187 events occurred on test apparatus and 62 events on the control (P=0.0014). We found no difference in mean perching duration between the test apparatus (21.1s) and control (30.6s) (P=0.86). While perching, 63% of the hens were facing towards the hens receiving the reward, while 45% of the hens faced the control's "reward" tube. The results indicate that the apparatus was

biologically relevant for the hens to use, since they figured out how to operate both apparatuses within the different roles (initiator/receiver), and that hens may have the ability to understand the purpose of using the apparatus. To the best of our knowledge, this is the first apparatus developed to test prosocial behaviour in laying hens within commercial groups in the absence of specific training.

Investigating the impact of early dietary interventions during stress on laying hen behaviour: insights from novel object and manual restraint tests

Annemarie J.W. Mens¹, Ingrid C. de Jong¹, René P. Kwakkel²

It's been noted that young pullets are susceptible to stress, potentially affecting their ability to cope later. Nutrition during this critical early period may play a role. We studied the impact of four diets administered during two key early rearing phases on layers' stress response, during the manual restraint (MR) and novel object (NO) test. We hypothesised that specific dietary compositions could mitigate stress later in life. Our study followed a 2x4 factorial design, involving stress by disruption during two sensitive periods (2-4 or 5-7 weeks of age; woa) and four diets provided during and two weeks after these periods. Each treatment combination was replicated six times with pen as the experimental unit. Disruption involved removing litter, perches, pecking blocks; leaving only slats. Diets comprised a control (CON) and three experimental diets: increased fat/fibre (FAT), additional energy (+100 Kcal; ME), or essential amino acids (+10%; AA). MR was conducted at 11, 19, and 35 woa, the NO at 10, 14, 22, 28, and 32 woa. Data were analysed using ANOVA for normally distributed data and GLMM for non-normally distributed data, with diet, disruption, and their interaction considered in the models. At 11 woa, during the MR, hens stressed during 2-4 woa showed more vocalizations (+4.9; P=0.022) and struggles (+0.3; P=0.031) than those stressed at 5-7 weeks. The ME-diet led to more vocalizations (+8.6; P=0.008) than CON- or AA-diets. At 20 weeks, AAfed hens stressed during 2-4 woa had a longer struggle latency (+60.7s; P=0.04) compared to FAT-fed hens stressed at 2-4 woa and ME- or AA-fed hens stressed at 5-7 woa. At 28 weeks, ME-diet led to lower latency (-25.2s, P=0.018) for the first 3 random hens to approach the NO within 10cm, compared to AA-diet. In week 22, the combination of stress at 5-7 woa and FATdiet tended to have a longer latency for the first 3 random hens close to the object compared to birds stressed at 5-7 and fed AA (-42.7s) or ME-diets (-43.4s) and stressed at 2-4 combined with CON-diet (-43.0s; P=0.06). Early rearing stress (2-4 woa) resulted in a more active later hen stress response during MR, with increased vocalisation and struggle frequencies. Little effect of the diets were observed. This study highlights the impact of early life stress on

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individual stress responsiveness, suggesting that more energy-dense diets negatively affect fear responsiveness at later life, while an increase in AA could have some beneficial effects.

Play ontogeny in young chickens - effects of domestication, tameness, and sex Rebecca Oscarsson¹, Per Jensen¹

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It is known that young chickens display different play behaviours, however, the research is scarce. Moreover, domestication is believed to have increased playfulness in multiple species, and tameness is hypothesized to be the main selected trait driving the domesticated phenotype. Furthermore, potential sex differences in play have not been investigated in chickens. Therefore, in three separate studies, we aimed to describe potential effects of (1) domestication, (2) tameness, and (3) sex, on chicken play ontogeny. First, we compared the ancestral Red Junglefowl (RJF) and the laying hen hybrid of White Leghorn (WL). Secondly, we compared Red Junglefowls selected for high (RJF HF) and low (RJF LF) fear of humans. Lastly, male, and female WL were compared. Incubation, hatching, and housing occurred under the same conditions. Groups of three to four chicks were created, and the same group were then moved to a play arena twice per week, from day 6 until day 53 post hatch. The frequency of 14 behaviours, categorized as locomotor play, social play and object play were recorded during 30 min. Every play group constituted the independent statistical replicates. In all three studies, the play frequency peaked between 25-40 days of age. In study (1), total play and object play were significantly more common in WL (total: F1, 252 = 106.1, P < 0.001; object: F1, 252 = 5.9, P < 0.05), whilst locomotor and social play were significantly more frequent in RJF, (locomotor: F1, 252 = 8.9, P < 0.001; social: F1, 252 = 25.4, P < 0.001). In study (2), the preliminary results show an effect of tameness, where RJF LF play more (total: F1, 269 = 63.509, P < 0.001). In study (3), total play, social play and object play were significantly more common in males (total: F1, 164 = 121.719, P < 0.001; object: F1, 164 = 54.167, P < 0.001; social: F1, 164 = 144.030, P < 0.001), whereas no difference in the frequency of locomotor play was found (F1, 164 = 1.375, P = 0.243). Hence, domestication has led to a higher occurrence of play, mainly directed towards objects. The finding that tame chicks play more indicate an important role of tameness for domesticated behaviour. Lastly, that males play more than females regarding social, and object play may be related to the large sexual dimorphism in the species. The results may inform future studies on the play-welfare relationship in young chickens.

Session 6: Education in Animal Welfare and Behavior

Tuesday, July 23rd, 17:15 – 17:30

Applicability of the Glasgow Feline Composite Measure Pain Scale and the Feline Grimace Scale in assessing pain in cats by veterinary medicine students, and the sociodemographic factors that influence their utilization

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The complexity of pain identification in animals, particularly in cats, is recognized. While there are validated methods for pain recognition, their knowledge and implementation are not widely used in veterinary medicine practice. The study aimed to estimate the agreement in the assessment of pain in cats and the sociodemographic factors influencing pain perception by veterinary medicine students using the Glasgow Feline Composite Measure Pain Scale (CMPS-Feline), and the Feline Grimace Scale (FGS). Ten female cats underwent elective orchiectomy and were video recorded after 2 hours at the veterinary hospital. The videos were edited to last 1.5 minutes, providing sufficient time to simulate the CMPS-Feline. Moreover, images were obtained from them. These audiovisual materials were utilized in on-site training sessions, where 76 veterinary medicine students (female = 63, male = 13) at the Universidad de Las Américas (UDLA), Chile, utilized the CMPS-Feline, and FGS mobile phone application. Cronbach's alpha reliability was also measured to testify internal consistency of both scales. The degree of agreement when using both scales was determined using the Fleiss' kappa coefficient (κ) for multiple raters. Logistic regression models with Poisson distribution were used to determine the sociodemographic factors influencing the evaluation. Statistical analyses for this study were performed using the SPSS version 26 software. The internal consistency of CMPS-Feline and FGS was high, with Cronbach's alpha values of 0.989 and 0.996, respectively. The CMPS-Feline showed greater variability in pain scoring and an agreement classified as "poor" (κ= 0.291), in contrast to FGS, where a higher and "fair to good" agreement was obtained (k= 0.662). Students with experience as cat owners assigned lower pain scores when using CMPS-Feline than those without experience (6.13 vs. 7.09; P = 0.014).

Furthermore, students who had more than 3 cats assigned higher scores, in contrast to those who have only one cat (7.46 vs. 6.85; P = 0.044). First and fourth-year students assigned higher pain scores (P < 0.05) on both scales than fifth-year students. The use of FGS, which utilizes facial expression and is a mobile application, appears to be a more reliable tool than CMPS-Feline for veterinary medicine students. Experience as a cat owner and year of the veterinary medicine program influence the perception and evaluation of pain. The need to integrate the use of validated pain assessment scales into academic training is emphasized, aiming to contribute to the understanding and management of pain in cats, thus promoting animal welfare.

Applied animal behaviour and welfare researchers' perspectives on the challenges they face while engaging in research

Bianca Vandresen¹, Emeline Nogues¹, Marina A.G. von Keyserlingk¹

Academics in the fields of applied animal behaviour and welfare science may face challenges while working within the constraints imposed on them by institutional and regulatory frameworks. However, to our knowledge no study has described the nature of these difficulties and whether there are regional differences. We recruited 47 delegates attending the 56th congress of the International Society of Applied Ethology (ISAE) held in Tallin, Estonia in August 2023 to participate in focus group discussions. Participants represented 33 countries, covering five continents. Of the 47 delegates, three of them participated twice, once as part of the Developing Countries Congress Attendance Fellowship (DCCAF) workshop, and then again two days later in a workshop open to all attendees. The DCCAF group had 15 participants, and the later workshop was composed of six focus groups where five groups had six participants and one group had five participants. Participation in the study was voluntary and anonymous, and all participants signed a consent form before the discussions started. All data collection methods were approved by The University of British Columbia Behaviour Research Ethics Board and adhered to the ISAE Ethical Guidelines. Using a semi-structured interview guide, participants were encouraged to discuss the challenges they face while engaging in research, possible solutions to the identified challenges, and the role international societies such as ISAE could play in overcoming some of these challenges. Each of these seven group discussions was audio-recorded and transcribed and subjected to thematic analyses by two coders that familiarized themselves with the transcripts prior to starting the analysis. Patterns in codes were identified as themes, and connections between codes and themes were described. Three main themes were identified: (1) the discipline of animal welfare and behaviour; (2) conducting and sharing research; and (3) researcher welfare and networking. Many of the challenges identified were shared among all participants, regardless of region but some issues were more strongly voiced by specific to socio-demographic groups. Many delegates described numerous barriers hindering their research process, originating from

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within their academic institutions, local governments and journal guidelines, but also arising from prejudice and other personal challenges. Whilst solutions were difficult to identify, the majority of participants communicated their willingness to collaborate as a first step to change patterns.

Evaluation of veterinary doctors and students of veterinary medicine on the factors that affect the welfare of dogs during veterinary medical care

Patrícia Arroyo Ruiz¹, Adroaldo Zanella¹

Veterinarians and Veterinary students can promote dog welfare in clinical practice when implementing best practices in their routine work. There are no published studies evaluating how frequent attitudes that enhance dog welfare happens in clinics. A multiple-choice questionnaire with 40 practices aimed at promoting dog welfare in veterinary consultations was applied to verify how Veterinarians (n=42) and Veterinary students (n=53) considered those practices important and frequent, classifying them using a Likert scale. The questionnaires (n=95) were distributed electronically to Veterinarians and students of the state of São Paulo. The Brazilian human ethical review board approved the protocol (number 5.894.436). Veterinarians worked either in public institutions (54.8%) or in private institutions (45.2%). A scale from 0 to 4 was attributed to the levels of frequency and importance on the qualitative data. Pearson's correlation coefficient and Student's t-test for paired samples were applied (Shapiro-Wilk test, p>0.05). Responses from Veterinarians and students were analyzed to verify which factors were considered most and least relevant, most and least frequent, and the correlation between them was assessed. The association between mean frequency and mean importance attributed to each practice (n=40), measured using Pearson's correlation coefficient was 0.42 (p=0.0063, 95%Cl=0.13-0.59) reveals a moderate association. The presence of many outliers contributes to the poor correlation. When comparing the means of importance with the means of frequency (paired Student's t-test), it was observed that there is a statistically significant difference between them (p<0.0001). Responders tended to attribute higher scores to importance (mean=3.21; 95%CI=3.10-3.32) than to frequency (mean=1.81; 95%CI=1.63-1.99), demonstrating that there is a disparity between the extent to which they assessed if the practices presented are relevant to the welfare of dogs and their application in veterinary clinical practice. Items that were related to a low stress environment such as separate dogs from cats in the waiting room and let the dog get used to the room before start examination, tended to be classified as low frequent and high important. Recognize signs off stress in dogs was also classified with high importance and low frequency.

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Most items classified as having high frequency and high importance were related to the veterinarian posture and conduct. Leaving treats, water, and toys available for the dog were items to which were attributed low frequency and low importance. The results indicated that the most critical deficiency is in actions, transforming knowledge into practice, and increasing the frequency with which these practices are applied.

Impact of observers' training on gender bias and reliability of the Angus calf grimace scale

Mostafa Farghal¹, Ed Pajor¹, Maria Camila Ceballos¹

This study explores the effect of observers' training on their reliability and gender bias of the Angus calf grimace scale (ACGS). ACGS is composed of six facial action units (FAUs) with a three-point scoring system. Videos of 34 Angus beef calves were recorded before, during, and after castration. To manage pain, calves received an oral dose of Meloxicam (1.0 mg/kg body weight) immediately following castration. Screenshots of calves' faces were extracted from each video before and after castration and meloxicam injection but prior to the effect of meloxicam is expected. Two different training sessions (TS1 and TS2), 4 months apart, were performed by reference observer (MF). Three scoring rounds were done using 70 images from calves before, during, and after castration (round 1 was done one week before TS1, round 2 was done 3 weeks after TS1, and round 3 was done 3 weeks after TS2). Images were renamed and randomized by a third party on each round to avoid observer bias. TS1 was conducted to six observers (3 females, 3 males); who scored images in round 1 using FAUs description scoring manual and round 2 after TS1. Due to low observers' reliability with MF after TS1, TS2 was conducted to seven observers (6 females and 1 male; 4 of them participated in TS1). Inter-(with MF) and intra-observer reliability were evaluated using the intraclass correlation coefficient (ICC) of a single total score ranging from 0 to 1 (sum of FAU scores/maximum score). Bland-Altman analysis and limits of agreement (LOA) were calculated using average scores of females and males separately using RStudio. Gender bias analysis was done for the first and second rounds when observers' gender was balanced. Inter-observer reliability with MF ranged from poor to good in the first (ICC = 0.20-0.75) and second (ICC = 0.11-0.77) rounds and moderate to very good in the third round (ICC = 0.51-0.90). Among observers who participated in all rounds, inter-observer reliability was 0.75 (0.64-0.84, P < 0.001), 0.53 (0.31-0.68, P < 0.001), and 0.88 (0.82-0.92, P < 0.001) at first, second and third rounds, respectively. Minimal gender bias was detected before TS1: -0.59 (-0.88, -0.30), with lower LOA= -2.98 and upper LOA= 1.8. This bias was reduced after TS1 to -0.29 (-0.60-0.02) with lower LOA= -2.86

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and upper LOA= 2.28. In conclusion, observer's training on the ACGS evaluation reduces gender bias and increases observers' reliability.

Member experiences from an Animal Welfare Education Community of Practice (AWECoP)

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Animal welfare science is a growing field in higher education, requiring professionals trained in teaching this complex topic. To develop a dialogue amongst educators within the field, we created an animal welfare education community of practice (AWECOP) for those teaching animal welfare science, applied ethology, and/or animal ethics and here we describe the history, objectives, and members' experiences within this community. AWECOP hosts 6-8 meetings annually for members to discuss topics relevant to our community and exchange teaching resources; within its first two years, the community has grown to 121 members representing approximately 70 institutions across 6 continents. A 12-question mixed-method survey was distributed to capture member demographics, engagement with AWECOP, motivations for joining, and self-evaluation of AWECoP's impacts. Quantitative data from the survey are presented descriptively, while reflexive thematic analysis was applied to the qualitative data. Survey respondents (n = 54) felt that AWECoP is a vital community and safe space for members to share their ideas and receive feedback, inspiration, information, and resources about subject-specific and broader pedagogical topics. As a result, a majority experienced professional (e.g., development of new contacts) and personal (e.g., increased feeling of belonging in their field) benefits, as well as impacts realized in their teaching practice. We conclude with an examination of challenges faced in ensuring AWECoP remains accessible to a growing membership and offer recommendations for facilitating similar communities to support fellowship and training in the teaching of animal welfare and applied ethology.

Observer bias in veterinary students: implications for welfare assessment of dogs from commercial breeding kennels and the role of veterinary medical education

<u>Shanis Barnard</u>¹, Margaret Pritchett¹, Jennifer E. Weller¹, Traci Shreyer¹, Candace Croney¹

Observational biases may result from a person's experiences, education, empathy for, and attitudes towards animals. Such biases can affect how animals' behavioral and emotional states are perceived, and in turn, influence how their welfare may be assessed. When observational biases occur in veterinary medicine, they may be particularly problematic, as correctly interpreting an animal's state is essential for providing good welfare. In this study, veterinary students from three Veterinary Medicine Colleges in the USA (n=190, 86% females, 25-29% enrolling in the first 3 years and 17% enrolling in the 4th year/clinical rotations) were asked to score the emotional states of dogs residing in USDA regulated commercial breeding kennels (CBKs) from videos (N=8, different breeds in each video), using a fixed list of qualitative behavior assessment (QBA) terms. Pen conditions were clean, and all dogs appeared clean and healthy. We investigated whether being informed about dogs' sourcing, observer empathy towards animals, and other factors, such as gender and education, influenced participants' interpretations of dogs' emotional states. Students who were informed that dogs were from CBKs scored dogs higher on the QBA term depression than the uninformed participants (t-test: F=9.61, p=0.002). QBA terms were reduced to three components (PCs) using principal component analysis, and linear mixed models were used to analyze the effects of different factors on these PCs. Female students (vs. male, p=0.036), those in their 2nd year (vs. 4th year, p=0.032), and those in the small animal track (vs. food animal, p=0.008) were more likely to score dogs as expressing higher levels of fear/stress-related states (PC2). These findings indicate that veterinary students' assessments of the emotional states of dogs originating from CBKs reflected some degree of bias, perhaps related to their perceptions of such kennels, though demographic and other factors also appear to have influenced their judgments. Because bias may introduce inaccuracy into assessments of dogs' states that potentially undermines the quality of care, interactions, and trust clients experience in

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veterinary medicine, these findings reiterate the importance of addressing bias in veterinary medical education curricula. Further, the results suggest that veterinary professionals, scientists, and others should pre-emptively quantify observer bias when integrating qualitative assessment tools into dog welfare assessment protocols given the need for valid and reliable assessments that are used to drive related decision-making.

Session 7: Farm Animals Behaviour and Welfare (Mixed)

Wednesday, July 24th, 10:45 – 11:00

Consistent individual differences in cattle behavior and how they relate to grazing distribution on extensive rangelands

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Consistent individual differences (CIDs) in beef cattle behavior have been studied using a variety of methods, but many require restraint (e.g. in a squeeze chute or similar apparatus). Cattle exhibit consistent grazing patterns on extensive rangelands that contribute to sustainability of land use for cattle production. If CIDs measured when cattle are handled or processed can predict grazing personalities while cattle are on rangeland, this would have implications for rangeland conservation. Cattle behavior was measured in repeated management assays over two years while cows were isolated and involved a handling procedure, chute, and hydraulic squeeze. Cows also participated in two preference assays, one which was a social-feed tradeoff task and one which incorporated response to a novel item. The same cattle (n=49) were also tracked with GPS collars over two summer grazing seasons (June-September in 2021 and 2022). We calculated repeatability of behaviors across repetitions of the management assay, which were: durations for cows to be handled and traverse the chute, squeeze, and upon exiting the squeeze. Daily rangeland use metrics of elevation, slope, distance traveled, distance from supplement, loafing sites, and water were averaged by week. Two rangeland use metrics were calculated with data from each entire grazing season: kernel density core home range and social network degree strength. Durations of repeatable behaviors from the management assay, and latencies to consume feed in the social-feed tradeoff assay and approach a novel bucket in the novel approach assay were standardized predictor variables in eight separate mixed models with the rangeland use metrics as response variables. Cows with a more passive response in the chute (took longer to traverse) were found at higher elevation (p=0.017), further from water (p = 0.043), and closer to supplement sites (p = 0.029), and tended to have wider core home ranges (p = 0.067), and use steeper slopes (p=0.060). Cows that had higher latency to supplement in the socialfeed tradeoff task traveled shorter distances on rangeland (p = 0.035). There was some

evidence that a more passive response to isolation and processing, possibly showing a reactive coping style, meant cows had more optimal grazing patterns on rangeland because they used more rugged terrain at higher elevations and did not clump near water. Cows' willingness to separate from conspecifics to consume supplement in a social-feed tradeoff assay was consistent in their willingness to travel further distances to forage while on rangeland.

Could environmental factors decrease resting time and impair welfare and physical performance in horses?

Tiago Oliveira¹, Angela Barbosa¹, Raquel Baccarin¹

The study of sleep patterns in horses is little explored and has great potential for improving performance and welfare. Some papers indicate that there is a great influence of the environment on the resting pattern of horses. In osteoarthritic horses monitored using cameras during the first 5 days of hospitalization, the total time of lateral recumbency and frequency of recumbency were greater after the 4th day of hospitalization, while the total time of sternal recumbency was greater after the 3rd day. The severity of the disease appeared to affect recumbency time, as horses with mild osteoarthritis spent more time in recumbency while those with severe osteoarthritis may have been partially sleep-deprived because they laid down less. Another study showed that the average times of sternal and lateral recumbency, and the number of times the horses were in sternal and lateral recumbency were lower on days when the horses were hospitalized, but baseline values were reached on the first night of returning to their stalls. Sub-optimal husbandry conditions, such as lights on at night and low bedding, can significantly reduce the horses' resting time. However, under these conditions, there was no negative impact on the horses' cognitive performance. In another study, REM sleep deprivation was achieved by not letting the horses attain sternal or lateral recumbency for 72 hours to investigate the impact of interference in sleep patterns on athletic and cognitive performance. There was a tendency to increase the time needed to resolve spatial memory tasks in the sleep-deprived group, suggesting an impact on equine cognition performance. These same horses were evaluated with Horse Grimace Scale (HGS), heart rate variability (HRV), and during a test competition. Regarding the HGS e HRV, there were no alterations in score between the control (not sleep deprived) and sleep deprivation groups. Furthermore, judge scores were not able to identify which horses were in the sleep-deprived group during the test competition, and the horses achieved similar scores when they slept and when they were deprived of REM sleep, without identifying improvement or worsening in performance during the competition test. Studies around horse sleep have been very

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promising and require more knowledge and technology to advance their application in the field.

Preference for different pecking blocks offered as pair-wise comparisons in White and Brown-feathered laying hens

<u>Tunmise Ehigbor</u>^{1,2}, Elijah Kiarie¹, Alexandra Harlander^{1,2}, Tina Widowski^{1,2}

Different commercially available pecking blocks (PB) have varying nutrient composition, appearance, and texture. However, the relative preferences of laying hens for different PBs are unknown. We housed 120 Dekalb White and 120 Bovans Brown in twelve (360cm x 116cm) test cages from 18 weeks of age. Test cages had two identical sections, each containing a nest, scratch mat, and container. Then, three types of PB (A, B, C) were offered in a series of pairwise comparisons over three 4-week periods. Block A mainly comprised mineral + grains, B of mineral + grain + molasses, and C primarily mineral based. PB use (disappearance) was measured by weekly weighing. Pecking and scratching behaviour was measured at the individual level by live observations of 7 focal birds (per cage) once a week (thrice daily). Behaviour at the group level was measured from video cameras mounted over each PB. The numbers of birds pecking or scratching (POS) at the blocks were counted every 5 minutes throughout the light period using instantaneous scan sampling (total = 169 scans per day, split into 3 times of day (TOD): a.m., mid-day, and p.m.). Data were analyzed using SAS Glimmix to determine fixed effects of strain, PB type, period, pair-wise combination, and time of day. Brown hens used the B block more than any other block and strain combination (p < 0.0001). The frequency of scratches/7 focal birds/time interval in the Whites was higher at C and B than at A block (p = 0.04); in the Browns, it was higher at B than at C and A block (p < 0.0001). At the group level, pair-wise comparison affected POS behaviour (p < 0.05). In the Whites (p = 0.04), the pair-wise comparison of blocks B and C resulted in more birds POS at C. The opposite was observed in the Browns, with more birds directing POS at B (p = 0.01). Interactions were also observed between the time of day and PB type (White: p < 0.0001, Brown: p = < 0.0001). The Whites pecked and scratched the C block at p.m. more than any other block at any other TOD. In the Browns, POS was directed at the B block at mid-day and a.m. more than any other block at any other time. The two common strains of laying hens in this study preferred different types of PB and used them at different times of day.

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Feeding horses in groups: do feeding strategies affect horse welfare?

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Finding feeding strategies that meet the needs of horses without compromising their body condition is essential to optimise their welfare. This is even more important for horses kept in groups, as hay availability is directly related to the risk of injury. Recently, two feeding strategies have emerged to improve horses' welfare: portioning the horses' daily intake into smaller, more frequent meals using timed hayracks, or slowing their intake using so-called "slowfeeders". However, little is known about the effects of such management practices on horse behaviour. We designed a cross-over study with 18 mares divided into four groups to compare three feeding strategies: "traditional" (3*2-hours meals/day, TD), "portioned" (6*1hour meals/day, PO) and "slowfeeding" (ad libitum hay covered by a net, SF). Each treatment consisted of 3 weeks of habituation and 2 weeks of data collection during which we observed each group for 15 hours. We continuously recorded social interactions and noted the position and activity of each mare every 15 minutes. We also recorded injuries at the beginning of each data collection period, as well as two and four days after. Finally, we measured the time spent lying during data collection using accelerometers. We analysed the effects of treatment on agonistic and affiliative behaviour within groups using generalised mixed model and selected the best model using AIC. We used the same procedure for the injuries and lying behaviour at the individual level. Horses in SF spent 70% of their time feeding (similar to natural conditions), suggesting that the presence of the net did not disturb the horses. In line with our hypothesis, fewer injuries and more affiliative behaviours were observed during SF (1.6 behaviour/horse/hour, vs. 0.4 for both PO and TD). Surprisingly, however, our results suggest that PO may be more frustrating for the horses than TD. Indeed, significantly less agonistic behaviours were observed at feeding times in TD (10.5 behaviours/horse/hour on average) compared to PO (+ 5.6, 95% Confidence Intervals = [1.7; 9.6]). Furthermore, lying behaviour tended to be impaired in PO (-11.3min/day, 95% CI[-25.8; 3.1]) compared to SF and TD (37.5 min/day on average). In our study, portioning into smaller, more frequent meals did not reduce the stress in horses, stressing the need for further research on portioning strategies to

find optimal management. Our results further highlight that ethological observations are an effective - non-invasive - way to assess the impact of human activities on equine welfare.

Using a real-time location system to assess how tail docking affects lamb activity and proximity to the dam

Jocelyn M. Woods¹, Sarah J.J. Adcock¹

Many lambs globally have a portion of their tail removed, a procedure known as tail docking, because it is believed to reduce fecal soiling of the wool. However, support for this rationale is sparse, and tail docking unequivocally causes acute pain, which can become chronic. The potential for chronic pain and impaired communication may reduce social behavior and activity during a critical developmental period as has been seen in other species, but this has not been explored in docked lambs. We aimed to test whether tail docking lambs with rubber rings impacted their proximity to their dam and activity during the preweaning period. Within 28 Polypay female twin pairs, one lamb was docked by placing a constrictive rubber ring on the tail between 24-48 hours of age while the other lamb's tail was left intact. Two days after lambing, lambs (n=56) and their ewes (n=28) were fitted with harnesses containing a Noldus TrackLab ultra-wideband real-time location sensor that continuously recorded the animal's x,y coordinates. The twin pairs and their dam were tracked for 57 days in 2 group pens within a barn (14 dam-lamb families/pen). Linear mixed models were used to determine if each lamb's proximity to their dam and their daily distances traveled, both calculated using Euclidean distance, were predicted by their docking status and days since docking, with individual ID nested within dam as a random effect. There was no significant difference between the docked and undocked lambs' proximity to their dam (mean 2 SE; docked: 9.47 2 0.35 m; undocked: 9.32 2 0.35 m; X2 = 0.77, p = 0.38). As time progressed, the distance between the lambs and their dam increased, regardless of treatment (X2 = 50.21, p < 0.001). The daily distance traveled did not differ based on treatment (docked: 6.7 2 0.2 km; undocked: 6.7 2 0.2 km; X2 = 0.005, p = 0.95) but did decrease over time (X2 = 18.82, p < 0.001). Our results do not suggest that tail docking with rubber rings within 24-48 hours of age influenced the lambs' proximity to their dam or their locomotor activity in the preweaning period. However, lambs spent less time with their dam and were less active as time progressed, providing insight into how activity patterns change with age. Further research on inter-lamb interactions would

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provide insight into how tail docking may influence other important social relationships in the preweaning period.

Session 8: Human-Animal Interactions

Wednesday, July 24th, 10:45 – 11:00

An integral approach to understanding rancher-animal relations in regenerative ranching

Matías Hargreaves-Méndez¹, Ethan Gordon², María José Hötzel¹, Hannah Gosnell²

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An integral approach was first developed to understanding real-world phenomena from four quadrants that contemplate everything about a subject. These quadrants gather human experiences, behaviors, culture, and systems. Regenerative ranching, understood as a livestock production that aims to restore ecosystems functions in vast extensions of land, shows some evidence reflecting strong rancher-nature connections. However, there is a knowledge gap on connections between ranchers and animals. This study explored rancheranimal relations in regenerative ranching and discussed implications for animal welfare. We conducted semi-structured interviews with 16 regenerative ranchers and 10 key informants (i.e., regenerative movement opinion leaders, including academics, NGO founders, training institutions) in the Pacific Northwest area of the US. We got ethical approval from Oregon State University's IRB; Protocol number: HE-2023-225. Using NVivo 13 (R1), we conducted a thematic analysis and created specific themes for the four quadrants of integral theory. The interviewees' main experiences were the instrumentalization of cattle and speciesism favoring horses and dogs. Practices like slaughtering required to be conducted with an attitude of respect for the animals, branding, and castration with a sense of professionalism, and lowstress animal handling techniques were used to move the cattle. Dehorning, calving, and weaning lacked a consistent narrative about how to conduct them. There was some intention amongst the interviewees to modify or avoid practices that inflict pain or stress on animals. However, some practices were perceived as unavoidable due to cultural and systemic barriers. Cultural barriers included branding traditions and peer pressure. Systemic barriers included branding laws and beef industry requirements. Rancher-animal relations could be improved around practices that inflict animal pain and stress. Methodologies widely used by regenerative ranchers like holistic decision-making lack explicit statements of animal's moral

considerations and how regenerative practices should be conducted to ensure good animal welfare. Regenerative ranchers are already leaders in ecosystem stewardship. These findings suggest they could also become role models in rancher-animal relations by promoting explicit consideration of animal sentience and highlighting the interconnections between animal welfare, human welfare, and environment conservation. By identifying these interconnections, regenerative ranchers can push cultural and systemic boundaries to show the importance of animal welfare to be truly regenerative. Adequate animal welfare standards in regenerative ranching would include identifying which practices should be revised. It would also anticipate potential public scrutiny regarding animal welfare.

Can I touch you? The effect of choice on dog behaviour during human-dog interactions

Amir Sarrafchi^{1,2}, Natassja de Zwaan^{1,2}, Maya Tucker¹, Katrina Merkies^{1,2}

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Choice and consent are important aspects of welfare, particularly during human-animal interactions. Dogs involved in therapeutic settings commonly experience human touch interactions where they lack control over the situation. The current study investigated the effect of forced and free touch treatments on stress-related behaviours and avoidance/approach tendencies toward participants/owners in therapy dogs during humandog interactions. A total of 18 therapy dogs (9 neutered males, 9 spayed females) handled by their owners and 44 humans participated in the study. Each human interacted individually with up to four therapy dogs and each dog participated in both forced and free touch treatments for 3min. During the forced touch treatment, the dog was on a leash held by the owner and the participant was instructed to touch the dog continuously. During the free touch treatment, the dog was loose in the pen and the participant, sitting in the center, was instructed to touch only if the dog came within arm's reach. Testing sessions were video recorded for retrospective behavioural coding and analyzed using a GLMM for repeated measures. Lip-licking (P=.5938), panting (P=.4090) and yawning (P=.8619) behaviours did not differ between treatments. Avoidance behaviours (moving/leaning away from participant) were more frequent during free touch (3.0\mathbb{2}0.31/3\min) than forced touch (0.4\mathbb{2}0.33/3\min) sessions (P<.0001). Conversely, dogs displayed more approach behaviours (actively engaged with the participant) during forced touch (17.620.04/3min) compared to free touch sessions (14.520.04/3min; P<.0001). Male dogs (2.720.40/3min) displayed a higher frequency of avoidance than female dogs (0.500.39/3min; P=.0032) regardless of treatment although there was no influence on approach behaviour toward participants (P=.3960). Treatment did not impact the frequency of owner interaction (dog focused on the owner) however male dogs (2.520.22/3min) tended to show a higher frequency of interaction with owners compared to female dogs (1.420.23/3min; P=.0604). The results demonstrate more avoidance behaviour in free touch interactions particularly in male dogs and more approach behaviour in forced touch

sessions. This highlights the therapy dogs' special training to stay calm and tolerate being touched by unfamiliar humans while they lack control over the situation. The findings emphasize the importance of choice/consent during human-dog interactions which should be considered by program facilitators to promote the welfare of therapy dogs.

Consistent individual differences in behavior among beef cattle in handling contexts and social-feed preference testing

Kristina Horback¹, Maggie Creamer¹

Consistent individual differences (CIDs) in animal behavior, also known as personality, refers to the relative stability of an individual's response to similar stimuli over time and across various contexts. For example, an animal may consistently be more active than others of the same age or sex during feeding, social interactions, or, environmental exploration. Depending on the species, knowledge of such CIDs could be predictive of immunity strength, reproductive success, and/or foraging patterns. These metrics are very important for selection practices among breeding livestock populations, such as beef cows. In order to identify cross-contextual consistency in behavior, 50 Angus x Hereford cows were observed across three contexts: interactions at a handling chute, choice in a social-feed tradeoff test, and response in a novel object test. All tests were repeated one year later in order to evaluate temporal consistency. Cows were consistent in time to traverse the chute, cross the squeeze mechanism, and exit the squeeze in the handling context, as well as in their latency to approach supplement bucket in social-feed tradeoff test and in the novel object test. Predictive models provided evidence that less active and less excitable cattle in handling context are more feed-centric in socialfeed tradeoff assays. Cows that were less active in the chute, slower to traverse the squeeze, and slower upon exiting the were more likely to choose supplement over conspecifics in the social-feed tradeoff assay, and vice versa, more active and excitable cows in the chute context were less likely to choose supplement over being in proximity to herd mates. access to conspecifics. We did not find a relationship between any component scores and behaviors in the novel approach assay. Cows that were spending the least amount of time in an isolated context (shorter duration in the chute, squeeze and exit) were choosing conspecifics in the tradeoff assay, which distinguishes cattle that had a more active response to isolation. object test and the other contexts. No relationship between the behaviors displayed in the novel object test were related the other contexts. Given the consistency of individual behavioral response within and between the years, it appears that breeding Angus x Hereford cows that display more retain active response to isolation during handling contexts could also have a

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higher motivation to be in proximity to herd mates. This could predict behaviors displayed when grazing on pasture or extensive rangeland, as choice to be near the herd could influence grazing patterns.

Zero abandonment project: use of play as a methodology to teach positive human-animal relationship

<u>Ana Vitória Furtado de Faveri</u>¹, Márcia Inês Grapéggia Zanella¹, Germana Vizzotto Osowski¹, Érika Lage de Macedo¹, Ana Paula Tarozo¹, Isabela Zanetti de Campos¹, Marina Batista de Sousa¹, Adroaldo José Zanella¹

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The Ministry of Health classifies the school as a facilitator in the exchange of knowledge and change in behavior, and teaching children helps in the perpetuation of knowledge, as they transmit what they learned to friends and family. Furthermore, new knowledge helps children to develop critical thinking, as well as respect for life. Therefore, the aim of the study was to evaluate whether play can act as a teaching methodology for complex topics, allowing the child to reach a level of learning that makes them a propagator of knowledge. We taught children in the 3rd year of Elementary School at a public school in the city Pirassununga, SP and evaluated the impact that the child's education has on their parents' knowledge. The methodology included games about the Animal Welfare Five Freedoms, animated videos about zoonoses, and the use of images and hands on games and crafts developing activities relevant for pets' behavior and body language. Despite the fact that the study involved 50 children, the statistical analysis was only possible for the parents of 16 students, as they were the only ones who had complete documentation with pre- and post-intervention questionnaires. The statistics included descriptive analysis and Chi-square test animal welfare, sentience, duty of care and zoonoses. No statistical differences were identified in the parents, when contrasting their knowledge in animal welfare, sentience and duty of care, before and after the intervention with their children (p > 0.05), indicating that the intervention did not reach its objective. However, when observing the descriptive analysis, we identified that there was prior knowledge on the part of the parents on the three topics (87.5%; 93.7% and 75% knew the themes and the numbers changed to 93.7%, 100% and 93.7% after the intervention, respectively). Surprisingly, regarding zoonoses, the p-value was significant (0.004), when contrasting the data collected from parents before and after the interventions. The descriptive analysis showed that this was the topic on which parents had the least prior knowledge (50%

pre-intervention and 100% post-intervention). Therefore, although the data and conclusion need to be understood with caution, due to the small sample size, this study raised the possibility of developing future research with a larger sample group, as the data left positive evidence that knowledge transfer can occur between children and their parents on animal related education strategies, based on play.

Assessing the impact of repeated exposure and movement on the fear response in broiler chickens

<u>Bethany Baker-Cook</u>¹, Alexandra Jackson¹, Marcela Quino¹, Anusha Gautam¹, Katie Still¹, Melissa Gilpin¹, Denise Landers¹

Fear tests are a common method used in research to assess the affective state of an animal. This study aimed to 1) understand the impact of repeated exposure to fear tests on fear response and 2) assess how the addition of movement to fear stimuli during novel object test (NO) impacts bird fear response. Over two trials, a total of 3,600 Ross 308 birds (1800 birds/trial) were raised until 42 days of age. At 23d, 30d, and 37d, three fear tests were performed in each pen, NO, human approach (HA), and observer response (OR) tests. NO was split into three movement treatments, traditional NO (TNO) without movement, intermittent NO (INO), and continuous NO (CNO). In INO, the NO was moved for 30s, then stationary for 30s, repeated for a duration of 180s. In CNO, the NO was continuously moved through the pen. OR test was performed before and after the other tests occurred. Data for NO and HA was analyzed as a one-way ANOVA for the main effects of treatment or age (PROC GLIMMIX with Poison Distribution), with pen as experimental unit. OR data was analyzed by paired Ttest (PROC TTest). Fear response decreased after multiple tests within the same day, with OR results before testing (66%) differing from after (42%, P<0.01). Fear response also decreased with repeated exposure across life. Latency to HA was longest on 23d (162s), then 30d (110s), and shortest on 37d (80s; P<0.01). Similarly, with NO the latency to interaction was longest on 23d (105s), then 30d (49s), and shortest on 37d (12s; P<0.01). For both HA and NO, at all-time points, the number of birds interested in the human or NO was higher on 37d than 23d (P<0.01). The addition of movement to NO increased the fear response. CNO had a longer latency to interaction than TNO (75s v 36s; P<0.01). The number of birds around NO was higher for CNO than TNO at all time points (P<0.01). Overall, the addition of movement to the NO added an additional dimension to the fear stimuli and increased fear response. The repeated exposure of birds to fear tests reduced the fear response, both when birds were repeatedly exposed to several different fear tests on a single day and when repeatedly to the

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same fear test multiple times across their lifetime. This impact of repeated exposure is important to recognize when designing experiments that utilize fear tests.

Session 9: Applied Technologies to Assess Animal Behaviour

Wednesday, July 24th, 13:45 – 14:00

Sound analysis during the lifespan of broiler chickens

<u>Patricia Sostser</u>¹, Tomasz Grzywalski², Pieter Thomas², Camila Lopes¹, Imad Khan¹, Bassem Khalfi¹, Kobe Buyse³, Dick Bottleldooren², Paul Devos², Maarten de Gussem⁴, Frank Tuyttens³, Ghunter Antonissen¹

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At least four distinct vocalizations of broiler chickens have been described: 1) distress calls (DC), characterized by a declining frequency with high energy; 2) short peeps (SP), featuring a declining frequency, low energy, and short duration; 3) warble notes (WN), which consist of repetitive bow-type elements with low energy; and 4) pleasure notes (PN), characterized by an increasing frequency and low energy. The objective of this study was 1) to develop and validate a broiler vocalization detector based on a fully-convolutional neural network, 2) to employ sound analysis to automatically identify four vocalizations in broiler chickens to understand patterns of vocalizations from 1 to 42 days in normal and in heat stress conditions. Any vocalization not falling into these categories was categorized as "other" (O). The present work was approved by the ethical committee of ILVO. The experiment involved four rounds, each with 560 male Ross 308 chickens, totalizing 2240 birds. The stable was divided into two compartments, each with climate control capabilities. Each compartment contained two pens of 36 m². One compartment remained in the thermoneutral zone, while the other underwent heat stress (HS) for ten days in the last two weeks. HS involved increasing the temperature from 22°C to 32°C for six hours. Vocalizations were identified through manual labeling. The broiler vocalization recognizer achieved a balanced accuracy of 87.9% with limited confusion. In the first day, PN were recorded for 4 hours, across the 24-hour periods, rapidly declining thereafter to approximately 0.5 hours by day 6 and 6 minutes after day 20. DC, initially noted at less than 1 hour per day, increased after day 12 of the experiment. SP peak between days 4 and 7, averaging between 10 to 15 hours per day, before gradually decreasing to 1 to 2 hours per day by the end of each round. WN appeared infrequent and were primarily observed in

young broilers with a minor peak of approximately 6 minutes per day around day 3, diminishing to less than 1 minute per day post day 10. O vocalizations increased in the last three weeks, suggesting that as broilers age, their vocalization repertoire expands. Currently, interactions with HS are still being analyzed. The developed method is sensitive and capable of detecting the four vocalizations in broiler chickens across different age stages. The next step of the present work is to perform a clusterization to further analyze O types of broiler vocalizations.

Computer vision benefits ethology by automating data collection and moving focus to analyses

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This study integrates computer vision into the workflow of behavioural observations. Traditional observations, which provide a rich contextual understanding, remain timeconsuming and variable across individual observers and over time. Computer vision has proven its accuracy, efficiency and consistency in identifying specific animal behaviours, yet its adoption in behavioural studies is limited. We propose to use pose estimation to pre-select video sequences for further observations, thereby reducing the viewing time required by observers. We illustrate this approach by detecting sequences where cows are likely to be using a mechanical brush after approaching it with their muzzle. We compare the reduction in viewing time against the potential information loss. One hour of video was recorded from a top-down view over a mechanical brush. A human observer annotated when cows were present in the vicinity of the brush and when they displayed the behaviour. The same recording was processed frame by frame with the Sony Multi-View System to produce object detection and pose estimation. In the initial filtering step, object detection identified 36:25 minutes of video with cows in the area of interest. There was a deviation of 20 seconds (0.9%) compared with human observations, stemming from differences in defining the area of interest. This means that, based on the absence of detected animals, a third of the video material could be confidently filtered out from further human observations. In a subsequent step, intersection between cow and brush bounding boxes and proximity of the head keypoint to the brush identified 25:06 minutes where a cow was likely interacting with the brush. Comparing to human observations, 5:19 minutes of these did not contain brush interactions. For 18 seconds of observed brush interaction, proximity was not detected. Using this method to identify sequences of interest to subsequent behavioural observations, the time without brush interaction merely represents additional viewing time, while the missed interactions

represent a loss of information of 1.5%. Although the method needs to be validated on more diverse video material, the results show that computer vision reliably detects sequences in which a behaviour is likely happening. This implies that using available pose estimation software and relatively accessible numerical methods to filter its output, observers can limit the video to be annotated to sequences of interest, with minimal information loss. Detecting the behaviour per-se requires fine tuning how the model output is translated into a behaviour, highlighting where human expertise is essential.

Exploring opportunities for automatic monitoring of laying hen vocalisations

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Laying hens are vocal animals and their vocalisations can inform us about their affective state. Both the type of vocalisation and the characteristics of the vocalisation, such as pitch frequency and duration, can inform on the specific emotional state and the intensity of the emotion. In human babies, similar approaches are taken to improve our interpretation of the sounds the babies produced. From that field, tools are available that may also be useful to detect and analyse different types of vocalisations in animals. Therefore, the aim of this pilot study was to explore the opportunities for automatic monitoring of laying hen vocalisations. To meet that aim, vocalisations of four groups of 10 birds were recorded with a Reolink security camera with microphone and analysed at three different ages: 2, 10 and 22 weeks of age. Two pens contained ISA Brown birds and two pens contained Dekalb White birds. Birds were housed in floor pens with litter on the floor, a perch, a raised platform and ad libitum feed and water. From each pen and age, 30 minutes of data (from 12:00-12:30 CET) were analysed with the Praat software package, developed for analysis of human vocalisations. Praat is able to select parts of the recording where vocalisations are detected and allows the comparison of call characteristics based on the sonogram. The pitch frequencies of 40 contact calls per age were compared statistically using anova, with age as fixed factor. The percentage of time spent vocalising decreased with age: at 2 weeks of age, vocalisations were detected during 79% of the time, compared with only 7 and 29% of the time at 10 and 22 weeks of age (overall effect F=579.29; P<0.05). This decrease is also related with the development of the vocalisation pattern from chick to pullet, to adult hen. There was no significant difference between brown and white birds. The mean pitch frequency of the contact calls dropped from 3508 and 3005 Hz at 2 and 10 weeks of age to 1774 Hz at 22 weeks of age (F=76.99; P<0.05). This pilot study indicates that the Praat software can be used to detect laying hen vocalisations and to analyse call characteristics, such as pitch frequency. This can be used to start a larger study where automatic analysis of vocalisation patterns is compared with other welfare indicators.

Exploring pose estimation as a tool for the assessment of brush use patterns in dairy cows

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Understanding brush use patterns provides useful welfare insights. Computer vision can potentially sustain large scale studies and provide continuous data with high granularity on brush interactions. Therefore, the objective of this study was to explore if pose estimation can be used to assess brush use patterns in dairy cows. The exploratory study was conducted at the Swedish Livestock Research Centre. 55 Cows were kept in a loose housing system with an automatic milking system, with access to two mechanical rotating brushes (DeLaval swinging cow brush SCB). Six hours of video was recorded simultaneously from two cameras, monitoring one brush located at a passage (5.28*2.43m) between cubicle rows. These included a top-down view (4.5 m) and a side view (3 m), referred to below as 1, and 2 respectively. A human observer annotated the full six hours of video on a second basis for: 1) number of cows in the area, 2) brushing, and 3) area of brushing defined as cranially of the poll, caudally of the tail head or between either two of successive anatomical landmarks: poll, neck, withers, T13, sacrum and tail head. The exploratory analysis using computer vision included a six-step approach: 1) object detection of cows and brush, 2) pose estimation of aforementioned anatomical landmarks, 3) discarding sequences of volatile brush movement, 4) calculating relative distance from brush bounding box centre to key points, 5) expressing the coordinate of the brush centre to the axis formed by the key points of the cow, and 6) comparing the estimated brush position on the body against that annotated. Comparisons between the human observer and machine were limited to sequences containing one cow in the passage. A total of 66 brush interactions were compared. Cohen's kappa was calculated to assess agreement between human observation and predicted brush use, for camera views 1 and 2. Cow detection was limited to the top-down view only (k1=0.87). For brush use (k1=

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0.48, κ 2=0.36) and anatomical area of brushing (κ 1=0.24, κ 2=0.54), all cameras were assessed. In conclusion, we explored pose estimation to pinpoint where the brush is used by a dairy cow. The preliminary results indicate that, amongst others, the following areas need to be addressed: data filtering, relating predictions to human annotations, understanding the link between anatomical landmarks and brush use, and differentiating static from volatile brush movements. Additionally, a 3D reconstruction approach could offer a good complement, and will therefore be explored.

Tag 'n' Track: facilitating validation of animal behaviour tracking technologies with automated annotation

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Recent technological developments offer new opportunities to explore animal behaviour. However, the validation of these developments, although essential, is often neglected due to the intensive nature of the labour, resources, and time it requires. Our study tackled the challenges associated with manual annotation in validating animal-tracking technologies. Specifically, we detail the implementation and demonstrate the effectiveness of an automated annotation method that utilises 3D ArUco markers. Based on an established, open-source computer vision technology similar to QR codes, the markers have a grid design and come with a unique ID. The 3D ArUco markers were attached to vests, fitted onto 21 chickens, to facilitate the validation of technologies for monitoring visits to indoor litter and perch areas in a research context. To assess the effectiveness of adapting this computer vision tool for automated tracking of individual animals, we used 70 10-minute video sequences recorded to capture individuals' presence within indoor litter and perch areas. The validation involved comparing the traditional manual annotations, made by two trained human observers considered here as the gold standard, with the automated annotations. Our automatic tracking method generates annotated videos, revealing the ID of subjects present in the regions of interest, as well as the start and end timestamps of their presence. Compared with conventional human observation, our method demonstrated excellent performance: high correlation (ρ =0.95, p<0.01), sensitivity of 94.37%, specificity of 99.90%, accuracy of 98.71%, precision of 98.80% and F1 score of 96.45%. In addition, our method, with the capabilities of the computer used, enabled the 70 10-minute video sequences to be annotated automatically in 13.05 hours, a time saving of 83.41% compared with the 78.67 hours that would have been needed for continuous on-screen observation by humans for manual annotation. The proposed method can complement or replace manual annotation, thereby simplifying the

validation process of new technological solutions (e.g., RFID, accelerometer) and encouraging adoption of these to advance animal behaviour science.

The development of a computer system to assess lameness in sows using dorsal view

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In commercial pig farms, 8% to 65% of animals suffer from lameness, compromising animal welfare and production results. The diagnosis of lameness is often subjective, as it is made through observation and depends on the training and experience of the observer. However, it is possible to accurately monitor, interpret and recognize images using computer vision. Computer vision uses kinematic analysis to assess pig locomotion, due to its ability to characterize and identify information about movement. The aim of this project was to develop a computer vision model to identify and track locomotion poses in sows in dorsal vision, through images using deep learning. The data were acquired on a commercial pig farm. Two RGB cameras captured 2D images corresponding to 565 lateral and 642 dorsal videos of moving sows. However, 40% of the videos were not used due to issues during filming. A total of 364 lateral and 336 dorsal videos were converted to the MP4 format. These lateral videos were evaluated by thirteen experts to identify the animals' locomotion scores using Zinpro's Swine Locomotion Scoring system allocating scores ranging from 0 (no signs of lameness) to 3 (severe lameness). The animals' skeletons were also defined by a set of keypoints marked on certain parts of the animal's body in each frame of the 106 videos used to develop the models. It was used the SLEAP (Social LEAP Estimates Animal Poses) software as a tool to label the keypoints and to estimate the pose, test, and train models. The skeleton with 10 keypoints in dorsal view and LEAP architecture showed a high accuracy in keypoint identification (Mean Average Precision (mAP) 0.72), a similarity between keypoints of 0.86 and mean Euclidean distance between labelled and predicted keypoints of 11.37 pixels. Based on the kinematic data of the combinations of angles and distances of the 10 keypoints, Weka software was used

as a tool to select attributes (characteristics to describe the data), test and train deep neural network models, using the Wrapper and GreedyStepwise methods. The Multilayer Perceptron algorithm was 68.18% accurate in identifying locomotion scores 0 to 2. Score 3 was difficult for the model to identify, so the algorithm developed to identify scores 0 to 3 had an accuracy of 59.09%. The model showed promise for developing a system to identify lameness in sows automatically.

Session 10: Integrating Animal Welfare & Perception of Welfare and Ethology

Wednesday, July 24th, 13:45 – 14:00

Stakeholder perceptions of animal welfare as a component of sustainable beef programs in the United States

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Cattle welfare is a critical component of sustainable beef production systems. This study aimed to understand how stakeholders in the United States beef industry incorporate animal welfare into their sustainability programs. A survey was administered online to the United States Roundtable for Sustainable Beef primary membership contact list serv (n = 142) from September to October 2023. The survey included multiple-choice, open-ended, and Likert scale questions about animal welfare inclusion, definitions, and indicators used in their companies' sustainability programming. A total of 27 surveys were included in the final analysis. Researchers performed descriptive statistics and thematic analysis. The majority of respondents were producers (n = 13, 48%) and the primary role represented was owner (n = 8, 30%). The majority of respondents (n = 24, 89%) agreed that animal welfare was a critical component of sustainability. Nearly all respondents (n = 26, 96%) had sustainability programs incorporating animal welfare. The most common factors selected influencing the incorporation of animal welfare into sustainability programs were cattle health (n = 20, 74%), cattle performance (n = 12, 44%), and consumer perceptions (n = 12, 44%). When asked how often they reported (e.g., internal report or assessment, external reporting to customer) animal welfare indicators per year, approximately half of the respondents (n = 10/19, 53%) indicated that metrics were reported once per year. Over half of the respondents (n = 16, 59%) believed that emphasizing one pillar of sustainability (i.e., environmental, social, and economic) could negatively impact either of the other two. However, almost all believed that animal welfare positively impacts environmental (n = 25/26, 96%), economic (n = 25/26, 96%), and social (n = 26/26, 100%) sustainability. Thematic analysis of 5 free response questions identified ten themes: Animal Care, Regulations and Guidelines, Responsibility, Consumers and Stakeholders, Performance and Efficiency, Financial Impact, Connectedness, Key

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Component, Animal-based Outcomes, and Employees. When asked how they define animal welfare, the most common themes in responses were Animal Care and Regulations and Guidelines. Findings suggest a widespread recognition of animal welfare's importance within sustainable beef production. The interconnectedness of the pillars of sustainability highlights the need for balanced approaches to integrating sustainable animal welfare solutions in beef systems.

Elementary school students' perception of dog behavior and welfare

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According to data from the Brazilian Institute of Geography and Statistics (IBGE), the country is home to 54.2 million pet dogs, while according to other sources, 20 million are abandoned. Among the main reasons for abandonment, behavioral and aggressive problems stand out and children appear to be the primary victims of dog attacks. The objective of the present work was to analyze the students' prior knowledge regarding the welfare and behavior of dogs and then raise awareness about these topics. The study was conducted in a Brazilian public school, with 31 students aged 13, 14, and 15 years old. Data were collected through questionnaires, including dog images in different states. Data from descriptive analysis demonstrates that the majority of students have companion animals (70.9%), with dogs being the most common (41.9%), followed by cats (29%). Additionally, students classified their animals as extremely important to them (54.8%), and 80% stated they could identify when their animals were happy, angry, or scared. Within this population, 77.4% of students have experienced accidents with animals, including bites and scratches. The frequency of accidents with animals is higher (66.6%) among individuals who have animals at home, suggesting that most accidents occur in a familiar environment. There was no difference between the gender of the students in the frequency of accidents. Regarding knowledge about animal welfare, 90.3% of students were already familiar with the topic, and 95% stated that animals have feelings. However, 70% admitted not being aware of animal sentience, and 100% of students emphasized the importance of respecting animals' rights. Regarding the interpretation of the dog images, students answered that it was extremely (19.3%) and very dangerous (54.8%) to approach the dog when looking at the aggressive behavior image. Considering the image of the dog showing play behavior, students answered that it was just a little dangerous (87.1%), middle dangerous (9.7%), and extremely dangerous (3.2%) to approach the animal. In conclusion, despite students having demonstrated prior knowledge regarding dog behavior

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and welfare, a considerable number of them experienced accidents with these animals, which suggests that efforts to enhance their understanding of recognizing emotions in dogs are essential to decrease accidents.

Feline stakeholder attitudes and opinions on community cat welfare

Nattawipa Ampaiwan¹, Jacquelyn Jacobs¹

Of the approximately 800 million cats living globally, 60% of them are free roaming. Freeroaming cats, or "community cats" as they're often referred to, include both stray (pets abandoned or lost) and feral cats (unsocialized domestic cats). Community cats experience different welfare challenges than owned cats, such as exposure to extreme temperatures, potentially unstable social groups, and nutritional uncertainty, among others. Assessing and monitoring the well-being of community cats is vital to ensuring their overall welfare, but accessible and validated tools are lacking, and the few available focus only on animal health. The ultimate aim of our study is to create a valid and reliable tool to assess welfare in community cats, however, understanding how welfare is currently being measured in the field is an important first step in the development of a more robust tool. Therefore, the objectives of the current study are: 1) identify currently utilized methods for assessing community cat welfare, and 2) evaluate demographic characteristics of participants that might influence stakeholders' perceptions and approaches. Through targeted snowball sampling methods, we are conducting two rounds of surveys using the Delphi technique, a widely used method to investigate agreement among experts. Data collection is ongoing, and round one of survey responses will be analyzed by May, with round two launched in June. As of mid-February, the first survey round had received 154 full responses from feline experts in various related fields. Participants are located throughout the United States and represent 23 states. When asked to identify their role in community cat care, the majority of respondents self-identified as caregivers (34%), volunteers at a TNR organization (32%), shelter staff (21%), and veterinarians (15%). Open-ended questions will be analyzed using qualitative content analysis when data collection in round one is complete. Preliminary analysis of the responses revealed some common themes, mostly related to resource availability and quality (e.g., food, water, shelter) and health (e.g., body condition, neuter and vaccination status). Surprisingly, the ability to express natural behavior and show signs of positive affect were rarely mentioned, even though they are important components of welfare. Results from the first round will be re-

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organized and sent back to participants for comments on the importance and relevance to community cat welfare. This study will provide a better understanding of community cat welfare criteria used among stakeholders, which will help inform future development of an assessment tool that expands upon currently utilized methods.

Investigating factors that influence handling techniques used by veterinary staff in North America on dogs displaying fearful and aggressive behaviors during routine veterinary examinations

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Restrictive restraint (e.g., full-body) during veterinary care is a stressor for dogs. Low-stress handling (LSH) is recommended, where handling techniques are based on evidence-based recommendations and behavioral assessments. Factors that influence LSH use on dogs is unknown; thus, we used a cross-sectional study to examine risk factors associated with minimal and full-body restraint of dogs. An online questionnaire was distributed to veterinary staff (i.e., veterinarians, technicians, and assistants) in Canada and the United States (N=687). Using manual stepwise forward selection, logistic regression models were created to examine associations between participant demographics (e.g., gender, age), veterinary-related factors (e.g., mixed or small animal practice, LSH certification), dog-related factors (e.g., behavioral history, size), well-being (e.g., compassion satisfaction and fatigue), and use of minimal and full-body restraint on dogs showing fearful and aggressive behavior. Univariate analyses were assessed to reduce tested predictors and confounders were statistically assessed and retained, if identified. Respondents were licensed veterinary technicians (35%, 238/680), veterinarians (32%, 219/680), and veterinary assistants (19%, 130/680), and worked in a mixed or small animal practice (84%, 561/665) and emergency clinic (15%, 104/665). When asked to rank factors to consider when conducting veterinary exams, 36% (223/617) of participants ranked staff safety as first most important, 37% (232/627) ranked animal stress second, and 33% (201/614) ranked client satisfaction third. The use of minimal and full-body restraint depended on dog behavior, with reports of minimal restraint decreasing and reports of fullbody restraint increasing, as behavior changed from calm (minimal: 83%, 565/680; full-body: 58%, 393/680) to aggressive (minimal: 52%, 352/675; full-body: 68%, 456/675). Regression results reveal that participants aged 25-44 years are more likely to use minimal restraint on fearful dogs (vs. 18-24; p=0.0135). Higher odds of using minimal restraint on aggressive dogs was associated with identifying as a man (vs. a woman; p<0.01), working at a mixed practice

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(vs. small animal; p<0.01), and having a LSH certification (vs. no certification; p<0.01). Higher odds of using full-body restraint was associated with identifying as a man (vs. woman, p<0.01), and graduating between 2015-2023 from their veterinary program (vs. 2004-2014, p=0.01) for fearful dogs, and working at a mixed animal practice (vs. emergency; p<0.01) for aggressive dogs. Participants reported that owner presence influences whether full-body restraint is used on fearful (p=0.018) and aggressive (p<0.01) dogs. Results suggest that full-body restraint is commonly used on dogs and highlights factors that may prevent adoption of LSH principles for further intervention research.

Management practices and welfare of equids involved in equine-assisted interventions: from practitioners' perception to field evidence

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Animal-assisted interventions (AAI) are based on activities involving a patient, a professional and an animal, equids being second after dogs. There are almost no studies on the chronic effects of the activity on the animals involved. EAI (equine-assisted interventions) equids are also submitted to management conditions that influence their welfare similarly as for other working equids. Life and working conditions depend on human decisions, that are influenced by equestrian cultures which shape the stakeholders' perception of equid welfare and appropriate management. We performed two studies in order to (1) gather information, through an on-line survey (51 responses analysed) for managers of facilities where EAI or conventional riding lessons (RS) were predominant; (2) obtain direct information, through an observational study (8 facilities, 174 equids: 25 EAI, 89 EAI-RS, 60 RS) on management and working practices and their consequences on equids' welfare. Welfare assessment was performed in the horse's home environment using validated behavioural, postural and health indicators. Statistical analyses were performed using R software using the catdes function (FactoMineR package) from the data for each individual. The results showed differences in managers' perception of good management practices that were related to the facility's involvement in EAI, with an increased awareness of equids' needs in terms of housing and feeding conditions when EAI was the main activity. Managers of EAI facilities that did not offer riding school lessons reported feeding their equids with more hay (p=0.05) than managers of EAI facilities proposing also riding school lessons. Observations confirmed that facilities more involved in EAI housed their equids mostly in groups outdoors with roughage available at all time. There were correlates between management decisions and welfare consequences for the equids: the higher the proportion of equids with mixed activities (EAI-RS equids), the higher the proportion of equids presenting stereotypic behaviors (rho=0.92; p=0.001), which was not the case for the proportion of RS equids despite the same management type. There were more EAI facilities practicing groundwork and bitless work, two parameters that seemed

to influence the equids' reactions to an unfamiliar human and to riding equipment (ex: proportion of bitted horses/ number of negative reactions at saddle test: rho=0.73; p=0.038). Given the particularities of EAI patients, conventional working modalities may be more at risk of increased discomfort for equids (ex: actions on bit by unbalanced/disabled riders). Overall, the survey and observational approaches converged, suggesting that observational studies on a larger number of facilities, allowing more multifactorial statistics, would be very promising.

Using the One Welfare framework and mixed methods to understand connections between dairy farmer, cattle, and wildlife welfare in Canada

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This study used qualitative and quantitative methods to explore how farm management, technology, and farmer well-being are interrelated with the management, health, and welfare of dairy and wild animals. Dairy farmers (n=86) on farms in British Columbia, Alberta, Saskatchewan, Manitoba, and Ontario completed an online survey regarding farm management, work-life balance, social support, financial concerns, animal health, perceptions of wildlife, and mental health using validated psychometric scales. On a subset of farms (n=66), we scored lameness, body condition, and hock/knee/neck lesions (30% of lactating cows/herd to a maximum of 69 cows). We conducted univariable linear and multiple factorial analyses (MFA) followed by hierarchical and k-means clustering methods. Farmers with <10% mastitis incidence had lower stress scores compared to those with ≥10% (P=0.02). Compared to <5% clinical lameness (≥3 on 5-pt scale in loose housing, ≥2 of 4 indicators in tie-stall), having ≥5% prevalence tended to be associated with lower stress (P=0.07) and anxiety scores (P=0.06), and higher resilience scores (P=0.10). Farmers who rated mice as more harmful/stressful reported higher stress (P=0.007) and tended to be less resilient (P=0.07) than those who rated mice as neutral. The MFA yielded 4 groups of farmers based on their role, work-life balance, concerns about finances, and how valued they feel by consumers. The cluster characterized by greater concerns about farm profitability, animal activists, and public perception also had the highest stress and depression scores. Semi-structured interview transcripts (n=30) were analyzed using an approach informed by reflexive and codebook thematic analysis. Common themes identified included the pressure and complexity of caring for livestock, which also gave them feelings of fulfilment, illustrating a paradoxical relationship of caretaking. Farmers identified their capacity to care for their herd to be influenced by climate-related feed concerns, the demanding nature of dairy farming, entanglement of work

and life, labour challenges, financial stress, and sociopolitical and sociocultural challenges. Our qualitative results highlight the entanglement of dairy farmer well-being, animal care, and the environment, and the need for resources with (agri)cultural competency to support farmers. However, there were limited and some unexpected quantitative associations between farmer and animal well-being, which could be due to the low prevalence of disease on participating farms as well as the highly stressful economic climate at present. By using mixed methods and the One Welfare framework, we gain a more holistic understanding of the stressors and challenges experienced on-farm to better support farmers and their animals.

Session 11: Animal Emotion and Cognition

Thursday, July 25th, 10:45-11:00

Evaluating fearfulness in three genetic strains of cage-free laying hens using a startle test

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Laying hens can experience unpredictable events in cage-free environments, which may cause shifts in behavior and fearfulness. Research using novel object and tonic immobility tests indicates that genetic strain influences the stress response. Our aim was to develop a startle test to compare behavioral and physiological responses in three genetic strains of laying hens. H&N White, Hy-Line Brown, and Bovan Brown laying hens were housed in 3.2 x 1.2m cagefree floor pens (25 hens/pen; 3 pens/strain). At 22 weeks of age, a startle test was performed on 30 hens/strain. Hens were habituated individually to a 16ft3 chamber containing a feeder with mealworms for 5 min/hen for three consecutive days. On testing day, hens were placed into the same chamber along with a portable speaker. After 1 minute, an auditory startle stimulus (450 Hz, 1 sec) was played twice consecutively and behavior was video recorded for 2 min. Temperature changes were assessed in a subsample of hens (5 hens/strain) with thermal images and ingested core body temperature (CBT) sensors before startle, at startle, and for 5 min after startle. Thermal images were taken every 30 sec to assess maximum comb, maximum head, and average eye temperature with FLIR software. CBT sensors collected data every 1 min. Behavior data (latency to eat, frequency of head bobs for the first 30 sec, and the frequency of vocalizations, pecking, and stepping over 2 min) were coded in BORIS software. Data were analyzed with linear mixed effects and generalized linear mixed effects models in R software. H&N White hens performed more head bobs than either Bovan Brown or Hy-Line Brown hens (p=0.006). H&N White hens tended to be more likely to resume eating post-startle than the brown strains (p=0.08). However, of the hens that resumed eating, there were no significant differences in latency to eat between strains (p>0.05). Strains did not differ in the number of vocalizations, pecking, or stepping (p>0.05). H&N White hens had significantly higher comb temperatures from 1 to 3 min post-startle (p=0.001) and significantly higher eye temperatures at 2, 3, and 4 min post-startle (p=0.02) than Bovan or Hy-Line Brown hens. There

were no significant differences between strain, time of test, or their interaction for head temperature or CBT (p>0.05). H&N White hens were more behaviorally and physiologically reactive to the startle stressor than both brown strains, which may have implications for how they cope with unpredictable stressors in cage-free environments.

Fear, frustration and motivation to explore in broiler breeder cockerels under qualitative feed restriction

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Broiler breeders regularly experience hunger and frustration stemming from unfulfilled behavioural needs for feeding. This is largely due to quantitative feed restriction which is common practice in the management of these birds, particularly in the rearing period. As an alternative, qualitative feed restriction, the reduction of feed nutrient content by adding poornutritious diluents to standard feed, allows a larger portion of feed to be provided without increasing the caloric intake. While several studies have assessed the effects of qualitative feeding on female broiler breeders, no such assessment has been done on broiler breeder cockerels. The aim of this study was to investigate the effects of feed dilution and roughage on the level of fear and motivation to explore of broiler breeder cockerels. 200 Ross 308 broiler breeder cockerels (5 to 10 weeks of age) were housed in 12 pens (6 pens/treatment), 17 birds/pen. The treatments were standard feed (Control) and feed diluted with 20% insoluble oat hulls and 150g of alfalfa roughage daily/pen (D+R). The D+R birds received 20% more feed per day. Novel object (NO) and frustration during thwarted feeding tests were performed in the home pen and the response recorded with video cameras. In addition, four birds from each pen were subjected to a tonic immobility test (TI). All behavioural tests were performed once a week at 6, 8 and 10 weeks of age. We expected the Control birds to be more frustrated and stressed, resulting in more time in TI and pecking at the feed container, and more motivated to explore the NO, keeping a shorter distance to it. The control birds showed a tendency to approach the NO faster than the D+R birds (P = 0.07) and were more likely to approach at 10 weeks of age (P = 0.006). In the frustration test, D+R birds spent less time pecking the feed container (P = 0.049), more time standing (P = 0.01) and tended to have fewer behavioural transitions (P = 0.09) than control birds, which indicates a reduction in frustration levels. In addition, Control birds stayed in TI longer than D+R birds (160.7 ± 15.5s and $98.1 \pm 12.s$, respectively. P = 0.005). The results show that the combination of feed dilution

and daily roughage can have positive effects on the welfare of broiler breeder cockerels by reducing the sensation of hunger as indicated by fear, frustration, and motivation to explore.

Fearfulness of individual Icelandic horses vary with the nature of the feareliciting stimulus — an investigation of behavioural reaction and heart rate

Johanna Stenfelt¹, Rebecca Grut¹, Vincent Bombail², Hanna Sassner¹, Maria Vilain Rørvang¹

The Icelandic horse is known for its friendly, calm and docile nature. Despite its popularity in riding, the Icelandic horse is underrepresented in the research literature, which mainly includes studies on warmblood horses. Previous research has indicated that Icelandic horses show lower behavioural reactions while still having strong physiological responses to feareliciting stimuli, unlike warmbloods. As such, more general research is needed on other breeds, especially those commonly used in leisure horse riding. As part of a larger study examining the effect of olfactory conditioning on the stress responses of horses during different aspects of handling, we aimed to investigate differences in emotional reactivity of Icelandic horses across different contexts. Forty-two privately owned Icelandic horses (30 geldings, 12 mares) of varying ages (range: 4-27 years, median: 14 years) from Sweden participated in the study. The study included a reactivity test battery aimed at assessing three different aspects of fearfulness in horses; fear of humans (HUM) in a forced human approach test with a novel person, fear of novelty (NOV) in a novel surface test where the horse was led over a white tarp, and fear of suddenness (SUD) in a startling test using a plastic bag tied to a rope pulled at ground level. The study measured behavioural reactions indicative of fear on ordinal scales (0-3) and heart rate (HR, mean and maximum) using the Polar Equine system during each of the three tests. Analyses with Spearman's rank correlation coefficient found a moderate positive correlation between behavioural reaction and maximum HR in SUD (rs=0.43, p=0.01), and a marginally significant correlation in NOV (rs=0.31, p=0.06), but not in HUM. Also, a marginally significant correlation between behavioural reactions was found for NOV and SUD (rs=0.31, p=0.06), but not for HUM. A mixed-effects linear regression analysing repeated measures on each horse found no effect of sex or age on maximum HR. Our results suggest that Icelandic horses may exhibit lower behavioural reactivity to fear-eliciting stimuli despite having strong physiological responses, especially during handling. This emphasises the

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importance of considering both aspects when evaluating fearfulness in this breed. Our findings further indicate that individual fear responses may differ depending on the nature of the fear-eliciting stimuli. We also discuss the limitations of this test battery and the adaptation of each test to the Icelandic breed, offering insights into considerations for optimising test batteries aimed at measuring temperament traits in the field.

Analysis of pig vocalizations using musical information extraction techniques to design musical stimuli

Natalia Álvarez-Hernández¹, Berardo de Jesús Rodríguez¹, Edimer David Jaramillo¹

Pig vocalizations can be categorized into different emission contexts, which can be grouped into three main types of emotional states: negative, positive, or neutral. Pig vocalizations have been utilized as welfare indicators on farms and during transportation through non-invasive methods. However, pig vocalizations have not been used for the creation of musical stimuli, which could serve as environmental enrichment on farms. This research aimed to propose a new type of analysis for extracting musical features from pig vocalizations, intending to create music that can be adapted to the needs of animals. The analysis of vocalizations was conducted using the Music Information Retrieval (MIR) extraction technique to evaluate acoustic parameters: Centroid, Spectral Deviation, High-Frequency Content, Amplitude, Dissonance, and Zero Crossing. The acoustic parameters were extracted using Sonic Visualizer version 4.5.2. In total, eight contexts (castration, crushing, holding, before and after nursing, huddling, isolation, reunion) each with two vocalizations, were grouped as negative, positive, or neutral. The recordings (original vocalizations) came from male and female piglets aged between 6 and 15 days, reared with the sow and littermates. Three synthesized vocalizations were created from the original vocalizations of the positive context using digital synthesis, sound manipulation, and sampling to determine if similar acoustic characteristics to those of vocalizations emitted in positive contexts could be emulated. ANOVA was implemented to assess the variability of the data obtained for each group (positive, negative, neutral). Multiple comparisons were carried out using the Tukey test, and the t-student test was performed to compare grouped data from original vocalizations and synthesized vocalizations, as well as non-parametric Spearman correlations. It was found that for the acoustic dissonance parameter, a significant difference was observed (P-value < 0.05) between neutral and negative contexts. Dissonant pieces can generate negative emotional states; therefore, the values of threatening contexts can be used as threshold values when adjusting the musical stimuli. When correlating the parameters of porcine vocalizations, it was found that certain parameters exhibited positive correlations with each other: zero-crossing and centroid (0.93),

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dissonance and zero-crossing (0.71), and centroid and spectral deviation (0.78). These parameters have been utilized in the classification of musical genres and could be similarly applied to the classification of pig vocalizations. The analysis of vocalizations using MIR techniques provides evidence of an alternative strategy for extracting acoustic parameters from pig vocalizations for the creation of musical stimuli. Increasing the number of vocalizations included in the analysis is recommended.

The effect of environmental complexity on the ability of calves to discriminate conspecifics and on sensitivity to reward

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Most dairy calves are raised in environments of low complexity. Providing environments that limit natural behavior is known to impair cognitive development and affective states. We explored the effect of environmental complexity on the ability of pair-housed calves to discriminate between conspecifics and on their sensitivity to reward. Holstein heifers (n=20) were housed in pairs from day 6 of life, and pairs were pseudo-randomly allocated to one of two treatments: 1) pair housing for 22.5 h/d with 1.5 h of daily access to an enriched pen which included 3 other calves and enrichment items (enriched calves, n= 5 pairs) or 2) pair housing for 24 h/d (control calves, n=5 pairs). After 10 days of treatment, calves were trained to discriminate between 2 calves in a Y-maze. Training sessions were performed once a day over 20 d, each consisting of 10 trials. The discrimination task was considered learned when calves performed 4 consecutive sessions above chance level. Twelve of the 24 calves tested reached this learning criterion, requiring 15.6 ± 2.59 (mean ± SD) training sessions. Treatment did not affect the number of sessions needed to reach the criterion. After 35 d of treatment, calves were subjected to a Successive Negative Contrast test (SNC). Calves were trained to approach a 0.5 L milk reward over 3 trials/day for 3 days. On the last training day, latencies to reach the reward differed between treatments (LMM, P = 0.005): enriched calves became slower over trials while control calves remained consistent. Starting on day 4, the reward was reduced to 0.1L of milk/trial for the 5 following test days. On test days, calves' latencies to reach the reward increased across daily trials (LMM, P = 0.01). Treatment did not affect latencies, but enriched calves exhibited more consistent latencies over test days (5.57 ± 1.99 s) than control calves (6.99 \pm 11.45 s). We hypothesize that shorter and consistent latencies exhibited by control calves on the last training day indicate greater sensitivity to reward in comparison to the increased latencies performed by enriched calves. Overall, our findings suggest that calves can discriminate conspecifics but temporary exposure to a complex environment did not impact this ability. Control calves' greater sensitivity to reward aligns

with previous work indicating that calves in housing of low complexity may experience a more negative emotional state. This study provides additional knowledge on how the environment impacts calf cognition and affective states.

Validation of an attention bias test for broiler chickens using an anxiogenic agent

Marconi Italo Lourenço da Silva¹, Alexandra Ulans², Leonie Jacobs²

Anxiety can have a negative impact on animal welfare because it is considered a negative emotional state. The objective of this study was to pharmaceutically validate an attention bias test (ABT) designed for fast-growing broiler chickens to measure anxiety, adhering to ISAE Ethical Guidelines. Two-hundred-and-four male Ross 708 broiler chickens were randomly assigned to the anxiogenic or control group at 25 days of age, with 102 chickens in each treatment. Chickens in the anxiogenic group received an intraperitoneal injection of 2.5 mg βcarboline-3-carboxylic acid-N-methylamide per kg of body weight and chickens in the control group received a saline solution at a dosage of 9 mg per kg of body weight. Both were administered at a volume of 1 ml per kg of body weight and underwent a 10-min wait period prior to the ABT. Two testing arenas contained pine shavings, and had a trough feeder with commercial feed and mealworms as positive stimuli. Birds were placed in the arena and a conspecific alarm call (the negative stimulus) was played for 8 sec, using a Bluetooth speaker (52 dB) near the arena. Both positive and negative stimuli were presented simultaneously. Two cameras were used for live (top-down view) and post-test (side view) behavioral recording. Chickens were tested in groups of three, with 34 groups per treatment. Latency to begin feeding, latency to first vocalization, latency to first step, and occurrence (yes/no) of vigilance behaviors (freeze, neck stretches, looking around, erect posture) were recorded. Data were analyzed in SAS using GLIMMIX followed by F-tests with treatment as predictor, and arena, group, and pen as random effects. Vigilance behaviors were assessed using GLIMMIX with a binary distribution. The control group was faster to begin feeding than the anxiogenic group (control = 442.6±9.4 sec; anxiogenic = 488.0±3.0 sec; F1,138 = 13.5, P < 0.001). The control group vocalized later (control = 80.9±10.9 sec; anxiogenic = 40.7±5.8 sec; F1,45 = 13.0, P < 0.001) and stepped later (control = 137.6±19.1 sec; anxiogenic = 80.8 ± 12.9 sec; F1,29 = 4.2, P = 0.050) than the anxiogenic group. The occurrence of vigilance behaviors

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did not differ between treatments (P \geq 0.145). This study successfully validated an attention bias test for fast growing broiler chickens, assessing three birds simultaneously. Our results indicated that the latencies to begin feeding, first vocalization, and first step were reliable indicators for quantifying anxiety.

Session 12: Miscellaneous Session

Thursday, July 25th, 10:45-11:00

A global assessment of fish welfare in aquaculture and fisheries: the fair-fish database

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In aquaculture and fisheries, fish welfare is a crucial issue that needs to be properly addressed. Scientific knowledge about the natural behaviors, farming conditions and welfare hazards of farmed and fished species are essential to provide reliable recommendations to improve their welfare. In this context, fair-fish created the fair-fish database, an online open access platform to collect welfare-related knowledge on aquatic animals in aquaculture (farm branch) and fisheries (catch branch), which are systematically categorized into profiles. These are created by searching the literature using specific terms related to each criterion of the profile, mainly focusing on most recent papers and considering a budget of about 80 h per profile. In the farm branch of this platform, by comparing fish behaviour and needs in the wild with what they are provided with in captivity, we assess their likelihood and potential of experiencing a good welfare level under basic and high-standard farming conditions, respectively, which are given in WelfareCheck profiles. For our assessment, we focus on 10 criteria we identified as most pressing regarding the welfare-related behaviour of aquatic species in aquaculture: home range, depth range, migration, reproduction, aggregation, aggression, substrate, stress, malformations and stunning/slaughter. We have published 85 WelfareChecks in the farm branch of the platform. In the catch branch, we assess 10 criteria covering welfare hazards during steps of the catching process: prospection, setting, catching, emersion, release from gear, bycatch avoidance, sorting, discarding, storing and stunning/slaughter. In this case, each created WelfareCheck corresponds to a profile of a species in relation to a specific method used to catch it. Since its publication in 2023, we have published three WelfareChecks, all concerning purse seine catching method. The goal is to expand these profiles for several fished species and other catching methods over the years. Both in farm and catch branches, we provide a final WelfareScore for each profile. This score works as a benchmark for assessing

and improving fish welfare in farms and in fisheries. In conclusion, the fair-fish database serves as an open-access source providing an overview about the welfare of a species given different farming or catching conditions, which may be used in different ways. It is a reliable tool helping to raise public awareness about fish welfare issues, giving scientists hints to knowledge gaps they can tackle, and providing practitioners with suggestions about how to avoid welfare risks and what alternatives to apply instead.

Can observers with different experience levels score similarly on a swine behavioral pain scale?

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Behavioral pain scales are widely used for assessing pain in farm animals and are generally applied by observers with significant experience working with swine. Due to high labor turnover on farm, it is very likely that caretakers are new to the position and have no previous livestock experience. Thus, understanding how varying experience levels can influence pain assessment is needed. This study aimed to evaluate the effect of observer experience on pain assessment scoring using a pain-altered behavior scale. We used a dataset collected from our previous study, contributing to the four R's of animal experimentation. Data was comprised of video recordings of 29 Large White x Duroc cross male piglets, filmed prior to castration, immediately following, and 24h post-castration. Six observers with varying experience levels (n=2 Extensive, n=2 Some, n=2 Little to No Experience) assessed pain using the Unesp-Botucatu Pig Composite Acute Pain Scale (UPAPS). Experience levels agreement for total pain scores were analyzed using Bland-Altman analysis. UPAPS total score was modeled using timepoints and observers' experience as predictors. Individual UPAPS sub-items were transformed into binary (dummy) variables, and modeled using observers' experience as predictors to identify which items were responsible for the disagreement. Descriptives are presented as mean ± standard error of the mean. Little to No Experience observers underscored UPAPS (Little to No: 1.09 \pm 0.14; Some: 2.02 \pm 0.23; Extensive: 2.25 \pm 0.22; p < 0.05) by scoring less on behavioral items composed by more than one behavior (attention to the affected area and miscellaneous behaviors). Bland-Altman analysis showed high disagreement between Little to no Experience observers and other observers (underscoring bias of 0.94 vs Some Experience and 1.17 vs Extensive Experience), while disagreement was

minimal (0.23) when comparing Some Experience vs Extensive Experience. The disagreement between Little to No Experience observers and other observers suggests that experience does impact pain diagnosis and assessment. Pain diagnosis would be uncertain with a bias near 1, and undertreatment may occur. It also suggests that some experience with pigs is sufficient to assess pain accurately compared to more experienced observers. Supporting this evidence, we identified specific sub-items that require some previous experience to be precisely scored. Further studies should consider higher sample sizes to precisely estimate experience influence on pain assessment.

How does sound a happy horse?

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Reducing discomfort is not enough to improve the welfare of a captive animal, the challenge now is to increase well-being even further. For this, reliable and practical indicators of the animal's internal state are needed, and things generally get more complicated when it comes to identifying those that characterize positive moods. Easily identifiable behavioral indicators include acoustic signals, known to convey emotional information. The signals that animals emit are typically divided into two categories: vocal signals, which call on the vibration of the vocal folds, and non-vocal sounds, which correspond to sonorous exhalations. We first investigated the acoustic production of more than 100 horses from 4 populations (riding centers, breeding farm, naturalistic conditions) in different contexts both at rest in their home environment (stall, paddock, pasture) or riding activity. Our attention was drawn to a nonvocal signal, the snort, very common in horses. Snorts occurred preferentially in preferred situations (outdoors vs stall), during preferred calm activities (foraging) and were typically associated with positive postures (ears forward). Separately, we conducted observation to assess the chronic welfare state of the horses. We found that horses with good welfare emitted more snorts than horses with compromised welfare. All this suggests that snorting is a way of expressing positive emotions, and that horses in good mood are frequent snorters. We then recorded the snort production of 22 mares (7-28 y.o.) living in a single group outdoor over three weeks in early Spring. A two-hour observation session was carried out each day, 5 days a week (8am-5pm). Meteorological data, as well as height and quality (legume/grass) of roughage, were collected. As predicted, snort prevalence (N=5673) increased with favorable environmental factors (warmer temperatures, higher quality roughage). During the session, outdoor temperatures varied from 4 to 20°C, and the median snort rate varied accordingly, increasing with temperature (at least until 20°C) (Spearman test, p=0.03, rs=0.61). Snort rate also increased with changes in grassland, corresponding to an increase in grass diversity (Wilcoxon test, Z=3.92, p<0.001). It is unlikely that this increase is explained by higher pollen load, as no signs of runny nose or coughing were observed. Horses are not the only animals

that snort and the few available studies on other species converge with the idea that snorts are principally emitted in pleasant contexts. This opens new avenues of research into animal welfare and the importance of preparing the ear for all forms of frequent noise emissions.

The electrolyte effect: investigating feeding behavior and activity following the replacement of a milk meal with electrolytes

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Calves are routinely provided electrolytes in place of a milk meal with the goal of increasing hydration, particularly during illness. However, the effect of withholding a milk meal with provision of electrolytes on calf behaviors remains unknown. Therefore, we aimed to determine if an electrolyte meal impacted calf motivation to consume milk, grain intake, and activity. A Latin-square study design was used, and all calves (n=100) received all treatment × test combinations (four combinations). Motivation to consume milk was measured through a 'test' of feeding bitter milk (0.35g/L quinine) vs. unaltered milk. At 0500h on test days, individually housed calves were provided the 'treatment' of either 2.7L of either milk or electrolytes. Calves would then receive calves either milk or bitter milk at 1700h and refusals were measured (g). Treatment application and tests occurred on days 1, 3, 7, and 11 after enrollment. The four treatment×test combinations included milk×unaltered milk, milk×bitter milk, electrolytes×unaltered milk, and electrolytes×bitter milk in a random order. Additionally, grain was weighed to calculate intake between meals (g), and a subset of calves (n=75) were equipped with a leg accelerometer to track their activity in the period between the morning and evening meals. Recorded activity included lying time (min), lying bouts (count), steps (count), and a motion index. Treatment had no effect on milk intake at the evening meal regardless of presence of bitter additive. However, calves fed bitter milk in the afternoon consumed less milk than calves fed unaltered milk (98.4% ± 3.1% vs. 51.4% ± 3.0%; P<0.01). Electrolyte calves consumed more grain than calves fed milk in the morning (135.4 ± 24.8 g vs. 82.4 ± 23.2 g; P<0.01). These results may indicate that calves fed electrolytes were hungrier than calves provided milk. Though the lack of difference in bitter milk intake may be due to a myriad of reasons, including the greater grain intake. Calves that received electrolytes had fewer steps (89.2 \pm 3.0 vs. 65.7 \pm 3.6; P<0.01), a lower motion index (568.8 \pm 15.7 vs. 452.2 \pm 19.2; P<0.01), and more time lying (390.2 \pm 3.5 vs. 369.7 \pm 5.0; P<0.01) than calves that

received milk in the morning. No difference in lying bouts was observed. This may imply that calves fed electrolytes instead of milk may be more fatigued than calves provided milk. Altogether, these results show that replacing milk with electrolytes affected both the intake and activity of calves.

Geolocation collars to monitor the Spatiotemporal Dynamics of livestock extensively reared in mountainous areas

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Precision Livestock Farming (PLF) technologies offer an opportunity to monitor livestock, enhancing farmers' decision-making for better control, improving animal welfare, and reduced environmental impact through proper management of pasture areas. The objective of this study was to assess the potential of data provided by commercial geolocation collars, along with environmental information such as natural habitats, topography, and weather conditions, to characterize the activity and behavior of mountain livestock. Twenty animals from three different herds and species were monitored (10 cows, 5 horses, and 5 sheep) during the pasture season (6 months) with geolocation collars in the Natural Park of Alt Pirineu (60,000 ha), located in Catalonia, Spain. Animal distributions were analyzed spatially and temporally in relation to different season moments. Localization data were used to study livestock preferences and rejections towards different types of vegetation, estimated by Jacob's index (JSI). Results indicated that cows positively selected meadows dominated by grasses (Festuca ovina) (JSI = 0.31, p < 0.05), horses preferred shrubs (Juniperus communis) (JSI = 0.25, p < 0.05), whereas pine forests (*Pinus uncinata*) were more selected by sheep (JSI = 0.24, p < 0.05). Moreover, all species avoided spruce-fir forests (JSI = -0.42, p < 0.05) or birch forests (JSI = -0.67, p < 0.05). These findings not only demonstrated interesting heterogeneity in distribution among species sharing a similar area, but also revealed differences depending on the season. In spring and autumn, livestock preferred lower altitudes in pine forests, whereas in the summer, they ventured into higher meadow areas. The use of the pasture area also varied among species. Cows and sheep showed a more homogeneous cover of larger areas, avoiding generally over-pasture; however, the monitored animals were in a larger (42 animals, in situ observations) and less dispersed groups comparing the average distance to every animal and the herd centroid (108 ± 52m). In contrast, horses were more heterogeneous occupying the area, staying more time in some specific zones, but the size of the group was lower (24 animals) and also more dispersed between them comparing to cows and sheep (390 ± 190m, p < 0.05). In conclusion, automatically collected data from geolocation collars show

great potential for studying animal behavior, especially in areas with difficult access such as high mountainous areas. This information provides a better understanding of interactions between livestock and the environment without the need for physical presence and can be easily applied to support silvipastoral management.

Perception of farmed salmon welfare and willingness to pay for welfare assured salmon products in Chile

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Fish farming is on the rise, with over half of aquatic products now coming from aquaculture. Chile ranks second globally in salmonid production, yet little is known about public perceptions of salmon welfare or willingness-to-pay for welfare-assured products. To fill this gap, we surveyed Chilean attitudes towards farmed salmon welfare and their willingness to pay for welfare-assured salmon products. We carried an online survey to Chilean consumers (currently, 1359 men and women, 18 to 100 years old independent of education level) throughout the country using a previously validated questionnaire. Our study found that most Chileans lack knowledge about salmon farming practices, with 52% having little to no understanding. Impressions of the industry were mixed, with 24% holding a favourable view, while 17% viewed it negatively, and 55% remained undecided about its overall impact. This contrasts with findings from Norway which showed a higher awareness of salmon farming practices. Also, 46% of Chileans recognized the importance of the salmon industry for food security, jobs, and economic benefits. However, concerns about environmental sustainability (81%) and operational transparency (77%) were widespread, differing from perceptions in Norway. Interestingly, despite reservations, most Chileans expressed acceptance, tolerance, and pride in salmon farming (83%, 67%, and 65% respectively), likely influenced by its economic and social contributions. Salmon welfare emerged as a significant concern for Chilean consumers, with 77% expressing its importance and 92% seeking information on salmon products. However, 90% of consumers feel inadequately informed and desiring more information about salmon welfare. These findings coincide with Chile's recent efforts to strengthen animal welfare regulations, potentially driving consumer interest in salmon welfare issues. However, when it came to willingness-to-pay, Chileans showed reluctance to pay more for sustainably certified (66%) or welfare-assured (63%) salmon products, with only a small minority (17% and 21% respectively) willing to pay up to 5%. This reluctance may stem from the already high prices of salmon products in Chile compared to terrestrial meats.

Chilean consumers recognize the economic importance of salmon farming, but concerns about environmental sustainability, operational transparency, and salmon welfare persist and consumer willingness-to-pay a premium for certified or welfare-assured salmon products is low.

Session 13: Miscellaneous Session

Thursday, July 25th, 15:00-15:15

The role of the quality of the human-animal relationship in shaping pigs' gut microbiota

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The microbiota-gut-brain axis is crucial in modulating pigs' stress response, behaviour, and overall welfare. Previous research has highlighted that stressful management practices can lead to gut microbiota (GM) dysbiosis, adversely affecting pigs' health and welfare. This study aimed to explore the impact of the quality of human-animal relationships on the GM composition of pigs, given the established bi-directional link between stress and GM composition. Thirty-six 21-day-old female pigs were randomly distributed across three experimental groups (12 pigs/group, 4 pens per treatment): Positive human handling (PHH), where animals experienced gentle tactile contacts consisting of stroking and scratching; negative human handling (NHH), where animals were subjected to chronic intermittent stress through various acute handling stressors (e.g., capture, pursuit, attempting to place a rope around the snout if animal contacted the handler); and a control group (CG), in which animals received minimal handling necessary for routine husbandry practices. Both PHH and NHH interventions were applied for 2 minutes, twice a day, five days a week, over six weeks (days 17-61 of the experimental period). Fecal samples were collected at baseline (T0, day 16), midstudy (T1, day 37), and the end of the study (T2, day 65) and analyzed using 16S rRNA gene amplicon sequencing. Alpha (Shannon and Inverse-Simpson indices) and beta diversity measures were assessed between handling procedures for each time point, using Kruskal-Wallis and PERMANOVA (with 999 permutations) tests, in addition to microbial differential abundance (DA) analysis using the MaAsLin2 package in R. There were no significant differences in alpha-diversity indices among groups at T0 and T1 (P>0.05). At T2, PHH pigs presented higher microbial diversity compared to NHH pigs, as indicated by Shannon

 $(\chi 2=8.815, P=0.009)$ and Inverse-Simpson $(\chi 2=8.812, P=0.010)$ indices, including differences in their community compositions, assessed by Bray-Curtis (P=0.036) and Jaccard (P=0.024) dissimilarities. Additionally, microbial DA differed between NHH and PHH pigs in four bacterial genera: Blautia (P=0.035), Subdoligranulum (P=0.035), [Eubacterium] hallii group (P=0.035), and Megasphaera (P=0.049). PHH pigs presented a higher abundance for all compared to NHH. These results highlight the influence of human-animal relationship quality on the composition and diversity of GM in pigs. Positive handling practices enhance gut health, promoting a more diverse and potentially resilient GM. This study emphasizes the intricate relationship between management practices and gut microbiota, suggesting that positive interactions with pigs could be pivotal in enhancing pigs' microbial health and overall welfare.

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Farmers' work satisfaction and the frequency of their interactions with pigs influenced the human-animal relationship in outdoor pig farming

Ophélie Menant¹, Laura Frigerio¹, Élodie Peyrard¹, Grace Caroll², Laura Boyle1, Keelin O'Driscoll¹

Positive human-animal relationships (HAR) improve farm animal productivity and welfare. The HAR is influenced by the frequency and nature of animal-human interactions, as well as individual farmer and pig characteristics. To investigate the HAR in outdoor pigs, we carried out a single Animal-Human Approach Test (AHAT) on 181 pigs in 16 farms from February to May 2023. The AHAT was performed on female, castrated and uncastrated pigs, aged from 3 months to 6 years (~10 pure/mixed breeds across all farms). The test consisted of 3 phases: the 'pig approach' phase (the animal had 30 seconds to approach an unfamiliar human located 2m away); the 'human approach' phase (the human approached the pig slowly if there was no previous contact); the 'scratch phase' (the human attempted to scratch the pig's forehead if there was previous contact). Scores were applied from 0-least positive (no contact) to 3most positive (scratch accepted). Farmers were interviewed, and we determined that their main interactions with pigs were "feeding", "petting" and "bedding/cleaning". Number of years in the industry, quality of life (WHOQOL-BREF, from 1-very unsatisfied to 5-very satisfied), mental wellbeing (WEMWBS, 1 to 75) and work satisfaction (1-very unsatisfied to 4-very satisfied) were scored using 3 different surveys. Spearman correlations were performed between all variables and AHAT scores, except for the WHOQOL and work satisfaction surveys, where Kruskal-Wallis tests were performed (R-Studio). The number of weekly interactions was positively correlated to the AHAT score (Rho = 0.28, p-value < 0.001), whether it was for feeding (Rho = 0.23, p-value = 0.002), petting (Rho = 0.21, p-value = 0.005) or bedding (Rho = 0.35, p-value < 0.001). Neither the number of years in the industry, the WEMWBS nor the WHOQOL had a significant impact on the AHAT score. However, work satisfaction had a significant effect on the AHAT score (H(2)= 8.01; p-value = 0.02). There was no effect of the pig sex on the AHAT score, but the AHAT score increased with pig age (rho=0.18, pvalue=0.015). We found that whatever the nature of the positive interaction, the frequency

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of the interaction improved the HAR. The pig age, and thus the duration of their experience also positively influenced the HAR. Interestingly, it seemed that as the level of farmer work satisfaction (which is a component of their welfare) improved, so did their relationship with the pigs.

Horse's perception of humans: a major site effect

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Horses' perception of humans is influenced by its welfare state and past experiences with humans during and outside work. These aspects are influenced by facility managers' perception of good management practices which is under cultural influences related in particular with the equestrian activity. There are for example expectations that the humanhorse relationship (HHR) may differ between racing and instruction horses. Few studies have examined horses' perceptions of humans at the facility level and tested the possible impact of work activity on HRR, despite the demonstrated importance of work type on chronic equine welfare status. Using standard experimental tests, we examined the response of horses to an unknown person and the possible relationship between these responses and the site and its main type of activity. 176 horses were tested that lived in 9 facilities differing in terms of equestrian activity (3 riding schools, 6 racing facilities: 3 trotting and 3 gallop). Three tests were used that were performed by an unknown experimenter to horses while the horse was in its box: approach-contact test (AC), equipment test (ET) (approach in stall with saddle or surcingle), corridor test (CT) (horse's reaction to a human passing in front of its stall). Measures were the number of positive (ears forward with or without approach), negative (ears backwards with or without approach, threats of biting or kicking) or no behavior change for the AC test, scores (A to F, from positive behavior with approach to possible threat and/or flight) for the two other tests. The test situations were not unusual for the horses and the reactions observed were close to that observed in response to daily human interactions: most horses (61%) showed positive reactions in to the CT and no reaction in the AC (72%). The results in the ET were more variable (40% positive horses, 29.5% fearful, 16% with no change in behavior, 14.5% aggressive). There were large individual variations: an ordinal regression revealed that there was a strong site effect that was more influential than work type (p=0.007). A PCA, followed by a hierarchical classification, grouped stables into 4 horses' profiles. Despite the limited number of facilities per discipline (but which are representative

of each discipline), these results show strong site effects, suggesting that managers' decisions may be highly individual but convergent with those of managers from other disciplines. Interestingly, racehorses did not express a more compromised perception of humans than riding school horses.

Behavioral evaluation of weaned piglets fed with diets supplemented with organic acids and essential oils

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In piglet weaning, challenges that diminish the feed intake and hinder performance can disrupt the gut-brain axis, potentially affecting animal behavior. To mitigate the use of antimicrobial growth promoters (AGPs) and avoid bacterial resistance, alternatives are being studied. Among them, we have organic acids (OA) and essential oils (EO). Therefore, the aim of the present study was to identify the possible effects of diets supplemented with OA (citric, phosphoric, fumaric, and malic acids) and EO (thyme and carvacrol), as an alternative to AGPs, on the behavior of weaned piglets through the behavioral tests of Open Field and Novel Object. For that, castrated males (n=60) and females (n=60) piglets, weaned at 21±3 days of age, were evaluated during the whole nursery phase (45 days). There were 5 treatments studied: T1) basal diet (no additive); T2) basal diet + 120mg of chlorohydroxyguinoline/kg of diet; T3, T4 and T5) basal diet + addition of 200g, 400g and 800g of OA+EO/ton of diet, respectively. For the Open Field test, piglets were placed in a 9m2 arena, divided into nine quadrants. Parameters such as exploration time, number of vocalizations, number of chewing behavior, presence of eliminatory behavior (urinating or defecating), and number of explored quadrants were observed for two minutes. For the Novel Object test, piglets were placed again in the arena, with an unknown object (box). Parameters such as latency to touch the object, frequency of touches, and total duration of contact with the object were observed for two minutes. All behavioral data was analyzed in R Studio software, through both linear and logistic regression, with T2 as front level treatment. For the Open Field test, animals in T1 (P<0.01) showed a higher number of chewing behaviors compared to T2, and no differences for the other treatments were observed (T3, T4 and T5: P>0.05). There were no statistical differences for the other parameters of the open field test (P>0.05). For the Novel Object test, no statistical differences were found for any observed parameter (P>0.05). Our findings

showed that the addition of OA and EO in the diet does not negatively affect piglets' behavior and can substitute AGPs.

A cohort study to understand the relationship between early life factors and the expression of stereotypies in foals

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Stereotypies are repetitive and unvarying behaviours with no apparent function or objective which are frequently manifested by individuals in compromised welfare conditions. Though these behaviours have been extensively described in horses, their causes and mechanisms are still the focus of debate. Abrupt weaning seems to be the starting point for expression of stereotypies in the species, however studies considering other early life events are scarce. The goal of this project is to investigate factors potentially involved in the determination of vulnerability or resilience to developing stereotypies in horses considering some predisposing factors to higher stress reactivity and neurological alterations associated to manifestations of repetitive behaviours, such as excessive fetal exposure to glucocorticoids, epigenetic modulations resulting from fetal programming and negative patterns of maternal interactions. For this purpose, a cohort of 70 foals of the Brasileiro de Hipismo breed born between August and December of 2023 is being followed from birth. All foals were born in the same farm in the state of São Paulo, Brazil, to surrogate mares as a result of embryo transfer procedures, and they share relatively similar genetic and environmental characteristics. At birth, samples were obtained from placental tissues immediately upon expulsion (n = 55), foal hair (n = 60) and surrogate mare hair (n = 15), for glucocorticoid quantification. Behaviour of the first 3 to 30 days of life was also recorded by cameras installed on the group maternity pastures (n = 45), which will be analysed for affiliative and agonistic maternal interactions, play behaviour and abnormal or repetitive behaviours. Additionally, information regarding surrogate mare characteristics, biological parents' characteristics, time until placenta expulsion, weight of fetal annexes, incidence of neonatal maladjustment syndrome, presence or absence of other foals in the pasture and size of pasture were also collected. All foals will be abruptly weaned in the first semester of 2024 in groups inside a small corral with an unfamiliar adult mare for 48 hours before moving to a group pasture, and behaviour during weaning will be recorded

to assess the expression of abnormal behaviours and stereotypies. Hair samplings will be repeated for glucocorticoid analysis, and placental samples of foals which perform stereotypies (n = 8) and don't perform stereotypies (n = 8) will be submitted to RNA sequencing and gene expression analyses to investigate potential epigenetic modulations. All information will then be correlated to data collected in early life.

Session 14: Miscellaneous Session

Thursday, July 25th, 15:00-15:15

Influence of hierarchy on the feeding behavior of growing pigs

<u>Gustavo Zigovski de Paula</u>¹, Juan Ochoteco², Raquel Rio Lopez², Adrià Clavell Sansalvador³, Yuliaxis Ramayo-Caldas³, Leandro Batista Costa¹, Antoni Dalmau²

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The aim of our study was to explore the impact of hierarchy on the feeding behavior of pigs subjected twice to a mixture during the growing period. At an experimental farm, 64 Duroc pigs (18±1.69 kg) were allocated to eight pens, each containing eight animals (four castrated males and four females). Four pens were mixed twice during the growing period and four were not mixed. The first mixing was executed after one month of arrival to the facilities (61 days old) and involved only the females of each pen and the second mixing was 22 days later (83 days old) and involved only the males. Hence, the study was divided into three phases: phase 1 - before any mixing; phase 2 - after the first mixing and before the second; phase 3 - after the second mixing. To ascertain animal hierarchies (dominant, intermediate, and submissive), pigs were tested in couples in an arena using sliced apples as a competitive resource. Dominance was primarily attributed to the pig that ate the apple first. After eating for 30 seconds, the pig was removed, allowing the other to eat, and after one minute, the first pig was reintroduced. Dominance status was confirmed if the reintroduced pig displaced the submissive from the apple feeder within one minute. Animals between dominants and submissives in the hierarchy were intermediates. The test was conducted for all combinations within each pen before the first mixing, after the first mixing, and after the second mixing. Parameters such as body weight, feeder visits, and feeding consumption and duration were assessed by means of electronic feeders in the pens throughout the entire experiment. This study was approved by the Ethical Committee (protocol number - 10329). Statistical analysis involved a linear regression model for body weight versus hierarchy. All analyses were conducted using R software. In phase 1, no differences were found in the evaluated parameters among hierarchical classes. In phase 2, submissive pigs consumed less feed (2476

 \pm 97.7 g/day) than dominants (2745 \pm 118 g/day) (P<0.05). The same occurred in phase 3, as submissive pigs ate 2854 g/day (\pm 110) while dominants consumed 3387 g/day (\pm 93.6), with submissive animals also showing lower body weight, less frequent feeder visits and feeding duration (P<0.05). The outcomes highlight the influence of hierarchy, as subordinate animals have low access to the feeders, whereas dominants show better performance and exhibit different feeding behavior.

Investigating dog walking as a form of human physical activity: The need for a One Health approach

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Dog walking is often proposed as a tool to promote human physical activity, yet we know very little about on-leash dog walking behavior beyond measures of duration and frequency. The goals of this study were to characterize the exercise intensity of dog walking using human physiological measures, and to describe differences in human exercise intensity based on walking behavior relative to dog sex. Between February 2020 and September 2022 in metropolitan Sydney, 43 dog owners (63% female) walked their dog on-leash for ≥20 minutes while wearing a validated chest strap to measure heart rate and carrying a phone to record distance, speed, moving time, elapsed time, and cadence using GPS and accelerometry. Mean values for each measure were then compared with established cut-off values and categorized as light, moderate or vigorous intensity. We also calculated the number of times that owners stopped at 0km/hr for ≥3 seconds as a relative average per 30 minutes, and the mean walking speed with and without stops. An independent samples T-test was used to compare the number of stops between male and female dogs. On average, the recorded dog walk was 2.34 km±0.84 and 34:44 minutes±10:15. Dog walking was classified as light intensity based on measures of heart rate reserve (mean 38.11%±10.81), heart rate max (mean 60.96%±7.18) and cadence (45.34 steps/min±8.40). Metabolic equivalents (mean 3.53 METs±0.61) and mean walking speed including (4.29 km/hr±0.82) and excluding stops (4.61 km/hr±0.73) indicated the walk was within the lower moderate intensity band. The mean number of stops per 30 minutes was 16±8.36 with no significant difference based on the sex of the dog (p=0.65). The total number of stops per walk ranged from 3 to 48. Overall, we found on-leash dog walking for ≥20 minutes fell within the upper end of light and the lower end of moderate intensity depending on the intensity measure used. Dog owners may need to participate in other forms of physical activity to meet current guidelines. There were vast differences in the number of stops across the sample, unrelated to dog sex. Other canine characteristics, such

as physical capacity, body condition, health status, breed, size, and behavior likely affect dog walking patterns and warrant further consideration, including measures of canine activity. Future studies should also consider the environment in which dog walking occurs as the density of urbanization may be a key determinant of human and canine walking behavior.

Thursday, July 25th, 15:30-15:45

Exploring animal faces: Mechanisms underlying child-animal interaction with autism spectrum disorder

<u>Manon Toutain</u>¹, Laurence Henry¹, Lou Léonard-Broissier¹, Marine Grandgeorge¹

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When interacting with an animal, we use several sensorial cues to communicate (e.g., visual, auditory). One of the most informative cues is the face, in particular the eyes. We know that typically developing (TD) people explore animal faces in the same way as they explore human faces: they pay more attention to eyes and secondarily to the mouth through a triangular movement. However, what happens when information extraction is disrupted, as in autism spectrum disorders (ASD)? On 2D stimuli (images/videos), children with ASD gaze more to the lower part of face and avoid eyes on human face compare to TD children, whereas with animals, ASD kids gaze more to the eyes. To date, we aim to know if, in a real encounter with animals, TD and ASD children explore faces similarly. Eight children with ASD (6M/2F, M=10.2±3.3yo) and 18 TD children (7M/11F, M=10.2±2.5yo) participated. The encounters followed a standardized scenario, in which each child wore eye-tracking glasses (60Hz). Each child met 4 individuals from different species: human, horse, dog and guinea pig. As done with 2D stimuli, each child was asked to face towards an animal or human, one after one (random order for 5 sec). Animal and human faces were divided into several areas of interest to quantify time spent exploring each of them (i.e., head versus outside head, eyes, lower face area). Fixation time per area was represented as a percentage. Mann-Whitney and Wilcoxon tests were used. As expected, in a real encounter situation, both TD and ASD children did not look at the human face in the same way; children with ASD looked more at mouth (P=0.007) and avoided eye area (P=0.014), whereas TD children did the opposite (both P<0.05). Furthermore, TD and ASD children had the same way of exploring animal faces (for all species tests, P>0.05). Moreover, there was variability in visual exploration between species: in the case of dogs and guinea pigs, TD and ASD children showed no difference in the duration of time spent on the lower face and eyes; whereas, they looked more at horses' eyes than at their snouts. In conclusion, visual exploration of faces by children with ASD, especially gazing

to eyes, is thought to be one of the underlying mechanisms explaining their ease of communication with animals, as they are able to "look for information in the right place", unlike with human faces.

Dissecting the how from the what: an investigation into the inner workings of Qualitative Behavior Assessment

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Qualitative Behavior Assessment (QBA) is a welfare assessment tool valued for its holistic approach to assessing an animal's emotional state. While implemented as part of many onfarm welfare assessment protocols, the method has been criticized due to its subjective method of capturing data. The objectives of this study were two-fold: 1.) To determine whether veterinary students could assess sow welfare using QBA similarly to swine experts and 2.) to investigate how welfare is assessed through the comparison of QBA and ethological outcomes. A video library of sows in a novel arena test was scored using QBA with predetermined emotional descriptors (e.g., happy, bored, etc.) by five swine experts and 14 vet students. Both groups of assessors were trained to assess welfare using QBA. QBA responses from experts and students were analyzed separately using principal component analysis. Two main principal components (PCs) were identified as the valence and arousal of each descriptor. PC loadings within and between groups for each video were analyzed using Kendall's Correlation Coefficient W and Pearson's Correlation Coefficient R, respectively. Experts displayed almost perfect agreement on PC1 (W=0.91), and moderate agreement on PC2 (W=0.66) while students displayed moderate agreement on both PC1 (W=0.63) and PC2 (W=0.72). A significant correlation was found between student and expert PC1 (r=0.927, p<0.001) and PC2 (r=0.958, p<0.001), indicating the two groups perceived observed animals similarly. Despite high agreement on PCs, descriptors that contributed significantly to each PC differed between the two groups, indicating that although animals may have been perceived in similar emotional states, each group selected different descriptors to describe these animals. For the second aim, the same videos were coded by six event behaviors to determine whether specific behaviors were likely to predict QBA outcomes. Each descriptor scored by both the experts and students was used as an outcome variable using a linear mixed model.

18 of the 20 descriptors were significantly predicted by at least one event behavior (p<0.05). These results suggest the performance of certain behaviors may be more likely to invoke the use of certain QBA descriptors. This warrants further investigation to better elucidate the specifics of these relationships and how assessors use different cues to assess the emotional states of sows under a controlled environment. This knowledge can improve QBA training and enable a more robust assessment.

Effects of Information Framing on Public Perception of Precision Dairy Technologies

Brad Kelly¹, Katy Proudfoot², Joao Costa³, Beth Ventura⁴

Certain management practices within the dairy industry, including increased reliance on technology use, have come under public scrutiny, posing a potential threat to the social sustainability of the industry if concerns cannot be reconciled. With increased interest in and use of precision dairy technologies (PDT) by producers, a clearer understanding of societal perceptions is needed. This study sought to characterize public perceptions toward PDT and describe how perceptions may be impacted by information framing about the potential effects of PDT usage. In an online survey distributed to US residents (n=2,398), participants were provided a neutral definition of PDT and then randomly received 1 of 8 vignettes, each containing either positively or negatively valenced information about hypothetical PDT impacts on data transparency, human-animal bond, and animal welfare. Here we present results of perceptions toward PDT as assessed via Likert item questions before and after vignette exposure. Linear models were selected using backwards selection to assess significant predictors of shifts in perceptions about PDT. Lower baseline perceptions of PDT were associated with more positive views of animals evaluated using animal attitude scores (p<0.001) and being politically independent compared to other political affiliations (p=0.006). Additionally, amongst dairy consumers, those who considered themselves 'extremely knowledgeable' about dairy farming had more positive baseline perceptions toward PDT compared to those 'moderately', 'slightly', and 'not at all' knowledgeable (p<0.02 or lower for all comparisons). Overall, information about data transparency did not shift perceptions of PDT in either direction, though dairy consumers were more sensitive to positive data transparency information compared to non-consumers (p=0.01). Those in the more liberal political party were more sensitive to negative human-animal bond information than political conservatives (p<0.001). The valence of animal welfare information strongly predicted the direction of perception shifts (p<0.001), such that negative information was a 'deal-breaker,' resulting in worse perceptions of PDT, even if participants received positive information about

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other attributes. We suggest that for the dairy industry to maintain its social license to operate, it will benefit from more closely engaging with public concerns around how PDT may impact animal welfare and the relationship between farmers and animals.

Poster + One-Minute Presentation Session 1: Farm Animals Behaviour and Welfare (Cattle)

Tuesday, July 23rd, 12:00-12:15

Implication of environmental enrichment on the performance of dairy heifers in the suckling phase

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Among the main requirements of animal welfare are adequate nutrition, performance, and good health. The lactation phase is an extremely important period in calves' lives, characterized by intense physiological, behavioral, and management changes. At this stage, using environmental enrichments has proven to be an effective strategy to help animals deal with environmental stressors and increase the satisfaction of their behavioral needs. This study aimed to investigate the impact of environmental enrichment on the performance of calves during the suckling period in an individual rearing systemin suspended cages. The experiment was carried out on a commercial farm, with twenty Holstein calves in the lactation phase up to 75 days of age, randomly distributed into two treatments: with environmental enrichment (EE; n=10) and without enrichment (WEE; n=10). Enrichment was used to improve the welfare of the animals, and this consisted of a ball, plastic chain, and ball with a bell, which were alternated every two weeks, and a brush fixed to the elevated calf box. The collected data was analyzed over time using PROCMIXED SAS and birth weight and growth variables were used as covariates in the model. The results indicated that EE had no significant effect on the variables (P>0.05) of weight gain (mean = 0.93, SEM = 0.02), weight (mean = 93.9kg, SEM=0.70), rump (mean = 19.20 cm, SEM = 0.33) and withers height (mean = 77.61 cm, SEM =1.12). However, an interaction was observed between treatment and time for length at 75 days of life (P=0.02), where animals with EE had an average of 78.5 cm, while animals with WEE had an average of 72.8 cm (SEM =1.6 cm). Although environmental enrichment showed no effect on most performance variables, a 5.7 cm increase was observed in the length of animals with environmental enrichment at weaning. This suggests that environmental enrichment may have a positive but modest impact on calf growth during the suckling period.

The effect of maternal contact on the feeding development of Norwegian Red calves

Laura Whalin¹, Kristian Ellingsen-Dalskau¹, Julie Føske Johnsen¹

Calves raised with peers may be more willing to eat concentrate, and try novel feeds than calves raised in solitude. Less is known about the effect of maternal access on calf feeding behaviour. Our aim was to describe when maternally raised calves consume concentrate daily, and try carrots in a food neophobia test. Twenty-four calves were enrolled for 10 wk. The study included 2 batches with 2 treatments: cow-calf-contact (CCC; 6 cow-calf pairs: freestalls, automatic milking (AMS), and calf access to a straw-bedded creep), or peer-housing (6 calves: straw-bedded creep, sawdust-bedded rubber mattress, and rubber slatted floors). CCC calves were free to suckle except when the cows were in the AMS, and peer-housed calves were fed whole milk to satiety from milk bars 4 times/d for a total of 4 h/d (average: 15 L/d/calf). All calves were given ad libitum access to concentrates from an automatic feeder (CCC in creep), hay, silage (CCC in creep and freestall area), and water. One CCC cow-calf pair was removed at 60 d of age due to mastitis. At 30 d post-calving, cows were habituated to chopped carrots near the AMS 8 h/wk for 4 wk. At 60 d of age, calves were randomly paired (n = 12), within their group, and habituated for 30 min to a test area with feed tubs. CCC calves were tested in pairs with their mothers. The following day, pairs returned to the test pen, and were given access to 2 kg of silage, and 2 kg of chopped carrots for 30 min. One trained observer scored all videos (IOR: kappa = 0.94). Descriptive data are presented as medians (Quartile 1 – Quartile 3). All cows approached the carrots. One peer-housed calf did not eat carrots. CCC calves took 1.5 (0.8 - 1.7) min to approach, and 1.6 (0.8 - 2.2) min to begin eating carrots. CCC calves spent 3.2 (1.1 - 5.5) min eating carrots. Peer-housed calves took 2.9 (1.3 - 7.0) min to approach, and 5.1 (2.5 – 17.3) min to begin eating carrots. Peer-housed calves spent 1.7 (0.7 - 3.3) min eating carrots. Peer-housed calves ate at least 200 g of concentrate by 56 (46 – 63) d, but only 3 of 11 CCC calves reached this target by 70 d. CCC calves appeared curious to try carrots with their mothers, but may have chosen to socially eat roughages instead of visit the concentrate feeder.

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Conditioned place preference four weeks after hot-iron and caustic paste disbudding dairy calves

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Disbudding, a painful procedure, permanently damages the horn-bud tissue either with a hot iron (heat) or caustic paste (chemical). The resulting wounds persist for an average 7-9 wk for hot-iron or 16 wk for caustic paste. Conditioned place preference for analgesia has been shown for hot-iron disbudded calves 3 wk after the procedure but is unknown for caustic paste or at later stages of healing. We evaluated conditioned place preference for analgesia 4 wk after sham handling (n = 23), disbudding with a hot iron (n = 21), 0.3 mL of paste applied to unshaved horn buds (n = 19) or 0.3 mL of paste on shaved horn buds (n = 18). Calves were conditioned to associate the effects of a lidocaine cornual nerve block with the location and pattern of a visual stimulus, and a control injection of saline with the contrasting stimuli. Four training sessions occurred over 4 d, 2 for each set of stimuli. On the 5th day, calves were provided both sets of stimuli for 5 min. The effect of treatment on latency to contact stimuli was evaluated using a generalized linear model. Linear mixed models were used to evaluate the effect of treatment on the proportion of time contacting each set of stimuli. We found no evidence of a preference for either lidocaine- or saline-paired stimuli (mean \pm SE proportion of time contacting lidocaine stimuli: Control: 0.46 \pm 0.07; Hot iron: 0.51 \pm 0.07; Unshaved 0.3 mL paste: 0.41 \pm 0.07; Shaved 0.3 mL paste: 0.47 \pm 0.08; P = 0.82). The latency to contact a lidocaine-paired stimuli was not different (mean seconds \pm SE: Control: 59 \pm 15; Hot iron: 59 \pm 16; Unshaved 0.3 mL paste: 43 \pm 12; Shaved 0.3 mL paste: 71 \pm 21; P = 0.68). Latency to contact the saline-paired stimuli also was not different (mean seconds \pm SE: Control: 68 \pm 14; Hot iron: 79 \pm 18; Unshaved 0.3 mL paste: 59 \pm 14; Shaved 0.3 mL paste: 60 \pm 14; P = 0.81). Our preliminary results show no evidence of a preference for analgesia by calves 4 wk after disbudding or sham handling. Analysis is planned to look at preferences for stimuli pattern during training sessions. Calf preferences may have been affected by the injection pain or ear

tag wounds, making the lidocaine-paired stimuli less rewarding, for example. Further analysis is needed to understand these unexpected results.

Cooling cows with sprinklers: body regions targeted during summer

Grazyne Tresoldi^{1,2}, Alycia M. Drwencke², Cassandra B. Tucker²

Sprinklers or soakers have been identified as highly effective in mitigating heat stress in dairy cows. However, limited research exists on how cows prefer to be sprayed. This study aims to describe how dairy cows utilize sprinklers to target specific body regions (i.e., position), and examine its relationship with air temperature (AT). We expected cows would shift their body position as temperature increased. A prospective study was conducted at the UC Davis Dairy Facility during the summers of 2016 and 2017. Both studies received IACUC approval. The preliminary dataset included 21 lactating dairy cows. Sprinklers were positioned at the feed bunk to reach the cows' withers; the distance that the spray reached was indicated on the ground. Continuous 24-hour surveillance cameras recorded the cows' behavior for 6-12 days, while AT was logged every 5 minutes in a nearby portable weather station. Six positions were recorded every 3 minutes when cows were observed at the feed bunk and sprinklers were cycling. Positions included: head through the gates and eating, head through the gates and not eating, horizontal to spray (left or right), perpendicular to spray but backward (posterior facing spray), and head facing spray but head outside the gates. Descriptive statistics, correlations, and paired t-tests were used for data analysis. Overall, cows spent approximately 26% of their time at the feed bunk engaged in behaviors other than eating. However, individuals varied in how they utilized sprinklers. For example, while some cows rarely had their heads through the gates without eating, others reached up to 17%. Additionally, some cows displayed a preference for wetting their right side (n= 8 cows), while others showed indifference. As AT increased throughout the day, cows shifted their body positions away from feeding. At peak heat (35.1°C), the frequency of feeding was 43%, compared to 88% when the AT was 26.8°C. In contrast, the frequency of backward and horizontal postures increased by 11% (0-11%) and 15% (2-17%), respectively, during this period. Cows also exhibited these postures more frequently as daily AT increased (r=0.83 and 0.63, respectively for horizontal and backwards positions). Our findings suggest that cows adopt distinct positions when utilizing sprinklers, and this relationship appears to be influenced by air temperature and

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individual preferences. We speculate that these distinct positions may provide cows with additional cooling benefits while avoiding wetting their heads.

Effect of a pre-slaughter walking on water intake and stress blood indicators in beef cattle

<u>Maria Eugênia Andrighetto Canozzi</u>¹, Anderson Saravia¹, Georgget Banchero¹, Eduardo Pérez¹, Juan Clariget¹

¹ Instituto Nacional de Investigación Agropecuaria (INIA), Estación Experimental La Estanzuela, Colonia, Uruguay Before slaughter, stressor experienced by cattle can impact welfare and meat quality. We evaluated the effect of a walking stress, as part of a bigger project evaluating different typical practices prior to slaughter in Uruguay, on water intake and hematological variables in beef cattle raised on pasture. A total of 170 steers from British breeds, weighing ~540 ± 35.0 kg and aged between 24 and 48 months, were evaluated. This experiment was replicated six times from November to December 2023. Two days before slaughter, cattle were individually weighed and randomly allocated to one of two treatments (n= 15 animals/treatment): without (0 km) or with (10 km) walking. On the day before slaughter, animals from "10 km" started the walk at ~6 AM, and cattle from "0 km" were kept in the handling facilities. The exercise sessions were conducted by three people on horsebacks and the speed and the distance data were recorded by a Garmin® watch. Once finished the walk, "10 km" steers stayed in nearby pens to those from "0 km". In the facilities, steers had ad libitum access to water, but not to feed. Cattle from both treatments were gathered in one pen 45 minutes before the loading, at ~5:30 PM. Cattle were transported to an abattoir 40 minutes from the farm. Slaughters were at 10:15 AM, following the commercial procedures and animal welfare standard. Water consumption was measured for each lot, at the farm, using a tank with meter. Blood samples were collected from each steer after throat cutting to measure hematocrit and metabolites concentrations. Data were evaluated as a completely block randomized, with each slaughter considered as a block and the group of animals as an experimental unit. Average overall period of fasting was 28 hours. Average duration and speed of walking was 2h27min and 4.28 km/h, respectively. Steers from "0 km" drunk less (P = 0.02) water than "10 km" (11.0 vs. 22.7 l/animal). The lactate dehydrogenase (LDH) concentration tended (P = 0.09) to be lower in "0 km" than "10 km" steers (1,761.2 vs. 1,821.1 U/L). There was no effect (P >

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0.05) of walking on hematocrit, albumin, globulin, total proteins, glucose and creatine kinase

concentrations. Although the higher water intake for steers from "10 km", the extra exercise

did not compromise blood parameters; however, the higher LDH could indicate a negative impact on welfare and a possible negative effect on meat quality.

Effect of analgesia on calves weight gain and animal welfare after castration

<u>Natalia Aguilar</u>², Valeria E. Pared¹, Marcelo L. Jara¹, Jose A. Chávez¹, Maria M. Pereira², Luis Gandara², Paolo A. Sussini², Valeria Borelli²

Castration of male calves intended for beef production is a common management practice, performed between 6 and 9 months of age, to mitigate agonistic interactions and unwanted sexual activity between animals. Previous studies have demonstrated that irrespective of the method, castration induces stress, fear, and pain in animals, resulting in decreased food and water intake during the initial post-intervention period, a significant animal welfare issue for the beef industry. While castration methods aim to halt testicular irrigation and remove the testicles, the traditional knife method prevails in northeastern Argentina. This study aimed to analyze the average daily gain (ADG, kg) during the first days of post-treatment of castrated calves using different methods with or without analgesic. The research was conducted at the National Institute of Agricultural Technology (INTA), Corrientes, Argentina, between September and November 2021, using 43 Braford cross calves unweaned (30±11 days old) and 59.24±12.31 kg average body weight (BW, kg). The animals were distributed at random into three groups: traditional knife castration performed by a trained worker (T1 n=8), surgical castration performed by a veterinarian with analgesia (T2 n=17), and a traditional knife castration with analgesia (T3 n=18). The day of castration, calves were separated from their dams and placed together in a pen, and the BW of all calves was identified and recorded. In addition, at T2 and T3, injected 3-5 min before castration, local anaesthesia (lidocaine HCl 2%) in the spermatic cord, around the neck of the scrotum and testicles. The procedures were executed with the calves restrained in lateral recumbency on the ground and held by two operators. After restraint, the scrotal region was cleaned with iodine solution and alcohol. Postoperative treatment included application of a nonsteroidal anti-inflammatory (Flunixin Meglumine), antibiotic, and ectoparasite repellent, except for the anti-inflammatory for T1. During the second day of collection, between 6- and 8-days post castration, the lesion was assessed, and the BW of each animal was recorded. For data analysis used the (InfoStat v. 2019e) statistical package, employing descriptive statistics, ANOVA, and means comparison

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tests, revealed statistically significant differences (p<0.05) between treatments for ADG values. Showing that ADG was lower in T1 (0.69 kg) compared to T2 (1.18 kg) and T3 (1.31 kg). This study contributes valuable insights into the impact of analgesics in various castration methods on ADG in calves, underscoring the imperative need to address pain management in this routine procedure in bovine farming.

Effect of milk supply methods on the behavior of dairy calves: a meta-analysis approach

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This review aimed to evaluate the effect of milk supply methods on the behavior of dairy calves, using estimates from the peer-reviewed literature by a meta-analysis approach. We searched on Web of Science, PubMed, and Cab Direct using the terms: (1) population: calf, calves, heifers, dairy, milking, (2) comparison: environmental enrichment, nutritional enrichment, feeding enrichment, feeding method, artificial teat, nipple, bucket, and (3) outcome: behavior. The resulting articles (n=540) underwent a four-step PRISMA appraisal process and were excluded when presented wrong population (e.g., goats), focused on other results (e.g., blood, glycemic index), or dealt with other subjects. We selected studies that compared the offer of milk in an open bucket with any other method that stimulated the calf's natural suckling behavior (e.g., nipple bucket, nipple bottle, rubber teat). This process resulted in a final sample of 13 studies that reported means and mean error. Then, we utilized the Der-Simonian and Laird methods to analyze the standardized mean differences between calves' behavioral outcomes across different milk delivery methods. Also, the between-study variance was evaluated using both the chi-square test of heterogeneity and I² statistics. Publication bias was evaluated using the funnel plot and outliers were removed when studentized residuals were outside of the 95% confidence interval. Among the 13 studies, 4 studies evaluated cross-sucking (non-nutritive sucking between calves), 8 studies evaluated self-grooming (the calf cleans itself for hygiene purposes or as a form of thermoregulation), 8 studies evaluated the duration of drinking milk (time that the calf spent to consume milk with the offering method), 6 studies evaluated lying behavior (the calf is resting on the ground; head may be supported or unsupported by the neck) and 10 studies evaluated interacting with pen fixtures (the calf is in contact with fixtures or biting any fixtures of the pen; it's

scratching or licking). Offering milk from artificial teats did not affect cross-sucking (-0.098; CI=-0.58–0.38; p=0.693) and self-grooming (-0.098; CI=-0.34–0.15; p=0.427), compared to calves that received milk from an open bucket. However, calves that receive milk from artificial teats spent longer drinking milk (0.643; CI=0.32–0.96; p<0.0001), lying (0.385; CI=0.04–0.72; p=0.025) and showed lower interaction with the pen (-0.239; CI=-0.46–0.01; p=0.035) than calves that receive milk from an open bucket. Although offering milk from artificial teats did not affect cross-sucking, this practice stimulates the calves' natural behavior during calf nursing and can show positive effects on their affective states.

Effect of tactile stimulation at birth on the expression of maternal protective behaviour by Nellore cows

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This study aimed to evaluate the effect of tactile stimulation on the expression of maternal protective behaviour by Nellore cows immediately after birth. The study was carried out on a private farm located in the Northwest of the state of Mato Grosso, Brazil, by scoring the maternal protective behaviour of sixty-six cows, randomly distributed into two groups, those that received tactile stimulation at birth (TS, n=33) and those that did not (NTS, n=33). Tactile stimulation was performed on the calves' third day of life for one minute after being separated from their mothers and subsequent manual restraint for navel healing and identification. The assessment of the maternal protective behaviour was carried out when each of the calves was separated from their mothers, in the maternity picket, with the application of one of the following scores: 1) the cow is distant or moves away from the calf at the moment the cowhand approaches and remains distant during handling; 2) the cow is distant or moves away from the calf when the cowhand approaches and remains distant, but keeps his gaze directed at the calf; 3) the cow is close to the calf and does not move away, remaining close to the calf most of the time, she can smell or lick the calf and does not threaten or attack the cowhand; 4) the cow does not move away from the calf when the cowboy approaches and shows threatening behaviour, but does not attack the cowhand and 5) the cow attacks the cowhand and does not let him approach the calf. Score 3 is considered the best expression of maternal protective behaviour. Data were analysed using the chi-square test (χ 2). Cows with scores 1 and 2 and 4 and 5 were grouped due to the low frequency of these scores in the TS group, thus defining three scores (1+2, 3 and 4+5). Maternal protective behaviour differed significantly between the TS and NTS groups (χ2=19.87, DF=2, p<0.001), with 24% of TS cows and 15% of NTS cows assigned with score 1+2; 70% and 55% of TS and NTS cows scored 3, respectively; and 6% and 30% of TS and NTS cows scored 4+5, respectively. We concluded that

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tactile stimulation at birth affects the expression of maternal protective behaviour by Nellore cows, being a relevant factor for the safety of cowhands during handling.

Effect of water trough location on the drinking behaviour of dairy cows

Day Teixeira¹, Sachini Weerasinghe¹, Akinropo Adebayo¹, Matt Bell¹

Designing better management strategies when offering water for dairy cows can guarantee that animals of different social rank can have access to fresh water and use it according to their needs. The objective of this study was to evaluate the association between social hierarchy, milk yield, drinking behaviour (frequency, duration) and water intake of dairy cows when they have access to three water troughs (A and B: 330 I, 1.8 m length, 0.42 m width, 0.40 m depth; C: 190 l, 1.8 m length, 0.42 m width, 0.20 m depth) positioned in different locations [one at each end (A and C) and one in the middle of a cubicle housing (B)] in a rectangular free-stall barn system. The hypothesis was that dominant cows would drink more often from the trough positioned close to the area where the herd spent more time. The study was conducted at Hartpury University Farm using 59 first lactation Holstein Friesian cows over a two-month period. Agonistic Success Index was used to calculate dominance from agonistic interactions; water intake and milking yield were obtained from the farm records; camera traps and direct behaviour observation were used to record the frequency and duration of drinking at each water trough. Descriptive statistics from the first 11 days of data collection are presented. Preliminary data showed that 15 cows were considered high-ranking animals (index of displacements above 0.6), 25 were considered middle-ranking animals (index between 0.4 and 0.6) and 23 were considered low ranking animals (index of displacements below 0.4). Social rank does not seem to be associated with milk yield (high-ranking: 30.2 I/day; middle-rank: 29.7 I; low-rank: 31.6 I). Personal observation seemed to indicate that the social rank of cows did not influence preference to drink from a specific water trough, but it suggested that cows preferred drinking more often from the smaller trough positioned at the end of the stalls (trough C). Our preliminary findings could suggest that the location of the water trough plays a more important role on drinking behaviour of dairy cows than the social rank, however, further data collection and statistical analysis are needed to confirm such suggestions. Addressing these correlations will help to improve the water supply for dairy cows to ensure equal welfare standards across the herd.

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Effect of weaning on use of physical enrichment items and gene expression of dairy calves

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Differences in gene expression have been linked to variations in the stress response of rats and humans. Lower expression of serotonin and glucocorticoid receptors are associated with mood disorders and risk of depression, which can trigger a response of anhedonia. Considering the challenge of the weaning period for dairy calves, we explore the effects of this stressful period on the use of physical enrichment items and gene expression dairy calves. This exploratory study was carried out at the Institute of Animal Science, SP-Brazil. Seventeen F1 Gyr x Holstein female calves were housed in a collective pen with a rest area, one water trough, one automatic milk feeder, and one feeder. The physical enrichment items (brush, scratcher, tire, and chain) were available at the rest area during all experimental periods. Data were collected in three periods: before weaning (BW81; 81 days old), the day after weaning (DAW95; 95 days old), and a week after weaning (WAW101; 101 days old). During the experimental period, the physical enrichment usage (number of events and duration) was observed by video recording, and blood samples were collected to determine the expression of two receptors (mRNA, 2-ΔΔCT method), glucocorticoid (GR) and 5-hydroxytryptamine 2B (HTR2B). The use of physical enrichment items is presented descriptively, while duration and gene expression were analyzed by a generalized linear model with Gamma distribution. For the models, we considered periods as fixed effects and animals as random effects. There were 152 events of enrichment usage on BW81, 104 events on DAW95, and 165 events on WAW101. An estimated increase (p=0.001) of 44% in the usage duration of enrichment items was observed at DAW95 and WAW101 compared to BW81. The highest (p<0.05) average usage duration was registered on the brush (57.1 ± 11.3s; range: 3-280s), followed by the scratcher (49.1 \pm 8.6; range: 2–289s), the tire (46.1 \pm 8.9s; range: 2–277 s) and the chain (26.1 ± 5.6s; range: 2–121s). There was an estimated reduction (p=0.01) of 39% in the expression of HTR2B at DAW95 (0.28 \pm 0.03) compared to BW81 (0.46 \pm 0.05); while similar levels

(p>0.05) of HTR2B were found between BW81 and WAW101 (0.42 \pm 0.04). There was no influence (p>0.05) of the period on the expression of GR. The reduction in HTR2B might be related to short-term negative emotions after weaning; however, when raised in a complex environment, calves can seek enrichment items when motivated to do so.

The influence of pairing age on the occurrence of abnormal behaviors in Holstein calves

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Few studies have evaluated the effect of calf pairing age on the occurrence of abnormal behaviors. The aim of this study is to analyze the effect of pairing age on total non-nutritive oral behaviors, stereotypies, and bedding ingestion in dairy calves during the milk-feeding phase. A total of 140 Holstein calves housed on a commercial dairy farm (Paraná, Brazil) were included in the study. Calves were individually housed in pens from day one and subsequently allocated to different treatments: Early (E, n = 48; paired at 5-7 days), Intermediate (I, n = 48; paired at 29-31 days), and Late (L, n = 44; paired at 49-51 days). Behaviors were assessed five days per week, twice daily, post-feeding (at 8:00 and 15:00). Observations lasted 30 minutes with a 5-minute interval between observations (totaling 1 hour of assessment per day and 14 behaviors evaluated per day per calf). A total of 97,713 scans were conducted throughout the experiment. Data were analyzed using R software with mixed linear models. Initially, data were aggregated by summing the number of scans for selected behaviors over the entire period. To understand the temporal dynamics of these behaviors, daily data were modeled. Due to varied study durations among animals, the total number of scans was included as a covariate in the models. Treatment had a significant effect on the frequency of bedding ingestion (P<0.001), stereotypies (P<0.001), and non-nutritive oral behaviors (P<0.05). Calves in the Early treatment showed fewer instances of bedding ingestion (0.878 ± 0.415) compared to Intermediate (4.414 ± 0.411) and Late calves (5.773 ± 0.433) . The number of stereotypies was lower for calves in the Early pairing group (0.24 \pm 0.502) compared to Intermediate (2.67 \pm 0.513) and Late calves (2.57 ± 0.535). Early paired calves also exhibited fewer non-nutritive oral behaviors (36.9 \pm 3.46) compared to Intermediate (50.7 \pm 3.73) and Late calves (52.3 \pm 3.93). In conclusion, early calf pairing had a positive effect on reducing bedding ingestion, nonnutritive oral behaviors, and stereotypies in Holstein calves during the milk-feeding phase.

What bucket height for feeding calves do they prefer?

Gabriela Jimenes Stofella¹, Luana Quirino¹, Michail Sabino Moroz¹, Ruan Daros¹

Concentrate intake of calves is influenced by many factors. Thus, it is fundamental to analyse the impacts of different feeding practices that influence intake. Usually in dairy farms calves are fed concentrate in buckets, but bucket depth in relation to the concentrate level has rarely been associated with calf preference. This study aimed to assess the preference of calves for different feeding bucket's depth. Twelve individually housed Holstein female calves were divided in two groups: shallow bucket (concentrate 10,5cm from the top) and deep bucket (concentrate 21cm from the top). In both treatments the concentrate, hay and water were offered ad libitum since the first day of life. Calves received 6 litters of milk per day (until 75 days). The preference test was divided in adaptation phase and free choice phase, starting between 37 and 51 days of life. The adaptation phase consisted in providing the shallow or deep bucket for 4 days each (totaling 8 days in the adaptation phase). After this period the free-choice began between 11am to 14pm; the two buckets were placed side by side for 3 days. In the free choice phase calves were observed for 2 hours in days 1, 2 and 3. The first bucket choice in day 1 and its latency, total time feeding and concentrate intake was assessed in all days of adaptation and free-choice phase. Statistical analysis were performed in R using linear models and mixed linear models. The concentrate intake in the free choice phase was biggest in the shallow bucket (P<0.05), compared to the deep bucket, 761 ± 54.6 grams and 505 ± 54.6 grams, respectively. There was no difference the latency to approach shallow or deep bucket and in feeding time in each bucket, in this stage were consider 9 calves. The latency to first approach the deep bucket was 15.1 ± 8.02 and 4.2 ± 8.02 to approach the shallow bucket. The concentrate feeding time was 1.21 ± 0.5 minutes for the deep bucket and 2.41 ± 0.5 minutes for the shallow bucket. This research is still ongoing, but preliminary results suggest that bucket depth influences concentrate consumption.

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Estimation of social hierarchy of socially stable riverine buffaloes by using elimination approach along with resource restriction

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Buffaloes, like other social animals, form dominant hierarchies after indulging in agonistic interactions for possession of resources when loose housed in groups. This natural hierarchy fosters overall group stability and resource distribution, crucial for their welfare and production. Dominant hierarchy is usually estimated by outcome of dyadic interactions while exposing animals to limited resource. This approach gives reliable estimates when there are fewer animals but may not be feasible or yield reliable outcome when done in a large socially stable group of animals. Aim of this study was to estimate social hierarchy of loose housed large group of riverine buffaloes using elimination approach after offering opportunities of multiple agonistic interactions. We enrolled 63 loose housed (covered area=250m2; open area=500m2) lactating Murrah buffaloes (BCS=3.25±0.16; 1-6 parity) maintained at ICAR-National Dairy Research Institute, Karnal, India for the study. Possession of manger space while eating concentrate mixture offered as a first meal at morning after overnight feed restriction was considered as limited resource for observing agonistic interactions. The manger length (length x width: 4.0 x 0.6 m) was restricted to allow only 10% of animals to eat simultaneously for inviting maximum aggression. All animals had the equal opportunity from a distance of 10m to possess manger space during opportunity 1 (Opp1). Buffaloes which actively participated in Opp1 were separated from the group. Remaining buffaloes were given Opp2 for interaction and buffaloes still passive after Opp2 were given Opp3. Cumulative ranking of Opp 1, 2 and 3 arranged in chronological order of linear succession for one trail constitutes the social hierarchy of the herd. All agonistic interactions were recorded by CCTV recordings by continuous sampling for 35 minutes after offering of concentrate mixture. The trail was repeated five times at 3 days interval to evaluate its repeatability. A socio-metric matrix of all agonistic interactions was prepared and dominance values were calculated and arranged in ascending order where ranks of opp1 followed ranks of Opp2 and then that of

Opp3. Mean participation of buffaloes in Opp1, 2 and 3 for possessing limited concentrate feeding space was 26.03, 33.90 and 40.0% respectively. Mean collegan effects recorded during Opp1, 2 and 3 were 1.0, 1.2 and 2.0 respectively. Spearman's rank correlation coefficient of 0.99 (p<0.01) indicated reliable rank order between trails. We concluded that elimination method along with concentrate as limited resource gives reliable estimates of social hierarchy of a socially stable group of lactating buffaloes.

Maternal behaviors and interactions of cow-calf pairs on pasture during early life

Shawna Siql¹, Jennifer Van Os¹, Megan Woodrum Setser¹, Katherine Creutzinger¹

There is increasing concern from the public surrounding the separation of dairy calves from their dam at an early age. Understanding the natural behaviors of cow-calf pairs can be applied to inform how to meet the needs of animals in husbandry settings. Thus, the objective of this in-progress study is to collect preliminary data to characterize the behavior of cow-calf pairs in a semi-natural setting during the 2 wk after calving. In the summer of 2023, pregnant dairy cows were placed on pasture (219 × 278 m) prior to calving and kept on pasture with their calves for 2 wk after calving in a dynamic group of up to 6 cows and 7 calves. Data collection was completed on 2 cow-calf pairs; continual enrollment will occur until the target sample size of 15 pairs is met. Trees lined the pasture and variable forage was in the pasture; distribution of tall grass-covered and open areas were dynamic with rotational grazing. Video was continuously recorded with 8 cameras surrounding 2 sides of the pasture with infrared lights to increase visibility at night. Behaviors were recorded from video using 10 min scan sampling intervals and were classified as cow- or calf-specific behaviors. Focal cow behaviors included eating, lying, standing, and close proximity (≤ 1 cow length) to other cows, unrelated calves, and their own calf. Focal calf behaviors included eating grass, suckling their dam, lying, standing, hiding in tall grasses or trees, and close proximity (≤ 1 cow length) to other calves, unrelated cows, and their dam. Preliminary data is descriptively summarized for the first 2 d after calving (day 1 = 0 - 24 h, day 2 = 25 - 48 h). Focal calves hid for 28.1% (calf A) and 26%(calf B) of observations on day 1 calving and 22.2% (calf A) and 31.3% (calf B) of observations on day 2 after calving. Focal calves were in close proximity of their dams for 20.1% (calf A) and 21.2% (calf B) of observations on day 1 and 12.8% (calf A) and 11.5% (calf B) of observations during day 2 after calving. Detailed behavioral descriptions will improve our understanding of the natural behavior of cow-calf pairs and support future research to identify suitable management practices for cow-calf contact systems.

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Effects of staggered integration of freshly calved buffaloes in pairs into resident buffaloes on behaviour and performance

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Dairy buffaloes face agonistic interactions during group formation and regrouping in loose houses for exerting dominance. This has implications for their welfare and production. At buffalo farms fresh buffaloes are usually introduced into the resident lactating groups after colostrum period of 4-5 days directly as they calve individually. Aim of this research was to investigate effects of staggered entry of post-partum buffaloes in pairs into an established group on their behaviour, stress levels and productive performance. A total of 36 freshly buffaloes allotted at random to two groups were introduced 5 days post-partum into resident group (RG) of 70 to 75 milking buffaloes. Buffaloes in T0 (n=12) were introduced directly full time on d1 individually into RG and buffaloes in T1 (n=24 in 12 pairs) were introduced into RG in a staggered manner in pairs spread over 4 days. On d1, 2 and 3 these buffaloes were kept in RG for only 2, 4 and 8 hours respectively and withdrawn thereafter and housed separately in loose house with similar housing and feeding opportunities. On d4, these buffaloes were left in into RG for full time. Significance of differences in means of experimental measures recorded daily continuously for first 4 days of introduction and then at weekly intervals continuously for 24 hours by Close Circuit Television Cameras over 100 days was tested by one-way analysis of variance in SSPS statistics 20. Overall, T₁ buffaloes exhibited higher (P<0.05) daily feeding time (340.81±6.55 min/d), rumination time (432.52±9.53 min/d) and total lying time (611.09±6.11 min/d) than TO (319.56±9.38; 390.66±15.04 and 558.50±10.86 min/d) respectively. T0 buffaloes exhibited higher (P<0.05) standing time (881.50±10.86 min/d) and idle standing time (355±23.53 min/d) than T1 (828.93±6.10; 283.22±25.69 min/d) respectively. Agonistic interactions (fighting, butting, pushing, threatening, avoiding) and plasma cortisol concentrations were higher (P<0.01) in T0 (12.21±0.80 ng/ml) than in T1 (6.59±0.59 ng/ml). Daily times spent on feeding, rumination, resting and number of agonistic interactions between two groups was similar after 3 to 4 weeks of introduction indicating their social integration into RG. T1 buffaloes demonstrated higher (P<0.01) daily milk yield (8.91±0.67 vs. 6.17±0.98 kg/d), higher (P<0.01) milk flow rate (0.977± 0.04 vs. 0.861±0.037

kg/min) and lower (P<0.05) somatic cell counts (2.01±0.11 vs. 2.58±0.13 lakhs/ml) than T0 buffaloes. In conclusion, staggered introduction of freshly calved buffaloes in pairs into resident buffaloes positively influenced behaviour and welfare, reduced social stress and improved their productive performance.

Poster + One-Minute Presentation Session 2: Wildlife Behaviour, Management and Welfare + Companion Animals Behaviour and Welfare

Tuesday, July 23rd, 12:00-12:15

Evaluation of the activity budget of Captive Olive baboon (*Papio anubis*) in Audu Bako and Aldusar Zoological Gardens in Kano and Katsina States, Nigeria

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Olive baboons (Papio anubis) are social primates found in various habitats in Africa. Understanding their activity patterns and budget is critical in comprehending their behaviour and ecology. The study aimed to explore the activity budget of Olive Baboon and the variation in the two zoological gardens. The study was conducted for 8 weeks between March and May 2022, totalling about 16 hours with 2 hours of weekly observation. The observation was done for one of the three distinct time spans each day: Morning (8:00-10:00 am), Afternoon (12:00-2:00 pm) and late afternoon (2:00-4:00 pm). Purposive sampling was utilized to select baboons, with a single observer engaging in focal sampling between the two gardens. In Audu Bako, a pair of adult male and female baboons were observed, while in Aldusar, three adult baboons, comprising one male and two females, were studied. Descriptive statistics were applied to analyse the data. Male baboons in Audu Bako exhibited sitting as the highest activity at 44.79%, with yawning being the lowest at 0.03%. Female baboons in Audu Bako spent 56.25% of their time sitting, while playing accounted for only 0.83%. In Aldusar, female baboons' dominant activity was sitting 32%, while vocalization was the least observed activity at 0.85%. Male baboons' primary activity in Aldusar was pacing at 22.22%, with teeth baring being the least at 1.94%. Activity budgets were compared between zoos and time periods. Locomotion was the dominant activity in Audu Bako during the afternoon (55%), transitioning to rest (38.83%) in the morning and subsistence activities (13.64%) in the late afternoon. In contrast, other behaviors prevailed in Aldusar between 8:00-10:00 am (65.67%), followed by locomotion (52.43%) in the late afternoon. The activity budget of the observed animals was found to exhibit some changes across the two study areas. Location, time and gender were

found to have an influence over the dynamics of the animals between the zoos and within species. Excessive sitting among the baboons was identified which highlighted some underlying welfare issues in the zoos as social behaviour which is commonly found among the baboon species was found to be rare. Effort to improve the welfare of the captive baboon species is needed to ensure the attainment of the conservation principles on which the zoos were established. 24-hours studies on the baboon and the provision of wider enclosures with enough environmental enrichment objects is encouraged.

Water Buffalo (*Bubalus bubalis*) as a spontaneous animal model of Vitiligo

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Vitiligo is a multifactorial disorder with spontaneous onset, yet existing laboratory animal models predominantly focus on transgenic or autoimmune aspects. This research underscores the significance of spontaneously occurring depigmentation in water buffaloes (Bubalus bubalis) as a near match to human vitiligo. The objective is to establish water buffaloes as a reliable animal model for studying the development of vitiligo and behavioural aspects. Female healthy adult buffaloes of Pandherpuri/Mixed/ breeds were selected for the study. Lesional and non-lesional skin samples were collected from each buffalo. Clinical diagnosis of vitiligo conducted was using Wood's lamp, followed by histopathology: immunohistochemistry and electron microscopy. Additionally, gRT-PCR was done for the genetic aspects of the disease. To observe the buffalo, we used cameras on the farm to capture video of their daily movement. For measuring milk production, we manually measured the weight of milk daily for two months. This comprehensive methodology aims to elucidate the similarities between buffalo depigmentation and human vitiligo, as well as their natural behaviour. Biopsies from lesional and non-lesional sites revealed a complete absence of melanin and melanocytes in lesional skin, confirmed by Masson Fontana staining and S-100 immunohistochemistry. During ultrastructural analysis, transmission electron microscopy revealed a few melanosomes in the affected skin layers. Gene expression analysis demonstrated reduced expression of key melanogenic genes in lesional skin. The study suggests buffaloes as potential models for vitiligo research. The study revealed that among the six buffaloes with vitiligo, 2 exhibited changes in their behaviour concerning light and dark cycles. Buffaloes with vitiligo tended to spend longer durations in dark or shady regions compared to healthy buffaloes (p<0.05), possibly due to vitiligo-related complications. The eating patterns of these two buffaloes were not significantly different. The vitiliginous buffaloes preferring dark had similar feed intake as those preferring light. However, a slight decrease in milk production was observed in the two buffaloes preferring dark compared to the buffaloes preferring light (p=0.08). Another exciting activity that was observed in my study

was related to the place of stay of buffaloes, buffaloes remain standing during their maximum day duration. They all used to stay on the flat ground during their leisure time. We present evidence suggesting that buffaloes could be a valuable animal model for investigating vitiligo. The comprehensive analysis, including histological, ultrastructural, cytological, immunohistochemical, understanding of innate behavioural changes, and molecular data, indicates that the naturally occurring depigmentation in buffaloes closely resembles human vitiligo.

Behaviour and brain activity (EEG) in Nile Tilapia exposed to different preslaughter procedures

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In Brazil, humane slaughter regulations do not cover fish, and most fish slaughterhouses use live chilling before slaughter. This study aimed to assess unconsciousness using behavioural and electroencephalogram (EEG) measures in Nile tilapia exposed to different pre-slaughter procedures. All procedures were previously approved by the Ethical Committee (4446150817/2017). Nile tilapia $(563.7 \pm 182.3 \text{ g}, 30.7 \pm 2.9 \text{ cm}, n=85)$ were divided into two pre-slaughter procedures for behavioural analysis: anaesthesia with Lippia alba (LA) (n=10) at 500 mL/L (65.2% linalool) until deep anaesthesia followed by 60 s of air exposure, and live chilling (LC) (n=10) in a tank containing water/ice mixture (1:2) at 1-2 °C for 20 min followed by 60 s of air exposure and euthanasia. Swimming, equilibrium, response to handling, response to painful stimuli, vestibulo-ocular reflex and breathing were assessed to establish the stage of deep anaesthesia. For EEG, fish were restrained, and lidocaine was injected before the insertion of electrodes. Baseline recordings were obtained from conscious fish in water, followed by exposure to 2-phenoxyethanol at 1 mL/L (PE) (n=21) for 10 min, or Lippia alba (LA) at 500 mL/L (n=23) for 10 min, or LC for 20 min (n=13). After recording, all fish were euthanized. PowerLab and the Labchart were used for EEG recording and data analysis (AD Instruments, Australia). For spectral analysis, the total power (Ptot) and spectral median frequency (F50) were calculated in one fish per group in epochs of 2 s from the filtered trace, excluding movements. The behaviour showed time required to reach the level of deep anaesthesia of 128.6 \pm 46.4 s for LA and 866.0 \pm 200.1 s for LC. In addition, 4/10 in the LC group reacted to a painful stimulus during the 60 s of air exposure, suggesting the induction of immobility rather anaesthesia. In groups exposed to PE and LA, both Ptot and F50 values exhibited a decrease at 3 min of exposure. In contrast, in LC group, Ptot decreased while F50 increased over time, indicating immobility with pain perception rather than unconsciousness. Anaesthesia in Nile tilapia using Lippia alba essential oil or 2-phenoxyethanol induced

unconsciousness after 180 s of immersion, as determined by EEG. The use of live chilling at pre-slaughter of Nile tilapia fails to induce unconsciousness and should be urgently replaced by methods that align with humane slaughter criteria. More research is needed to replace the use of LC with humane methods on a commercial scale.

Behavioral repertoire of Brown dog tick (*Rhipicephalus sanguineus*) and climate change

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The Rhipicephalus sanguineus tick is one of the world's most relevant transmissible disease vector, both to humans and domestic animals. The main transmitting diseases includes Ehrlichiosis and the Mediterranean and Rocky Mountain Spotted fevers. The species has gone through a recent taxonomical review, considered now to be divided in at least two lineages with different geographical ranges (tropical and temperate) and possible contrasts in habits and behavior. This study aimed to stablish and compare the behavioral repertoire of two lineages of R. sanguineus, focusing on evaluating the rates of spontaneous host seeking behavior. A total of 24 focal ticks were observed in 10 sessions per day, throughout 24 days from September 22 to November 29, 2022. Ticks were kept in six different terrariums, three containing individuals from the tropical lineage and three from the temperate lineage. Inside each terrarium a piece of cotton was maintained wet each week for hydration along with a "plant model" containing four branches, each with a different colored leaf. The data collected was analyzed through linear mixed regression models on R Studio software. The tick's position and behavior were tracked along with the temperature and air humidity at the laboratory in the moment of the observation and how it influenced the tick's behavior. None of the positions or behaviors analyzed showed to be influenced by the tick's lineage. Out of the covariates considered, only the increasing of air temperature influenced the tick's towards placing themselves at the lid (p=0.03) and at the base (p=0.04) of the terrarium instead of the walls or the plant-model. These results show that the R. sanguineus behavior as quite similar between the two lineages when not exposed to an artificial bait, which contrasts with their clear distinct questing patterns when the bait is present, as established in previous analysis in this research project. This type of inquiry may prove to be relevant due to the expansion of the geographical range of R. sanguineus due to climate change and the unknown consequences that it should imply, along with the lack of behavioral analysis of this species post its systematic review.

Assessing the severity of Sporotrichosis on feline exploration and human-animal interaction

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Sporotrichosis is a zoonosis that severely affect cats and requires the isolation of infected animals during long treatment periods. This study aimed to evaluate how treatment time and lesion severity affect the behaviour of hospitalized cats. Eight cats (male = 5, female = 3) were observed at the university veterinary clinic's infectious diseases isolation ward. Cats were housed individually in stainless steel cages under the same environmental conditions and medical treatment routine. All cats were submitted once an open arena, novel object, and unknown human test. The tests occurred in a 3.50m² arena, divided into 49 quadrants, located apart from the isolation ward; cats were carried in individual carrier boxes by the researchers. All tests were video recorded and BORIS® software was used to evaluate the total of quadrants explored, number of vocalizations, latency to approach a novel object, unknown human, and duration of contact in seconds. Pictures of the injuries were taken two weeks before experimental period and used to measure lesion severity and the extension using a lesion score (0 = no lesion; 30 = severe lesions in all body parts) developed by the authors. Nodules, crusts, and ulcers scored one point, while mucosal or tumour-like lesions, presence of necrosis, and muscular or bone tissue exposure scored two points reaching a maximum of 30 points. Based on the average number of injuries, cats were categorized into two groups: mild $(\mu < 8.5)$ and severe $(\mu > 8.5)$. Cat's time under medical treatment was evaluated in weeks to assess behavioural changes. Subsequently, cats were divided based on the average duration into short (μ < 10.25 weeks) or long (μ > 10.25 weeks). Statistical analyses were performed in R using linear regression. Cats with severe lesions explored fewer quadrants than cats with milder lesions (p-value = 0.10). Similarly, the group with a longer treatment time explored fewer quadrants than the short treatment time (p-value = 0.14). No cats presenting severe lesions approached the human in the unknown human test, while all cats with mild lesions approached the unknown human. There were no significant differences found in other analyses. We observed that cats with severe lesions show cautious behaviour, exploring less

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and avoiding unknown humans, while longer treatment times are linked to reduced exploration; overall, lesion severity and treatment duration influence the behaviour of infected cats, suggesting potential behavioural changes from the disease and treatment.

Can environmental enrichment improve cat behavior? A perspective from veterinarians and managers of Brazilian shelters

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Animals in shelters commonly face low welfare and environmental enrichment activities (EEA) may be useful to improve this condition. We studied how often EEA are offered to cats in Brazilian shelters, whether there is an assessment of these enrichments, and if there is a relationship between EEA and behavioral problems. An online questionnaire was applied to veterinarians and managers of shelters through social networks, email and phone calls, with 116 valid responses. Overall, 51.7% of respondents reported performing EEA, while 42.2% did not, and 6% were unable to answer; 59.9% of respondents reported that one or more cats in their shelter presented behavioral problems, 39.6% that no cats presented such problems, and 0.9% that they did not perform behavioral assessment. The statistical analysis was carried out using Pearson's chi-squared test, considering 95.0% confidence, with a value of P<0.05. There was a positive association between behavior problems and EEA (x^2 , p=0.004) and a trend for a negative association between EEA assessments and behavior problems (x^2 , p=0.052). As for EEA assessment, 43.3% performed an assessment by observing the cat's behavior and checking for interaction with the enrichment, 41.7% of respondents reported that they did not perform any and 15.0% reported that they observed the cat's behavior, checked for interaction with the enrichment, and recorded the behaviors. As a large proportion of respondents are not monitoring EEA results, there is scarce information on their effectiveness. The positive correlation between behavioral problems and EEA may be related to either the use of EEA as a treatment for cats previously diagnosed with behavioral problems or a lack of behavioral assessment in places that do not perform EEA, or both. Thus, the frequency of EEA provision and the guarantee of its effectiveness through monitoring seem critical points for improved welfare of sheltered cats in Brazil.

Exogenous cortisol and residency affect the aggressive behavior in yellow tail lambari (*Astyanax altiparanae*)

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Social dominance hierarchies have been observed in several gregarious species, including aquatic animals. Aggressiveness is often used to establish and maintain these hierarchies, which determine which animals have privileges over group resources. However, the causes and consequences of aggression in fish are not well understood. Cortisol, which is involved in the stress response, is also involved in neural mechanisms, and can increase aggressive responses in groups of animals. In addition, residency in a particular environment can also increase aggressive behavior, as fish have a greater capacity and motivation to fight for an environment they have already conquered. Intra-specific aggressive behavior is an important factor in the expanding Brazilian fish farming industry. The yellowtail lambari (Astyanax altiparanae), which is increasingly produced in country, exhibits aggressive behavior, especially in females, resulting in economic losses. Here we evaluated the effects of exogenous cortisol and previous residency on the aggressive behavior of female yellowtail lambari. Juvenile fish $(9.2 \pm 6.4 \text{g})$ and $8 \pm 0.3 \text{cm}$ were divided into four experimental groups: RT1 (residency without hydrocortisone), NRT1 (non-residency without hydrocortisone), RT2 (residency with hydrocortisone) and NRT2 (non-residency with hydrocortisone). The fish received a peritoneal injection (no mortality was found after the injection) of either T2 hydrocortisone or T1 vehicle (control) and were isolated in individual aquarium for residents (where they stayed for three days for acclimatization after the injection) or re-grouped in novel tanks for non-residents (acclimatization for three days before being transferred to a novel aquarium). After the acclimatization period, the fish were directly confronted with another conspecific that had not been treated or conditioned. The confrontation was recorded for 20 min to analyze aggressive behavior using a species-specific ethogram. The fish were then euthanized with an overdose of anesthesia and blood was collected for analysis of plasma cortisol concentrations. The study showed that elevated levels of the hormone in blood

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caused by hydrocortisone administration, and prior awareness of the environment, caused by the housing condition, increased the aggressiveness in the species. In contrast, non-resident animals were less aggressive, which may be related to the alertness induced by the new environment to which the females were exposed. These results suggest that cortisol and housing conditions directly influence the aggressiveness of female yellowtail lambari. Further research is needed to establish strategies to reduce this aggressive behavior, such as environmental enrichment, and improve farming conditions and animal welfare, as well as reduce economic losses in fish farming.

Identification of most motivating resource for estimation of social hierarchy in water buffaloes

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Buffaloes are social animals and live in herds under natural settings. Under husbandry conditions they are loose housed in groups for providing them opportunity for expression of social behavior. Construction of social hierarchy and knowledge of its determinants is warranted for determining the optimum group size and formulating strategies for shifting of animals from one group to another. Social hierarchy is normally estimated by recording agonistic interactions among all animals for occupancy of limited resource. Aim of this study was to find out the most limiting resource for estimation of social hierarchy in riverine buffaloes. The study was conducted on 63 loose housed (covered area=250m²; open area=500m²) lactating Murrah buffaloes (BCS=3.25±0.16; 1-6 parity) maintained at ICAR-National Dairy Research Institute, Karnal, Haryana, India. Three limited resources considered for observing agonistic interactions were i) seasonal green fodders ii) concentrate mixture and iii) occupancy of space under the sprinklers during summer season (11:00; 07:00 and 14:00 hours respectively). The manger length was restricted to allow for 10% (length x width: 4.0 x 0.6 m) of animals to eat simultaneously and 10% of sprinklers (3.7 m² space for 2 sprinklers) were operated to provoke maximum aggression. Each restriction was carried out once daily in the same group. Access to resource was allowed from 10 m to ensure equal opportunity for occupancy. Agonistic interactions (physical- fighting, bunting, pushing and non-physicalthreatening, avoiding) for each resource were recorded manually and by Closed Circuit Television recordings five times at weekly intervals by continuous sampling for 1 hour. A sociometric matrix of all agonistic interactions was prepared and pooled data was analyzed using one way ANOVA in SPSS version 22. Participation of buffaloes for occupancy of limited resource was higher (p<0.01) for concentrate mixture (40.32±0.86%) than for green fodders (25.08±0.66%) and sprinklers (12.38±1.80%). Number of agonistic interactions were significant (p<0.01) for occupancy of concentrate (25.4±0.54) followed by green fodders

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(15.80 \pm 0.42) and sprinklers (7.80 \pm 1.13). Number of physical and non-physical interactions were higher (p<0.01) for concentrate (439.4 \pm 13.49, 368.8 \pm 12.16) than green fodder (126.6 \pm 9.49, 147.8 \pm 90) and sprinklers (36.90 \pm 7.22, 83.5 \pm 6.61). We concluded that concentrate restriction motivates the most for expression of agonistic interactions for resource occupancy in a socially stable lactating buffaloes.

Poster + One-Minute Presentation Session 3: Farm Animals Behaviour and Welfare (Pigs)

Tuesday, July 23rd, 16:15-16:30

A pilot study: Are environmental enrichment and feeder interaction in early nursery indicators of successful pig transition?

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When transitioning into the nursery, some pigs fail to thrive. These pigs often need additional support such as antibiotics, additional heat sources, or movement to specialized care pens. Early identification of individual pigs that require additional support may improve their welfare and survival. Producers are trained to observe pig behavior during daily walkthroughs. III pig behaviors include decreased environmental interest and reduced feeder usage. It's unclear if there is a link between observation of these ill pig behaviors and pigs unsuccessfully completing nursery. Therefore, the objective of this study was to determine if pigs that were unsuccessful in nursery, were less likely to interact with enrichment and feeder during the first hour after enrichment placement for the first three days of nursery. The project was approved by the animal care and use committee. Twelve mixed-sex pigs (Camborough 1050 X 337, PIC) were retrospectively allocated to a pulled treatment, defined as caretaker intervention (n = 6), and a success treatment, defined as no caretaker intervention (n = 6) at nursery conclusion. Each pulled pig was matched to a successful pig in the same pen based on their body weight at nursery placement (19 to 24d). Pigs were randomly sorted into six pens (~0.29 m²/pig). Each pen was equipped with a 4-hole, dry selffeeder (0.61 m long). Each feeder had two cotton ropes, strung with two enrichment biscuits, dipped in a jam solution. Enrichment and feeder interaction were collected using continuous sampling, and a one zero recording method. A pig was considered interacting with the enrichment if they had snout contact with the enrichment device. A pig was considered interacting with the feeder if their head was over the feeder tray and nose down in the feeder. Individual pig frequency of enrichment and feeder use was calculated using the FREQ procedure in SAS. On day one, 3 (50%) and 5 (83%) of pulled pigs interacted at least once with

enrichment and feeder respectively, compared to 2 (33%) and 6 (100%) successful pigs. On day two, 5 (83%) and 4 (66%) of pulled pigs interacted at least once with enrichment and feeder compared to 6 (100%) and 6(100%) successful pigs. On day three, 1 (16%) and 4 (66%) of pulled pigs interacted with enrichment and feeder at least once compared to 4 (66%) and 4 (66%) successful pigs. In conclusion, interacting at least once with the feeder appears to be a potential indicator of pig nursery success over this enrichment.

Does olfactory enrichment affect pigs' reaction to a novel food item?

Cady W. Chan¹, Johanna Stenfelt¹, Maria Vilain Rørvang¹

Pigs are known to be hesitant or cautious when trying new food items, a phenomenon known as food neophobia which can be defined as weariness or refusal of ingesting strange feed. Pigs may be more curious to novel food if provided enrichment stimulating the gustatory senses. This MSc-thesis project developed a Novel Food Test to assess pigs' reaction toward a novel food item, and investigated if pigs' reactions differed consistently between treatments. The study included 800 pigs housed in pens with 10 littermates. Half of the pens had odourised straw; straw sprayed with one of five different odours: ginger, lavender, thyme, pine, or aniseed, with either odour changing weekly (O1: n=20 groups) or odour changing daily (O2: n=20 groups) provided in a rack during the growing period (13-15 weeks). The other half of the groups had odourless straw (mineral oil) either provided on the floor (C1: n=20 groups) or in a rack (C2: n=20 groups). Treatment pens were not isolated. The pens were tested once in week 7-10 of the growing period (mean: 8.5 weeks), and behaviour was recorded during the 5 minutes test duration (from delivering the novel food item to 5 minutes after). The test consisted of distributing 10 equally large carrots in the feed trough of the pig pen (carrot to pig ratio 1:1) as carrots were assumed to be biologically relevant to pigs yet novel for commercial pigs. The pigs' behavioural reaction was scored on a scale from 1=quick interaction to 5=no interaction, and pigs not engaging (not looking or moving) were excluded (n=75 pigs). The hierarchy within the pen was not analysed. The majority of pigs received a score 1 (70%), and only 0.4% received a score 5. As pigs' reactions within a pen were not independent, the scores were summed and a mean reaction per pen was calculated. Data was thus analysed at pen level (ntotal=80, 20 pens per treatment), using a two-way Anova with reaction as the response, and treatment and pen ID as the factors. No significant effect of treatment or pen ID was found on pigs' reaction towards the novel food (p-values > 0.1). These results indicate that odourised straw did not impact finishing pigs' reaction towards carrots as a novel food item. The Novel Food Test serves as a starting point in developing

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testing regimes to investigate the impact of odourised enrichment on growing pigs' behaviour.

Gilts prefer an open pen to a stall

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Stalls or crates are a very common type of housing used on pig farms that restrict an animal's movement. How this confinement impacts the animal's affective states is seldom investigated. As preference testing is a common way to investigate affective states, our objective was to quantify the preference of gilts between an open-area and stalls when free access is provided. A secondary objective was to study the relationship between this preference and personality traits. We conducted a preference test over 7 days where trios of gilts (n = 10 trios, 27.4 ± 1.5 weeks old) had continuous free access between individual selfclosing stalls (~1.2 m2) and a shared open area allowing 2.8 m2 /animal. Gilts had access to ad libitum feed and water both inside the crates and in the open area. After seven days, personality traits of the animals were assessed with open field (OF) and novel object (NO) tests. While a small fraction of the gilts divided their time between pen and stall, most of the animals spent the majority of their time in the open area. The median time outside of the crate was 95.2%. Gilts, however, were likely to spend less time in the open area at night compared to day (Odds Ratio = 0.49, 95CI= [0.40, 0.60]), as well as experimental days passed (OR = 0.70, 95CI = [0.66, 0.73]). Average preferences for the open area were highest during the first night (93.8 \pm 22.2%) and lowest during the seventh night (68.7 \pm 44.8%). Analysis of personality tests through Principal Component Analysis (PCA) yielded two main components which we defined as Passivity and Engagement. Passivity had no relationship with preference for the open area, but engagement during OF/NO was associated with less use of the open area (OR = 0.39, 95CI = [0.25, 0.60]). We conclude that gilts have a clear preference for an open area, but not a total aversion for stalls when free access is provided between the two, perhaps as stalls provide opportunities for voluntary social separation. This preference appears to be influenced by both intrinsic and extrinsic factors.

Impact of air quality on damaging behaviour, lesions, stress indicators, and growth in weaner pigs

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Air quality (high levels of ammonia (NH₃), carbon dioxide (CO₂), methane (CH₄), hydrogen sulfide (H₂S), or suspended particulate matter (PM)) can negatively affect pig health, and consequently, their welfare and productivity, being also considered risk factors for damaging behaviours such as tail biting. The objective of this study was to evaluate the effect of air quality on the welfare of weaner pigs, specially focusing on damaging behaviours. A total of 810 piglets divided into 9 batches were evaluated in two indoor weaner farms at the beginning (4 weeks of age) and end of the weaner phase (8-9 weeks of age). For each batch, three identical rooms were used, and 30 pigs/room were individually identified (90 pigs/batch) and housed in two fully-slatted floor pens (15 pigs/pen) at 0.22 m²/pig. The air quality parameters assessed inside the barn once a week were: NH₃, CO₂, CH₄, H₂S, PM_{2,5} and PM₁₀. The animal welfare measures were adapted from the Welfare Quality protocol and assessed at the individual level (90 pigs/batch) were: weight, body condition score, skin condition, tail and ear lesions, and manure on the body and at pen level (6 pens/batch): thermal comfort, respiratory, and digestive measures. Salivary samples for cortisol and Adenosine deaminase (ADA) analysis were taken for 5 pigs/pen (30 pigs/batch). Linear mixed-effects models were employed to analyse the difference in welfare indicators between the two evaluations. For CO₂ and NH₃, the mean concentration for each room was calculated and threshold levels were used to classify the pens as being "over" level (average concentration above 3000 ppm for CO₂ and 20 ppm for NH₃) or "under" level. For all the gases, data was analysed for the linear relationship with welfare indicators. The key findings revealed a significant increase in diarrhoea (marginal effects: -0.24 "under", 0.37 "over", p.adj.<0.01), skin lesions (0.04 "under", 0.74 "over", p.adj.<0.05), and ear lesions (0.19 "under", 0.96 "over", p.adj.<0.01) with regards to CO₂ levels. Both skin (coefficient: 3.23×10^{-4} , p.adj.< 0.05) and ear (3.38×10^{-4} , p.adj.< 0.05) lesions were also found to linearly increase with CO₂ levels. Coughing rate was also found to increase in a linear

fashion with higher concentrations of PM_{10} (coefficient =0.11, p.adj.<0.001). No significant effect was found for the "over" NH_3 group (p.adj.>0.05). These results indicate a potential correlation between CO_2 levels and specific animal welfare indicators, one of those being ear lesions, which highlight the importance of developing strategies and technologies to reduce indoor gas levels.

Impact of confinement on the behavior and cortisol chronobiology on gilts

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Common husbandry practices on farms today have the possibility to compromise animal welfare. Confinement can inhibit the expression of species-specific behavior and challenge natural homeostatic activities. Many breeding females on pig farms remain confined in individual stalls or crates. These crates serve to restrict the animal's social, explorative, and maternal behavior and likely induce stress. Here, we studied how the behavior and circadian rhythm of cortisol (CR) in naïve gilts entering the herd were impacted by individual stall housing. Twelve replacement gilts (TN70-Topigs) were enrolled in a within-subject repeatedmeasure designed study. Gilts were housed in individual crates for four weeks (Wk1 to Wk4). Twice per day, saliva was collected (7:00 and 18:00), and behaviors were observed (7:30 and 17:00) on three consecutive days in each of the four weeks. Salivary cortisol concentrations were determined using a Cortisol Enzyme Immunoassay Kit (Salimetrics®) and a ratio calculated between morning and evening samples. Three 2-minute videos taken during each observation session (totaling 12 minutes/animal/day) were coded using focal continuous recording. The behaviors observed were standing, sitting, lying down, moving back and forth, pawing vertical surfaces, rooting attempts, biting, and abnormal oral behaviors. Linear mixed models were used to analyze outcome variables with 95% confidence, using each behavior as the dependent variable, multiple variables as fixed effects (week, day, and reproductive state), and individuals as a random factor. CR appeared disrupted Wk1 with ratios near to 1 and were significantly less than values at Wk3 and Wk4, suggesting a normalizing of cortisol chronobiology over the time following the initial introduction to the stalls. However, standing, moving, and rooting increased, while lying down decreased in the morning during the study, whereas in the afternoon, the pawing of vertical surfaces increased. No other behavioral differences were observed. The behavioral drive for species-specific behavior, such as walking and rooting, seemed to increase during the study despite the confinement of the crate thwarting the realization of these behaviors by the gilts. It is interesting that the disruption to the animal's physiology appears to be affected immediately by confinement and ameliorates

over time, whereas its impact on the psychological health of the animal compounds as they become more restless with time in the crate.

Judgment bias testing in piglets using an automated versus a conventional method

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Emotions can be assessed using physiological, behavioral and cognitive methods. The objective of our study was to develop and validate an automated version of the active judgment bias test (JBT), in which weaned piglets voluntarily interact with a test apparatus in their home pen. We predicted that automated JBT results would replicate previous studies, in which both pen enrichments and positive human interactions induced positive (optimistic) judgement bias in conventional JBTs. After piloting and refining the automated apparatus over 16 pens of 4 piglets, we implemented standard (STA) and enriched pen treatments (ENR: peat moss and toys), and regular positive (HAR+: gentle contacts and speak) or negative (HAR-: rough contacts, chasing) human interactions, in a factorial design (16 ENR/HAR+ piglets, 16 STA/HAR+, 15 ENR/HAR-, 16 STA/HAR-). Half of the piglets (n=31) were tested with the automated JBT, and the others (n=32) with a conventional active JBT. For the automated JBT, no piglet reached our learning criterion of 80% success (46±10% success rate (mean±sd)). During the learning phase, voluntary interaction with the apparatus decreased over time (143±212 trials initiated on the first three days, 107±106 over three days mid-phase, 64±65 on the last three days). Trials were very frequently interrupted by another animal entering the apparatus (41±13% trials). Neither enrichment nor HAR affected learning success (ENR/STA Mann-Whitney test U=121, P=0.98; HAR+/HAR- U=116.5, P=0.90) or the number of trials initiated per animal (963 \pm 678 trials, ENR/STA: U=139, P=0.46; HAR+/HAR-: U=120, P=1.0). The conventional JBT required on average 13 habituation sessions to the test pen, so the learning phase began only after the two-week period in which HAR+/HAR- treatments were applied. Subsequently, analyses below focus on ENR/STA treatment effects. STA piglets learned the positive stimulus-reward association more rapidly (after 9 sessions, 9/16 STA vs. 3/16 ENR piglets had reached the learning criterion, U=116, P=0.01). However, ENR piglets performed better when the negative stimulus was introduced (ENR: 36±8% success, STA: 11±5%, U=96, P<0.001). We hypothesize that higher motivation to participate among STA piglets promoted

initial learning, but that they were less cognitively flexible when a second contingency was added. Enrichment influenced learning in the conventional JBT, but this could not be assessed in the automated JBT. Ongoing efforts to find ways to keep piglets motivated to learn, and prevent interruptions, will help to reach the learning criterion and assess emotions efficiently and continuously using the automated JBT.

Housing systems for boars alter vocalization patterns of their offspring in behavioural tests

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The environment of male parents can alter their offspring's development through epigenetic mechanisms and possibly influence their coping behaviors and affective states. This study aims to assess the emotionality of piglets sired by boars reared under different housing conditions. Adult male pigs were placed in either crates (C; N=6), pens (P; N=6), or enriched pens (E; N=6). After four weeks, semen was collected, pooled across all treatments, and used to inseminate 13 group-housed gilts in an outdoor system. Male piglets (N=52) born from these matings were tested at 25 days of age. As vocalizations provide one avenue of insight into an animal's affective state, acoustical recordings were conducted during a variety of behavioral tests including open field (OF), novel object (NO), and elevated plus maze (EPM) tests. Two trained researchers used Raven® Pro 1.6 to quantify and to classify 33,876 piglet vocalizations. Experimenters were blinded during analysis, as treatment allocation was determined at the end of the study by a post-hoc paternity test that linked piglets to their father's housing condition. Linear mixed models were used to identify differences (p<0.05) in vocalization duration during the tests contrasting boar housing conditions. The call durations were longer in the piglets sired by boars from treatment C than in E piglets during EPM (p<0.001) and trend in the contrast of the same treatments in the OF (p=0.07). No difference was found in the NO test. The consistent long-duration nature of vocalizations by offspring from boars housed in crates suggests a more aversive experience for these animals during the behavioral tests. Classifying calls into different types can help to understand better the emotions of piglets at the interface of context and genetics. Our study showed for the first time that boar housing can influence vocalization performed by their piglets during behavioral tests and suggests a less effective coping style for pigs whose fathers were reared in confinement.

Use of probiotics and their effects on the health of sows in different reproductive phases

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Sows often experience various stressful factors that disrupt their intestinal microbiota, potentially altering health parameters and, consequently, behavior. Therefore, incorporating probiotics into the diet of these females may help mitigate these adverse effects by modulating the microbiota. The objective of this study was to evaluate the effect of a probiotic on health parameters of sows at different stages of reproduction, using observations based on the Welfare Quality® protocol. For that, forty-seven sows housed in collective pens were divided into 4 treatments: T1) control group (no additives in the diet [n=11]); T2) probiotics added to the diet throughout the gestation period (from day 1 post-insemination) and during lactation (n=12); T3) probiotics added to the diet from the second third of gestation (day 38 of gestation) and during lactation (n=12); and T4) probiotics added to the diet from the last third of gestation (day 76 of gestation) and during lactation (n=12). The probiotic (Enterococcus faecium, Lactobacillus acidophilus, and Lactobacillus plantarum) was administered at doses of 1g/animal/day during gestation and 6g/animal/day during prepartum and lactation phases. The sows were evaluated three times for health parameters: at the beginning of gestation, and at the beginning and end of lactation. Parameters assessed included body condition, presence of lesions and bursitis, lameness, enteric diseases, skin inflammation, and dirtiness. Microbiological analysis of feces was also conducted during the same evaluation periods. Data were analyzed using Statgraphics Centurion XVI.I software, employing multiple linear regression. No significant differences were observed among treatments for health parameters during gestation and lactation phases (P>0.05). However, by the end of lactation, treatments T1 and T2 showed lower dirtiness scores compared to treatments T3 and T4 (P=0.020), potentially linked to higher counts of thermotolerant bacteria found in microbiological analyses of T3 (8.309 CFU/g; P<0.05) and T4 (9.390 CFU/g; P<0.05), as well as higher temperatures inside the barn, increased water consumption, among other

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factors. In conclusion, sows receiving treatments T1 and T2 exhibited lower dirtiness scores and lower counts of thermotolerant bacteria. Nevertheless, further variables such as temperature, air humidity, diet composition, among others, should be analyzed to fully assess the effectiveness of probiotics provided during each phase.

Environmental enrichment for Intensive Pig Farming: a systematic review of Interventions, Welfare Indicators, and Production Stages

Letícia Bicudo Nogueira¹, Vitória Ágatha Alves¹, Maria José Hötzel¹

The Normative Instruction 113 of the MAPA (Ministry of Agriculture, Livestock, and Supply) from December 2020 aims to align Brazil with global advancements related to legislation on good practices in commercial pig farming. The requirement to use environmental enrichment (EE) in confined housing creates a demand for scientific and technical knowledge on the subject. Following the PRISMA model, this systematic review synthesized the main types of enrichments tested in intensive pig farming systems, the animal welfare indicators assessed, the main alterations promoted, and the stages of production involved. Experimental, peerreviewed studies comparing the application of interventions with a control group without enrichment use and that did not present confounding bias (different concurrent environmental modifications), were selected from searches in the Scopus (387) and Web of Science (686) databases. Out of the 64 included studies, 55% used "toys" (non-fragmentable structures, 92% of them suspended from the floor) and 44% used substrates (mostly straw and shavings). Other interventions included music (4 articles), cognitive enrichment using feeders (2), ice (1), and ramp (1). Outcomes were identified by evaluations of behaviour (60 articles), productive performance (15), injuries (15), cortisol (12), immunity (9), and cognitive functions (2). Most studies evaluated animals in the production phase (piglets in maternity, 14%; and in growth and termination, 76%). The remaining studies evaluated gestating females (8) and in maternity (3), gilts (2), and breeding males (1). Out of the 64 articles, 51 concluded that there was improvement in at least one of the welfare indicators assessed. Environmental enrichment increased playful and exploratory behaviours and reduced stereotyped behaviours, aggression, belly-nosing, tail-biting, and idle lying time. An increase in behavioural flexibility, reduction in bodily injuries, and increased immunoglobulins were also reported. Studies with inconclusive results (7) referred to changes in physiological biomarkers; 4 did not report changes, 1 reported increased competition in the growth phase, and 1 reported an increase in cortisol and neutrophils. Although no study directly addressed indicators of emotional valence, the diversity of variables measured indicates the perception of the need

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for a comprehensive evaluation in the investigation of animal welfare status. Several EE strategies have proven effective in promoting welfare improvements and can be applied to comply with Brazilian regulations. Animals used for reproduction were less represented in the studies, indicating a possible information gap.

Wound healing and mortality in tail-docked and castrated piglets with or without environmental enrichment

Katarina Buckova¹, Alexa Marie Newgard¹, Bozhao Zhang¹, Anna Johnson¹

Most neonatal pigs are routinely tail-docked and castrated without the use of pain relief. Recently, there have been the efforts to refine or replace these painful husbandry procedures by more welfare-friendly and sustainable practices. To achieve these goals, policy-makers will need reliable scientific information on routine tail-docking and castration. Therefore, we investigated: (a) Effects of environmental enrichment on tail-docking and castration wound healing, (b) how long tail-docking and castration wounds take to heal and, (c) mortality rate after surgical castration performed without pain relief. The study was carried out at the lowa State University Swine Teaching farm. 185 piglets were born either into enriched (n=14 litters) or standard (n=13 litters) farrowing stalls. Enriched farrowing stalls consisted of four physical enrichment items: a jute bag, Rubber Ball, Easy Fix Toy, and a Biting Ring. All piglets were taildocked and males were castrated at 7 days of age. Tail-docking and castration wounds were inspected in one male and one female per litter 1, 3, 5, 7, and 12 days after processing. Wounds were scored 1 to 5 for tail-docking and 1 to 6 for castration (1 – completely healed, 5/6 – open wound). Enrichment effects on tail-docking and castration wound healing were analyzed using SAS GLIMMIX procedure with multinomial distribution. Enrichment provision had no effect on tail-docking (F1,40=0.09, P=0.77; odd ratio: 0.9) or castration (F1,38=0.0, P=0.96; odd ratio:1.02) wound healing score. D 12 only one pig had completely healed for taildocking while other piglets received scores 2 or 3 (80% and 18% of scored piglets, respectively). A total of 9% of males scored one on d12 for castration and 91% scored 2. Seven (7.9%) out of 89 males died or had to be euthanized within 12 days from processing. For the comparison, the female mortality rate was 5.2%. Our findings imply that tail-docking and castration wounds take more than 12 days to heal. Although enrichment improved a pig ability to cope with stress in previous studies, the enrichments used in our study did not facilitate healing process. Males that underwent castration without pain relief did have a numerically higher mortality rate. Therefore, further investigation into (a) other enrichment is warranted and (b) to further consider a link between castration and mortality in the pre-

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weaning phase is advised. The study was funded by the Artz Chair for Faculty Excellence in Animal Science, Iowa Pork Industry Center and the Iowa Farm Bureau Federation.

Poster + One-Minute Presentation Session 4: Farm Animals Behaviour and Welfare (Mixed)

Tuesday, July 23rd, 16:15-16:30

Perceived producer barriers to the implementation of best management practices for the transportation of lactating cull cows

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Our objectives were to understand dairy farmers' awareness, perceptions, and barriers to implementation of best management practices (BMP) for the transportation of lactating cows. In this context, transportation was generally of cows being culled and moved to auction or abattoir. In total, 28 dairy producers (from 25 farms) in Ontario, Canada participated in 5 semistructured focus groups. Participants identified as 21 men and 7 women, on average were 42 years old and had 93 lactating cows on farm. All focus groups were audio recorded, transcribed verbatim, and analyzed with applied thematic analysis. Four themes were developed from the data: 1) transfer of responsibility, 2) interpersonal relationships, 3) juggling priorities, and 4) complexities of long-term planning. While individual participant knowledge varied, all focus groups described and discussed perceptions of transport that were inaccurate (e.g., underestimated duration, unawareness of distances or multiple destinations). Participants did not agree whether producer's responsibility ended once the cow left their property or if shared responsibility remained between all stakeholders from the farm until the final destination. Many focus groups discussed the importance of trusted relationships with local cattle transporters, other farmers, and veterinarians to ensure best practices are shared (e.g., peer discussions, veterinary clinic client workshop) and followed. All focus groups discussed how business pressures (e.g., production demands, space limitations) often challenge their ability to dry off lactating cows before transport but the degree of milk production may influence the animal's destination (e.g., direct to slaughter for animals with high milk production). Some participants elaborated that there is a dynamic balance between business pressures and potential for declining welfare of the animals under consideration to be culled

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during lactation. Poor herd management, being overwhelmed by daily workload, and producers nearing retirement were all identified as having a negative impact on long-term planning and may increase transportation of lactating cows. Finally, the lack of access to transportation direct to slaughter was identified as an important barrier for adherence with the BMPs for transportation of lactating cows. In conclusion, most participants did not have an accurate understanding of what happens to cull cows and were unsure if they maintained a level of responsibility for that animal after being transported from their farm. Given the priority placed by participants on trusted interpersonal relationships, greater awareness and training of farmers and transporters, and structural changes to increase local slaughter capacity may increase compliance with BMP and enhance cull cow welfare.

Strengths and weaknesses of 21 dairy farms in Chile, to obtain the animal welfare certification "Welfair®"

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In recent decades, animal welfare has been crucial for the dairy food chain affecting product quality. Producers have begun implementing welfare measures and seeking certifications, such as Welfair® to differentiate their production systems and be recognized by consumers. In October 2023, the first dairy company in Chile obtained this certification, marking a milestone in the region. The present study aimed to determine the strengths and weaknesses of Twentyone dairy farms in southern Chile, across the regions of La Araucanía and Los Lagos of varying sizes, breeds and production systems, according to the Welfair® certification system. According to PCA1 analysis, the criteria of expression of social behaviours, expression of other behaviours, absence of hunger, and ease of movement, collectively explain 45% of the variability of this component, differentiating between certified and uncertified farms. 100%, 76%, 57.14%, and 95.23% of the farms obtained an excellent rating in expression of social behaviours, expression of other behaviours, absence of hunger, and ease of movement, respectively. On the other hand, 95% of the farms scored below the required level in the flight test; however, 57.15% and 42.85% obtained scores rated as good and sufficient, respectively, in the criterion of positive emotional state. PCA2 analysis shows that the indicators of absence of thirst and absence of diseases were the other two criteria that differentiated the farms, in this case, with low evaluation. In absence of thirst, 71.4% of the farms were in the insufficient category, 9.52% in sufficient, 14.28% in good, and 4.76% in excellent; while in absence of diseases, 14.28% obtained an insufficient score, 71.42% sufficient, and 14.28% good, with no farms in the excellent category. The study deduces that farms gained animal welfare certification mainly due to factors like social and other natural behaviours, absence of hunger, and movement freedom, underscoring cows' need for expressive environments such as pastures. Notably, the human-animal relationship emerged as a critical shortfall, with 95% of farms underperforming, revealing a systemic threat due to inadequate evaluation metrics for pasture-based interactions. PCA2 revealed thirst and disease as additional deficiencies among

the farms, highlighting the importance of accessible water and clean troughs for certification. In essence, Chilean farms' strength lies in pastoral systems that facilitate natural behaviours and proper nutrition. However, disease management requires improvement, and drinker management shortcomings adversely affect dairy cow welfare. The most pressing challenge identified is the absence of a reliable measure for assessing human-animal relationships.

The presence of brushes increases activity and reduces visits to the feeder in pre-weaned dairy calves

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The environments in which farm animals are raised are often barren and lack many of the stimuli present in the natural environment. Therefore, environmental enrichment is often used to encourage natural, adaptive behaviours to be performed. In the UK farms that house cattle indoors are sometimes required to provide brushes as enrichment as their presence is believed to positively affect welfare. However, the effects of brushes on calf behaviour have not yet been explored. Here, we used precision livestock technologies (ultra-wideband location sensors and automatic milk feeders) to determine the effects of stationary brushes, on the movement and feeding of 75 dairy calves ranging from 23 to 65 days of age at the start and monitored continuously for 72 days. The trial period was divided into 48h blocks, half of which were randomly allocated to having 3 stationary brushes fixed to the walls of the pen, and we used mixed-effects models to test if and how the presence of brushes affected a range of calf movement and feeding behaviour. On average calves spent 11 more minutes in areas where brushes were placed per day (p<0.01) and entered these areas an additional 11 times (p<0.01), compared to the same locations when brushes were absent. Whilst brush use declined over time, suggesting habituation to the novel item, individuals showed high engagement with the brushes throughout the trial and showed limited inter-individual differences in brush use. The presence of the brushes significantly increased the daily distance calves travelled by 26 meters (p=0.02) and reduced their residence time (defined as the time an individual spends inside a circle centred around its location) by 17% (p=0.01), indicating higher activity. Calves also changed their feeding behaviour while still consuming the entirety of their milk allowance. Feeding rate was reduced by 14.3 ml/min in the presence of the brushes (p=0.05), which would lead to better nutrient absorption. There was also a 16% reduction in the number of rewarded visits to the automatic feeder (p<0.01) and an increase in their duration, indicating uninterrupted meals. Overall, adding stationary brushes as an element of environmental enrichment, increases activity in all calves and reflects positively in calf feeding behaviour.

Offloading Practices by cattle handlers at Akinyele International Cattle Market, Ibadan, Nigeria

Priscilla Komolafe¹, Olaniyi Babayemi², Kehinde Thomas³

Concern for animal welfare is fast gaining attention in developing countries including Nigeria. Whilst some animal welfare legislation exists in Nigeria, awareness and compliance to humane handling is very low. Although many may be ignorant about the need for humane handling, it is also likely that some who is knowledgeable simply do not consider that animals should be handled humanely. One area of particular concern is offloading practices at cattle markets. Thus, the objective of this study was to describe the condition of cattle upon arrival at the Akinyele International Cattle Market, which is centrally located in the southwestern zone of Nigeria; a market that facilitates the sale of high numbers of cattle. The study was carried out for a period of 5 consecutive weeks, where daily (Monday to Sunday) transporters arriving at the marker between the hours of 7am and 7pm were observed. With the aid of a check list, a camera and a recorder, direct observations were undertaken and the following information was logged: Stocking Density-SD (m2), Physical status of cattle at offloading (Standing, Assisted with Tail-AwT (tail is pulled up to raise), Assisted by Wood raising-AwW), Carted (nonambulatory)), Transit time and cattle injured in transit-CIT (Bruised skin, Twisted neck, injured leg and injured horn). Data were analysed using descriptive statistics and Pearson's correlation at α0.05. Findings indicated that there were 11,634 loads of cattle that arrived the study area. The stocking density in vehicles at offloading time ranged between 1.2 and 1.8 m². The 598 loads of vehicles collectively had transported 11,635 cattle of which 64.2% were able to stand and walk after offloading (53% male, 47% female); 35.8% cattle (48.5% males, 51.5% females) were non-ambulatory, requiring assistance to walk; 18.1% walked by pulling up their tail AwT (64.1 male, 35.9% female); 8.6% assisted to stand with wood-AwW (39.2% male, 60.8% female) and 9.1% carted (non-ambulatory) as they could not respond to all aids rendered (40.2% male, 59.8% female). The minimum transit time was 4.39±0.4 while maximum is 28.55±0.2. Total cattle injured in transit was 42.1% with bruised skin constituting 68.5% other

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injuries were Twisted neck (18.7%) injured leg (8.5%) injured horn (4.3%). The study revealed that there is a strong correlation (p<0.01) between the transit time and injuries sustained in transit. The inability of cattle to stand on hoof during the offloading practice indicates that the welfare of the animals was compromised.

Musical information retrieval: an attempt to study interspecies communication in Colombian cattle herding songs

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In Colombia, milk and meat production relies on Holstein cattle and dual-purpose breeds, including Bos indicus varieties, normally raised in pasture. Traditional cattle herding songs are used in various scenarios, including herding, transhumance, night care, and milking. Similarly, in Scandinavia, Kulning is used to call livestock from the mountains. Acoustic characteristics of the latter have been explored, but Colombian herding songs and their similarities and differences with cattle vocalizations are unexplored. Our objective was to determine acoustic similarities and differences between bovine vocalizations from different contexts (positive and negative) and human vocalizations of herding songs. Musical information retrieval (MIR) was used to analyze five acoustic parameters from cattle vocalizations, Colombian herding songs, and Kulning, as an initial step to grasp their acoustic relationships and craft music for environmental enrichment for cattle. Recordings of Bos indicus and Bos taurus cattle vocalizations during grazing and pre-slaughter were subjected to acoustic analysis. Cattle vocalizations (n=36) were captured via a microphone attached to a Zoom H6 recorder, while Colombian pastoral songs (n=20) and Kulning (n=3) were sourced from CDs and the internet, respectively. Acoustic parameters including spectral deviation, high-frequency content, highest, dissonance, and zero crossing were extracted using Sonic Visualizer V. 4.5.2. Mean differences of these acoustic parameters between herding songs and vocalizations were analyzed via one-factor ANOVA. Acoustic dissonance was higher in vocalizations emitted at the abattoir for both B. Indicus (0.776 \pm 0.044) and B. Taurus (0.280 \pm 0.050) compared to grazing vocalizations ($P \le 0.0001$). Conversely, dissonance was lower ($P \le 0.0001$) in all herding songs compared to all cattle vocalizations. Similarly, centroid values were lower (P≤0.0001) during grazing than before slaughter. No significant differences (P > 0.05) were observed for

high-frequency content, spectral deviation, and highest parameters between cattle vocalizations and herding songs. Stressful situations, such as pre-slaughter, prompted higher-pitched vocalizations with elevated centroid values and acoustic dissonance. The lower values for these two parameters and similarities in other acoustic parameters between cattle vocalizations and herding songs imply cowboys' thoughtful incorporation of bovine-like traits into songs to influence animal behavior. The proposed musical information retrieval not only provides an alternative to exploring interspecies communication, but also serves as a valuable tool for discriminating negative and positive vocalization contexts. These elements are essential for the creation of music tailored to each species.

Selection gate passage in cows housed with their calves in an automatic milking unit

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The practice of rearing dairy calves together with their dams is currently increasing in Europe. Cow-calf contact (CCC) in automatic milking systems (AMS) gives the cows more agency compared to other dairy management systems, potentially benefitting their welfare. In AMS, cows are often steered by automatic gates to different areas of the pen, depending on time since last milking. It is important that passage through these gates is not obstructed, so that the cows can move freely within the pen. This study aimed to explore descriptively whether CCC affects how the cows move through a selection gate located by the milking robot. Gate data from four batches of cow-calf pairs housed in the same free-stall AMS between August 2019 and June 2021 was analysed. Three of four batches also included conventional cows (CONV) kept in the same AMS, but without physical calf contact. Two periods were compared: 1) before (CCC: 68 cows, 2931 cow-days; CONV: 49 cows, 2326 cow-days) and 2) after (CCC: 19 cows, 588 cow-days; CONV: 17 cows, 554 cow-days) the calves were separated. During the pre-separation period, CCC cows did not directly pass the selection gate to the milking robot on 55% of occasions, but instead retracted their heads when the gate opened and then tried to enter the gate a median 3 (range 1-261) more times before passing. For CONV, the corresponding proportion was 25%, with a median 3 (1-85) tries. When cows were directed to the resting area, CCC cows did not pass directly 5% of occasions [median 1 (1-90) tries] while the values for CONV were 13% of occasions with 1 (1-79) tries. For occasions with 30+ head retractions, the gate directed CCC cows to the milking robot 95% and CONV cows 42% of the occasions. During the post-separation period, CCC cows directed to the milking robot did not pass directly on 27% of occasions [median 2 (1-36) tries], values for CONV were 19% of occasions and median 2 (1-55) tries. Values when directed to the resting area were similar in both groups [CCC: 12% of occasions, median 2 (1-39) tries; CONV: 14% of occasions, median 1 (1-29)]. Our preliminary data indicates that CCC does influence how selection gates are used in AMS, which could affect cow-traffic negatively. We suggest that future research explores

where these gates should be placed in relation to other resources to minimize effects on cowtraffic in CCC barns.

Providing water in nipple bucket reduces the cross-sucking of dairy calves housed in groups in a pasture area

João Pedro Donadio¹, Karolini De-Sousa², Maria José Hotzel⁴, Teresa Alves⁵, Matheus Deniz³

Calves raised in natural settings, i.e., alongside their mothers, typically do not exhibit crosssucking behavior. Non-nutritive oral behavior, such as cross-sucking, is considered an abnormal behavior for calves in the dairy industry and can cause negative effects on their welfare. Thus, we evaluated the effect of providing water from nipple buckets on daytime behavior and cross-sucking on calves housed in groups in a pasture area. The study was conducted at Embrapa Pecuária Sudeste (São Carlos/SP, Brazil). Twenty-four Jersey x Holstein calves were enrolled in a replicate longitudinal study from birth to weaning (60d). Groups of 3 calves (n = 8 groups) balanced by age $(\pm 7d)$ were randomly assigned to one of two treatments: waterT, where calves had access to an open water trough (capacity of 90L, 60cm diameter, and 48cm height); waterN, where calves had access to a water trough with nipples (capacity of 30L and 5 nipples positioned a 55cm from the soil). All calves were kept in outdoor paddocks with access to pasture, concentrate, and shade. Whole milk was offered twice a day (3L/calf/ meal) in nipple buckets (same as waterN) on the opposite side of the water supplier. Behavioral observation lasted 10 h (7 am to 5 pm) and was performed once a week from wk 1 to wk 8. The daytime behavior (active, standing, and lying) was recorded through 5-minute scans, while the frequency and duration (s) of cross-sucking and drinking behavior were recorded continuously. The cross-sucking behavior was defined as when a calf sucking another calf in any body part. The group's water intakes were measured 3x a week by hydrometers. The data were analyzed by generalized linear mixed models including treatment as fixed effect, group and week as random effects. There was no difference in active (p=0.323) and lying (p=0.113) behaviors between treatments, but the calves on waterN were less likely to spend time standing (36%; p=0.047) than calves on waterT. Calves on waterN were less likely to perform cross-sucking (47%; CI: 0.29 – 0.93; p=0.027), and the duration was shorter for

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each event (41 \pm 71 s vs 69 \pm 96 s; p=0.007). The water intakes were similar (p=0.07) between treatments (waterT: 1.73 \pm 1.1 L/day vs waterN: 1.50 \pm 0.7 L/day); however, calves on waterN (111.6 \pm 124.2 s) spent longer (p<0.05) drinking water than calves on waterT (76.2 \pm 75.6 s). Our results suggest that the use of nipple buckets for water supply can reduce cross-sucking behavior, probably related to the motivation satisfaction of sucking behavior.

Salivary cortisol concentration and milking parlor behavior of first lactation horned and dehorned dairy cattle

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Most US dairy producers remove horns due to safety concerns. Dehorning remains a significant welfare issue in US organic dairy production due to limitations on approved organic pain mitigation and hesitancy among organic producers to utilize effective synthetic substances to relieve disbudding pain. Raising horned cattle may more closely align with organic values of natural living while removing a welfare concern in early life. The purpose of this study was to compare the behavior and salivary cortisol concentrations of dehorned (N = 37) and non-dehorned (N = 14) first lactation Holstein and crossbred dairy cattle at the University of Minnesota West Central Research and Outreach Center organic dairy in Morris, MN. Dehorned cattle were disbudded via hot-iron between 5 to 7 weeks of age. Cattle were milked twice daily in a nine-swing parabone milking parlor and observed during the first 8 days of lactation. Handlers scored ease of milking parlor entry and behavior in the milking parlor, and recorded occurrences of kicks and stomps in the milking parlor. Immediately after milking, cattle were released into a chute for collection of saliva samples. Linear mixed models were used for analysis of salivary cortisol and logistic regression was used for analysis of behavior with the fixed effects of dehorning status, days in milk, time of milking (am or pm), and the interaction of dehorning status and days in milk, with cow as a random effect. Dehorned cattle (163.3 pg/mL, 80% CI = 156.1 to 170.8) had higher salivary cortisol (P = 0.037) than horned cattle (142.3 pg/mL, 80% CI = 132.9 to 152.4). Mean salivary cortisol was lower (P < 0.001) for increased days in milk from 203.2 pg/mL on day 1 to 140.4 pg/mL on day 8. Cortisol was higher (P = 0.006) during the morning (μ = 157.7 pg/mL, 80% CI = 151.0 to 164.6) than during the evening ($\mu = 147.4 \text{ pg/mL}$, 80% CI = 141.2 to 153.8). There was no effect of dehorning status, days in milk, or time for parlor entry or behavior scores or the occurrence of kicking or stomping in the milking parlor. Dehorned dairy cattle in the first lactation had higher salivary cortisol and similar behavior in the milking parlor during the first week of lactation compared to non- dehorned dairy cattle. The results indicate that dehorning may influence animal wellbeing later in life, but further research is needed.

Use of enriched cages for does during gestation and lactation: behavioral effects on offspring

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As experiências intrauterinas e início da vida pode impactar no comportamento futuro por meio de heranças epigenéticas. O estudo investigou o impacto da utilização de gaiolas enriquecidas e convencionais em coelhas durante a gestação e lactação no comportamento da F1. Foram utilizadas 16 coelhas multíparas da raça Nova Zelândia, divididas em quatro grupos de tratamento com base nos períodos de gestação e lactação: enriquecidoenriquecido, enriquecido-convencional, convencional-enriquecido e convencionalconvencional. As coelhas foram alojadas individualmente em cada tratamento de forma aleatória após a confirmação da gestação por ultrassom no 10º dia após a reprodução. No 28º dia de gestação, foram transferidas para as gaiolas correspondentes ao seu tratamento. As gaiolas convencionais mediam 80x60x40 cm, enquanto as enriquecidas eram oito vezes maiores, com áreas de esconderijo, plataformas para descanso, espaço para ficar de pé e uma caixa com maravalha. Os filhotes foram desmamados e transferidos para gaiolas convencionais no 35º dia de vida, alocados em duplas da mesma ninhada. Aos 90 dias, uma das duplas contendo somente fêmeas aleatoriamente selecionadas para testes comportamentais: campo aberto (CA), novo objeto (NO), reconhecimento de novo objeto (RNO) e interações sociais. Os testes foram conduzidos individualmente em uma arena de 4 m². Cada teste foi realizado em intervalos de 24 horas, gravados e analisados por dois observadores. No teste de interação social, a latência do primeiro contato, o tipo de contato (positivo ou negativo), frequência e duração dos contatos foram observadas ao longo de 5 minutos. Os comportamentos observados foram analisados através do programa Rstudio, utilizando regressão linear mista para avaliar os efeitos dos tratamentos no comportamento das ninhadas, a matriz e o reprodutor foram considerados como efeitos aleatórios; as premissas dos modelos foram avaliadas graficamente. No teste de CA, o tratamento convencional-enriquecido tendeu a explorar mais quadrantes do que o convencionalconvencional (20.9±2.96 vs. 15.4±3.01; P=0.07). No teste de NO, houve diferença no número

de vocalizações e bateção de patas entre o tratamento enriquecido-convencional e o convencional-convencional (1.5±0.17 vs. 0.25±0.182; P<0.01) e (1.12±0.09 vs. 0.0±0.09; P<0.01). No teste de RNO, coelhas do tratamento enriquecido-enriquecido tocaram o objeto novo por mais tempo do que as do tratamento convencional-convencional (8.8±2.3 vs. 2.2±2.41; P=0.04). Na interação social, as coelhas do tratamento convencional-enriquecido brigaram mais do que as do convencional-convencional (3.5±0.82 vs. 0.0±0.83 P=0.003). Portanto, o estudo destaca a influência de gaiolas enriquecidas durante a gestação e lactação de coelhas no comportamento da prole.

Comparative analysis of nest quality in primiparous rabbits

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In the routine of rabbit breeders and researchers, analyzing nest quality is essential for the development and well-being of rabbit kits. However, the methods available in the literature employ different criteria for assessing nest quality, making the choice challenging. This study aimed to analyze the nest quality of primiparous does based on two different methods and compare the agreement between them. The study was conducted at the Rabbit Breeding Sector of the Gralha Azul Experimental Farm (FEGA) of the Pontificia Universidade Católica do Paraná, located in Fazenda Rio Grande, Paraná, Brazil. Twelve primiparous New Zealand White rabbits were used, reproduced by natural mating. The nests were made of wood with dimensions of 40 cm x 23 cm x 8 cm, placed on the 27th day of gestation, along with hay. Two analyses were conducted based on photographs taken of the nests on the day of birth and four days later. Two trained observers performed the photograph analyses, following the methods of Sawin & Carry (1959) and Blumetto et al. (2010). In the Sawin & Carry method, nests are evaluated for the level of excavation and compaction, being classified into categories such as "none," "poor," "fair," "good," "very good," "excellent," and "packed." In the Blumetto et al. method, the evaluation focuses on the level of mixture between hay and fur, classified on a scale from 1 to 4. For statistical analysis, each classification was converted into a numerical score, where values below 3 were defined as 0 and values equal to or greater than 3 were defined as 1. To assess the agreement between the methods, the Kappa test was performed using RStudio. The comparison between the two methods on the day of birth showed no agreement (Kappa = -0.51, P = 0.07), as well as on the fourth day after the birth of the kits (Kappa = 0.11, P = 0.7). The results indicate that the methods are different and have particularities regarding the evaluation criteria. Therefore, they cannot be considered as substitutes. This finding reflects the need for further studies that assess nest quality and integrate all criteria into a single method.

Effect of abnormal behaviour on memory, judgment and attention abilities of mares housed in individual boxes for three consecutive days

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Horses are commonly housed individually in boxes which has been associated to the development of abnormal behaviours in some individuals. Nonetheless, abnormal behaviour may be used as a coping mechanism to improve the animal's emotional state. This study aimed to investigate whether mares that exhibit abnormal behaviours when housed individually show more positive responses in emotional tests. Fifteen mixed breed adult mares, housed outdoors, were housed in individual stalls for three days and after this isolation period, memory bias, judgment, and attention tests were conducted. Each mare was recorded in the stall during seven 5-min focal continuous sessions per day through a video, yielding 21 scans per horse (105 min observation) looking for abnormal behaviours. At the end of the third day, each mare underwent three tests in the stalls. For the memory test, mares were trained before the isolation period to associate a black bucket with food and a white bucket without food, in a test arena. To assess mares' discriminatory memory at the end of the third day in the stall, both buckets were placed simultaneously inside the stall, and investigating the black bucket first was considered a positive memory bias. Next, a grey bucket was added between the two buckets for the judgment test, for five minutes. The mares' looking inside the grey bucket was considered a positive bias. For the attention test, the intensity and duration of the behavioural reaction was evaluated based on ear position, neck height and body posture for five minutes following an iron bar thrown to the ground. The same researcher analysed all tests. Thirteen mares exhibited at least one episode of box walking during the three days of housed. The results of box walking were ordered according to the total frequency observed in each mare. The median of these values was used to separate the mares with and without abnormal behaviour. For the emotional tests: nine mares answered the memory test correctly, twelve mares investigated the grey bucket and four mares maintained a low level of attention when evaluating body posture. There was no association between the frequency of box walking and the positive responses in the memory, judgment

and attention tests (P>0.1). The effects of individual housing may not have affected the mares as expected due to the short period in the stalls and *ad libitum* amount of roughage provided, or our sample size was not enough to detect differences.

Perching behavior and lameness in pregnant dairy heifers

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Perching is commonly observed in cows housed in freestalls and is defined as when the animal supports its front limbs on the bedding while keeping its hind limbs on the ground. In lactating cows, this behavior has been identified as a risk factor for lameness. However, there are no studies on this behavior in dairy heifers. Here we determine the association between perching behavior and lameness in pregnant dairy heifers. A longitudinal study was conducted with 77 Holstein heifers, aged 13 to 18 months, housed in a freestall system. Only heifers that did not show lameness in the first gait scoring assessment were included in the study. Data collection on locomotion and perching behavior was performed in monthly visits and lasted 12 months. Gait scoring was recorded for two consecutive days in each visit to minimize observation errors. The behavioral assessment involved three observations in one day using the scan sampling method, which consisted of observing and recording perching behavior at approximately 09, 10, and 11 am. Heifers with a locomotion score of ≥3 for two consecutive days were considered lame. Lameness was interpreted as moderate for heifers scoring 3 and severe for those scoring 4 or 5. The study data was analyzed using R Studio software and mixed effects logistic regressions were used. We observed a tendency in the association between perching and lameness (p=0.08), suggesting that lame heifers are twice as likely to exhibit perching behavior when lameness is present (OR = 2.03; 95%CI: 0.90-4.76). However, there was no association between previous perching and subsequent lameness (p=0.48). The study results suggest that perching may be an indicator of the presence of lameness in pregnant dairy heifers, but not a predisposing factor.

Insights into the impacts Facebook can have on management decisions for (recreational) sport-horses in New Zealand

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The aim of this study is to investigate the impact of casual advice exchanged on horse management in New Zealand-based Facebook groups, with a specific focus on recreational and sport horses. It seeks to understand how New Zealand horse owners utilise equinethemed Facebook groups to make "informed" decisions regarding horse husbandry, and the impacts of doing so. A literature review exposed that previously published theory on New Zealand Facebook users and their motivation neglects to address advice sought for animals and particularly horses. The first stage of this sequential mixed methods study demonstrates to what extent and in what circumstances Facebook users in New Zealand equestrian-themed groups are likely to ask for or give advice, by inviting horse owners to complete an anonymous online survey on appropriate Facebook groups. The survey received responses from 160 participants, providing a small but valuable dataset (n.156) for analysis. In the second part of the study, the research explores reasons why professionals appear unable or unwilling to interact or respond to posts seeking or giving advice, even when the advice given may potentially compromise the welfare of the horse. The ramifications of not interacting with advice given in equine-themed Facebook groups that the professionals have been tagged in, or that they come across in their personal Facebook dealings is examined as well. A variety of professionals were invited to in-depth interviews, with five professionals from various regions and disciplines in New Zealand responding. This mixed-methods approach allows for both quantitative data collection and autoethnographic learning through collegial discussions, providing a comprehensive understanding of Facebook users' advice-seeking behaviors within the New Zealand equestrian community, while maintaining conciseness. Autoethnography is integrated through the researcher's active participation in New Zealand equestrian-themed Facebook groups under investigation. By actively engaging with these online communities, the researcher gains firsthand experience and insight into the dynamics of advice-seeking behaviors among horse owners. This immersive approach allows the researcher to provide personal reflections and observations, enriching the overall understanding of the

phenomenon being studied. Additionally, autoethnography is employed during the qualitative interviews with equine professionals. The researcher's own experiences and perspectives as a participant in the digital equestrian community inform the interview process and shape the discussions with professionals. This study was conducted in accordance with ethical guidelines and received approval from Otago Polytechnic ethics committee. All participants provided informed consent, and measures were taken to ensure confidentiality and anonymity.

Trends and perspectives from a scientometric analysis on the impact of heat stress in confined dairy cows

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This study aimed to investigate state-of-the-art research involving heat stress in lactating cows housed in compost barns, free-stall, and tie-stall over the last 22 years (January 2000 to April 2022). The scientometric review was conducted according to the guidelines of the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) protocol. A systematic search was performed by two authors on the Web of Science. The terms used for the search were TS (Topic Search) = (dairy AND cow) AND ("heat stress" OR "thermal stress") AND ("compost-bedded pack barn" OR "compost barn") AND (free-stall OR "free stall") AND (tiestall OR "tie stall"). A total of 2.245 scientific studies were saved for further selection. The screening was composed of four stages. Stage 1: Studies written in all languages and document types were considered. Stage 2: Studies published before 2000 were excluded. Step 3: Titles and abstracts were assessed to identify and remove studies that did not use lactating cows, confinement systems and heat stress. Step 4: Finally, the complete studies were read in detail. Studies that did not address the impact of heat stress on lactating cows in a confinement system were excluded. The remaining studies (n = 604) were included in the scientometric analysis. For data analysis, co-occurrence and cluster analysis were performed using the CiteSpace software. The co-occurrence network of the countries identified 72 nodes and 812 links, with a modularity index of 0.5357 and an average silhouette of 0.8783, showing consistency in our data. The United States, Germany and Israel showed burst (8.00, 3.61 and 3.83, respectively). The United States and Israel were the countries that showed the coolest colours among the links, indicating that these countries contained the oldest studies under investigation in this study area. Clustering of the keywords and titles identified eight clusters, with silhouettes ranging from 0.812 to 1.000. The keyword cluster #7, hair cortisol, and the title cluster #7, production variation, were the oldest areas of study, while #0 behaviour and

#0 heat stress were the most current areas in our research. The terms behaviour and heat stress are the trends in our study. In future perspectives, more studies evaluating the effect of heat stress on the behaviour of lactating cows in confinement systems are needed.

Poster + One-Minute Presentation Session 7: Farm Animals Behaviour and Welfare (Mixed)

Wednesday, July 24th, 12:00-12:15

The addition of a rider increases stride length and ground force in horses used for Equine Assisted Services

Bethany Baxley¹, Nichole C. Anderson¹

Equine Assisted Services (EAS) involve qualified professionals using horses to address physical, cognitive, emotional, or social needs of people. People with physical disabilities may place uneven pressure on the horse's back requiring the horse to adjust its motion which could lead to pain-associated gait asymmetry or lameness. Our objective is to understand the influence of riders on the horse's gait during ridden EAS sessions. The hypothesis is that riders will cause the horse's gait to change when compared to the gait with no rider. Horses (n = 22) from four EAS centers who had worked in EAS for at least a year, were outfitted with inertial measurement units on all 4 distal limbs. The gait of each horse was analyzed without and with a rider on three different days thus representing a repeated measure design. Riders were individuals receiving EAS sessions and riders of any age were enrolled. Data was collected on the horse's first session of the day. Using a linear mixed model, where a single leg was the outcome, rider was a fixed effect and horse, day, and center are random effects. We found that left front (LF), right front (RF), left hind (LH), and right hind (RH) stride length were influenced by the presence of a rider. A rider increased the average LF stride length by 0.10 m (P = 0.02) and RF length by 0.12 m (P = 0.01). The LH length increased by 0.09 m (P = 0.03) and the RH increased by 0.08 (P = 0.04) when compared to a horse's gait without a rider. Right front, LH, and RH peak vertical ground reaction force (GRF) was also influenced by a rider. Our analysis of RF peak GRF revealed that in the absence of a rider, the estimated GRF is 54.32 newtons (CI: 46.02-62.62), which increases by 2.73 newtons (P = 0.04) when a rider is present. Similarly, LH peak GRF is estimated to be 44.97 newtons (CI: 40.06-49.89) without a rider, increasing by 2.73 newtons (P = 0.02) with a rider. Lastly, RH peak GRF is estimated to be 45.34 newtons (CI: 41.19-49.50) without a rider, increasing by 2.28 newtons (P = 0.04) with a rider. Overall, the presence of the rider increased stride length and downward force to the ground

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on 3 of the legs. This modification could increase the likelihood of lameness, resulting in negative welfare to the horse.

A systematic review of reporting on the living conditions of stallions used in semen studies published in 2017-2022

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Semen, a bodily fluid produced during ejaculation by male mammals, is influenced by the environmental and physiological conditions of the animal that produces it. Inadequate housing, feeding, social contact, health status and other stressors that impair welfare are commonly observed in semen producers, the stallions. Some of these stressors are detrimental to semen quality and contribute to the development of behaviours like aggression and stereotypical behaviours. This systematic review aimed to understand the extent to which horses' needs are addressed within the equine semen research and identify information on stallions reported in scientific articles focused on semen quality. Studies published in 2017-2022 were identified via searches in the Scopus and Web of Science databases. Our final sample consisted of 211 studies (2499 stallions; 1-241/study), from which we extracted information regarding ethical approval; age/breed of stallions; housing, environmental enrichment, feeding, health, social contact; behaviour. The most reported information was stallions' age (68%) and breed (54%). Housing information was absent in 81% of the studies; none of the studies presented information on environmental enrichment; most studies did not indicate if concentrate (84%) or roughage (77%) were provided. Of the studies that did, over 59% did not specify amount; only 27% mentioned ad libitum roughage and 99% did not mention access to pasture. Most studies (87%) provided no information related to social contact. Few reported the semen collection method (28%). Most studies had no information on stallions' health (86%). Only 46% were approved by an Animal Care Committee, 36% did not provide information on the issue and 18% cited some rule, law or guideline on animal welfare. None of the studies presented information about behaviours or temperament of the stallions, nor reported the presence of abnormal behaviours. Overall, the studies failed to report aspects of the living conditions and of the stallions that produced the semen used in studies. Particularly underreported were the opportunities for expressing highly motivated behaviours such as grazing and social interactions, thus we conclude that the welfare conditions of these stallions are mostly unknown. Lack of information on the conditions under

which the semen was produced undermines the quality of the publications. Careful management of horses used in experiments is essential to comply with ethical standards of care and to allow reproducibility and generalization of research results. We call for scientists, scientific journals and Animal Care Committees to discuss these issues and update their scientific and publishing practices.

Brushing pregnant ewes during the last third of gestation enhances their maternal motivation immediately after lambing

<u>Livia Pinto-Santini</u>¹, Valentina Reyes¹, Mónica Hernández¹, Rodrigo Valerio¹, Josefina Mañana¹, Jimena Fernández¹, Messy Pantoja¹, Karina Neimaur, Rodolfo Ungerfeld¹, Aline Freitas-de-Melo¹

Lambs born to calm ewes have a greater survival rate than those born to nervous ewes. Brushing causes a calming effect, improves the welfare of ewes and lambs, and promotes a better human-animal relationship. Therefore, the hypothesis tested in this study was that brushing pregnant ewes during the prepartum period triggers a greater maternal motivation, so the aim was to compare the ewe and lamb behaviors of brushed and non-brushed ewes immediately after birth. Forty-eight pregnant multiparous Australian Merino ewes carrying single fetuses were shorn at 104 days of gestation, and 30 days before the estimated date of birth, they were assigned, according to body condition score and weight, to one of the following treatments: A) 24 ewes were individually brushed for 5 min/day during 21 consecutive days (from day 124 until day 145 of gestation). Briefly, manual brushing was of the dorsal region in cranial-caudal direction using a brush with semi-hard bristles by trained brushers (group BRU); B) 24 ewes remained unbrushed in an adjacent pen while BRU ewes were brushed, i.e., they had visual and auditory contact with the brushers (group CON). Ewes were monitored continuously for 24 hours throughout the lambing period by trained observers, and the following data were recorded: a) length of the second stage of parturition; b) time from lambing to the mother beginning licking its lamb, and the licking' duration; c) the number of low-pitched bleats of the mother from lambing until the first sucking of its lamb; d) the lapse from lambing to lamb' first stand up, and the number of attempts to achieve it; e) the time to first suckling after birth and its duration, and f) the lamb' birth weight 24 h after lambing. All the variables were analyzed by proc mixed (SAS on Demand for Academic). For this purpose, the variables that did not had normal distribution were normalized. The model included the position of the lambing (standing up or lying down) and the sex of the lambs as random effects for the ewe's and lambs' behaviors, respectively. The number of low-pitched bleats emitted by BRU was greater than CON ewes (217.1 \pm 38.7 vs. 93.7 \pm 38.7) (P = 0.03), with no differences between treatments for any other variable evaluated. The greater display

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of vocalizations of BRU ewes -more than double- indicates a stronger motivation for establishing a bond with their lambs, which is relevant for lamb survival.

Can sleep pattern interference irritability be measured in dressage horses by an approach test?

<u>Ângela Perrone Barbosa</u>¹, Tiago Marcelo Oliveira¹, Sarah Raphaela Seidel¹, Paula Keiko Anadão Tokawa1, Fernando Mosquera Jaramillo¹, Neimar Vanderlei Roncati², Raquel Yvonne Arantes Baccarin¹

¹ Faculdade de Medicina Veterinária e Zootecnia (FMVZ) - Universidade De São Paulo (USP) – Departamento de Clínica Médica (VCM) - São Paulo - SP - Brazil, ² Grupo de Educação UNIEDUK - Gestor - São Paulo - SP - Brasil Horses adapted their polyphasic sleep pattern by dividing it into small fragments and alternating between REM phase (Rapid Eyes Movement) and NREM phase (Non-rapid Eyes Movement). REM sleep only occurs when the horse is in recumbency. In humans, sleep pattens disorders can cause irritability. Some studies have attempted to measure horse's irritability with an approach test, inferring the horse's comfort and readiness in being caught. The objective of this study was to analyze if irritability by REM sleep interference can be asset by an approach test. Ten Lusitano Pure Breed horses, age from 3 to 7, all Dressage athletes, were used in this randomized crossover experiment. REM sleep deprivation was accomplished by an observer not letting horses enter in sternal or lateral recumbency for 72 hours. All control horses exhibit REM sleep. This work was approved by the Ethics Committee on Animal Use (CEUA/FMVZ) with protocol number 6584300721. After 72 hours, an approach test was done by an unknow observer in both control and sleep deprived moments. Animals were approached in the stall only once and evaluated using the following parameters: 1 indifferent, 2 – turn the head 3 – come to the person. Then the observer approached with a halter and following parameters were observed: 1- does not allow approximation 2- moves away at the step 3- moves the head 4- does not show resistance. There was a pattern of horses advancing toward the unknown evaluator during the approach test. Differently from what was proposed by other studies, animals were evaluated in the stall as opposed to a round pen. In general, horses did not show changes in behavior between two experimental conditions, apart from four animals. Second and tenth horses, which preferred not to approach the evaluator when in sleep deprivation condition, and the fifth and seventh that showed a reverse behavior, not approaching evaluator while in control condition. It is worth discussing here how much this can be correlated with irritability or tiredness, since these horses associate human presence with leaving the stall. For this population, leaving the stall is associated with work,

whether mounted or in the round pen. Literature in general does not bring many references on how to quantify the irritability of horses, but maybe some indications, such as ear position, hematological data or diseases could be used to infer welfare and irritated behaviors associated with sleep deprived situations. New tests should be conducted to enhance our understanding of irritability in horses.

Lambs born to multiparous mothers respond more intensively to weaning than those born to primiparous ewes

<u>Aline Freitas-de-Melo</u>¹, Daniela Casuriaga¹, Magdalena Nin¹, Maria Jesus Frisch¹, Valentina Arbuet¹, Agustína Velázquez¹, Florencia Corrales-Hlinka², Raquel Pérez-Clariget², Rodolfo Ungerfeld¹

Lambs born to multiparous ewes establish a stronger bond with their mothers than those born to primiparous ewes. Therefore, the aim of this study was to compare the behavioral response to artificial weaning of lambs born to multiparous and primiparous mothers. The study was performed with 50 Corriedale ewes and their single lambs: 25 born to multiparous and 25 born to primiparous ewes. Lambs were weaned abruptly 80 days after birth. Lambs' behaviors (standing up, lying down, walking, pacing, grazing, vocalising, and ruminating) were recorded from 3 days before until 4 days after weaning. The frequency of each behavior was compared between treatments with a mixed model, including the treatment, the time and the interaction between the treatment and time as main effects in the model. The day of weaning, lambs born to multiparous mothers were more frequently observed standing up (71.2 ± 2.3 % vs. $60.0 \pm 2.4 \%$, p= 0.009), grazing ($60.4 \pm 2.6 \%$ vs. $45.7 \pm 2.7 \%$, p= 0.001), walking (5.0 ± 0.8 % vs. 0.8 ± 0.8 %) and vocalising (13.8 ± 1.5 % vs. 5.3 ± 1.5 %, p< 0.001) than those born to primiparous mothers. The percentage of observations in which lambs were recorded lying down (23.0 \pm 0.1 % vs. 38.4 \pm 0.1, p< 0.001) and ruminating (9.9 \pm 1.3 % vs. 20.0 \pm 1.3 %, p< 0.001) was lower in lambs born to multiparous than those born to primiparous ewes at the same day. In conclusion, artificial weaning triggered stronger behavioral responses in lambs born to multiparous mothers than those born to primiparous. These results highlight the need for further studies to determine practical strategies for weaning lambs, considering the category of the mother.

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Using a real-time location system to detect behavioral changes in ewes with subclinical mastitis and their lambs

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Subclinical mastitis, the asymptomatic inflammation of the udder, is a major welfare concern in meat breed flocks, causing pain, decreased milk yield, and increased mortality in ewes and lambs. Cases typically remain undetected until more severe stages of infection, highlighting the need to investigate early detection methods such as automated behavioral monitoring. Our objective was to measure the daily activity of and proximity between ewes affected by subclinical mastitis and their lambs compared to healthy cohorts using an ultrawideband realtime location system. We predicted sick ewes and their lambs would be less active and spend less time near each other than healthy ewes and their lambs. To detect subclinical mastitis, duplicate milk samples were collected weekly from each udder half of 24 meat breed ewes from 2 to 7 weeks postpartum. A ewe was considered infected if ≥ 100 CFU/mL were isolated in at least one udder half in absence of clinical symptoms (69 cases/133 possible). Each ewe and her lamb(s) wore a harness that held a sensor positioned at the base of the neck. T recorded x, y coordinates every 0.05 seconds while the ewes and lambs were housed in a group pen. We ran 3 linear mixed effects models with fixed effects of week postpartum (2-7), weekly diagnosis (sick or healthy), their interaction, and a random effect of animal ID nested within dam ID. Outcomes included average daily ewe-lamb distance and average daily distance travelled by ewes and by lambs. In weeks 5-7 postpartum, the average daily distance between sick ewes and their lambs was smaller compared to healthy cohorts (mean ± SE; Sick: 7.89 \pm 0.58m; Healthy: 10.38 \pm 0.67m; Week*Diagnosis: F = 33.37, p < 0.001). In the same postpartum period, sick ewes traveled greater distances each day than healthy ewes (mean ± SE; Sick: 4.1 ± 0.3 km; Healthy: 2.9 ± 0.3 km; Week*Diagnosis: F = 21.30, p < 0.001). The same was true for lambs reared by sick ewes (mean \pm SE; Sick: 4.3 \pm 0.2 km; Healthy: 3.2 \pm 0.3 km; Week*Diagnosis: F = 19.96, p < 0.001). Contrasting our predictions, our results indicate that ewes with subclinical mastitis spend more time with their lambs than healthy ewes and that both sick ewes and their lambs are more active. Future research investigating pain and nursing

behaviors in ewes with subclinical mastitis could provide insight into potential mechanisms behind the observed behavioral changes.

Supplementation of yeast-based additives positively impacts welfare indicators of broiler chickens challenged with Eimeria

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The yeast-based postbiotics and prebiotics improve the intestinal quality of broiler chickens. This improvement might support responses to infection and protect intestinal segments, especially the jejunum and ileum. The production of serotonin, which can improve the welfare of animals, is influenced by the quality of the intestine, as around 95% of it is produced there. This study was conducted with the aim of investigating the effect of supplementation of Saccharomyces cerevisiae yeast-based additives on welfare, intestinal histomorphometry and plasmatic serotonin levels in broiler chickens challenged with Eimeria. For this study, 1,890 one-day-old male chicks were used, with an average weight of 45g (+/- 10%), housed in 42 pens (45 birds/pens, 7 replications), in completely randomized design. To promote the health challenge by Eimeria, at 4 old-days, the broilers were challenged with 20x Eimeria vaccine dosage. The dietary: T1 - negative control (NC, no additives); T2 - positive control (PC, whit salinomycin); T3 - NC + 500 mg/kg yeast cell wall; T4 - NC + 455 mg/kg of yeast cell wall and 45 mg/kg of free nucleotides; T5 - NC + 500 mg/kg of autolyzed yeast cell wall associated with postbiotic; and T6 - NC + 250 mg/kg of autolyzed yeast cell wall associated with the postbiotic. Intestinal quality (duodenum, jejunum, and ileum) was assessed by histomorphometry (villus height, crypt depth and villus:crypt ratio). Plasma serotonin levels were measured using ELISA commercial kit. At 42 days-old, blood was collected from the ulnar vein of broilers. Welfare was assessed through walkability (gait score - GS), resilience (Latency to lie - LTL) and reactivity (approaching test). The data were submitted to ANOVA using PROC GLM (General Linear Models) of SAS 9.4 and compared for Tukey test (p<0.05). All treatments containing prebiotics (T3, T4, T5 and T6) improved intestinal histomorphometry and serotonin levels in broilers (p<0.0001). The T6 group exhibited a tendency to improve walkability, which was highlighted

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by a GS p-value of 0.0651. It is important to note that broilers supplemented with yeast-based additives (T3, T4, T5 e T6) had lower gait score 2, compared to those receiving an antibiotic diet (T2). There was no difference for LTL and reactivity between groups (p>0.05). The Eimeria challenge causes damage to the intestinal mucosa, but supplementation with yeast-based additives improves the integrity of this mucosa, resulting in higher serotonin levels and better welfare in broiler chickens. FAPESP #2020/12641-4, São Paulo State Research Support Foundation.

Unveiling Personality: a comparative analysis of Chilean Creole vs commercial laying hens

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Personality significantly influences how animals cope with stress, whether it's short-term or long-lasting. Laying hens show varied personality traits and previous studies have revealed that these traits, proactive or reactive, tend to be specific to certain breeds considering that a hen's personality isn't just shaped by its environment but also by its genetics. This study aimed to compare the personality traits exhibited by Chilean creole laying hens and Isa Brown laying hens through two behavioral tests. All birds were housed in an aviary system (0.4 m²/hen). Creole (n = 17) and Isa Brown hens (n = 60), aged 16 weeks, were subjected to two individual behavioral tests: an open-field test and a novel object test. In the open-field test, each hen was placed alone in a novel black square wooden box (125 cm × 125 cm × 125 cm) for three minutes. Subsequently, in the novel object test, the same experimental arena was used, but this time, a novel object was introduced, and the hen's behavior was observed for two minutes. The variables measured in both tests were latency to walk and latency to vocalize. In the novel object test, the latency to approach the object was also added. Behavioral test scores underwent principal component analysis (PCA) for each breed group, revealing intertest relationships across two factors of correlated test scores in both breeds. These factors were interpreted as representing distinct personality traits within the respective breed groups. In the creole group, PCA identified two distinct personality traits: "Hesitant Investigative" and "Quiet Movement." Within the former, hens exhibited delayed responses in both walking during the open-field test and vocalization during the novel object test yet displayed a notably quicker approach to the object in the latter test. Conversely, the latter trait, "Quiet Movement," was characterized by prolonged vocalization latency in the openfield test but shorter walking latency in the novel object test. In the commercial breed group, PCA delineated two personality traits: "Talkative" and "Anxious Reactivity." Hens characterized

by the "Talkative" trait showed reduced latency to vocalize during the novel object test. Conversely, those exhibiting "Anxious Reactivity" displayed extended latencies in both vocalization during the open-field test and walking during the novel object test. These findings underscore the divergent responses of the two groups to stress-inducing situations and novel surroundings, emphasizing the need to consider genetic diversity and rearing environment when studying personality traits in laying hens.

Validating behavior sampling periods for turkey pecking behaviors

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Turkeys frequently peck at conspecifics and objects in their environment, which can lead to animal welfare challenges. Pecking is understudied in turkeys, and insights may improve management strategies. Continuous behavior sampling is time intensive, so our objective was to validate sampling periods for estimating the rate of pecking in turkey toms. Nicholas Select toms (22 toms/pen, n=6 pens) were video recorded for 60 min at 4 weeks of age. The frequency of pecking another turkey (back- and head-directed), pecking the floor (foraging bout), and pecking the feeder (feeding bout) were recorded continuously for all turkeys in each pen for 60 min in BORIS software. Data were summarized to determine the rate (count/min) for each behavior during the first 5, 10, 15, and 20-min periods, as well as across the whole 60 min. Rates for each period were compared to 60-min rates with correlation and regression analyses. For regression, three criteria assessed the accuracy of each period's estimations: R²≥0.90, slope=1, and intercept=0. Coefficients of variation (CV) were calculated across pens for each period. Turkeys performed 17.1 ± 3.1 (mean ± SD) back pecks/min/pen, 4.3 \pm 0.9 head pecks/min/pen, 3.3 \pm 0.2 foraging bouts/min/pen, and 3.5 \pm 1.1 feeding bouts/min/pen in the 60-min period. Estimates from the 20-min period correlated with estimates from 60-min for back pecking, feeding, and head pecking (r=0.90, 0.89, and 0.78; p=0.015, 0.019, and 0.068, respectively). The slope and intercept met criteria for nearly all periods and behaviors, but the R² did not. The R² was typically highest for the 20-min period for all behaviors (R²=0.76, 0.73, 0.51, and 0.17 for back pecking, feeding, head pecking, and foraging, respectively). The CV across pens was low to moderate for each behavior at 60-min (5, 18, 22, and 33% for foraging, back pecking, head pecking, and feeding, respectively). However, the CV was often the largest at the shortest sampling period and decreased as the sampling period became longer (e.g., 70 vs 20% for back pecking at 5 vs 20-min periods), suggesting that pecking rates fluctuated across the 60-min period. Overall, our results demonstrate that pecking occurs at high rates in 4-week-old turkeys. Shorter observation

periods could not accurately capture 60-min performance. This may be due to variability in pecking throughout the observation period or limited sample size.

Well-being of broiler chickens supplemented with bacilli-based probiotics

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Probiotics maintain the intestinal integrity of broiler chickens and thus increase serotonin levels and decrease corticosterone levels. The production of serotonin improves animal welfare, and the integrity of the intestinal mucosa is related to the levels of this neurotransmitter, as more than 90% of serotonin is produced in the intestine. This study was carried out with the objective of evaluating bacilli-based probiotic supplementation and the behavioral response of broiler chickens. When spraying the probiotic, still in the hatchery, early colonization occurs in the gastrointestinal tract by the probiotic bacilli as the chicks interact and peck each other and, in this way, ingest the microorganisms. A total of 2,000 1day-old male chicks were housed in 40 boxes (50 birds/box, 10 replications) in a completely randomized design. Intestinal integrity was assessed by histomorphometry, plasmatic serotonin and corticosterone levels using a commercial kit, and behavioral evaluation assessed by resilience (latency to lie) and reactivity (grab test and touch test). The dietary treatments were: T1 – probiotic spraying when sent from the hatchery; T2 – probiotic spraying when sent from the hatchery, associated with the probiotic in the feed; T3 – probiotic in the diet from the 1st to the 42nd day; T4 – negative control, without performance-enhancing antimicrobial or probiotics. Samples for evaluations were collected at three times: 7, 27 and 42 days; Behavioral response were made on the 42nd day. Data analysis was performed using the SAS 9.4 program (2013), analyzed by ANOVA and compared using the Tukey test and Chisquare test (P<0.05). Regarding intestinal histomorphometry, it was observed that the use of probiotics (isolated or not) was able to maintain the intestinal integrity of the broilers chickens justified by the higher vill:crypt ratio when compared to the negative control (P<0.05). Correlated to the maintenance of intestinal integrity, in general, serotonin and corticosterone levels were better for treatments containing probiotics (P<0.05). The T1 and T2 treatments

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showed better results for the behavioral response, approach test (P=0.020) and catch test (P<0.05), with the birds being less reactive. However, the mean values found for latency to lie within Gait Score (GS) 0 and 1 did not differ between treatments (GS 0 mean = 00:02:28, P=0.652, GS 1 mean = 00:01:18, P=0.751). Thus, we can infer that the use of bacilli-based probiotics, regardless of the mode of administration, promotes improvement in intestinal integrity in broiler chickens and consequently better levels of serotonin and serum corticosterone demonstrating better animal well-being.

Characterizations of equine sounds as a form of expression and communication

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Acoustic signals are forms of communication and expression in horses that are still poorly understood by humans. Therefore, this study describes sounds emitted by horses associated with their emotions in response to the immediate context (stimuli) in an equestrian center. We present data from 10 stabled horses (2 females, 2 males, 6 geldings) of different breeds (2 Arabians, 2 Lusitanos, 2 crossbreeds, Criollo, Quarter Horse, English Thoroughbred, Mangalarga), 5-26 years-old, receiving water ad libitum and fed with commercial feed plus hay offered in four meals into the stall, and released daily for 5-7 hours in an individual paddock. Only one person observed each horse separately for five days and in two locations (stall and paddock)—five repetitions of 20-minute sessions in the stall and the paddock, totalling 10 observations per horse. Each sound's type, frequency and stimuli with associated ear positions were recorded individually. The types of sounds observed were snort, neigh, short neigh, and blow. The ears' positions during sound emissions were forward, sideward and backward, respectively, associated with attentive, relaxed or aggressive emotional states. The stimuli for sound emissions by horses were groomer with food, wallowing, self-grooming, interaction with another horse, presence of dogs, presence of another horse, and groomer without food. Using the Kruskal-Wallis test, there was a difference in the distribution of the types of sounds produced by horses between the two locations, stall and paddock (H= 104.01 p <0.01), and regarding the position of the horse's ear during the emission of sound (H = 170.05 p <0.01). Furthermore, the Spearman correlation showed that there is a high positive correlation between the type of sound emitted by the horse and the position of ears at the time of emission of a specific type of sound ($\rho = 0.816$, P < 0.01), regardless of the place (stall or paddock) where the horse emitted the sound. In the stall, the most observed expression was the short neigh, associated with the attitude of attention to the groomer with food; on the paddock, the most observed expression was snorting, associated with a relaxed attitude without interacting with another individual. More studies must be carried out to understand

the influence of different environments and stimuli on the acoustic communication of horses and what the emotional attitudes of horses are towards these stimuli, with a consequent improvement in the human-horse relationship.

Case study: Is shade distance from barn relevant for free-range laying hens' behavior?

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Further understanding of the behavior of laying hens in outdoor areas of free-range systems is needed. This study evaluated the use of the shade area in relation to its distance from the exit from the shed in a free-range system. A total of 60 H&N Brown Nick layers at 17 weeks of age was randomly split into three indoor boxes (20 birds/box at 0.14m2/bird). Each box had access to a corresponding outdoor area (2m x 8m; 0.8m2/bird) covered with perennial grass, fitted with an 1m high polyethylene mesh (80% retention) of 2mx2m (4m2) for shade provision. In each outdoor area, shade coverage started at either 2m (S2), 4m (S4) or 6m (S6) away from the shed exit and hen access was granted daily (07h45h to 18h30) from week 20 to 26 of age. Outdoor temperature and relative air humidity (RH), as well as the number of birds under and outside the shade per treatment were evaluated at 09h00, 12h00 and 15h00 daily for 42 days. Correlation analysis was performed with a significance level set at P>0.05. As a result, the birds chose to remain in the shade starting nearest (at 2m/10% more hens remained at 2m from the shed exit; P<0.05) to the shed exit. Total number of birds outdoors in relation to indoors and under the shade area was progressively higher the closer the shade was from the exit (P<0.05). The number of birds outside but not under the shade area was similar in all treatments (P>0.05). Even when provided with a shaded area, hens used outdoors mostly at the coolest times of the day. Total hens outdoors were inversely proportional to the ambient temperature; this behavioral profile was observed for both conditions, hens not under the shade (52m r = -0.48, 54r = -0.44 e, 56m r = -0.52; P<0.001), as for those foraging under the shade (S2m r = -0.18, S4 r = -0.21 e, S6m r = -0.18; P<0.05). In conclusion, it was observed that the hens reared under free range systems significantly preferred artificial shade areas located at 2m from the shed exit mainly during morning hours, compared to further distances of 4m and 6m. This pattern emphasizes the importance of providing adequate and accessible shade for hen protection and to optimize animal welfare in alternative housing systems.

Assessment of stress levels in horses subjected to different blood collection schemes for the production of antivenom

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Snakebite envenoming, a global public health concern, necessitates the use of antivenoms, often derived from horse blood serum, raising animal welfare concerns. This study aimed to compare the impact of two blood collection schemes on antivenom serum-producing horses. Stress biomarkers (serum and fecal cortisol levels, body temperature, body condition scores (BCS), hematocrit, and hemoglobin) were assessed. Two groups, LS (Long Scheme, 60-day interval, two inoculations, three consecutive blood collections) and SS (Short Scheme, 15-day interval, one inoculation, one blood collection), were compared. Measurements were taken at various time points: baseline (start of experiment), inoculation (snake venom injected), sampling (pre-blood collection for hematocrit and hemoglobin), and blood collection (4 to 6 liters collected from jugular vein). Mann-Whitney U tests and multivariate analyses of variance (MANOVA) were used to assess the dependent variables (N, LS=26, SS=22 horses). Serum cortisol levels differed significantly between groups across all periods (p < 0.001; LS mean = $39.29 \,\mu\text{g/dL}$, SS mean = $18.98 \,\mu\text{g/dL}$), favoring the SS scheme. Fecal cortisol levels also differed significantly in the baseline and sampling periods (p < 0.001; Baseline LS mean = 7.70 μ g/g, SS mean = 2.92 μ g/g; Sampling LS mean = 5.03 μ g/g, SS mean = 2.08 μ g/g). An interaction between the period and group was significant for serum and fecal cortisol (p < 0.001). Group differences were observed for rectal temperature during sampling (p = 0.011; LS mean = 37.33°C, SS mean = 37.55°C) and blood collection (p < 0.001; LS mean = 37.58°C, SS mean = 38.09°C). BCS remained consistent throughout the study. Hematocrit (p = 0.037; LS mean = 30.09%, SS mean = 36.64%) and hemoglobin (p = 0.018; LS mean = 28.22 g/dL, SS mean = 36.85 g/dL) levels indicated differences between groups. No significant differences were found in behavioral observations between groups across all assessed variables (Kicking, Scratching the ground, Head movement, Visible Eye sclera, Eye wrinkles, Vocalizations, Ears backward, Prominent jaw, Tense mouth, Tense nostrils, Closed eyelids, Tension above the eye area,

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Flehmen Reflex, Yawning, Urination, Defecation, jaw movement; all p > 0.05). The behavioral observations were assessed on 5 horses of each scheme. The study underscores the importance of optimizing blood collection procedures for ethical animal treatment in antivenom production. The shorter and more frequent collection scheme (SS group) demonstrated reduced stress, contributing to enhanced animal welfare and potentially improving antivenom quality.

Poster + One-Minute Presentation Session 8: Human-Animal Interactions

Wednesday, July 24th, 12:00-12:15

Beyond pet therapy: innovative approach to addressing mental health among Muslim college students through alternative animal-assisted interventions

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Animal-assisted interventions (AAIs) involve trained animals interacting with patients, complementing conventional therapies through the human-animal bond. Dogs are the most commonly used animals in AAI due to their availability, trainability, and predictability in therapeutic settings. However, dogs are not suitable for all identity groups, such as Muslims, due to perceptions about their impurity stemming from historical views about contagion and disease related to dogs' saliva. This study piloted a novel approach to identify a group's unconscious preferred animal using total time spent looking at an animal (dwell time). The study protocol, including the survey instrument, was approved by the University of Minnesota's Institutional Review Board under protocol 00022290. To test this approach, 9 white-identifying college-age student participants aged 18 to 22 completed a test using webcam-based eye-tracking software (GazeRecorder: Malopolskie, Poland) to determine stare time among animals with the assumption that greater staring time means greater preference. The eye-tracking software analyzed participants' gazes while using 10 slides, each containing two images of animals (dog, cat, hen, rabbit, mini horse) vertically split. Each slide was present for 15 seconds, and the software recorded dwell time measurements. Each species was paired with a different species until participants saw all possible pairs of companion animals. A simple linear regression model was used with dwell time (seconds) as the outcome and animal as the explanatory variable. The results showed that students displayed the highest dwell time for dogs at 8.42 seconds (95% CI: 7.40, 9.43), followed by mini horses (8.05 seconds: 95% CI: 7.03, 9.06), rabbits (7.48 seconds: 95% CI: 6.47, 8.50), hens (7.16 seconds: 95% CI: 6.15, 8.18), then cats (6.39 seconds: 95% CI: 5.37, 7.41). These results indicate that participants spent more time staring at dogs, which are the preferred animal of white-identifying college students. This preliminary data demonstrates that the eye-gazing

software is a sufficient tool in identifying preference in therapy animals. Therefore, identifying a therapy animal that is better suited for Muslim college students is possible using this software and methodology.

Death and pets: characteristics of grief among Brazilian dog and cat owners

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Death of a companion animal can be a stressful factor for owners and, in many cases, risk factor for depression. Grief is seen as the set of physical, emotional, and behavioral responses to a significant loss for the individual. Little is known about the characteristics of grief due to pet loss and studies of this nature are important for a better understanding of the humananimal relationship. We aimed to evaluate the characteristics of grief due to the loss of companion animals in a sample of Brazilian dog and cat owners. We used social networks to distribute an online survey to pet owners. A questionnaire was developed containing 36 qualitative questions related to feelings of grief, as well as socio-demographic questions of the respondents. Data was collected between Nov/2022 and Feb/2023. A total of 1,454 valid responses were obtained, of which we calculated absolute and relative frequencies. We also used Factor Analysis to extract the main factors (dimensions of grief). The analysis resulted in four factors, classified according to the nature of the feelings. Factor1 (Resignation), explained 19.01% of the total data variance (eigenvalue 3.99); Factor2 (Yearning), explained 17.29% of the variance (eigenvalue 3.63); Factor3 (Regret), explained 7.64% of the total variance (eigenvalue 1.61); Factor4 (Veterinary), explained 6.35% of the total variance (eigenvalue 1.33). Resignation ranged from higher loadings for more positive issues, such as acceptance of death and gratitude to the veterinarian who took care of the animal, in relation to high negative loadings for sentences that portray sadness and pain. The Yearning factor was composed of sentences that denote yearning for the pet, such as "I miss him when I think about him". The Regret factor was composed of higher loadings in sentences that denote feelings of regret, such as "I wish I had given him more love". The Veterinary factor was expressed by a higher positive loading for the sentence "I feel gratitude to the veterinarian", and a higher negative loading for "The veterinarian could have done more for my animal". In short, the grief profile for the loss of a companion animal differed between more positive and more pessimistic feelings. According to the literature on grief due to loss of loved ones, positive and negative feelings are part of the stages of grief. More studies are needed to verify

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the impact of pets' loss on people's mental health, and its influence on the human-pet relationship.

Effects of handling strategies on the reaction of goat kids to human restraint

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Positive interactions between human and dairy goats can bring benefits by facilitating routine milking and improve animal welfare. This study aimed to evaluate the long-term effects of three handling strategies during the suckling period of goat kids on the reactions of weaned goats when restrained by humans. The study was carried out at the Goat Teaching and Research Center at UNESP, Jaboticabal, SP, Brazil. Forty-seven goat kids, 30 Saanen (S) and 17 Anglo-Nubian (AN), 24 males (M) and 34 females (F) were subjected to one of three handling strategies as follows: NS = natural suckling, kids remained with their mothers from birth until weaning in a group pen (N=14; 9 S and 5 AN; 6 M and 9 F); NSS = natural suckling with separation, kids spent nine hours a day with their mothers from the 2nd day of life until weaning in a group pen and the rest of the time in a group cages (N=13; 8 S and 5 AN; 7 M and 6 F); and AS = artificial suckling, kids were separated from their mothers shortly after birth, and kept in group cages with three kids and fed with cow's milk in buckets with nipples (N=20; 13 S and 7 AN; 12 M and 8 F). After weaning (60 days of age), each goat kid was restrained by a familiar person, holding it in her arms for 60 seconds at 60, 75, 90, 105, and 120 days of age. Their reactions were scored from 1 (calm) to 5 (attempting to escape). Data were analyzed using a generalized linear mixed model, considering the fixed effects of handling strategy, days of age, sex, breed and animal as a random effect. There were significant effects in all fixed effects (p<0.01). The NS group showed higher reactivity to restraint (2.28±0.18) when compared to AS (1.67±0.13). At 75 days of age (2.42±0.23) was found higher values than 60 days of age (1.42±0.17). Females (2.42±0.15) were more reactive than males (1.55±0.12). And Saanen goats (2.23±0.12) showed higher values than Anglo-Nubian (1.69±0.14). We conclude that goat kids from the AS group appeared more

comfortable when restrained by humans and the reactions to human restraint vary according to the age, sex and breed of the goat kids. Therefore, establish a good human-animal interaction is important to optimize routine milking and consequently improve dairy goats' welfare.

Factors that affect handling quality during the auction sales of beef cattle in Panama

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Human-animal interaction is an important component of animal welfare assessments, with implications for the productive efficiency of beef cattle farming. However, there is a gap in knowledge regarding the quality of management during the sale of animals at auctions. This study aimed to evaluate the factors affecting the quality of beef cattle management at auctions in Panama. Videos available on YouTube were evaluated, covering 75,501 animals during the commercialization phase in four locations across the country. The quality of handling was measured by the frequency of negative tactile interactions, indicated by the presence (1) or absence (0) of blows, with sticks of wood or flags to move each animal during marketing. The reactivity of the animals was evaluated by assigning scores as follows: calm (1): the animal walks slowly and without behavioral changes, does not show aggression or sudden movements, head and neck low and relaxed; reactive (2): walks or runs continuously, with vigilant behavior, alternating the position of the legs, head and neck slightly elevated, staring and moving the ears in the direction of the noise; very reactive (3): remains with head up, very attentive to the environment, presents violent, continuous, and agile movements, turning or struggling violently, trying to pass (under, cross, and jump) the barrier. The data were analyzed using generalized linear models with sex, genetic group, animal category, reactivity, time of year, and location as fixed effects. We found that 91% (68,736) of the animals were subjected to blows during marketing. Sex did not have a significant effect (p=0.37). Taurine animals had a higher chance (OR=1.33, p<0.001), while zebu animals had a lower chance (OR=0.85, p<0.001) of receiving blows compared to crossbred animals. Cows

(OR=0.87, p=0.01), bulls (OR=0.73, p<0.001), breeding males (OR=0.79, p<0.001), and heifers (OR=0.79, p<0.001) were less likely to receive blows than were calves. Cattle classified as reactive (OR=1.25, p<0.001) and very reactive (OR=1.91, 95% p<0.001) demonstrated a greater probability of being targets of blows than those classified as calm. Animals sold during the dry season had a lower probability (OR=0.95, p=0.06) of receiving blows compared to those sold during the rainy season. No significant differences (p>0.05) were identified in the presence of negative tactile interactions between the evaluated locations. In conclusion, factors such as genetic group, animal category, reactivity levels, and time of year significantly affect the quality of management during the sale of cattle at auctions.

Human-animal interactions, cattle reactivity and gait incidents in Colombian livestock market events

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Transit of cattle through market events is stressful, facing unfamiliar environments, people and animals, which, along with other factors such as transportation, can negatively impact their welfare. The objectives of this observational study were to 1) identify the type of humananimal interactions that occur in markets, 2) determine associations between human interactions with bovine reactivity and gait incidents, and 3) investigate whether the time elapsed from the moment cattle leave the farm until they are unloaded at the market event has an impact on their reactivity and gait incidences. Thirteen (13) markets were visited (4 fairs, 4 auctions and 5 farmers markets) between June-November, 2023. Observations were made during the unloading, exhibition and uploading of animals. The type of auditory, visual and tactile human interactions (negative vs. neutral/positive) and the presentation of cattle gait incidents (no incidents, trips, slips or falls) were evaluated along with animal reactivity using a 5-point scale, where 0=calm and 4=very excited/trying to flee. During the unloading, 101 lots of cattle were observed (11 cattle/lot on average). For the exhibition and loading processes, 75 lots (5 cattle/lot on average) and 102 lots (9 cattle/lot on average) were observed, respectively. At unloading, the main human interactions were negative-tactile (35.7%) and negative-auditory (18.6%). The reactivity of the majority of cattle lots was calm (56.7%); 31% reacted by running. The most common gait incidents upon unloading were trips (23.7%) and slips (14.4%), followed by falls (9.3%). During the exhibition, negative-tactile human interactions occurred entirely in auctions and took place in 13% of the observed lots. Of the lots exhibited, 37.3% reacted by running. During loading, the main human interactions were neutral/positive-tactile (62.7%) and neutral/positive-auditory (42.2%), and the majority of cattle walked up the vehicles calmly (68%) and without gait incidents (77%). Negative-tactile human interactions were directly associated with greater reactivity of bovines when unloading (Odds ratio [OR] = 4.3; 95% CI: 1.8-10.6) and with a greater presentation of gait incidents when loading (OR = 2.6; 95% CI: 1.0-7.0). Lots of cattle with falls at unloading were lots that had a longer elapsed time between leaving the farm and being unloaded at the

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market (i.e., sum of transportation time to the market and the pre-unloading waiting time after arrival) (13.7 h [25^{th} - 75^{th} percentile: 1.2-6.1 h] vs. 2.5 h [4.6-16.4 h]; X^2 =8.2; p-value<0.01). The findings show the importance of promoting positive human-animal interactions in livestock markets and reducing transportation times.

Behavioural changes of lactating cows in a compost barn system under heat stress: a systematic review

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Here, we aim to identify behavioural changes in lactating cows housed in compost barns under heat stress. A systematic review was conducted according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) protocol guidelines. Systematic searches were carried out on Web of Science and Scopus. The search terms used were (cattle OR cow OR milking OR lactating OR "lactating cows" OR dairy OR "dairy cow" OR "dairy cattle") AND ("confined system" OR "confinement system" OR "compost-bedded pack barn" OR "compost barn" OR "compost bedded barn system") AND (climate OR heat OR "heat stress" OR "thermal comfort" OR temperature OR "relative humidity" OR "temperature and humidity index" OR THI) AND (behaviour OR "lying down" OR feeding OR "standing up" OR walking OR "water intake"). The Web of Science search returned 20 studies and Scopus 18. All the results (n = 38) were exported to Mendeley® software to remove duplicates. The remaining studies went through a four-step screening process. Step 1: Studies written in languages other than English, review articles, theses, dissertations, book chapters and conference papers were removed. Step 2: Titles and abstracts were assessed to identify and remove studies that did not use lactating cows, compost barn systems and heat stress. Step 3: The above-selected studies were reviewed and identified from their titles and abstracts. These studies that assessed the behaviour of lactating cows in the compost barn system were chosen for step four. Step 4: Finally, the studies were read in detail. Studies that did not address behavioural changes in lactating cows due to heat stress in compost barns were excluded. The remaining studies (n = 7) were included in the systematic review. The main breeds identified in the herds were Holstein and Jersey, and only one study evaluated Girolando cows (six studies number of animals between 10 - 147). The main indicators of thermal comfort were the temperature and humidity index (n = 6, stress when THI was above 74). The behavioural changes observed were reduced feeding events and lying down when the temperature was above 25 °C. In addition,

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increases were observed in the number of steps (20% rise) and agonistic behaviour followed during visits to the trough. Heat stress had a negative impact on the behaviour of lactating cows housed in composting sheds, emphasising behavioural repertoires around resting and social interaction with members of the same species.

Proposal to adapt the Welfare Quality® protocol for beef cattle raised on pasture in Brazil

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As the world's largest beef exporter, Brazil, which serves more than 150 countries, still has a long way to go to comply with the animal welfare conditions established by Brazilian legislation. The European Welfare Quality® project has developed protocols to assess animal welfare, which are based on the following principles: nutrition, housing, health and behaviour. However, the protocol was developed for cattle in confined systems, which would suit few Brazilian farms. The aim of this work is to propose an adaptation of the Welfare Quality® protocol for the pasture cattle scenario, since they are completely different systems, in addition to the different structure such as: feeders and drinkers, the form of assessment proposed by the original protocol is hampered by the relief of the paddocks. In order to adapt the protocol, 20 extensive beef cattle farms located in the north of the state of Mato Grosso were visited, chosen for their convenience and the producer's willingness to receive the researchers. The additions made were based on the principles of the original protocol: Good feeding: the use of salt as a supplement and the quality of the pasture, a pasture considered bad includes the presence of areas of exposed soil and pests. Good housing: type of Brazilian feeding trough (consider whether it has a cover or not for the integrity of the supplementation offered to the animals), presence of shading for thermal comfort (consider whether there is enough shade to shelter all the animals), type of water source (all the properties had a water source from a stream or pond). Good health: assess iron marking separately from other clinical lesions and assess all animals that can be visualised with binoculars (minimum number of 25 animals as suggested in the original protocol). Adequate behaviour: standardise the time at which the Qualitative Behavioural Assessment (QBA®) is applied. As a suggestion, the whole process should be carried out inside a vehicle, without the animals seeing the assessors. A mathematical model is being developed so that all the variables collected can be weighted with a greater or lesser impact on animal welfare, all the weights will be calculated based on

the literature. A level of animal welfare (good-medium-bad) will be generated for each principle, after which the producer will be able to carry out mitigation on the property. A protocol adapted for extensive systems could provide greater precision in animal welfare, benefiting producers and animals.

Quantitative and qualitative analysis of the behavior of racing horses from a horse club in São Paulo city

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The of horses' domestication was carried out with the aim of facilitating management and individual housing became very prevalent. Such strategies deprived the animals of showing social behaviors, grazing and an environment for exercise and energy expenditure (Vieira, 2015). Because of this restriction, stereotypies emerged, which are repetitive, invariable behaviors that do not have an apparent function, usually observed in horses under confinement and restricted forage supply (Broom and Kennedy, 1993; Houpt and McDonnell, 1993). The aim of our work was to quantify and define the most frequently observed stereotypies in high-performance horses housed in a private club in the city of São Paulo. There are few reports of similar studies, especially for athletic horses. The study is ongoing and to date we have studied 7 English Thoroughbred horses, aged between 2 and 10 years old. Despite a large variation in the age of the animals, all animals were subjected to the same diet and exercise training routine. The animals were observed for 1 hour in the afternoon (time of greatest daily activity) once a week for 6 months, totaling 27 observations in which behaviors were measured by direct observation (Ad libitum) with the help of an ethogram (Martin and Bateson, 2021). Each animal was observed for 5 minutes during three different moments during the same hour, totaling 255 minutes per individual. The animals were observed before feeding with concentrate. The most frequently observed abnormal behaviors were head shaking (17.95% of total time observed), biting door/pen (11.6% of total time observed). Other observed abnormal behaviors involved pacing (7%) and licking an empty feeder (2.6%). Although the observation was concentrated in just one hour during the day, the found prevalence of abnormal behaviors was relevant. High-performance athletic animals are normally housed individually, which could be a great challenge for sociable species. Furthermore, the lack of environmental enrichment for individuals is very common. Often, space restricted and the absence of social interactions can act as a causal factor for the manifestation of the abnormal behaviors observed. Therefore, a review of accommodation

systems and daily physical activity routines and practices is necessary, aiming to minimize the absence of social interaction and, consequently, abnormal behaviors.

Consumer perception on animal welfare certification in southern Brazil – Rio Grande do Sul

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Animal welfare has gained attention across various sectors, including academia and agriculture, emerging globally as a critical factor influencing consumer purchasing decisions for animal-derived foods. This research aimed at evaluating consumer perceptions of animal welfare (AW) and AW certified products in a supermarket in Marau, Rio Grande do Sul-Brazil, during the months of June and July 2023 (Human Research Ethics Committee/CEP approval n. 67670123.0.0000.8024). The interviewed consumers' knowledge of an international known certification scheme, their willingness to pay for AW certified products and the main mindset factors related to AW and purchasing were assessed. A qualitative, descriptive, and objective research methodology, with a questionnaire consisting of 15 open and closed questions was used to inquiry those who were seeking to purchase pasta, mayonnaise, and chicken meat, not necessarily with the Certified Humane logo (AW certification). The same researched was responsible for interviews consumers that approached the supermarket shelves with the cited products. Demographic information, consumer perceptions about AW concepts, and values they attribute to certified products were analyzed. Demographic data were computed (SAS Statistics) with chi-square distribution analysis adjusted by Pearson's test; correlation analysis was used to evaluate demographic and AW certification-related responses, considering a significance level of P<0.05. Seventy-five consumers were interviewed (61% women/39% men), mostly over 50 years old (45%). Of the respondents, 43% had a higher education degree, mainly in Humane Sciences (40%) and Agricultural Sciences (36%). Women showed greater concern for animal care (66%) compared to men (34%), associating AW concepts with proper care and, consequently, higher food quality. After questioned on the relevance of AW certification, the interviewed consumers revealed willingness to start consuming the studied products, if did not do at that moment. The qualitative questions intended to understand respondents' association between AW and AW certification indicated that "care" and "responsibility" were mostly responded. A total of 97% of the interviewed consumers stated

they consider AW certification important to ensure the care of animals that produce their food. They primarily associated AW certification with "product quality" and "responsible company" emphasizing the relevance for the corporative image. It is concluded that, to this group of consumers interviewed, the AW certification plays a crucial role in increased awareness towards ethical care of animals and promoting transparency of husbandry practices. Consumers perceive as a positive outcome for companies undertaking efforts towards social responsibility, resulting in high-quality food and ethical standards from the animal well-being perspective.

Poster + One-Minute Presentation Session 9: Applied Technologies to Assess Animal Behaviour

Wednesday, July 24th, 15:30-15:45

Age Matters: Validation of a Behavioral Monitoring Collar for Dairy Calves

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Validating the precision and accuracy of behavioral monitoring technologies is crucial for continuous and reliable assessment of animal behavior patterns in dairy farming. Despite the importance of such technologies, there remains a scarcity of validation studies concerning the use of behavioral monitoring collars (BMC) in monitoring dairy calves. Furthermore, no studies investigated if animal's age affects the BMC assertiveness. This study aimed to validate the BMC (Cowmed, Santa Maria, Brazil) for assessing activity behavior in dairy calves. Twentythree Holstein dairy calves, aged between 15 and 90 days, were randomly selected from a commercial farm. Calves were divided into three groups according to their age: 15 to 22 days (age group 1), 30 to 40 days (age group 2), and 70 to 90 days (age group 3). BMCs were placed on the calves' custom collar on the 10th day after birth. Behavioral data, including rumination, feeding, and idle time, were concurrently recorded using BMCs and visual observation. Rumination, feeding, and activity times were summed and considered as general activity. Visual observations were conducted at 1-minute intervals to align with the BMC's data logging setup. Each calf was observed for 360 minutes per day over four days, totaling 1440 minutes. Statistical analyses were conducted comparing BMC recordings and visual observations using Pearson's correlation, Lin's concordance correlation coefficient, and linear regression to validate precision and accuracy. We conducted the statistical analysis only with general activity because idle time by default is a mirror variable of general activity in this study. For age group 1, initial findings revealed promising correlations: Pearson correlation coefficient (r) of 0.36. For age group 2, correlations strengthened significantly: Pearson correlation coefficient (r) of 0.66, suggesting a more pronounced relationship between BMC measurements and observed

behavior. The enthusiasm peaks for age group 3, where correlations are higher: results for Pearson correlation coefficient (r) of 0.90. Linear regression analysis revealed insights into the trends of activity across different age groups. The slope for age group 1 was 0.38 (p=0.37; intercept = 98.05), 2 was 0.55 (p=0.10; intercept = 96.87), and 3 was 1.09 (p=0.005; intercept = 4.72), indicating accelerated behavioral changes as calves approached maturity. These robust correlations, accompanied by significant linear regression parameters, underscore the BMC's unparalleled accuracy in capturing behavioral dynamics in older calves. These findings underscore the importance of considering age-specific behavioral dynamics in dairy calf management strategies.

Deep machine learning for farm animal behavior studies with the YOLOv8 algorithm: automated facial part detection and segmentation for facial expression and distress inference in sheep

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Animal welfare assessment often relies on skilled observers to interpret subtle cues reflecting emotional states and distress levels. However, manual evaluation can be time-consuming, inconsistent between raters, and not scalable. Computer vision provides an opportunity to automatically analyze animal facial expressions and contribute objective insights. The primary objective of this study was to precisely detect and segment facial components, including the ears, eyes, and nose and mouth, as individual bounding boxes and segments for making inferences about the emotional states or distress of the animals. The state-of-the-art YOLOv8 (You Only Look Once version 8) algorithm was employed to detect and segment facial components in 546 sheep images from multiple breeds (train = 382, test = 164) from the Sheep Facial Expression Primary Dataset*. Target facial parts included ears, eyes, and nose and mouth. The authors manually annotated the images for each facial part class, which were used for model training. The dataset encompasses diverse background conditions and animal poses. With optimally tuned parameters, the best YOLOv8 model achieved an overall box precision of 0.988, recall of 0.969, and mask mAP50 of 0.989 across all 164 sheep images (784 instances). For the ear class (164 images, 324 instances), box precision was 0.984, recall 0.959, and mask mAP50 0.991. The eye class (164 images, 297 instances) achieved a box precision of 0.991, recall of 0.953, and mask mAP50 of 0.982. For mouth and nose (164 images, 163 instances), the precision was 0.988, recall 0.996, and mAP50 was 0.993. The inference speed was 5.4ms per image. The results indicate the feasibility of precisely localizing and segmenting key sheep facial parts. This approach, while focused on sheep in this study, has broader implications for livestock welfare assessment. Although distress inference was not explicitly performed in this study, the precise facial part detection and segmentation facilitated the

analysis of facial expressions for distress inference, a crucial step in understanding animal welfare. Future research will explore keypoint detection to further enhance automated facial analysis. This study demonstrates the feasibility and effectiveness of employing YOLOv8 for automated multi-facial feature detection and segmentation. The methodology provides a foundation for scalable and less biased livestock welfare assessment, contributing to advancements in real-time animal welfare monitoring and management practices.

Identification of dairy cattle from barn cameras

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The automatic recognition of dairy cattle using overhead aerial cameras in barn settings holds considerable promise for agricultural technology applications. Deep learning offers a viable solution for identifying cattle in these intricate environments. However, it requires extensive data for training, and the procurement of annotated data on a large scale is challenging due to the impracticalities of annotation in barn environments. Although unlabeled data can be easily amassed through ongoing camera surveillance, it lacks annotations, making model training difficult. A common workaround is to gather data in structured environments, such as milking parlours, where RFID scanners and cameras collect synchronized image and RFID data. This method, however, falls short as it does not represent the full range of behaviours, movements, lighting, and perspectives encountered in an open barn setting. As a result, models trained exclusively in the milking parlour environment perform poorly when applied to barn conditions. This research introduces novel self-supervised learning strategies and automated data processing techniques to effectively utilize unlabeled barn data. Our approach uses a tracking algorithm to generate localized images of cows across several video frames, creating a scalable, weakly labelled dataset linked to tracking IDs. These weak labels are then used in a triplet loss function for self-supervised model training. We assessed the model on a fully labelled image dataset comprising 1785 cows captured in the easily annotated milking parlour from the same farm where the videos were captured. The models trained with our self-supervised method achieved over 63% accuracy on the milking parlour test set, despite no previous knowledge of this domain. Additionally, our method is able to identify cattle in the barn by matching their images to those from the milking parlour tagged with corresponding RFIDs. This study highlights the potential of self-supervised learning to address the challenges of cattle identification in barn settings, thereby advancing livestock identification and welfare monitoring technologies.

Something is different in their voice: discriminating different broiler breeds using vocalization features

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A thorough analysis of the distinctive vocalization features is important to decipher the acoustical code for a specific breed of broiler and also to understand the evolution of communication signals in different breeds. Vocalization is a non-invasive tool for broiler breed identification. This study examined the effect of breed on vocalization features and checked the possibility of using these vocalization features to discriminate different broiler breeds. A total of 140 broilers (35 Hubbard Flex, 38 Arbor Acres Plus, 30 Ross 308, and 37 Cobb 500) were used for the experiment. The vocalization of individual birds was recorded, and vocalization features were extracted using SAP2011. The vocalization features (mean frequency, peak frequency, pitch, amplitude modulation, frequency modulation, amplitude, goodness, and entropy) were subjected to a general linear model procedure and multivariate discriminant analysis. The amplitude differed between Arbor Acres Plus and Hubbard Flex (p<0.001), while there was no difference for other breeds. There was a breed effect on goodness (p=0.02) with Cobb 500 and Hubbard Flex having different goodness (p=0.01). The mean entropy differed between Cobb 500 and Arbor Acres Plus (p=0.01), while there was no difference between the entropy of Hubbard Flex and Cobb 500 (p=0.42), Hubbard Flex and Ross 308 (p=0.61), Ross 308 and Arbor Acres Plus (p=0.93) as well as between Hubbard Flex and Arbor Acres Plus (p=0.26). Six vocalization features (amplitude, entropy, mean frequency, goodness, amplitude modulation, and peak frequency) out of the original eight features were effective in discriminating the four broiler breeds. Three canonical variables that accounted for 58.43%, 34.51%, and 7.05% of the total variation were identified. Amplitude was highly correlated with CAN 1. Entropy was highly correlated with CAN 2, while mean frequency, peak frequency, and amplitude modulation were moderately correlated with CAN 2. Goodness was highly correlated with CAN 3, while peak frequency was moderately correlated with CAN 3. The shortest Mahalanobis distance was observed between Ross 308 and Hubbard Flex, while

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the longest was observed between Hubbard Flex and Arbor Acres Plus. 65.00% of Arbor Acres Plus, 70% of Cobb 500, 60.00% of Hubbard Flex, and 55.00% of Ross 308 were correctly assigned to their respective breeds. The accuracy of the result obtained in this study may be improved using a higher sample size. The results obtained from this study revealed that the vocalization feature is a promising non-invasive tool that can be used for broiler breed identification.

Validating the use of RFID technology to monitor drinking behavior of broiler breeder pullets

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Drinking behavior patterns of poultry can inform water management practices to ensure adequate hydration and minimize water spillage and wet litter. Automated behavior monitoring with radio frequency identification (RFID) technology is a possible management tool. We aimed to validate the use of ultra-high frequency RFID technology to monitor drinking behavior of broiler breeder pullets. Sixty Cobb 500 female pullets were housed in a floor pen, fed a daily restrictive diet, and provided ad libitum water through a drinker line mounted with two RFID readers. RFID leg bands were attached to 10 colored focal birds. At 4 weeks of age, RFID readers and video cameras simultaneously recorded the pen (8hr photoperiod). Drinking duration and frequency were downloaded from the RFID software, while a trained observer coded the same behavioral data continuously from video footage. Values obtained for drinking behavior were compared between RFID and video observations via correlation and regression analyses. For regression, RFID was considered to accurately estimate drinking behavior if three criteria were met: R²≥0.90, slope not statistically different from 1 (p>0.05), and intercept not statistically different from 0 (p>0.05). From video only, we assessed the effect of the time of day on drinking duration and frequency with linear and general linear models and the coefficient of variation (CV) across focal birds. Pullets spent 16.1±4.8 min/bird (mean±SD) drinking over 8hr. They visited the drinker 212.8±64.3 times/bird for 4.5±0.2 sec/bout/bird. There was moderate variability across pullets for drinking behavior (30% CV for duration and frequency). One hour after feeding, pullets spent the most time drinking and made the greatest number of visits to the water line for a 60 min period, then decreased to a consistent pattern for the remainder of the photoperiod (p<0.0001 for both models). Drinking behavior had a weak, negative correlation between the RFID and video observation methods (r=-0.34 and -0.33 for duration and frequency, respectively), which was also reflected in the adjusted R² (0.002 and -0.0005). RFID

consistently underestimated drinking behavior compared to video observations as indicated by different regression intercepts (duration:1014.6, p<0.0001; frequency: 243.1, p=0.0001) but the slope did not differ (duration: -2.6, p=0.20; frequency: -5.8, p=0.28). The validity of RFID may have been poor for this pen due to the position of birds while drinking, the position of the readers, interference between readers, short drinking bouts, or movement of the suspended water line. Further investigation into refining these factors should be pursued to improve validity.

Can a machine learning algorithm adjust castration-induced swine pain diagnosis by non-experienced observers using a behavioral scale?

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Domestic swine is an important species to livestock and research. In both situations, pigs are exposed to painful procedures, and monitoring pain is critical from a welfare standpoint. Unesp-Botucatu Pig Composite Acute Pain Scale (UPAPS) has been employed worldwide to assess pain in pigs through behavior. UPAPS was built based on experienced observers, and therefore, the lack of experience level may interfere with pain diagnosis. Artificial intelligence tools have been widely employed to refine pain diagnosis in farm animals. However, no studies are trying to fill the gap from differences in observers' experience levels in pain diagnosing. We aimed to investigate if a random forest algorithm can improve the swine pain diagnosis by non-experienced observers using UPAPS. In the first step, we used a database of 48 weaned and 16 pre-weaned pigs assessed by five experienced observers using UPAPS pre- (free-pain condition) and post-castration (painful condition), which was split into training (70%) and testing (30%) sets. The random forest algorithm target variable was the free-pain and painful conditions, while UPAPS behaviors were used as feature variables using the training set. Algorithm hyperparameters and cross-validation parameters were optimized using the grid search technique testing 3,888 algorithms. The algorithm with the highest accuracy contained two feature variables in each decision tree, 501 trees in the forest, four-folds, and nine repetitions, which was assumed for the study. The probability of the pig being painful and its threshold were calculated in the testing set. Pain diagnosis quality of the algorithm was

assessed by the area under the curve (AUC) in the testing set. In the second step, UPAPS of 10 weaned and 10 pre-weaned pigs in pre- (free-pain condition) and post-castration (painful condition) scored by six non-experienced observers from another database were input into the algorithm to obtain a painful indication according to the algorithm threshold. The algorithm had an AUC of 91.38%. From non-experienced observers' scores, the original scale and algorithm had a specificity of 47.5 and 42.5%, and a sensitivity of 76.6 and 78.3%, respectively. The algorithm improved sensitivity by 1.7% and diminished specificity by 5%. From a practical point of view, low sensitivity could be a concern, meaning that non-experienced observers are less likely to recognize the swine in painful conditions. In conclusion, the algorithm could not appropriately adjust pain diagnoses by non-experienced observers scoring UPAPS. More studies should address preventing misdiagnosed pain enlarging the sample size and advancing pigs' welfare.

Random forest algorithm identifying best-ranked pain-altered behaviors to improve acute pain diagnosing in castrated swine - preliminary results

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Surgical castration is a standard painful procedure performed on swine farms to avoid boar taint. In 2020, the Unesp-Botucatu Composite Acute Pain Scale for Pigs (UPAPS), a speciesspecific behavioral scale, was validated for diagnosing acute pain in pigs after surgical castration. Current work utilizes random forests to rank behaviors and improve pain diagnosis for pain assessment scales for sheep, horses, and humans. This algorithm can capture a pattern in the data after training using several decision trees, improving accuracy. However, this approach has yet to be explored in pigs. Therefore, we aimed to rank each UPAPS painaltered behavior based on the random forest algorithm and compare the algorithm's predictive capacity of pain diagnosis with the original UPAPS. Pain scores were used from a previous study evaluating pain in 45 weaned piglets and 14 pre-weaned piglets. Videos were collected at two timepoints: pre-castration (pain-free condition) and post-castration (painful condition). Five observers masked to timepoint were trained and assessed all the videos using the UPAPS. A random forest algorithm was applied using UPAPS pain-altered behaviors scored by observers as feature variables and conditions as a target variable. To train the algorithm (training base), 70% of the pigs were selected randomly, while the predictive capacity was evaluated with 30% of the pigs (test base). The algorithm was optimized by adjusting the random forest hyperparameters and cross-validation parameters using the grid search technique. The random forest algorithm with the highest accuracy (94.26%) contained two feature variables in each decision tree, 501 trees in the forest, four-folds, and nine repetitions. DeLong test was used to compare the area under the curve (AUC) between the algorithm and UPAPS. Our results indicated that the five best-ranked pain-altered behaviors were

respectively, 'Head down' (100%), 'Sits with difficulty', (93.33%), 'Wags tail' (91.11%), 'Interaction 2: occasionally moves away from the other animals, but accepts approaches; shows little interest in the surroundings' (86.67%) and 'Activity 1; moves with less frequency' (64.44%). These behaviors may represent an evolutionary adaptive value to avoid activities not essential for survival, conserving energy, and protecting the injured area. Furthermore, the AUC of the algorithm (91.38%) and of the UPAPS (90.58%) had "good" predictive capacity and were statistically equivalent (p=0.85). In conclusion, the algorithm made it possible to identify a ranking of pain-altered behaviors without impairing the predictive capacity. Future studies could explore behavioral ranking based on random forest algorithm to refine pain scales across species.

"Bite-sized" observation windows misrepresent 24-h oral behavior performance in dairy calves

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Dairy calf normal and abnormal oral behavior is often measured as a noninvasive indicator of welfare. Research has previously investigated whether time sampling could be used as a reliable, timesaving alternative to continuous observation, but limited efforts have validated sampling over shorter time periods, compared to 24h. We assessed whether common observation windows could reliably estimate 24-h performance. Twenty-seven Holstein female calves were housed individually and limit-fed milk (3.8-5.6L/d step-up), common US practices, via bottle at approximately 0915 and 1615h. Calves received water and grain (n=9 calves) with additional access to long chopped mountaingrass hay (n=18 calves) from wk 0-7. Feed and water were topped up at 0800, 1200, 1600, and 2000h to maintain ad libitum levels. Oral behavior was recorded via direct observation using 1-0 sampling at 5-s intervals for 24h. One calf was observed at a time for a continuous 24h in wk 4 and 6, generating 54 calf-days of data (2d/calf). We measured 6 oral behaviors, presented with 24-h mean±SD as context of their prevalence: rumination (22±7%), eating (5±3%), drinking water (0.6±0.2%), selfgrooming (3±1%), non-nutritive oral manipulation of pen structures (NNOM; 7±3%), and tongue rolling (0.02±0.07%). We used these 24-h data to determine proportion of time engaged in each behavior over daylight hours (0800-2000h) and 1 or 2h after the end of each milk feeding. We calculated proportions by dividing the number of 5-s intervals in which each behavior occurred by the total number possible in the observation windows. Proportions generated from each window were compared via a linear regression against 24-h performance. Observation windows were considered to accurately represent 24-h values if the $R^2 \ge 0.9$, slope=1, and intercept=0. No behavior met all 3 criteria in any window <24h. Rumination happened to a greater extent overnight (31% from 2000-0800h vs. 12% from 0800-2000h) and all observation windows underestimated it. Observing for 1h after milk feedings led to estimates of 0% rumination for 48% of calf-days. All observation windows overestimated NNOM and eating with increasingly large discrepancies in individuals that did

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these behaviors more. Grooming met criteria for slope and intercept, but windows <24h did not predict 24-h values well ($R^2 \le 0.57$). Both tongue rolling and drinking water were rare, making reliable estimation difficult. Daylight observations were more correlated to 24-h values than observations after milk feeding in all behaviors. Overall, our results suggest that observation windows <24h, while helpful in reducing labor, misrepresent 24-h calf oral behavior.

Poster + One-Minute Presentation Session 10: Integrating Animal Welfare & Perception of Welfare and Ethology

Wednesday, July 24th, 15:30-15:45

Affective state (anxiety) in six broiler chicken strains when housed with A-frame huts or platforms

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Fast-growing broiler chickens can experience poor welfare associated with rapid weight gain. To mitigate this, producers may use slower-growing strains or add resources such as platforms or huts. Affect (long-term mood state), including anxiety (negative affective state), may be impacted differently when strains with different growth rates are raised with these resources. The objective was to assess the impact of resource type on anxiety in three fast- and three slow-growing broiler strains. All procedures complied with ISAE's ethical guidelines. In a 2×6 factorial approach, two resources were provided to birds of six broiler strains (bird n=1584), resulting in 12 treatment groups with 6 replicates (71 pens; only 5 replicates for 1/6 strains). Pens contained a plastic platform with ramps or an A-frame plastic hut. The growth rate ranged between 78-85g/day for fast-growing strains: Cobb 500, Ross 708, and Ross 308, and between 38-51g/day for slow-growing strains: Freedom Ranger Color Yield, Hubbard Redbro Mini, and Hubbard Ja57ki. Six birds/pen underwent the attention bias test at 3.7 kg liveweight. Three birds were placed in an arena with shavings and a feeder with feed and mealworms. Positive (feed) and negative (8-sec conspecific alarm call) stimuli were simultaneously presented and attention bias (indicative of anxiety) was quantified. We recorded the number of birds that began feeding for all birds (n=432) and the proportion of time birds spent vigilant for one of three birds per test (n=136). Fewer birds feeding and prolonged vigilance indicate a greater bias towards the negative stimulus, thus more anxiety. Data were analyzed in JMP Pro 16 using a nominal logistic regression, and a mixed model with resource and strain as predictors, and pen as the random factor. Resource type did not impact the proportion of birds feeding (hut: 20.8%, platform: 19.4% of birds; p=0.834) or vigilance behavior (hut: 42.0%,

platform: 40.8% of time; p=0.700). The proportion of birds feeding differed between strains (p<0.001), with Redbro Minis (36.4%) and Ja57kis (38.9%) having greater odds (OR≥3.54) of feeding than other strains (9.7%-14.1%). Redbro Minis (35.4% of time) spent less time being vigilant than Ross 308s (53.2% of time; p=0.026). These results indicate that fewer birds of the two slowest-growing strains showed anxiety compared to all other faster-growing strains. Resource treatments did not impact anxiety. Therefore, producers may reduce the number of birds experiencing anxiety, thus improving broiler chicken welfare, by producing Redbro Minis or Ja57kis.

Barriers and facilitators of adoption of alterative pig farming systems on the island of Ireland

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In Ireland, the majority of pig production occurs within conventional indoor systems. However, interest in alternative pigs farming systems is increasing across Europe and further afield, in response to concerns around consumer preferences, sustainability, animal welfare, and human health. Theoretical frameworks from the social sciences can be used to understand human behaviour in relation to animal welfare. Utilising a theory-informed approach, the aim of this research was to gather and interpret information on the barriers to, and facilitators of, high welfare pig production systems in Northern Ireland (NI) and the Republic of Ireland (ROI). Semi-structured interviews were conducted with conventional indoor (CSF) and alternative (free-range and organic) system (ASF) pig farmers to identify individual-level barriers and facilitators of higher welfare pig production. Additionally, two focus groups were held for conventional indoor farmers and processors, and alternative outdoor farmers and processors to identify industry-level barriers and facilitators. The COM-B model, Theoretical Domains Framework [TDF], and the One Welfare concept (Animal Welfare [AW], Human Wellbeing [HW] and Environmental Conservation [EC]), were used to structure the interview and focus group schedules. Participants were recruited using purposive sampling, and interview and focus group discussions were recorded, and transcribed verbatim. A deductive Framework analysis approach was utilised. Nine participants took part in the focus groups (Conventional system, n = 5, Alternative system, n = 4) and 19 interviews were conducted (Conventional system, n = 9; Alternative system, n = 10). Barriers to uptake of alternative systems related to AW [CSF perceived welfare as being compromised outdoors), HW (poor CSF wellbeing currently without asking further changes to farming practices to be made) and EC (fears of soil damage and soil leaching). Facilitators to uptake of alternative systems related to AW (freedom of the pig to express natural behaviour), HW (ASF enjoyment of working outdoors) and EC (growing feed on-farm). There were some perceived conflicts within One Welfare (e.g.

Natural motivation of the pig to root leads to soil damage and leaching) as well as synergies (e.g. Natural motivation of the pig to root for use in woodland management). The findings from this study can be used to inform the direction of efforts to improve pig welfare and encourage high welfare pig production systems on the island of Ireland and beyond.

Evaluation of the welfare of dairy cattle in year-round grazing systems in the Azores

Eveli Somelar¹, David Arney¹, Francesca Carnovale¹

The practice of year-round grazing for dairy cattle should ensure high-quality welfare and dairy products, with "happy cows" labelling on pasture milk products aligning with consumer expectations. However, the actual effects of all-year grazing on dairy cow welfare have not been thoroughly investigated and putative judgement on this is not based on scientific evidence. This project's aims were to assess those behaviours of grazing dairy cattle which external grazing factors may influence. The study was carried out between August 2023 and January 2024, on Terceira Island, Azores, during both the summer (hot) and winter (wet) seasons. The Azores, a subtropical region in Portugal, representing 30% of the total Portuguese dairy sector, is known for lush green pastures, and dairy farms' use of year-round grazing, adopting sustainable practices and promoting animal welfare. Behaviour was assessed individually with the Avoidance Distance (AD) test and in herd groups together with qualitative behaviour assessment (QBA) and social behaviour assessment (SB). Fourteen different dairy cattle farms were assessed with an average of 2.7 visits to each farm and herd sizes ranged from 21 to 303 lactating cows. A total of 1,554 cattle were subjected to two avoidance tests for a total of 1.9 visits for each farm; around 60% (AD1) and 63% (AD2) of cattle allowed themselves to be touched by the assessors, the rest of the animals tested had an avoidance mean 57.4 cm ± 1.6 SE (AD1) and 64.11 cm ±1.8 SE (AD2). The QBA assessment of the herd showed that most of the cattle were calm, content, positively occupied and friendly. Less than 20 % showed frustration, fear, or irritability. The cattle during the social assessment behavioural observations spent most of their time standing or feeding and less than 2% of the time was spent on social licking or other social activities. The behaviours observed during this study show that in this grazing system, all year-round grazing, the dairy cows were mostly happy and calm. These results verify that year-round grazing can be beneficial to the welfare of dairy cows, and this can contribute to improvement in the wellbeing of the animals.

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Animals and Society: A systematic review of public opinions toward management practices adopted on dairy farms

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The opinions of society have the power to impact the profitability of the dairy industry. Then, to maintain a social license to operate dairy systems, animal management practices must be accepted by society and meet the behavioral needs of animals. We carried out a systematic review to identify the public opinion in the literature regarding animal management practices adopted on dairy farms. This review employed the PICO framework (Population, Intervention, Comparison, and Outcome) for its search strategy and was conducted on Web of Science and Scopus. The search terms were applied to the titles, abstracts, and keywords with the following combination: ((perception OR opinion OR perspective OR attitude OR view) AND (citizens OR public OR society OR consumer OR 'lay public') AND ('dairy cattle') AND (practices OR management)). Studies published in English and peer-reviewed (n=266) underwent a 4step evaluation process, following PRISMA protocols and guidelines. Thirteen articles that evaluated the public opinion of at least one animal management practice on dairy farms, published between 2000 and 2024, fitted our selection criteria. These surveys were carried out mainly in the United States (7/13), Canada (4/13), Germany (2/13), Brazil (2/13) and United Kingdom (2/13). Among the studied management practices, we found articles (8/13) that evaluate public opinions regarding cow-calf contact, where public attitudes and perceptions of animal welfare were more positive when cow-calf were not separated. Six studies evaluated the access to pasture areas, the results showed that the public reject systems where cows had no access to pasture. Some studies (3/13) evaluated practices that

public had negative attitude such as dehorning without pain mitigation and calves housed individually, while the use of antibiotics in milk production and genetic modifications for reproductive purposes were considered as unnatural (2/13). In general, the public showed a more positive attitude toward systems that gave the animals the possibility of living in situations closer to natural, which makes the ideal scenario for dairy animals. Being aware of public opinion toward management practices used on dairy farms can contribute to predicting future changes and making then in advance. Regardless of the importance of public opinion, few studies were carried out, and are concentrated on developing countries. Studies in different world regions are necessary to boost changes in the dairy industry to improve communication about positive practices to better meet different populations needs and also understand different public perception about the topic.

Public perceptions of free-roaming dogs and cats in India and the United States

Brittany Watson¹, Reshmi Sensharma, Chelsea Reinhard¹, Lauren Powell¹

A cross sectional study was developed to describe differences between India and the United States in public perceptions of free-roaming dogs and cats, concerns related to free-roaming animals, and preferred strategies for veterinary medical interventions and population management. This study was reviewed and approved by the Institutional Review Board at the University of Pennsylvania (protocol number 849324) and informed consent was obtained from all participants. Between August 2021 and February 2022, 498 individuals, including 210 Indian respondents and 288 American respondents, completed a 58 questions online survey. Results were analyzed with Pearson Chi Square and Mann Whitney U tests. Free-roaming cats and dogs were largely perceived as community animals among Indian respondents (median of 1.00, IQR 1.00-1.00 in cats in India versus 3.00, IQR 2.00-4.00 in cats in US; median of 1.00, IQR 1.00-1.00 in dogs in India versus 4.00, IQR 2.00-5.00 in dogs in US), with significantly more respondents indicating they should be allowed to roam freely (median of 2.00, IQR 1.00-2.75 in cats in India versus 4.00, IQR 2.00-5.00 in cats in US; median of 1.00, and IQR 1.00-3.00 in dogs in India versus 5.00, IQR 4.00-5.00 in dogs in US). Respondents from both countries were concerned about animal welfare (56.2% in India, 85.8% in US for cats; 60.5% in India, 93.4% in US for dogs), although Americans were significantly more likely to list animal welfare (X2= 54.26, p<0.01 in cats, X2= 80.84, p<0.01 in dogs), public health (X2= 64.65 in cats, p<0.01, X2= 55.66, p<0.01 in dogs) and wildlife risks (X2= 114.09, p<0.01 in cats, X2= 46.89, p<0.01 in dogs) as significant concerns. American respondents were more likely to support adoption for sociable animals and euthanasia for unsociable animals (72.4% for cats, 89.5% for dogs in US; 46.2% for cats and 55.4% for dogs in India), whereas Indian respondents were more likely to support spay/neuter, vaccinate and release strategies for both dogs and cats (36% for cats, 28.7% for dogs in India; 15.4% in cats, 2.8% for dogs in US). Findings speak to the importance of implementing tailored strategies for free-roaming cat and dog management based on local cultures and community perceptions. By recognizing and responding to global variations in perceptions of animals, policymakers, veterinarians, and animal welfare organizations could

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develop more context-specific and effective approaches for improving the well-being of animals, promoting public health, and reducing human-animal conflicts.

Training to recognize pain-related behaviors improves students' decision-making to indicate analgesia in farm animals

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Many painful husbandry practices are routinely conducted on farm animals. Behavioral changes are key components to identifying pain in animals. Pain identification is crucial for decision making in providing appropriate treatment. Behavioral changes are easy to identify and can be observed remotely and non-invasively. However, a limitation is that inexperienced evaluators or those who are not aware of which behaviors are relevant indicators of pain, may not be able to correctly identify pain in animals. This study aimed to compare the indication of analgesic intervention by students in cattle and pigs before and after training. After watching videos (up to 5 minutes), first- and third-year veterinary medicine students (n=60) were asked whether they would provide analgesia for a bull and a pig suffering intense pain before and after students were trained to identify pain-related behaviors. The training was led by a senior professor (expert) with over 30 years of experience in animal anesthesia and pain assessment. Training consisted of watching tutorial videos of relevant pain behaviors according to the validated behavioral pain scales (posture, locomotion, interactive behavior, activity, appetite, attention to the affected area, tail wagging, foot stamping, head and hind limbs position, difficulty in overcoming obstacles, lying in ventral recumbency, and biting bars or objects) for cattle and pigs (videos are available on the www.animalpain.org website or Vetpain application). Before and after training, students watched one video of each species and indicated whether they would provide analgesia according to their opinion. First-year students evaluated the bulls, and third-year students evaluated the pigs. Chi-Square test was used to compare the students' indications for rescue analgesia before and after training. According to the expert's assessments all animals were suffering intense pain and required analgesia. Students recommended fewer analgesic rescues before training compared to after

training for both species (84.61% vs. 100.00% for bulls, p = 0.03362; 71.43% vs. 100.00% for pigs, p = 0.02747). After training, all students recommended analgesic rescue for the animals, just like the expert. In conclusion, after a brief training students recognized better the need for analgesic intervention in cattle and pigs experiencing intense pain, which highlights the importance of educating veterinary medicine students about animal pain behaviors to improve the welfare of cattle and pigs.

Sentience and animal use: What shapes students' ethical perspectives

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We investigated the influence of several variables on the attitude of undergraduate students to the attribution of sentience to animals and the use of animals for different purposes. Students from 68 undergraduate courses at the Federal University of Santa Catarina, Brazil (2015, n=1007; 2022, n=959) answered an online questionnaire addressing attitudes towards animal use (zoos, teaching, biomedical research, animal production, cultural traditions, recreational fishing, subsistence and recreational hunting) and the attribution to feel five different emotions to animals, using a Likert scale from 1 to 5. Demographic data (sex, age, region of the country, rural/urban living); undergraduate course; year of participation in the survey (2015 or 2022); having a pet or farmed animal; experience with animal use were also collected. Data were submitted to a Multiple Correspondence Analysis and Hierarchical Clustering using the software R. We discarded 27 individuals considered outliers. Cluster 1 (n=1112) had a predominance of females who lived most of their lives in big cities, were arts and biological science students and had the lowest contact with animals and experience with animal use. This group attributed the highest capacity of sentience to animals. Cluster 2 (n=552) had an expressive contribution from younger participants who had lived in big cities and were STEM students. Their attribution of sentience to animals was the lowest among the clusters. Cluster 3 (n=275) concentrated participants that reported the highest contact with animals, including farmed species, more experience in different forms of animal use, had lived in rural or small towns, and were enrolled in agricultural courses. This group attributed a lower capacity of sentience to animals than Cluster 1 and higher than Cluster 2. Support for animal use was lowest in Cluster 1. Cluster 3 reported similar support as C2 for most animal uses (except higher support for food production and sports hunting) despite the higher attribution of sentience to animals than this group. Our findings indicate that a combination of belief in animal minds, demographic characteristics and experience with animals and their use

underlie support for animal use. In the search for a more ethical world for non-human animals, increasing awareness of animal sentience can be an essential tool for transformation, especially among young people. However, besides belief in animal minds, other cultural factors associated with rural living and experience with animal use may underlie utilitarian views that justify support for animal use.

Does science subsidize policies for promoting a sustainable future in livestock?

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It has been 60 years since the book Animal Machines (1964) was published by Ruth Harrison, which describes how the animals were treated in the industry, under extreme confinement and the impossibility to express highly motivated behaviors selected throughout the years by evolutionary mechanisms. After over a decade of looking into the issues of industrial farming, a document proposing minimum standards was created and later named the Five Freedoms (which was the basis for the creation of the Five Domains). From that moment to now, the scientific evidence showing that industry was imposing pain, and suffering on sentient complex beings, is increasing fast and worldwide, with a growing and irreversible paradigm shift regarding the cognitive complexity of non-human animals. However, after six decades the world's default system for producing animals is still the most harmful. Looking back at the problems brought about by the book Animal Machines, we realize they were essentially the same animals are currently facing (castration and tail-docking without pain management and intensive confinement, to name a few). Even though science has already proven animals are sentient beings, the industry fails to provide sufficient conditions for them to live a life worth living. It is estimated that around 23 billion animals are on factory farms, many of them in extreme confinement conditions. In February 2024, 146,166 published scientific papers were found in just one database with the combination of words "animal welfare", but despite the scientific knowledge available, obvious practices in non-compliance with the interest of animals to not suffer are still present in the daily routine. Only considering laying hens regarding fractures during the productive cycle, the accumulated pain is estimated to be approximately 159 hours of disabling pain, 2,248 hours in painful pain, and 1,812 hours in annoying pain, which correspond to a substantial fraction of the laying cycle. Despite the suffering that must be considered on an individual level, if we consider the number of animals across the world the total hours in pain is huge. Another example of the mismatch of science and what happens in the animal industry is the misuse of antibiotics in non-therapeutic

protocols. Antimicrobial resistance poses a considerable threat to public health, with an estimated 4.95 million deaths associated with bacterial antimicrobial resistance in 2019. Based on these few examples, it is time to rethink the effectiveness of science influencing and impacting public policies.

Harmony in husbandry: the nexus of animal health, welfare, and sustainable practices in Nigeria

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Animal welfare in Nigeria faces numerous challenges, notably the absence of specific policies and guidelines, rendering existing general policies on animal care ineffective due to noncompliance and non-enforcement. Consequently, the advancement of animal welfare has largely relied on non-policy means. This abstract delves into the intricate interplay between animal health and welfare, spotlighting their symbiotic relationship as a compelling catalyst for the promotion of comprehensive welfare practices. The narrative centers on a case study involving a pasteurellosis outbreak on a rabbit farm, underscoring the pivotal role of diligent welfare measures in averting and managing health crises. In a breeder farm in Ile-Ife, Nigeria, thirty-five apparently healthy female rabbits underwent three successful kindlings and subsequent mating in September 2023. The abrupt loss of one hundred and thirty-seven litters within a week raised concerns. Mating in October also resulted in repeated fatalities, by December there was mortality of seventeen breeders that displayed symptoms of nasal discharge, ocular conjunctivitis, torticollis, and weight loss. Veterinary examination and postmortem analysis confirmed pasteurellosis as the diagnosis, prompting an evaluation of health and welfare parameters on the farm. The rabbits were individually housed in cages with wooden frames and wire mesh cage of 24by 30 inches, with feeders for pelleted rations and automatic freshwater drinkers. Despite adherence to a commercial pellet diet meeting National research council guideline (NRC), stress induced by sudden environmental changes emerged as a potential factor compromising the rabbits' immune systems and making them prone to disease. Monthly weather records revealed extreme conditions, particularly in January with an average temperature of 40.1°C. The wettest month, September, recorded highest temperature of 35.5°C and average temperature of 31°C and 94.69% humidity, while the driest month, December, saw a highest temperature of 44.5°C, average temperature of 38.8°C and 54% humidity. Educational interventions included implementing cooling systems, adjusting feeding schedules, and modifying breeding practices to mitigate heat-related stressors. Quarantine and biosecurity lapses were identified, prompting recommendations

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for foot-dip stations, reduced contact with wildlife, and repairs to broken netting allowing entry of pests. Advocacy for regular veterinary check-ups and swift attention to signs of illness for early disease management was emphasized. Additionally, introducing enriching elements to reduce boredom and stress. No more mortality was recorded after intervention and the remaining animals kindled by March, 2024. By elucidating the tangible benefits of conscientious husbandry practices, the synergistic relationship between animal health and welfare as a pivotal selling point was identified.

Farmers perception of lameness on pasture-based dairy farms

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Lameness in dairy cows affects both cow welfare and farm economics. This study assessed lameness awareness and management of 48 dairy farmers operating small-scale grazing herds in Minas Gerais, Brazil. On each of these farms, all lactating cows were mobility scored by trained assessors (using a 0 to 3 scale) and farm managers completed a questionnaire. Farms size ranged from 18 to 130 lactating cows, with an average milk yield of 12.7 L/cow/day. The farmers' estimated lameness prevalence averaged 9.9%, versus 16.1% as assessed by researchers, but these estimates were positively correlated (r = 0.66). Lameness was cited as a primary concern by 37.8% of farmers, but many farms appeared to rely on treatments of questionable efficacy, such as parenteral antibiotics (52.2%) and various disinfectant solutions (19.6%). Farmers cited structural inadequacies (86.1%), skilled labor shortages (74.4%), and insufficient information (72.1%) as major barriers to lameness management. Multivariable analysis showed that recognizing lameness as a key issue and allocating more resources for its management were associated with higher reported and observed lameness prevalence both by researcher and farmer. However, no variables accounted for the discrepancy between the prevalence estimates of researchers and farmers. We conclude that the dairy farmers we surveyed underestimated lameness prevalence in their herds; this result is consistent with findings from other regions. However, the correlation between lameness prevalence estimates of the researcher and farmers suggests that farmer estimates are at least based upon the true prevalence. We were unable to find reasons for the discrepancy between prevalence estimates of the researcher and farmers, but this could be due to both the lack of farmer training in lameness assessment, and that research estimates were based upon a single point in time assessment. We also conclude that many farms used what are

likely to be ineffective treatments. This finding, combined with the barriers we identified, highlight critical areas for intervention to reduce lameness on dairy farms.

The quality of life of animals used in veterinary education: Perspectives of 4th year veterinary students

Sherry Khoddami¹, Caroline Ritter¹, Beth Ventura², Kathryn Proudfoot¹

Veterinary students often practice their clinical skills on institution-owned animals as part of the curriculum, which may subject the animals to some level of pain or distress. However, there is limited insight into students' perspectives on the quality of life (QoL) of animals used in their education ("teaching animals"). Our study aimed to describe: 1) veterinary students' definition of a good QoL for all animals, and 2) their perception of the QoL of teaching animals. We invited 4th year veterinary students participating in clinical rotations at two Canadian veterinary colleges (including internal and external rotations) to engage in focus groups or interviews using a semi-structured interview guide. We conducted 10 focus groups and 2 interviews with students from 9 veterinary schools. Data were coded inductively and analyzed using template analysis. Definition of a good quality of life for animals varied, but generally included meeting the animals' basic needs (e.g., adequate food and shelter), freedom from pain and disease (e.g., access to veterinary care), mental stimulation (e.g., enrichment, social interactions), agency (e.g., control over their situations), and the ability to express natural behaviours. For teaching animals specifically, the importance of recognizing an animal's tolerance and suitability for handling was highlighted. Some students viewed teaching animals as having a good QoL despite some harm that is inevitable in an educational setting. However, others identified areas of concern for teaching animals, such as a need for further enrichment and mental stimulation to prevent boredom and behavioural problems. Teaching animals kept in more natural environments were perceived to have a better QoL than those in more restricted housing (e.g., access to pasture or loose housing for horses and cows versus restricted housing). However, practical limitations of a teaching environment were discussed as potential obstacles to incorporating these environments. Students considered poor QoL to be linked to health issues (e.g., laminitis, obesity) and behavioural problems (e.g., stereotypies, animal or human-directed aggression) in teaching animals. Some educational procedures were perceived to negatively impact animal QoL, with students discussing

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variations in animals' tolerance to these procedures. Students positively viewed QoL when animals were removed from teaching situations that caused them distress. These results suggest that veterinary students have complex views on the quality of life of animals, including those used within their veterinary curriculum. Understanding student perceptions of animal welfare could help veterinary colleges improve the care they provide to animals used in education.

Investigating dynamics of behavioral reactivity in dairy cattle using Chute and Exit Score Tests

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Current methods to evaluate cattle temperament in production settings include numerous subjective measurements which don't require extra equipment, data being thus easy and inexpensive to collect. The aim of this study was to determine whether cows' temperament remains stable over time, testing the reliability of the subjective measurements and investigating habituation to handling in adult cattle. Our hypothesis was that temperamental cattle acclimate more substantially to repeated handling. The study was conducted at the Research&Development Institute for Bovine Balotesti Romania, on 64 multiparous Romanian Black Spotted dairy cows, housed in a tie-stall barn, evaluated for their behavioral reactivity in the weighting platform. Temperament tests (chute score & exit score) were undertaken by a single observer over two distinct years (Sept. 2021 & Sept. 2022). The chute score (CS) was assessed using a 4-point subjective scale (1-calm to 4-extremely reactive), in increments of 1, with each cow being restrained for one minute in a Patura® A5000 chute (2.20/0.82 m). The exit score (ES/gait) was evaluated subsequent to the CS, analyzing each animal's manner of exiting. The scoring system ranged from 1 (walk) to 4 (jump), reflecting various levels of excitement during the exit phase. Cows were classified as either 'calm' (scores 1&2) or 'nervous' (scores 3&4), while analyzing the results of the CS and ES tests separately. The intraclass correlation for the CS response, was found to be poor (ICC2 = 0.28, p < 0.0082) and poor to moderate for ES response (ICC2= 0.35, p<0.00019). The scoring dynamic for 'calm' animals showed an increase of 0.37±0.18 points for CS response and an extremely small estimate (1.598e-16) for ES, indicating limited to no practical change in scores over time. For 'nervous animals', the scoring dynamic showed a decrease in scores of -1.19±0.19 points for CS and -1.30±0.18 points for ES response. The evidences from this preliminary pilot study show that 'nervous' animals exhibit a more pronounced decrease in scores over time, suggesting that temperamental cattle and those handled more frequently habituate the most. More research is needed in order to validate chute and exit scores as indicators of dairy cattle temperament.

While conventionally accepted, these tests may not solely reflect innate traits, being caused by contextual responses as well. This broader perspective encourages future research to develop more nuanced temperament evaluation tools, incorporating objective measures such as cortisol levels, to foster comprehensive insights into dairy cattle management practices.