



International  
Society for  
Applied  
Ethology

# ISAE 2020 Global Virtual Meeting Online Abstract Book

ISAE is going virtual!

6-7 Aug 2020

Due to the outbreak of COVID-19, the ISAE congress in Bangalore India was postponed to August 2021. In the meantime, we would still like to provide an opportunity, especially for students and early career scientists, to present their work. We therefore hold a virtual meeting on 6-7 August 2020. This meeting features different ISAE regions from six continents to showcase the diversity within our society, running as a 2x12h online webinar marathon!

As a scientific society, ISAE recognises the importance of supporting and promoting diversity in its membership and the wider community, in gender, age, ethnicity, geographical representation and other factors. Therefore, we always welcome and encourage people from all backgrounds to submit an abstract for a talk and participate in our virtual meeting. The meeting is free of charge and open to members (with early-bird registration) and non-members alike.

This meeting will not only facilitate networking of applied animal behaviour and welfare scientists around the globe, it will also be the perfect chance to learn more about the latest research happening in each ISAE region, so do join us in this first time ISAE global virtual meeting!

## Important Dates:

22 June: Early bird member registration open / Abstract submission open

29 June: Public registration open

10 July: Abstract submission close

21 July: Speakers notified

28 July: Registration close

30-31 July: Zoom links to each session sent to registrants

31 July: Deadline uploading talks

6-7 Aug: Meeting time! (12 h Thursday & AGM I & ECR workshop + 12 h Friday & AGM II)

## ISAE 2020 Global Virtual Meeting Programme

All times are shown in UTC (Coordinated Universal Time), please convert to your local time zone.

Day 1: 6 Aug 2020

Time (UTC)	Title of talk	Presenter
Australia/New Zealand – moderators: Kris Descovich & Amanda Doughty		
03:00 – 03:05	Welcome video by ISAE president Susanne Waiblinger	
03:05 – 03:25	Curiosity, fearfulness and use of aviary space in commercial free-range hens	Manisha Kolakshyapati
03:25 – 03:45	Paddock behaviour, production and personality of pasture-based dairy cattle	Heather Neave
03:45 – 04:05	Light preference testing for ultraviolet spectrum and intensity in laying hens	Md Sohel Rana
04:05 – 04:25	Correlations between dog behaviour assessments and the pulling behaviours of shelter dogs during on leash walking	Hao-Yu Shih
04:25 – 04:45	Characterising piling behaviour in Australian free-range commercial laying hens	Maxine Rice
04:45 – 05:00	Closing remarks/break & session change	
South/East/Southeast Asia (1) – moderators: Jashim Uddin & Laura Whalin		
05:00 – 05:05	Welcome video by ISAE president Susanne Waiblinger	
05:05 – 05:25	Assessment of welfare of cattle in Gaushalas (stray cattle shelters) in India	Madan Lal Kamboj
05:25 – 05:45	Automated behavioural quantification of beef cattle exposed to heat load conditions	Musadiq Idris
05:45 – 06:05	Effect of frequency of feeding colostrum and milk on behaviour, growth and health of crossbred dairy calves	Mamta Ssahu
06:05 – 06:25	Prevalence of Cattle Lameness and Associated Risk Factors in Small Dairy Farms of Bangladesh	Solama Akter Shanta
06:25 – 06:35	Break	
06:35 – 07:05	Invited talk: Scenario of food animal and poultry slaughtering across Bangladesh	Prof. Dr. Md. Jalal Uddin Sarder
07:05 – 07:25	Relationship between exploring behavior of captive giraffe and number of visitors	Maho Yamanaka
07:25 – 07:45	CalliFACS: A new muscle-based coding system to measure Common marmosets facial movements	Catia Correia-Caeiro
07:45 – 08:00	Closing remarks/break & session change	
Africa/Central Asia/Middle East (1) – moderators: Mabel Aworh-Ajumobi & Tozie Zokufa		
08:00 – 08:05	Welcome video by ISAE president Susanne Waiblinger	
08:05 – 08:08	Introducing plenary speakers	
08:08 – 08:33	Invited talk: Participatory animal welfare within the Southern African context	Dr Quixi Sonntag
08:33 – 08:43	Questions/Answers	
08:45 – 09:10	Invited talk: Starch level and free-choice provision of straw in the diet of fattening lambs: Effects on performance meal pattern, feed sorting, and chewing behavior	Dr Mohsen Sari
09:10 – 09:20	Questions/Answers	

09:22 – 09:37	Controlling dog overpopulation in Israel by a unique national DPM (Dog Population Management) program	Liat Morgan
09:40 – 09:55	Behavioural differences of two breeds of domesticated chicks to feed and alarm call playback	Iyasere Oluwaseun
09:55 – 10:00	Closing remarks/break & session change	
South/East/Southeast Asia (2) – moderators: Vijay Pal Singh & Jashim Uddin		
10:00 – 10:05	Welcome video by ISAE president Susanne Waiblinger	
10:05 – 10:35	Invited talk: A dog's life in the urban jungle	Dr Anindita Bhadra
10:35 – 10:55	Behavioral and physiological changes in the formation of all-female groups of pygmy lorises ( <i>Nycticebus pygmaeus</i> )	Josue Alejandro
10:55 – 11:15	Effect of bull biostimulation on growth performance and ingestive behaviour of Murrah buffalo heifers	Sunil Dutt
11:15 – 11:35	'Unseen' adoptions: UK owner satisfaction and experiences of rescue and adoption processes of imported Romanian dogs	Parizad Baria
11:35 – 11:55	Effect of mother contact and voluntary colostrum suckling on behaviour and health of Murrah buffaloes and their calves	Sanjay Choudhary
11:55 – 12:00	Closing remarks/break & session change	
Europe (1) – moderators: Ellen Williams & Emma Fàbrega		
12:00 – 12:05	Welcome video by ISAE president Susanne Waiblinger	
12:05 – 12:25	Preliminary results of part-time group housing for does: effect of kit age on skin lesions	Liesbeth Van Damme
12:25 – 12:45	Evidence for sex differences in behavioural and neural correlates to tickling in young Wistar rats	Emma Tivey
12:45 – 12:55	Break	
12:55 – 13:15	Behavioural assessment of capuchin monkeys ( <i>Sapajus</i> spp.) in a rehabilitation programme	Guillermina Hernandez Cruz
13:15 – 13:45	Speed talks (5 mins per talk)	
13:15 – 13:20	"Inactive not alert" as a new indicator of enduring negative affective state in laboratory macaques	Janire Castellano Bueno
13:20 – 13:25	Agreeable horses experience elevated hair cortisol concentration	Aurelie Jolivald
13:25 – 13:30	Sensor laterality and attentional state in horses coping with an unexpected stimulus	Chiara Scopa
13:30 – 13:35	Development of the joint-log-lift task in pigs: hints on cooperation?	Jean-Loup Rault
13:35 – 13:40	Influencing elimination location in the domestic cat: a semiochemical approach	Naïma Kasbaoui
13:40 – 13:45	Effects of the COVID-19 pandemic on animal behaviour and welfare researchers	Irene Camerlink
13:45 – 13:55	Speed talks Q&A	
13:55 – 14:00	Closing remarks/break & session change	
14:00 – 15:00	ISAE annual general meeting (AGM 1)	
15:00 – 16:30	Early Career Researcher workshop Please sign up here <a href="https://tinyurl.com/ISAEvirtual-ECR">https://tinyurl.com/ISAEvirtual-ECR</a>	

Day 2: 7 Aug 2020

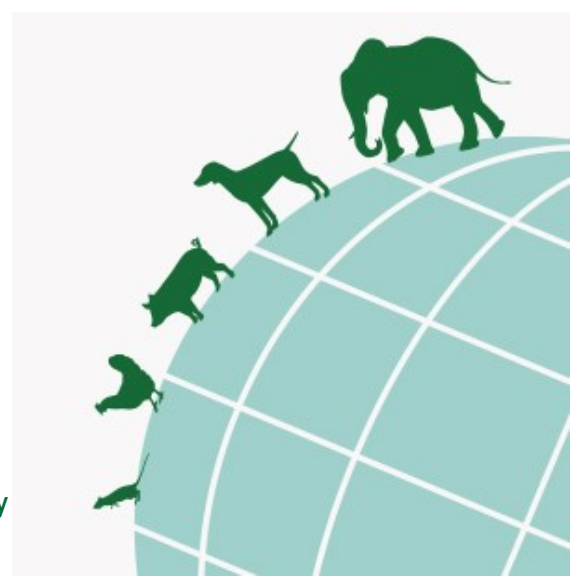
Time (UTC)	Title of talk	Presenter
08:00 – 09:00	ISAE annual general meeting (AGM 2)	
Africa/Central Asia/Middle East (2) – moderators: Mabel Aworh-Ajumobi & Seun Iyasere		
11:00 – 11:05	Introducing plenary speakers	
11:05 – 11:30	Invited talk: Animal Welfare Infractions of Working Donkeys in Kasena Nankana West and East Districts, in the Upper East Region, Ghana. A case study.	Dr Anthony Nsoh
11:30 – 11:40	Questions/Answers	
11:40 – 12:05	Invited talk: Challenges of implementation of Animal Welfare standards in the Middle East	Dr Rachel Dodeen
12:05 – 12:15	Questions/Answers	
12:15 – 12:30	Welfare and ethical issues on offloading of cattle at Akinyele market, Ibadan	Valentine Obiasogu
12:30 – 12:45	Comparison between working and unworking donkeys welfare issues in Nyala City: South Darfur, Sudan	Saber Yagoub Adam Abakar
12:45 – 13:00	Sleeping behaviour of Nigerian indigenous chickens	Ajayi Oreoluwa Doyinsola
13:00 – 13:15	Behavior of white Fulani calves as affected by plant species, age and spacing	Enwete Foluke Eunice
13:15 – 13:30	Improving pig welfare, production and farms' economic, by avoiding husbandry mutilations in piglets and providing environmental enrichment	Liat Morgan
13:30 – 13:45	Tigernut milk ( <i>Cyperus esculentus</i> ) enhances the welfare of broiler chickens	Olaleye Moshood Olayinka
13:45 – 14:00	Closing remarks/break & session change	
Europe (2) – moderator: Ellen Williams		
14:00 – 14:05	Welcome video by ISAE president Susanne Waiblinger	
14:05 – 14:25	Impact of epilepsy on judgement bias in a population of companion dogs	Rowena MA Packer
14:25 – 14:45	Exposure to controlled challenges increases stress resilience in dog puppies	Stefanie Riemer
14:45 – 14:50	Short break	
14:50 – 15:15	Speed talks (5 mins per talk)	
14:50 – 14:55	Slow and steady wins the race: a comparison of standard commercial broilers against a slower growing alternative	Laura Dixon
14:55 – 15:00	Effects of temporary-crating systems on the welfare of lactating sows	Heng-Lun Ko
15:00 – 15:05	Detection of dairy cows' hind-legs activity during milking using a 3D-accelerometer attached to the milking cluster	Camille Raoult
15:05 – 15:10	Social behaviour in pigs: are littermates nicer to each other than to strangers?	Christina Proßegger
15:10 – 15:15	Stockperson attitudes and on-farm handling of Swedish finishing pigs, and influence on animal welfare	Sofia Wilhelmsson
15:15 – 15:25	Speed talks Q&A	
15:25 – 15:35	Break	

15:35 – 15:55	Electronic sow feeder (ESF) use patterns are associated with locomotory ability and stereotypic behaviour in pregnant gilts	Martyna Lagoda
15:55 – 16:15	Cognition in the barn: range use and its relation to cognitive performance in free-ranging broiler chickens ( <i>Gallus gallus domesticus</i> )	Vitor Hugo Bessa Ferreira
16:15 – 16:35	Maternal behaviour of buffaloes and effect on calves' vitality	Lydia Lanzoni
16:35 – 16:55	Dairy cow motivation for access to open lying space	Laura Shewbridge Carter
16:55 – 17:00	Closing remarks/break & session change	
Latin America (1) – moderator: Maria José Hötzel		
17:00 – 17:10	Welcome video by ISAE president Susanne Waiblinger	
17:10 – 17:50	Invited talk: Progesterone, physiological reproductive status, stress response and reactivity in female ruminants	Dr Aline Freitas de Melo
17:50 – 18:15	Lambing season affects ewe milk production, lamb growth and its behavioural response to weaning	Ophélie Menant
18:15 – 18:40	Behavior by individual sows at the entrance of an electronic sow feeder (ESF) is impacted by feed order	André Alves de Albuquerque
18:40 – 19:05	Effects of early familiarization of cattle with feedlot diet and social group on feed bunk attendance and postural behavior during the finishing period in feedlot	Lucas Roberto Batista Ruiz
19:05 – 19:30	Does ponceau red artificial dye influence dog behavior?	Suellen Scheibel
19:30 – 19:55	Personality in Brazilian show jumping horses ( <i>Equus caballus</i> ): a methodological comparison study	Anna Carolina Nogueira Borzani
19:55 – 20:00	Closing remarks/break & session change	
North America – moderators: Rachel Park & Jen-Yun Chou		
20:00 – 20:05	Welcome video by ISAE president Susanne Waiblinger	
20:05 – 20:25	Evaluating self-isolation in group-housed dairy calves following disbudding through provision of a shelter	Katie Gingerich
20:25 – 20:45	The impact of providing periodic exercise on behaviour of stall-housed gestating sows	Mariia Tokareva
20:45 – 21:05	Assessing owner's ability to detect fear and aggression-related behaviours in the domestic cat	Kristina O'Hanley
21:05 – 21:15	Break	
21:15 – 21:35	Effects of milk- and starter-feeding methods on the performance and behavior of individually and pair-housed calves in outdoor hutches	Rekia Salter
21:35 – 21:55	Larger Visitor Numbers Do Not Correlate with Observed Aggression in Three Species of Primates at a Zoological Facility	Jocelyn Woods
21:55 – 22:00	Closing remarks/break & session change	

Latin America (2) – moderators: Alejandra Feld & Maria José Hötzel

22:00 – 22:40	Invited talk: Human-animal interactions and their effects on the welfare of farm animals	Dr María Camila Ceballos
22:40 – 23:05	Freedom to move versus piglet crushing: citizens' attitudes towards farrowing housing systems.	Bianca Vandresen
23:05 – 23:30	Application of different loading densities during transport to the slaughter plant and their effects on pigs' behavioral response	Valentina Montoya Urrea
23:30 – 23:55	The importance of behavioural observations in assessing the welfare of animals under human care: an example with elephants.	Débora Silvia Racciatti
23:55 – 00:00	Closing of the meeting by ISAE president Susanne Waiblinger	

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### Additional information:

- The meeting format will be pre-recorded talks with live Q&A on Zoom.
- English is encouraged as the official language for presentation, but in order to facilitate regional exchange and promote regional students/scientists, there is an option to give talks in the local language with slides in English.
- For presenters: Instructions have been sent to the presenters on how to pre-record voice-over presentations, upload the talks and participate in the Q&A during your session.
- For participants: Once registered, further details on how to connect to each Zoom session will be sent to your email closer to the meeting time. Do [check our website](#) and your email for the most updated information. Each session will have a unique Zoom link, so please check your inbox for emails from ISAE Virtual <no-reply@zoom.us> which contain the Zoom link to connect to each of the session you signed up, how to join and how to import into your calendar.
- All event emails will be from [isae.virtual@gmail.com](mailto:isae.virtual@gmail.com) so be sure to check they do not end up in your spam folder.

Please email [isae.virtual@gmail.com](mailto:isae.virtual@gmail.com) for any inquiries about this event.

We look forward to welcoming all of you!

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## Acknowledgements

### Organising committee

Jen-Yun Chou (lead), Christian Nawroth (concept), Emma Baxter, Irene Camerlink, Cathy Dwyer, Marisa Erasmus, Christy Goldhawk, Giuliana Miguel, Bas Rodenburg, Mark Rutter, Rebecca Sommerville, Tamara Tadich, Heleen van de Weerd, Jennifer Van Os, Susanne Waiblinger, Alexandra Whittaker

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Jashim Uddin (South/East/Southeast Asia)

Mabel Aworh-Ajumobi (Africa/Central Asia/Middle East)

Ellen Williams (Europe)

Rachel Park (North America)

Maria José Hötzel (Latin America)

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Tozie Zokufa

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# Abstracts

\*Due to the short time in organising this virtual meeting, all abstracts published hereafter are in their original states upon submission without further revisions.

## Curiosity, fearfulness and use of aviary space in commercial free-range hens

Manisha Kolakshyapati<sup>1</sup>, Peta Simone Taylor<sup>1</sup>, Adam Hamlin<sup>2</sup>, Terence Zimazile Sibanda<sup>1</sup> and Isabelle Ruhnke<sup>1</sup>

<sup>1</sup>*School of Environmental and Rural Science, Faculty of Science, Agriculture, Business and Law, University of New England, Armidale, 2351, NSW, Australia;* <sup>2</sup>*Faculty of Science, Agriculture, Business and Law, School of Science and Technology, University of New England, Armidale, 2351, NSW, Australia; mkolaks2@une.edu.au*

Fearfulness and exploration have been associated with the time that free-range hens spend on the range. Further understanding of these traits and the use of distinct areas within a shed may help to improve facility design and management techniques to benefit poultry welfare. The aim of this study was to assess the relationship of fear, exploration and use of key specific resources by individual laying hens on a commercial free-range housing system. The upper and lower chain feeders and the nest box areas in a 3-tier aviary system were monitored in 3 Lohmann Brown flocks from 18-72 weeks of age using a custom-built Radio Frequency Identification system. At 72 weeks of age, 769 hens were selected and subjected to a novel arena test (NA) and novel object test (NO). For NA test, hens were placed into the centre of a square arena (1.7m<sup>2</sup>) and left for 8 min. Immediately after, a NO was introduced through a small door and left with the hen for 5 min. Hen behaviour was continuously recorded using an overhead video camera and later analysed by ANYmaze tracking software and one observer blinded to treatment. Latency to step, number of lines crossed and escape attempts were assessed as indicators of fear during the NA test. Time spent within three zones (avoidance, approach and interaction) and the number of escape attempts were assessed as indicators of neophobia and exploration during the NO test. Statistical analysis was carried out using a machine learning approach (random forest). The model used 13 features to generate the most important variables for predicting the time spent at each location followed by GLIMM analysis to determine the significant association between factors. Time spent on the upper feeder was associated with less time spent in the NO interaction zone ( $F_{(1,758)}=6.26$ ,  $P=0.013$ ) and less lines crossed in NA test ( $F_{(1,758)}=12.1$ ,  $P=0.001$ ). Time spent at the lower feeder was associated with more time spent in the NO interaction zone ( $F_{(1,750)}=8.71$ ,  $P=0.003$ ), more lines crossed ( $F_{(1,750)}=7.35$ ,  $P=0.007$ ) and vocalisation ( $F_{(1,750)}=14.3$ ,  $P=0.001$ ) during NA test. These results indicate that hens that spent more time on the upper feeder were not curious while the lower feeder tier were more curious as indicated by the approach to novel stimuli. Hens may occupy key specific resources in the shed depending on their temperament. Further studies are warranted to determine if those areas promote exploration and positive welfare state or serve as a refuge.

## **Paddock behaviour, production and personality of pasture-based dairy cattle**

*Heather Neave<sup>1</sup>, Helen Thoday<sup>2</sup>, Katie Saunders<sup>2</sup>, Paul Edwards<sup>2</sup>, and Jim Webster<sup>1</sup>*

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In pasture-based systems, cows dedicate their time to resting, grazing and ruminating, but time for each behaviour may be affected by factors including walking distance to milking, time off-paddock (walking and waiting time for milking), and a cow's individual personality. This study investigated how each of these factors affected daily duration of grazing, ruminating and lying behaviours in the paddock. Cross-bred dairy cows (n=87) were managed in 3 rotationally-grazed herds (n=29 each) on the same farm and milked twice daily. A triaxial ear-tag accelerometer on each cow recorded daily duration of grazing and ruminating, and a leg-based accelerometer recorded daily lying time, for 17 days. GPS units on 6 cows per herd recorded daily time off-paddock (total time walking to and waiting for milking) and walking distance. Personality traits of each cow were assessed on an ordinal scale for investigative and reactive behaviour toward a novel object and novel human after exiting the milking parlour, reactive behaviour during restraint in a crush, flight distance in the paddock, and step-kick behaviour during milking. A mixed repeated measures model tested how time off-paddock, walking distance and personality affected daily paddock behaviours; age, breed, milk yield and max ambient temperature were covariates. Cows that were more afraid of humans (reactive to novel human; high avoidance distance) had lower lying time ( $P=0.02$ ), and tended to have greater grazing time ( $P=0.08$ ) and lower ruminating time ( $P=0.09$ ). Cows that were more calm and exploratory (calm in crush; investigated novelty) had greater grazing time ( $P=0.05$ ) and milk yield ( $P<0.01$ ). Cows that were more reactive to milking (during cup attachment) tended to have reduced milk yield ( $P=0.09$ ). When walking distance increased, cows spent more time grazing ( $P<0.01$ ) and less time ruminating ( $P=0.05$ ), and when time off-paddock increased, cows spent less time lying ( $P=0.02$ ). These results indicate that paddock behaviours are affected by off-paddock and personality factors; these automated behavioural and short personality assessment tools may identify individual animals or farms that may be at-risk for reduced animal welfare, especially during changing conditions.

## Preference testing for ultraviolet light spectrum and intensity in laying hens

Md Sohel Rana<sup>1,2,3\*</sup>, Andrew M. Cohen-Barnhouse<sup>2</sup>, Caroline Lee<sup>2</sup>, and Dana L.M. Campbell<sup>2</sup>

<sup>1</sup>School of Environmental and Rural Science, University of New England, Armidale, NSW, 2351, Australia;

<sup>2</sup>Agriculture and Food, Commonwealth Scientific and Industrial Research Organisation (CSIRO), Armidale, NSW, 2350, Australia ; <sup>3</sup>Department of Livestock Services, Ministry of Fisheries and Livestock, Dhaka, 1215, Bangladesh;

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Sunlight intensity and ultraviolet (UV) radiation may affect free-range hens' use of the outside range, particularly during summer months in Australia when sunlight is intense with a high UV index. However, it is uncertain what aspect of sunlight (brightness or UV) may be most aversive to the hens to discourage them from leaving standard indoor lighting conditions to venture outdoors. A controlled indoor-based choice study was conducted to determine whether hens show preferences for different light wavelengths and intensities that may affect outdoor range usage. A total of 84 cage-reared ISA-Brown laying hens at 44 weeks of age in 3 groups (28 hens/group) were tested for preferences of indoor standard LED-white light (control) versus one of three different spectral lights: (i) visible spectrum plus infrared wavelengths (VIS); (ii) visible spectrum plus UVA wavelengths (UVA); and (iii) visible spectrum plus UVA and UVB wavelengths (UVA/B) presented successively at low, medium, or high levels of intensity. Hens within each group were individually tested for 2 h in an apparatus with two compartments (control vs treatment) connected by a tunnel on both sides. Videos of hens' time spent in each compartment and behaviours were decoded and analysed using GLMMs. Results showed that hens had a significant preference towards UV spectrums over the control light. Hens spent more time under the low intensity of UVA/B light (62.48%) followed by low intensity of VIS light (60.69%), and medium intensities of both UVA/B light (60.64%), and UVA light (59.02%) compared to control light ( $P < 0.05$ ). Hens showed a significant preference to spend time directly under the light source at the low intensity of the UVA/B light (treatment:  $33.5 \pm 3.99$  min, control:  $17.0 \pm 3.93$  min) and medium intensity of the UVA light (treatment:  $37.1 \pm 5.49$  min, control:  $19.6 \pm 5.49$  min). Hens showed more foraging, ground pecking, and preening behaviours under low and medium intensities of UVA/B light than control light ( $P < 0.05$ ). The study suggests that UVA/B light (sunlight) may have positive effects for hen range use, but during peak sun intensities hens may need additional measures (e.g. shelter) to protect themselves. Confirmation of these findings in a free-range setting is warranted.

## Correlations between dog behaviour assessments and the pulling behaviours of shelter dogs during on-leash walking

Hao-Yu Shih <sup>1\*</sup>, Fillipe Georgiou <sup>2</sup>, Mandy B.A. Paterson <sup>1,3</sup> and Clive J.C. Phillips <sup>1</sup>

<sup>1</sup>. University of Queensland, Centre for Animal Welfare and Ethics, Gatton, QLD 4343, Australia; <sup>2</sup>. University of Newcastle, School of Mathematical and Physical Sciences, Callaghan, NSW 2308, Australia; <sup>3</sup>. Royal Society for the Prevention of Cruelty to Animals, Brisbane, QLD 4076, Australia; haoyu.shih@uq.net.au

Dogs sometimes lunge forwards during on-leash walks which may damage the soft tissue of their neck and their trachea. There is also evidence that the pressure exerted during the lunge can cause increased intraocular pressure. In shelter dogs, lunging behaviour, defined here as maximal tension on the leash, is suspected as being correlated with high arousal (excitement) and even aggression, concerns for new owners after adoption. However, behaviour assessments conducted in shelters often fail to identify this lunging behaviour. Therefore, it is important to develop an objective and accurate method of measuring dog lunging. In this study, a leash tension meter was invented (the first of its kind) which measures the leash tension while dogs are being walked on-leash. Three-hundred-and-seventy walks, involving 111 dogs and 74 volunteers were measured and analyzed. All walks were in the same designated area away from possible distractions. All dogs wore a neck collar and a front-clip harness. The leash tension meter was attached to a 1.4 m long lead and was connected to both the collar and the harness while volunteers held the other end of the meter. The leash tension and pulling events, defined here as tension greater than 0.1% of the dog's bodyweight, were correlated with the behavior assessment conducted in the shelter 3 to 5 days post-entrance. The behaviour assessment consisted of seven relevant tests: socialization to humans, toleration to humans' physical contacts, engagement in playing with toys by themselves or with humans, responsiveness and gentleness when humans are running and stopping suddenly, friendliness toward a model toddler, tendency of resource guarding and potential of separation anxiety. Generalised linear mixed models revealed that dogs exhibiting more reactions to time spent alone created higher mean ( $p=0.04$ ) leash tension and pulling frequency ( $p=0.04$ ). Dogs that were more engaged in playing with toys by themselves or with humans had less pulling frequency ( $p=0.04$ ). However, dogs that were more friendly to a model toddler had more pulling frequency ( $p=0.02$ ). In conclusion, there were correlations between behaviour assessment results and the leash tension during a walk which may provide additional information that is useful in predicting lunging behaviour in dogs.

## Characterising piling behaviour in Australian free-range commercial laying hens

Maxine Rice<sup>1</sup>, Ritu Acharya<sup>1</sup>, Andrew Fisher<sup>1</sup>, Peta Taylor<sup>2</sup> and Paul Hemsworth<sup>1</sup>

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In Australia, 47% of all egg sales are from free-range hens. Anecdotal industry reports indicate that smothering can account for up to 40% of mortality in Australian free-range laying hens. Smothering occurs when birds mass together, often on top of each other, resulting in death from suffocation, but not all piling results in smothering. The present preliminary observations, which are part of observational and epidemiological studies on hen smothering in Australian free-range farms, aimed to characterise the piling behaviour of a flock of 22,514 Hyline-Brown laying hens housed in a free-range aviary system over a randomly selected 9-h observation period. Video observations were conducted within a 5 x 5 m<sup>2</sup> field of view (FOV) located inside the shed on litter. A pile was defined as a minimum of 10 hens pressed together for at least 1 min and not performing any other discernible behaviour. Twelve piling events were observed, 6 which began outside the FOV but spread into the FOV. The average duration of the piling events was 27 min and 19 sec, with a minimum and maximum duration 3 min 26 sec, and 1 h 31 min. At their peak, the piling events occupied an average space of 6.54 m<sup>2</sup> with the largest pile spanning 15 m<sup>2</sup> and the smallest pile spanning only 2 m<sup>2</sup>. Of the 12 piling events, based on vision and sound, 6 had no obvious cause, 3 involved a stockperson moving through the shed, and 3 appeared to be due to birds moving towards shards of light on the litter. Only 3 of the piling events were dispersed gradually with no obvious reason: intervention by the stockperson was responsible for breaking up 6 of the piling events, with the remaining 3 dispersed when the pop-holes opened, automatic feeding lines started or hens appeared to be startled by an unknown event. These limited data suggest considerable variation in piling events within this commercial flock in terms of their duration, size and factors associated with their establishment and dispersal. Observations on four flocks throughout a production cycle are presently underway. Understanding the behavioural patterns and human, climatic and physical events preceding piling and smothering events is important in understanding and reducing smothering in commercial free-range laying hens.

## Assessment of welfare of cattle in Gaushalas (stray cattle shelters) in India

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Gaushalas are animal welfare institutions for providing shelter and succour to homeless, stray, infirm and abandoned cattle. Management and welfare of cattle in Gaushalas is under great focus of animal welfare organizations and Government agencies due to influx of a huge number of cattle in recent years. The aim of study, therefore, was to assess level of welfare and to identify indicators which compromise welfare the most. For this 30 Gaushalas from Haryana state were grouped into 3 herd size categories as small (S) (n=100-500), medium (M) (n=501-1000) and large (L) (n>1000). Welfare was assessed on 20 input and output based indicators using Integrated Diagnostic System Welfare (Calamari and Bertoni, 2009) protocol as modified by Kamboj and Kumar (2014). Data were collected on housing, feeding, breeding and healthcare practices by on-site measurements and by using a pre-testing structured schedule. Significance of differences among means of study parameters was tested using ANOVA in SPSS Version 22 software. Floor space (sq.ft) per cow was significantly ( $P<0.05$ ) higher in M ( $139.59\pm 29.67$ ) and L ( $130.36\pm 14.56$ ) as compared to S ( $55.46\pm 6.97$ ). Availability of green and dry fodders in all categories of animals was significantly ( $P<0.05$ ) higher at L as compared to S. Mean availability of green fodders, dry fodders and concentrate mixture in S, M and L was  $11.50\pm 0.65$ ,  $13.88\pm 0.74$  and  $17.11\pm 1.18$ ;  $5.20\pm 0.17$ ,  $4.80\pm 0.12$  and  $4.46\pm 0.11$  and  $0.70\pm 0.12$ ,  $1.29\pm 0.10$  and  $1.65\pm 0.11$  kg/day respectively, being significantly different among 3 categories of Gaushalas. Overall mean welfare score out of 100 in L ( $58.86\pm 5.07$ ) was significantly higher than in S ( $42.59\pm 4.72$ ) and M ( $48.47\pm 5.91$ ). Gaushalas ranked as good, average and poor welfare were 20.0, 53.3 and 26.6 % respectively and only 20% had acceptable welfare (score > 60). PCA revealed 4 components which explained 73.85, 78.72 and 76.77% of total variation in S, M and L respectively. Type of floor, cow comfort index, availability of feeds and fodders, system of housing, floor space, facilities for rescue, transport and treatment, type & height of roof, feeding practices, body condition score, breeding practices and cow cleanliness were the most compromised welfare indicators. We concluded that cattle welfare was unacceptable at all small Gaushalas and most of medium Gaushalas whereas it was acceptable only at about a half of large Gaushalas.



### **Automated behavioural quantification of beef cattle exposed to heat load conditions**

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Behavioural responses of beef cattle to hot environmental conditions were studied to investigate whether video digitised image analysis was associated with their behavioural responses. It was further explored whether a substituted diet (grain substituted with forage) would affect the behavioural responses to heat stress, measured by digitised movements. The 13 cattle were exposed in two cohorts, the first cattle (n = 6) fed a standard finisher diet based on high percentage of cereal grains, and the second cattle (n = 7) receiving a substituted diet in which 8% of the grains were replaced by an isoenergetic amount of lucerne hay. Ethical approval for the study was obtained from The University of Queensland Animal Ethics Committee (SAFS/460/16). The behavioural responses of cattle were recorded using 24-hour camera surveillance of all individually penned cattle 13 beef cattle in a climate-controlled chamber during exposure to a simulated typical heat event in Queensland, Australia. Open-source automated behavioural quantification software was used to record pixel changes of 13 beef cattle from the recorded videos. The data obtained was analysed, using mixed effect model in Minitab 18 (Minitab® 18.1 Inc. USA) for Windows using two different statistical approaches to compare thermoneutral, hot and recovery periods. Stepwise regression analysis in general linear model was used to understand the relationship of digitized movement of standing animals with stepping and grooming/scratching activities. Higher digitised movement (approximately 150,000 pixels) was observed in cattle during the period of hot environmental temperature ( $P \leq 0.001$ ). The cattle on substituted diet displayed less digitised movements (approximately 55, 000 pixels) than cattle receiving standard finisher diet (approximately 170, 000 pixels) on exposure to heat ( $P \leq 0.001$ ), suggesting less discomfort during hot conditions. The digitized movement was higher ( $P \leq 0.001$ ) in the standing animals (approximately 560, 000 pixels) compared to lying animals (approximately 80, 000 pixels), which was related to stepping and grooming/scratching activities in standing animals. The results suggest that cattle exposed to heat, display increased movement that could be detected automatically by video digitisation software, and that a replacement of some cereal grain with forage in the diet of feedlot cattle may reduce the measured activity responses to the heat.

## Effect of frequency of feeding colostrum and milk on behaviour, growth and health of crossbred dairy calves

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Under natural conditions dairy calves suckle their mothers for 7 to 10 times daily and consume substantially more milk. Under most modern husbandry conditions calves are allowed to suckle or artificially fed colostrum/ milk only twice a day. This causes quicker ingestion of colostrum/milk which does not satisfy their innate suckling instinct leading to emergence of abnormal sucking behaviours. This work aimed at studying the effect of feeding colostrum and milk at different intervals and frequencies on the behaviour, health and growth of crossbred dairy calves. For this, 20 calves were selected at birth and randomly allotted to 3 groups (Control=6; T1 and T2=7 each) for a period of 90 days. Calves in C were offered colostrum/milk twice-a-day at unequal intervals of 8 and 16 hours. Calves in T1 were also fed colostrum/milk twice-a-day but at equal intervals of 12 hours each. Calves in T2 were offered colostrum/milk thrice-a-day at equal intervals of 8 hours each for up to one month of age and thereafter twice-a-day at equal intervals of 12 hour each. The treatment means were compared for significance by test statistic one-way ANOVA in SPSS software. Colostrum intake on first day of birth was  $1.60 \pm 0.22$ ,  $1.64 \pm 0.05$  and  $2.16 \pm 0.14$  kg (6.52, 7.09 and 8.53% of calf weights) in C, T1 and T2 respectively; being significantly higher ( $p < 0.05$ ) in T2 than in T1 and C calves. Overall daily colostrum intake was  $2.17 \pm 0.12$ ,  $2.16 \pm 0.07$  and  $2.45 \pm 0.08$  kg in C, T1 and T2 calves respectively; being significantly higher in T2 than in other 2 groups. Time taken for ingestion of colostrum and milk was significantly higher ( $p < 0.01$  and  $< 0.05$ ) for T2 as compared to C and T1. Mean time spent on different abnormal behaviours was similar in all 3 groups. Daily weight gain for C, T1 and T2 was  $429.10 \pm 21.52$ ,  $410.81 \pm 23.15$  and  $509.19 \pm 22.23$  gm respectively which was significantly ( $p < 0.05$ ) higher in T2 group than 2 groups. Total immunoglobulins concentration after 48 hours of birth was  $28.64 \pm 2.15$ ,  $31.66 \pm 1.03$  and  $35.40 \pm 2.24$  mg/ml for C, T1 and T2, respectively which did not differ among 3 groups. Incidence of faecal scours was the higher in C (16.6% of calf days) as compared to T1 (2.85% of calf days) and T2 (2.85% of calf days). It was concluded that feeding of colostrum/ milk at equal intervals and increasing the frequency of feeding from twice to thrice daily from birth to one month of age improved the growth performance and health of crossbred calves.

## **Prevalence of Cattle Lameness and Associated Risk Factors in Small Dairy Farms of Bangladesh**

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Lameness is a severe welfare problem and a production-limiting disease in dairy farming and took the third place in causing economic loss to dairy farmers next to infertility and mastitis. The objectives of this cross-sectional study were to determine the prevalence of lameness, identify the associated risk factors and assess the lameness effect on milk production. A purposive sample of 10 Holstein-Friesian cross bred cows were selected from each of 43 randomly selected dairy farms from Baghabarighat Milk Shed Areas of Sirajgonj district in Bangladesh. Animals were observed during in motion for detection of any kind of abnormalities in locomotion. Milking and pregnancy status, milk yield before and after disease, feeding, body condition score, parity, floor type, frequency of floor cleaning, bedding type, herd size were hypothesized to be risk factors for lameness and statistically tested. Out of 430 dairy cattle examined, 58 (13.49%) showed lameness in different grades, with 78.0% slightly, 17.4% moderately and 4.6% severely lame. Lameness of one or more animal was detected in 32 (74.42%) of the 43 visited farms. Lameness was significantly associated with milking status, parity, injured hocks and dirty legs ( $P>0.05$ ). Lameness was more frequent in hind limbs than in forelimbs. In milking dairy cows, the mean daily milk yield (av. 11.46 L/day/cow) was significantly reduced (av. 9.38 L/day/cow) after the onset of lameness. The study showed that lameness is associated with milk yield and an economically important welfare problem in studied dairy units. Therefore, farmers should give attention to lactating cows for early detection and prevalence of lameness to minimize the economic loss.

DRAFT

## Scenario of food animal and poultry slaughtering across Bangladesh

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Around 3.5 million cattle, 15 million goats and 332.9 million poultry are slaughtered annually in Bangladesh for food. The aim of this study was to investigate the overall slaughtering situation of food animals and poultry at different places in Bangladesh focusing welfare related issues. We investigated the slaughtering of food animals and poultry in the slaughter houses regulated by City Corporation, Department of Livestock Services and private facilities over a period from January to December 2017. Data were collected by physical inspection, taking photographs, recording videos, and interviewing the butcher, veterinarians, and related people. In the capital city of Bangladesh “Dhaka” there are 2 city corporations; South and North. South City Corporation has 5 zones for slaughtering of food animals but no slaughter houses in North. Rajshahi City Corporation has one abattoir for large animals and one for poultry located at SahebBazar. Other cities have few or no slaughter houses. Only 10 to 15% of the animals are being slaughter at the abattoir while the remaining slaughters occur in open places, beside the roads and drains adjacent to the meat shops. Meat inspection by veterinarian are limited in all slaughter houses. Butchers bring their animals early in the morning to these slaughter house and slaughtered without stunning and animals are being slaughtered one by one in front of others. Moreover, existing slaughter houses have no proper facilities of lairage, slaughter hall, skinning tools, chilling, detention and condemnation room, or cleaning of ingesta. Additionally, there are many self-made field abattoirs in rural and urban areas, small towns where animals are slaughtered by unauthorized butchers. There are neither any humane methods of slaughter nor any ante- and post mortem examinations. In recent times, some entrepreneurs have taken initiatives modern private slaughter houses in Bangladesh. Therefore, in general in, slaughtering of food animals and poultry lack standards of welfare and are not maintained by proper sanitary situations. These situations may hamper the meat quality and public health in Bangladesh.



## Relationship between exploring behavior of captive giraffe and number of visitors

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It has been reported relationship between visitor and behavior of exhibited animal in zoo. This study aimed to investigate the relationship between the exploring behavior of captive giraffe and the number of visitors in giraffe exhibition area. Three giraffe (*Giraffa camelopardalis reticulata*, average age 4 years) rearing at Ishikawa Zoo (Ishikawa Prefecture, Japan) were used in this study. We measured the frequency and time of exploring behavior of captive giraffe and the number of visitors on weekdays (Monday to Friday) and holidays (Saturday, Sunday, and public holiday). First, we investigated the difference weekday and holiday by Mann-Whitney U tests. Second, we examined the relationship between exploring behavior of captive giraffe and the number of visitors using Kendall's rank-order correlation ( $\tau$ ). Third, we analyzed the factors directly affect exploring behavior with multiple stepwise regression analysis. Frequency of exploring behavior was significantly higher on holidays than on weekdays ( $P < 0.01$ ). Time of exploring behavior was significantly higher on holidays than on weekdays ( $P < 0.05$ ). There was significant negative correlation between frequency of exploring behavior and the number of visitors per hour on weekdays ( $P < 0.01$ ) and holidays ( $P < 0.05$ ). The result of multiple stepwise regression analysis indicated that frequency of exploring behavior on weekday was directly affected by time of exploring behavior of captive giraffe and number of visitors. Therefore, it is necessary to consider the affecting by the number of visitors to repertoire of captive giraffe behavior.

## CalliFACS: A new muscle-based coding system to measure Common marmosets facial movements

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Facial expressions are subtle signals, central for communication and emotion in mammals. Traditionally, facial expressions are classified as a whole (e.g. happy, bared-teeth), due to the automatic face processing in the human brain, i.e., humans categorize emotion globally, but do not see subtle cues such as a brow raise. Moreover, the same facial configuration (e.g. lip corners pulled backwards exposing teeth) can convey widely different information depending on the species (e.g. human: happiness; chimp: fear). The gold standard FACS (Facial Action Coding System) for investigating human facial behavior, avoids mistakenly assumptions of meaning, by objectively measuring observed movement linked to facial muscles coded as independent movements or Action Units (AUs). FACS was adapted before for seven different species (available at [www.animalFACS.com](http://www.animalFACS.com)), and here we follow similar methodology to create the CalliFACS for the common marmoset. First, we determined the facial muscular plan of marmosets by examining dissections from the literature and diffusion MRI scans. Second, we recorded 27 individuals in a variety of contexts (e.g. grooming, feeding, play, human interaction, veterinary procedures), at the Primate Research Institute, Kyoto University, and selected some clips from online public databases (e.g. YouTube). Hence, all video used here was either purely observational (e.g. passive filming in home enclosures) or collected during other research projects with previously scheduled procedures; hence no negative/stressful context was created exclusively for this project. Individual facial movements were classified according to appearance changes and the corresponding muscles. A diverse repertoire of AUs was identified by analyzing 3.5 hrs of spontaneous marmoset facial behavior on video (frame-by-frame): 15 AUs (independent muscle movements), 8 Action Descriptors (by non-mimetic muscles) and 4 Ear Action Descriptors. Although these results show a reduced range of facial movement when compared to humanFACS, marmoset's range of facial movements was larger than predicted before, which indicates its importance for this species social interactions. CalliFACS is a scientific tool to better understand the common marmoset's communication and expression. As the common marmoset has been hailed the new ideal laboratory animal model, from neuroscience to cognition, CalliFACS can be an important tool to evaluate marmoset welfare, particularly in captivity.

## **Participatory Animal Welfare in the Southern African Context**

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Welfare of animals is affected by several factors including animal-based and environmental conditions, and the way humans view, engage with and care for animals. A study focusing on the human element in animal welfare investigated the use of participatory reflection and action (PRA) in a rural community in South Africa and found that perceptions, knowledge and attitudes regarding rabies in a rural community in South Africa could change over a period of time. As the participants' knowledge about rabies control improved, a positive change in general attitudes towards dogs became evident. As a methodology for changing human perceptions and behaviour, PRA has the potential to improve animal welfare by changing views and attitudes, through empowerment and taking ownership of issues. Africa has many animal welfare challenges. Guidelines formulated by the World Organisation for Animal Health (OIE) on animal welfare are either partially implemented or not at all in many parts of the African continent. In order to investigate possible barriers and solutions to implementation of animal welfare standards, a new study aims to investigate how PRA may be utilised at different levels and with a variety of stakeholders. The study will entail a collaboration between approximately 7 countries in the Southern African Development Community (SADC). Through participatory engagement between country representatives it is envisaged that participants will share knowledge, experiences and resources to facilitate a better understanding of animal welfare issues at grassroots levels and develop the skills to improve animal welfare where it matters most. Follow-up engagements will enable participants to compare progress and learn from one another. This project is coordinated through the Candidate OIE Collaborating Centre for Animal Welfare at the Faculty of Veterinary Science, University of Pretoria, South Africa. Possible outcomes may include a participatory model for dealing with other similar topics (e.g. where new legislation needs to be implemented or awareness levels raised), the creation of an Afrocentric open educational resource for animal welfare-related topics, improved harmonisation of animal welfare standards and implementation across a region and a better understanding of the complexity of animal welfare, particularly in the Southern African and continental context. This may potentially expose a new African animal welfare perspective.



**Starch level and free-choice provision of straw in the diet of fattening lambs: Effects on performance meal pattern, feed sorting, and chewing behavior**

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This experiment was conducted to investigate the effect of starch level on growth performance, feed sorting behavior and preference toward straw in lambs fed forage and concentrate *ad libitum*. Twenty-four Arabian-male lambs with average live weight of  $18.8 \pm 2$  kg were used in a completely randomized design with 3 treatments and 8 replicates for a period of 84 days. Experimental treatments were included 3 levels of starch (27.1, 35.4 and 43.6 percent of ration dry matter (DM)) and wheat straw fed to lambs as a free-choice provision. Results showed that final body weight, concentrate consumption and total DM intake were not affected by the treatments. Feed conversion ratio improved linearly as level of starch in the diet increased ( $P=0.03$ ). Average daily gain linearly increased with increasing level of starch in the diet ( $P=0.02$ ). Increasing level of starch in the diet enhanced preference for wheat straw ( $P<0.05$ ). In all diets, lambs sorted for short particles of wheat straw against long particles. Sorting against long particles of wheat straw decreased with increasing level of dietary starch concentration. Time spent eating decreased linearly with increasing level of starch concentration in the diet ( $P=0.01$ ). Highest intake rate (g DM/min) observed in the first 2 hours after feed delivery. Intake rate (g DM/min) tended to decrease with increasing starch level of the diet at 1 hour after feeding ( $P=0.06$ ) but tended to increase 3 hours after feeding ( $P=0.09$ ). Ruminating time per gram of DM ( $P=0.04$ ) and NDF ( $P=0.01$ ) intake increased linearly with increasing starch level of the diets. Standing time increased linearly as starch content of the diets increased ( $P=0.04$ ). The hourly feeding time was consistently greater in lambs fed lower starch diet. The ruminating pattern showed that at 300, 400 and 600 hours, the ruminating activity was greater in lambs on high starch diet. The experiment indicated that in *ad libitum* concentrate feeding it was possible to maintain performance by using a lower starch concentrate. In addition, lambs did sort for the long hay regard to concentrate starch level, which may have been an adaptive reaction to lower ruminal pH caused by *ad libitum* intake of high starch concentrate.



**Controlling dog overpopulation in Israel by a unique national DPM (Dog Population Management) program**

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Abandonment of dogs is a global concern as it results in increasing numbers of free-roaming dogs and unwanted dogs in shelters. This problem is associated with impaired animal welfare as well as serious public health risks. Furthermore, it may be related to disease transmission and attacks of livestock, wild animals and humans. According to the Israeli “Dog Control Law, 2002”, dogs must be annually licensed, microchipped, rabies vaccinated and registered on a national governmental database. In addition, since 2012, a unique online searchable database has been gathering most homeless pets offered for adoption from non-profit organizations and municipal shelters, aiming to increase adoption rates (<http://Yad4.co.il>). In order to build a broad national, scientific-based, DPM program in Israel, a three years study has been conducted (2016-2019). The objectives were (1) to investigate the registered dog population in Israel and to assess its association to the abandoned dog population; (2) to reveal the risk factors for a dog to be adopted or to stay at the shelter and (3) to explore and characterize the diverse attitudes towards dog overpopulation and the possible solutions, among the general public, animal welfare organizations as well as municipal veterinarians. Data analyses included 758,288 registered dogs, 22,545 adoptable dogs, 1,236,532 online searches, 11,914 online adopt requests and feedback forms and more than 10,000 successfully completed questionnaires. Multi-Variate Linear Regression revealed that the number of abandoned dogs significantly increased by the numbers of active registered dogs and the dogs which were suspected to be not under responsible ownership ( $p < 0.05$ ). Among the abandoned dogs (average age when abandoned:  $1.7 \pm 2.12$  years), 94% were adopted, when published on the national adoption website. The risk factors to stay at the shelter were when the dogs were described as: “mix-breed”, “male”, “suitable for senior” or “for athletes” ( $p < 0.05$ ). In conclusion, a governmental national database is an important dog population management tool, with the potential to predict the number of abandoned dogs. In addition, a national online database may successfully improve dog adoption rates; however, dogs’ descriptions online have a significant impact on the chances for adoption. Based on this study, 1.2 million US\$ has been invested annually by the Ministry of Agriculture in encouraging responsible ownership, sterilizations and adoptions.

**Behavioural differences of two breeds of domesticated chicks to feed and alarm call playback**

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In a natural environment, the hen aids chicks in finding feed or avoid predators by vocalizing. In large farms, natural brooding is not commercially viable and so chicks are hatched in large incubators and reared artificially. It is unknown how different chick breeds respond behaviourally to feed and alarm call playback sounds. The experiment was carried out at the Poultry Unit of University Farms (DUFARMS), Federal University of Agriculture, Abeokuta, Ogun State, Nigeria. Thirty-two day old chicks of each breed (Nigerian indigenous chicks and commercial broilers) were randomly assigned to four replicates with each replicate having eight chicks. The playback experiment was conducted in a test pen mainly made of ply wood partitioned into two equal halves. Replicates of each breed were tested simultaneously in the two cells. The behavioural responses of the two breeds of chicks were recorded during the feed and alarm call playback period. At day 1, the eight birds/replicate were acclimatised to the test arena for a 30 min period where they had access to feed served in tray feeders and water in bell drinkers. On the 2<sup>nd</sup>, 4<sup>th</sup> and 6<sup>th</sup> day of age, chicks were exposed to feed call playback (lasted for 1 minute), four times per day within a 40 min period, at 10 min interval (data were collected on the number of chicks pecking, foraging and feeding). On the 3<sup>rd</sup>, 5<sup>th</sup> and 7<sup>th</sup> day of age, chicks experienced the alarm call playback, four times per day within a 40mins period, at 10 min interval (data were collected on the number of birds that crouched and ran). Data obtained were subjected to non-parametric test (Mann Whitney U test) using IBM SPSS 23. There was no significant effect of breed ( $P>0.05$ ) on the percentage of chicks feeding, pecking and foraging. On the other hand, a greater percentage ( $P<0.05$ ) of the broiler chicks crouched in response to the alarm calls compared to the Nigerian indigenous chicks. The percentage of chicks that ran at the sound of the alarm call were similar ( $P>0.05$ ) in both breeds of chicks. This study has shown that both chick breed responded to both the feed and alarm call playback but with some differences in the behaviour displayed.

**A dog's life in the urban jungle**

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A large body of research is focused on understanding the ability of dogs to communicate with humans. However, these studies are based on pet dogs, which are raised and cared for by humans. Free-ranging/stray dogs that are present in many parts of the world, on the other hand, depend on humans as resources and interact with people on a regular basis. Hence, they can provide interesting insights into the nature of dogs and give pointers to how dogs might have evolved from wolf-like ancestors to become man's best friend. We have been engaged in studying the free-ranging dogs in India for 11 years, delving into their ecology, behaviour and cognitive abilities. I will give a brief overview of our understanding of how free-ranging dogs survive in the human-dominated urban environment, co-existing with our species.

DRAFT

**Behavioral and physiological changes in the formation of all-female groups of pygmy lorises (*Nycticebus pygmaeus*)**

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The formation of iso-sexual pairs can be a behavioral management strategy to improve the quality of life of captive primates by meeting their social needs while also being a non-invasive way of controlling the captive population. For lorises, which are targets of various anthropogenic activities that threaten their survival in the wild, iso-sexual groups can also serve as a method to maximize space which increases the number of animals that can be rescued. Recent work on this species has revealed that they may be more social than originally reported and we tried social housing as a method to improve their welfare and captive management. At the Slow Loris Conservation Centre at the Japan Monkey Centre, we documented the formation of all-female groups by looking at behavioral and physiological parameters before, during, and after the group formations. The subjects were 8 female pygmy lorises, in 3 distinct social groups (two pairs and one quadruple group), moved from individual caging to enriched social housing. All-female groups were successful in social housing, with 6/8 females having long term pairing success. For activity budgets, moving time decreased significantly ( $V=0$ ,  $N=6$ ,  $P=0.03$ ) after social housing, and in positional behaviors, sitting increased, while no effects on other behaviors. All individuals preferred to be close and nest together (WRST Close-Far;  $P=0.03$ , Nest-Far;  $P=0.03$ ) rather than staying far from each other or sleeping in other available empty nests. The animals were less stressed after socializing, as there was a significant decrease in fecal glucocorticoids from individual housing to socially enriched housing ( $W=2266$ ,  $P=0.0014$ ). Our results indicate that whenever possible, same-sex female group formations can be a way to decrease stress while maximizing space at a loris rescue center.

DRAFT

## Effect of bull biostimulation on growth performance and ingestive behaviour of Murrah buffalo heifers

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Dairy buffaloes grow slower and attain sexual maturity at later age than cattle leading to greater cost of rearing until calving and shorter productive life. Biostimulation has been reported to improve growth in Sahiwal (*Bos indicus*) heifers, juvenile opossums and mice. The aim of this study was to investigate the effect of bull biostimulation on growth performance of Murrah buffalo heifers through fenceline and direct bull contact. For this 24 pre-pubertal heifers were allotted to 3 groups of 8 each on the basis of age ( $16.09 \pm 0.17$  months) and body weight ( $210.88 \pm 2.68$  kg). In no bull exposure (NBE) group, heifers were not exposed to bull; in fenceline bull exposure (FBE) group, heifers were exposed to an intact bull through a fenceline contact round-the-clock and in DBE (direct bull exposure), heifers were exposed to intact bull through direct contact twice daily for 6 hours (6.00-9.00 am and 4.00-7.00 pm). Heifers were fed similar rations (ICAR, 2013) to achieve daily weight gain of 600 gm. The significance of differences among the mean values of variables was tested using one-way ANOVA in SPSS version 22 software. Mean body weights after 8 months and mean daily weight gain differed significantly ( $p < 0.01$ ) among 3 groups; DBE ( $310.34 \pm 8.02$  kg;  $0.764 \pm 0.02$  gm), FBE ( $289.33 \pm 5.19$  kg;  $0.678 \pm 0.02$  gm) and NBE ( $271.07 \pm 4.49$  kg;  $0.545 \pm 0.01$  gm). Mean daily dry matter intake was significantly ( $p < 0.05$ ) lower in NBE ( $7.99 \pm 0.31$  kg) than in DBE ( $9.48 \pm 0.62$  kg) and FBE ( $9.23 \pm 0.58$  kg). Mean feeding time (minutes/day) in DBE and FBE ( $282.67 \pm 4.46$  and  $273.10 \pm 3.02$ ) was higher ( $P < 0.01$ ) than NBE ( $261.35 \pm 3.95$ ) while feeding frequency was similar ( $19.90 \pm 0.44$ ,  $18.92 \pm 0.63$  and  $19.21 \pm 0.57$  respectively). Mean rumination time (min/day) in NBE ( $344.76 \pm 6.75$ ), was lower ( $P < 0.01$ ) than in FBE ( $361.61 \pm 5.58$ ) and DBE ( $368.79 \pm 6.30$ ). Rumination frequency was higher ( $P < 0.05$ ) in DBE than in FBE ( $20.00 \pm 0.59$  vs.  $18.43 \pm 0.63$ ) but did not differ from NBE ( $19.74 \pm 0.41$ ). Mean water intake time (min/day) in DBE and FBE ( $16.86 \pm 0.90$  and  $16.32 \pm 0.94$ ) was higher ( $P < 0.05$ ) than in NBE ( $14.36 \pm 0.77$ ). Mean daily water intake frequency did not differ among the 3 groups ( $10.81 \pm 0.68$ ,  $12.24 \pm 0.80$  and  $11.65 \pm 0.72$  in NBE, FBE and DBE respectively). We concluded that biostimulation by limited direct contact or by fenceline bull contact greatly improved the growth rate and increased budget time for ingestive behaviours of buffalo heifers

## **'Unseen' adoptions: UK owner satisfaction and experiences of rescue and adoption processes of imported Romanian dogs**

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Adopting a dog 'unseen' from another country suggests a relatively risky human behaviour that may reduce likelihood of successful adoption. This study aims to describe experiences of UK adopters importing Romanian rescue dogs unseen and their satisfaction with the adoption process. In June-July 2019, a questionnaire was distributed via social media to UK owners who had adopted a dog from Romania from January 2014 to March 2019. Results are presented with preliminary univariable analyses undertaken using chi-square and reported with odds ratios. Of 1727 responses, 63.5% (n=1097) dogs were in Romanian centres or foster homes when the prospective UK-based owner decided to adopt, hence were adopted 'unseen'. Despite this, 62.7% respondents were extremely satisfied, 29.3% were satisfied and only 8% were unsatisfied/extremely unsatisfied with the rescue process, with 97.4% satisfied/extremely satisfied with the dog adopted. Pre-adoption home checks were undertaken in person (88.2%) or virtually (5.4%). Common information taken from potential adopters that significantly improved rescue process satisfaction included prior experience with dogs (79.9%, OR (95% CI): 2.77 (1.75-4.37),  $p < 0.001$ ), time the dog would spend alone during the week (77.6%, OR (95% CI): 2.12 (1.34-3.36),  $p = 0.002$ ), details of property type/permissions (73.3%, OR (95% CI): 1.82 (1.16-2.86),  $p = 0.12$ ), details of children at home (70.3%, OR (95% CI): 2.85 (1.83 -4.42),  $p < 0.001$ ). Asking for veterinary surgeon details (49.6%, OR (95% CI): 2.67 (1.65-4.31),  $p < 0.001$ ) and sitter arrangements (32.2%, OR (95% CI): 2.45 (1.39-4.34),  $p = 0.002$ ) also improved process satisfaction. Most respondents were given detailed information about the dog's behaviour around other dogs (75.6%) and familiar adult humans (73.4%), but less frequently about strangers (53.3%), children (51.5%) and cats (51.4%). Fewer respondents were given sufficient and accurate information about the dog's health (43.9%) and behaviour (41.6%) at the time of adoption. Insufficient/inaccurate health (OR (95% CI): 2.23 (1.09-4.55),  $p = 0.028$ ) and behaviour (OR (95% CI): 139.09 (19.3-1002.54),  $p < 0.0001$ ) information significantly reduced process satisfaction. Adopting Romanian rescue dogs 'unseen' is often a positive experience. However, this can be improved by providing detailed, accurate information about dog's behaviour and health and by pre-adoption screening. Inability to account for confounding and collinearity is a limitation of the study.

**Effect of mother contact and voluntary colostrum suckling on behaviour and health of Murrah buffaloes and their calves.**

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Separation of calves & mothers soon after birth & restricted artificial feeding has been implicated for hampered growth & behavioural stress. Aim of this study was to investigate effect of direct mother-calf contact & ad.lib. natural colostrum suckling by calves on behaviour, health & performance of Murrah buffaloes & their calves during golden period (0-5 d of birth). For this 24 mother-calf, pairs were blocked at parturition into 3 groups of 8 each (parity 1-3; lactation yields  $2460.25 \pm 173.76$  kg). In group NC; calves and mothers were separated at birth & offered colostrum twice a day by bottle (1/10th of body weight). In group RC; calves were housed separately, & allowed restricted contact & natural suckling of their mothers twice daily. In group FC; calves & mothers were given round-the-clock full contact by housing them together & allowed to voluntarily suckle their mothers. Behavioural parameters were recorded using 24 hours CCTV camera recording. Mean values were analysed using one-way ANOVA in SPSS. The average birth weight of calves was  $31.47 \pm 1.24$ ,  $29.41 \pm 1.31$  &  $30.25 \pm 1.04$  kg in NC, RC & FC groups. The average colostrum intake was ( $p < 0.05$ ) higher in FC & RC than in NC calves on d1 ( $2.06 \pm 0.15$  &  $2.05 \pm 0.16$  vs.  $1.05 \pm 0.06$ ), d2 ( $2.60 \pm 0.14$  &  $2.56 \pm 0.12$  vs.  $1.90 \pm 0.10$ ), d3 ( $2.96 \pm 0.16$  &  $3.15 \pm 0.19$  vs.  $2.23 \pm 0.06$ ), d4 ( $3.25 \pm 0.10$  &  $3.46 \pm 0.14$  vs.  $2.32 \pm 0.12$ ) & d5 ( $3.70 \pm 0.16$  &  $3.72 \pm 0.18$  vs.  $2.44 \pm 0.11$ ). Average daily gain (gm) differed ( $p < 0.05$ ) among 3 groups (FC  $660.00 \pm 0.02$ ; RC  $450.01 \pm 0.03$ ; NC  $350.10 \pm 0.02$ ). Mean suckling frequency & duration (min/day) in FC group on d1 was  $19.50 \pm 0.62$  &  $49.87 \pm 1.69$  which declined linearly to  $7.87 \pm 0.29$  &  $32.65 \pm 2.41$  on d5. Stress biomarkers viz., heart rate (beats/min, before & after reunion with mothers), neutrophils, lymphocytes count & cortisol level were ( $p < 0.05$ ) lower in FC than in RC & NC calves. Number of diarrheal incidences were 3, 46 & 33 in FC, RC & NC calves. Mean time spent on eating of buffaloes (min/h) was ( $p < 0.05$ ) higher in FC ( $347.25 \pm 9.40$ ) vs., RC ( $325.23 \pm 10.64$ ) & NC ( $319.54 \pm 11.75$ ). Mean time spent on lying was ( $p < 0.05$ ) higher in FC ( $805.91 \pm 9.67$ ) than in RC ( $623.17 \pm 12.96$ ) & NC ( $609.20 \pm 11.26$ ) dams, whereas time spent on standing was ( $p < 0.05$ ) lower in FC ( $629.58 \pm 10.83$ ) as compared to RC ( $814.73 \pm 13.62$ ) & in NC ( $829.23 \pm 12.43$ ) dams. We concluded that full mother-calf contact & voluntary colostrum intake greatly improved growth, health of calves & reduced the behavioural stress in both calves & mothers. The experimental mythology procedure for this research work was approved by the Institutional Animal Ethics Committee of ICAR-NDRI Karnal under the approved ongoing research project No. B-49.



**Preliminary results of part-time group housing for does: effect of kit age on skin lesions.**

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In rabbit farms does are housed individually even though they may benefit from group housing in enriched parks (larger area and social interactions). Research on continuous group housing, however, showed welfare implications mainly caused by aggressive behaviour of does at late gestation and around parturition. Part-time group housing aims to circumvent the most severe consequences of such aggression by delaying the grouping of does until the kits are older and more resilient. This study assessed aggression, using skin lesions as indicator, when does are grouped at different ages of the kits. Multiparous does (n=240) were housed in parks in groups of four. During five reproduction cycles each park was randomly assigned to one of the following treatments: grouping when kits were 22, 25 or 28 days old. The group housing phase ended at weaning when kits were 35 days old. Skin lesions were counted and scored one, four and six days after grouping and at weaning using a tagged visual analogue scale (0cm = no skin lesions, 10cm = death by lesions). No significant treatment effects were found for lesions on the does. Kits grouped at 22 days showed a lower total number and total lesion score compared with kits grouped at 25 (P=0.02 and 0.02, respectively) and 28 days (P=0.003 and 0.01, respectively). One day after grouping, 60% and 16% of does and kits respectively showed skin lesions, increasing to respectively 79% and 39% at weaning. For the kits, grouping at an earlier age seems more beneficial but the total percentages of wounded does and kits are still high. Agonistic behaviour serves as the base for establishing a social hierarchy which may lead to aggression related skin injuries. Park design should be adapted to reduce aggression and a better understanding of the social structure of rabbits is needed.



**Evidence for sex differences in behavioural and neural correlates to tickling in young Wistar rats**

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Positive welfare is considered to be not simply the absence of suffering but also the presence of positive experiences. ‘Tickling’ has been shown to induce positive affective states in laboratory rats as evidenced by the production of appetitive 50kHz ultrasonic vocalisations (USVs). However, few tickling studies have used female rats, thus whether females and males respond differently to tickling is not well understood. It was hypothesised that female rats would exhibit different behavioural responses and neural correlates to tickling than male rats due to sex-specific neural regulation of positive affective states. Wistar rats (n=32/sex) were placed in an arena for 2 min/day for 10 days. Tickled animals (n=16/sex) received alternating 15 seconds of tickling and 15 seconds of rest over 2 minutes. Control animals (n=16/sex) were placed in the arena for 2 minutes; the rat received no hand contact. Play behaviours (hops and darts) and 50kHz USV production were quantified. After 10 days of testing, whole brain was collected for analysis of the neural correlates underpinning observed behavioural responses. Double-labelled immunohistochemistry was used to quantify c-fos (a marker of cellular activity) expression in oxytocinergic neurons of the paraventricular nucleus of the hypothalamus. Behavioural data were analysed using a general linear model and Tukey pair-wise comparison. A multiple regression analysis was used to identify behaviours that explained neuronal activity in key brain regions. It was found that tickled rats, regardless of sex, performed more hops (p=0.006) and darts (p<0.001) than control rats. Female tickled rats produced significantly more 50kHz USVs than any other group (p<0.001). 50kHz USV production in tickled females significantly explained c-fos immunoreactivity in dorsal parvocellular oxytocinergic neurons in PVN (p=0.005), but not in tickled males (p=0.663). Whereas, c-fos immunoreactivity in medial parvocellular oxytocinergic neurons was explained by 50 kHz USV production in tickled males (p=0.040). These results provide evidence for a sex-specific behavioural and neural response to tickling in adolescent rats. Tickling was successful in inducing higher levels of 50kHz USVs in females and this may be explained by activity of parvocellular oxytocinergic neuronal populations in PVN. Elucidating the neurobiological basis of positive affect will develop our understanding of the relationship between tickling and positive affect.

## Behavioural assessment of capuchin monkeys (*Sapajus* spp.) in a rehabilitation programme

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Survival skills proficiency is a prerequisite for primate reintroductions. One way to assess the progress of rehabilitant individuals is by measuring changes in activity budgets across time. Here we evaluated the progress of 18 brown capuchin monkeys (*Sapajus* spp.) confiscated from the illegal trade and housed in 25m<sup>3</sup> enclosures in a Brazilian rescue centre. The monkeys were part of a rehabilitation programme that consisted of providing an adequate diet and housing whilst maintaining the animals in species appropriate social groups and avoiding human interaction as much as possible. We hypothesised there would be an increase in genus normative behaviours (i.e. foraging, locomotion, social behaviours, solitary play, vigilance, inactivity) and a decrease in human interaction and stress-related behaviours (i.e. motor stereotypies, self-directed and self-injurious behaviours) when comparing before and after rehabilitation. We performed 54 hours of observation divided into three phases (baseline, second, and final). We found that foraging and social positive behaviours increased significantly ( $X^2 = 23.44, p < 0.01$ ;  $X^2 = 15.75, p < 0.01$ ), while human interaction decreased significantly ( $X^2 = 7.28, p < 0.05$ ). The other behavioural categories remained stable. Our results reveal the rehabilitation is helping the monkeys to gain foraging and social skills as well as reinforcing human dishabituation but is not enhancing other important survival skills such as locomotion. We propose the use of environmental enrichment to enhance these skills in future rehabilitant individuals.

DRAFT

**"Inactive not alert" as a new indicator of enduring negative affective state in laboratory macaques**

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The lack of an objective method to assess enduring negative affective states is a current obstacle to improving the welfare of non-human primates in research. We propose the behaviour "Inactive not alert" as a welfare indicator of enduring negative state in rhesus macaques. The behaviour is defined as sitting or lying stationary with no visual contact with objects or individuals and not engaged in any other action for at least 10 seconds. In the past, this behaviour has been experimentally induced by exposing macaques to stressors known to cause depression in humans and has been pharmacologically validated with antidepressants. Therefore, literature suggests that the display of this behaviour is related to a negative affective state associated with low arousal (depressive-like state). Using video recordings of home-cage behaviour of 29 laboratory rhesus macaques housed in a relatively enriched environment, we report the presence of this behaviour, with an important variability in its frequency between and within individuals. These results pave the way to future investigation of factors responsible for this variability. Furthermore, results show no correlation with an "Inactive alert" behaviour, in which the subject is stationary but not withdrawn from its environment. We therefore suggest that the "Inactive not alert" behaviour should be differentiated from other inactive behaviours that are not known to be associated with a negative affective state. Limitations of the use of "Inactive not alert" as a behavioural welfare indicator include the current lack of a standard definition across the literature, its unknown sensitivity to negative affective states associated with high arousal and its potential sensitivity to factors unrelated to affective states (e.g. ageing).

## Agreeable horses experience elevated hair cortisol concentration

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Hair cortisol concentration (HCC) is increasingly used as a long-term biomarker of HPA axis activity and stress. HCC has been linked with personality in several wild and captive animal species. However, in equines the literature on HCC is sparse and the link between individual differences in behaviour and chronic HPA axis activity not well documented. This study aimed to explore the relationship between subjectively rated personality and HCC in horses, while accounting for additional covariates of HCC identified in other species (age, sex, hair colour, sampling location). HCC was assayed in mane hair from the withers, midpoint of the neck and poll of 24 riding school horses (66% male, mean age  $13 \pm 4$  years). Three caregivers provided scores for Agreeableness, Neuroticism, Extroversion, Gregariousness towards People and Gregariousness towards Horses for all subjects using a validated equine personality questionnaire. A gamma GLMM was used to model HCC as a function of personality scores and covariates. Individual horses were fitted as random intercepts in the model. Manual backward selection was used to identify the best-fitting plausible model. Mean HCC was  $3.8 \pm 1.2$  pg/mg. The final model only retained Agreeableness and sampling location as explanatory variables of HCC. HCC was significantly positively associated with Agreeableness ( $t=2.7$ ,  $p=0.01$ ). There was a weaker but significant impact of sampling location ( $t=2.1$ ,  $p=0.03$ ; higher HCC at the poll). The lack of association between Neuroticism and HCC was unexpected and indicates that trait stress-sensitivity may not be reflected in the activity of physiological stress pathways. The positive association between Agreeableness and HCC suggests that more agreeable horses experience higher levels of chronic HPA axis activity. This unexpected result may be because more stress-sensitive horses are more motivated to avoid pressures associated with the use of negative reinforcement in equine training, resulting in agreeable behaviour. Compliant behaviour may also result in increased exposure to aversive management or training practices in more agreeable individuals. This result has important implications for welfare in the context of the horse-human interaction and warrants further investigation to clarify whether a causal link exists between agreeable behaviour and elevated HCC in the horse.

## Sensor laterality and attentional state in horses coping with an unexpected stimulus

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Stimuli perceived from the environment are classified through attentional mechanisms, which in turn trigger behavioral responses. Attention can be exogenous (involuntarily directed towards the stimulus) or endogenous (voluntary initiated). The emotional content of a stimulus is a further aspect of these attentional processes as attention is typically focused on the stimulus that arouses the strongest emotional response. Here, we investigated exogenous attention to an unexpected stimulus in 57 Italian saddle horses across seven different stables. Horses' eyes are laterally positioned on the head, making them a good model for investigating visual laterality and attention. Each eye projects primarily to the contralateral hemisphere, allowing eye use to be a proxy for preferential processing in one hemisphere of the brain. In this study, we instigated the rapid appearance of a stimulus (an inflated balloon) designed to induce an avoidance reaction and elicit a negatively valent emotional state. A remote-controlled device with a video camera positioned in line with the stimulus, allowed us to avoid perspective errors in eye use recordings. Because of the emotionally arousing nature of the stimulus, we predicted that horses would primarily use exogenously directed attention mediated by their left eye and right hemisphere. Horses typically use the left eye when facing unfamiliar objects and often show escape responses when viewing a frightening stimulus with the left eye. We analyzed both monocular- and selective binocular attention to the balloon. The frequency, percentage duration and the length of the single bout of attention between right, left and binocular attention were significantly different (Friedman Test,  $p_{\text{freq}}=0.012$ ;  $p_{\%d}=0.001$ ; One-way ANOVA,  $p_{\text{Md}}=0.000$ ) showing a higher rate of the lateral attention compared to binocular attention (right vs binocular:  $p_{\text{freq}}=0.030$ ,  $p_{\%d}=0.006$ ,  $p_{\text{Md}}=0.000$ ; left vs binocular:  $p_{\text{freq}}=0.039$ ,  $p_{\%d}=0.002$ ,  $p_{\text{Md}}=0.017$ ). However, no difference was found in the preference for the right or left eye in any of the parameters (Friedman Test,  $p_{\text{freq}}=1.000$ ;  $p_{\%d}=1.000$ ;  $p_{\text{Md}}=0.2469$ ). Our results confirm a lateralized eye use tendency when viewing emotionally arousing stimuli in horses, in agreement with previous findings. However, there does not seem to be any alignment of lateralization at the group level, suggesting that previous reports of population level lateralization in horses may have been overstated.

## Development of the joint-log-lift task in pigs: hints on cooperation?

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Prosocial behaviours have mainly been studied for their proximate mechanisms and evolutionary functions, especially in cognitive biology and behavioural ecology, but their implications for animal welfare remain poorly understood. This is due to a lack of research rather than null findings. We developed a paradigm to encourage cooperative behaviour in order to study its welfare implications. Our first aim was to develop a biologically relevant task. The joint-log-lift task is a social foraging paradigm in which pigs have to lift jointly and simultaneously a wooden log with their snouts to each obtain a food reward. The task relies on an obligate strategy, meaning that the only way to benefit is to cooperate. Our second aim was to test it as a freely-accessible apparatus that allows testing in the home pen ad libitum, letting the animal control when and with whom to cooperate. The test apparatus consisted of a self-contained box, fixable to a pen wall, with a wooden log behind a panel and two small openings where each of two pigs could lift a side of the log. Pigs received a reward after two of them simultaneously and successfully lifted the log against a magnet. Four studies were conducted, building upon each other to refine the testing apparatus, testing 5-9 week-old farm pigs. In the first study, 5 out of 7 pairs of pigs successfully solved the task with a prototype apparatus in as little as 5 min. In the second study, pigs maintained interest in the task for 10 days, although interest and success varied considerably between pens and individuals. Success rate progressively increased while failed attempts decreased, suggesting learning. Some dyads were more successful than others, hence preferential social associations emerged. In the third study, the pigs showed moderate interest in the device but never succeeded in lifting the log together. This was possibly due to the younger age (5 wk) or lack of experience of the food reward. Therefore, a food preference test was performed to determine the best rewards, and in the fourth study, habituation to the food reward and apparatus was offered prior to testing. These pigs showed continued interest and success at the task: the majority (> 50%) of pigs were successful after 1 week, reaching for the most successful pens (9-14 pigs per pen) between 68 and 100 successes over a 30 min testing session. Overall, farm pigs were able to spontaneously solve the joint-log-lift task. Some pigs succeeded from the first day, and the most successful pigs lifted the log up to 2 times a minute. This allowed us to investigate the effects of social factors such as littermates vs. mixed groups and dyads' characteristics related to success at the task. Further research is required to investigate the cognitive mechanism at play. This apparatus may offer the first cooperative task in farm animals to allow studying its social underpinnings and welfare implications.

### **Influencing elimination location in the domestic cat: a semiochemical approach.**

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Amongst undesired behaviours in cats, one major issue is unwanted toileting in people's gardens or in the house. This may lead to a breakdown of the cat-owner relationship, resulting in euthanasia or relinquishment of the cat. It may also lead people to use aversive methods to address this issue, potentially endangering the cats' welfare. The aim of this study was to assess the effect of a cat's anal glands semiochemical composition on the elimination behaviour of domestic cats. The study was conducted in four catteries, which housed 33 cats (male and female, entire and neutered), using 37 litter trays. The study followed a randomised crossover design using the litter tray as experimental unit and was approved by the IRSEA ethics committee. During two weeks, twice a day, litter trays were cleaned, filled up with fresh litter substrate and sprayed with treatment or placebo according to the randomisation list. Parameters studied included daily elimination (urine plus stools) weight, urine only weight, elimination type and urine/stool quantity's scoring. Parameters were analysed using GLMM with SAS 9.4 software. Three out of five parameters studied showed a treatment effect, all consistently in favour of cats defecating significantly less in the litter trays sprayed with the active ingredient versus litter trays sprayed with the placebo (elimination weight **p=0,0199**; elimination type **p=0,0251**; stool quantity **p=0,003**). The last two parameters quantified urine production. These results demonstrate that an intraspecific message can be used to influence cats' defecation location, potentially helping to manage unwanted toileting, thus decreasing the risk that the uses of aversive methods pose to cats' welfare.

DRAFT



## Effects of the COVID-19 pandemic on animal behaviour and welfare researchers

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The COVID-19 pandemic has resulted in many changes in the way research is conducted. Some specific groups (e.g. women) and activities (e.g. teaching) may have been disproportionately affected. It is therefore relevant to investigate and acknowledge its consequences for researchers. Our aim is to assess the impact of the COVID-19 pandemic on animal behaviour and welfare researchers' work experience and productivity. In June 2020, an online survey link was distributed by email. The survey included questions on, among others, childcare, research and teaching load and changes due to the pandemic, and included the Perceived Stress Scale (PSS) and the Inventory of Socially Supportive Behaviours (ISSB). This abstract reports preliminary findings based on 109 survey responses, originating from 27 countries. Respondents mainly worked at universities (77%) and were PhD students (35%), researchers (41%) or senior researchers (24%) working on animal welfare (71%) or animal behaviour (26%). More women (79%) than men responded, and the average age was  $38.5 \pm 0.9$  years. Thirty-five percent of the respondents were taking care of children ( $n=27$  women and 12 men), with childcare increasing for 79% of the parents during the pandemic. At the time of completing the survey, 88% were working from home, of which 62.3% in lock-down. The number of working hours either reduced (33%), remained the same (31%), or increased (24%) during the pandemic, and the same figures for time spent on education were 25%, 41%, and 18%, respectively. Time spent on research mainly remained the same (48%) or reduced (43%), whereas writing papers and grants increased for some (36%), and for others it decreased (31%) or was halted completely (12%). The PSS was on average  $21 \pm 6.5$ , indicating greater perceived stress compared to the reference value (13.0). The ISSB scale was on average  $40 \pm 11.6$ , out of a maximum level of social support of 95. The PSS and ISSB were unrelated to gender and childcare (all  $P > 0.05$ ). Most variables did not differ between men and women, but 89% believed that female researchers faced more challenges during the lock-down than male researchers. Responses varied greatly regarding productivity and preference to remain working from home. Overall, the preliminary data leans towards a bimodal distribution of researchers' experiences and work output during the pandemic.



**Animal Welfare Infractions of Working Donkeys in Kasena Nankana West and East Districts, in the Upper East Region, Ghana. A case study.**

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In Ghana, the highest population of donkeys is in the Savannah agro-ecological zone. This zone borders with Togo to the east, Côte d'Ivoire to the west and Burkina Faso to the north. With reported outbreaks of Strangles in donkeys in neighbouring countries (Burkina Faso, Mali, and Togo)<sup>1</sup>, and confirmed cases of African Horse Sickness in Accra, Ghana<sup>2</sup>, the health and wellbeing of donkeys across the country is of increased animal health concerns. Donkeys are used in transportation and labour, playing an integral role in the farming systems in the Upper East, Upper West, and Northern regions in Ghana. These animals are essential in fulfilling the economic needs of the local communities. Unlike sheep and goats which have deep cultural significance for sacrifices, dowry, as well as animal sourced protein needs, donkeys are largely kept as working animals which are thought to be used with limited rest and denied various critical welfare needs. The purpose of this study is to determine the current socio-economic uses for donkeys, evaluate welfare conditions for shelter, feed, water, veterinary care delivery, and assess the knowledge, attitudes, and practices (KAPs) of the donkey owners for sustainable production practices in the Upper East region of the country. Seven communities will participate in the study. These are Yua, Sirigu, Natugnia, Manyoro, Mirigu, Kandiga, and Nabango, all in the Kassena-Nankana West and East Districts of the Upper East Region. Fifteen donkey owners from each community will be given a questionnaire containing open and closed-ended questions to assess the five freedoms of animal welfare in relation to their donkey(s), uses of their donkey(s), and access of animal health care delivery. The fifteen owners from each community will be randomly selected by the community opinion leaders, veterinary technical officers, or assembly-men in each community. Responses will be evaluated to determine problem areas to be addressed such as shelter, feed and water access, vaccination and/or veterinary care, and workload.

<sup>1</sup>Verbal communication with Dr. Awuni at Accra Veterinary Lab, Accra, Ghana June 7, 2019

<sup>2</sup>Verbal communication with Dr. Awuni at Accra Veterinary Lab, Accra, Ghana June 7, 2019

## Challenges of implementation of Animal Welfare standards in the Middle East

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Animal welfare is a complex subject with scientific, ethical, economic, cultural, religious, and political dimensions. eight animal welfare standards were adopted by animal health organization in 2005 covering the following areas: transport of animals by land; - transport of animals by sea; - transport of animals by air; - animal slaughter for human consumption; - animal killing for disease control purposes; - the use of animals in research and education; - animal welfare and beef cattle production systems. There are many challenges that face animal welfare in MENA region but the main challenge is to reach a consensus amongst all OIE Members, the sole common point being the exclusive use of a scientific basis to establish standards for adoption. animal welfare still not integrated as a core subject in veterinary education and to promote applied research as the scientific basis for standards and regulations. Moreover, here the major challenges in front of animal welfare in the region; national legislations are not yet updated in some countries, no sufficient support from political and high-level leadership within OIE Member Countries, Resources and access to technical expertise and scientific information are not enough. No actively involvement of Stakeholders, including farmers, industry, NGOs, academia, religious authorities, and the veterinary profession in the process of animal welfare. Implementation approaches of the general policies and standards of the OIE are not consistent. There is no effective and professional coordination and communication between countries. research priorities for the region is not identified for the relevant international developments in animal welfare. There are no identifiable and clearly defined regional standards and guidelines. Animal health in the countries still not well improved. Sustainability of activities regarding animal welfare is questionable. Very little efforts carried out in terms of animal welfare awareness. Veterinary service not developed and suffering from problems.

## **Welfare and ethical issues on offloading of cattle in Akinyele market, Ibadan**

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Transportation of animals is accompanied by many stressful events, which affect animal welfare and uncover ethical issues about animal transportation. The study was carried out at the International Cattle, Sheep and Goat market, Akinyele, Ibadan to uncover unethical issues of cattle offloading using a questionnaire. The average number of cattle carried by a canter, single and trailer are 17, 28 and 51 respectively. The average time taken for cattle offloading from a canter, single and trailer is 19, 28 and 49 minutes respectively. Sokoto state had the least amount of cattle and Kebbi state had the highest amount of cattle offloaded at The International Cattle, Sheep and Goat market, Akinyele, Ibadan at 0.2% and 65.8% respectively. The population of the breeds of cattle offloaded at the International Cattle, Sheep and Goat market, Akinyele, Ibadan stood at 1%, 31.8%, 32.1%, and 35.8% for Kuri, Bunaje, Red Bororo and Gudali breeds respectively. The total number of cattle offloaded at the International Cattle, Sheep and Goat market was proportioned at 39% bulls and 61% cows. 60.7% of the cattle offloaded were meaty or in good body condition while 39.3% of the cattle offloaded were rickety or debilitated. 60.7% of the cattle offloaded were without carcass bruises while 39.3% of the cattle offloaded were with carcass bruises. 1.7% of the cattle were dead at offloading, 4.4% of the cattle were slaughtered at offloading, 6.8% of the cattle were wheeled, 7.6% of the cattle were supported with therapy, 12.7% were supported with wood 29.1% of the cattle were standing, 37.7% of the cattle were supported with tail during offloading. 3% of the cattle offloaded had injured leg, 3.2% of the cattle offloaded had injured horn 7% of the cattle offloaded had bruised skin, 78.9% of the cattle offloaded was without defect. Single, Trailer and Canter vehicles were used for cattle transportation at 26.2%, 31.7% and 42.1% respectively. It was concluded that cattle is dragged, beaten and unethically handled during offloading.

**The comparison between working & unworked donkeys welfare in Nyala city, South Darfur, Sudan.**

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Donkeys in developing countries are contributed in people's livelihood. The welfare of animal in Sudan, regarding donkeys is neglected if compared with developed countries. However, no information about donkeys welfare condition in Darfur. The aim of present study is welfare assessment of donkeys, animals were divided into two groups (n=50 of each), working and unworking donkeys. The parameters investigated included physical, emotional, manual parameters and owner questionnaire. There were significant differences ( $P>0.001$ ) in body condition scores (sound=84%, (working 37%, unworking 47%), emaciated=16%, (working 13%, unworking 3% ), lameness 5% working, (hoof problem 33% working, 19% unworking), coat state=8% erected (7% working, 1% unworking), coat appearance not bright =46% (36% working , 10% unworking), orifice discharge(ocular=18% ( 15% working, 3% unworking), nasal 7% working but not unworking), scare=26%, (16% working, 10% unworking), wounds=28% (25% working, 3% unworking), and behaviors response 7% depressed, 5% aggressive, was observed in working donkeys only. In addition, respiration rate 50% polypnea (42% working, 8% unworking donkeys) and pulse rate 37% increased (31% working, 6% unworking donkeys) and only hydration= 11% dehydrated was observed in working donkeys. Moreover, there was significant differences in hitting tools = 24% whip (17% working, 7% unworking donkeys), stick=34% (33% working, 1% unworking donkeys), and feeding time=82%, one to three times daily (50% working, 32% unworking donkeys), 18% free access only observed in unworking donkeys. Yet, there were no significance differences ( $P<0.06$ ) in emotional parameters. We conclude that working donkeys are suffering from multiple welfare problems. Awareness and education service are important to improve donkeys welfare.

### **Sleeping behaviour of Nigerian indigenous chickens**

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Sleep is of great significance as it relates to the welfare of the animal; it ultimately increases their rate of survival. Local chickens still exhibit their natural behaviours of which sleeping is one of them. Despite their large population, little research work has been undertaken on the sleeping behaviour of Nigerian indigenous chickens, hence the need for this study. Sixty four birds (twenty four cocks and forty hens) were randomly assigned to eight pens with three cocks and five hens in each pen. Each pen had a perch (111cm above ground level) for the birds to roost on. The perch consist of three tiers; the lower, middle and upper tiers which were 20cm, 50cm and 90cm above the ground level, respectively. The sleeping behaviour of two focal birds per pen (1 cock and 1 hen) were monitored in terms of roosting location on the perch (lower, middle and upper tiers), neck position (neck stretched out, neck in normal position and head tucked under wings), sleeping direction (facing forward or backward) and sleeping posture (standing or crouching) at 8.00pm daily for twelve consecutive days. Data were analyzed using descriptive statistics. For the 12 days of the study, only three birds (1 hen and 2 cocks) were consistent (100%) in the use of the upper tier of the perch. Two hens used the upper tier (83.3%) and middle (16.7%) while two cocks used the upper tier (91.70%) and middle (8.30%). One hen was observed to be more consistent on the middle tier (91.7%). Four birds (1 hen and 3 cocks) were the most inconsistent in the perch use (used the three tiers of the perch). In terms of sleeping direction, only one cock was highly consistent (100%) facing back on the perch. Two cocks showed 91.7 and 83.7% consistency facing front. Five cocks and four hens showed 58.3-75% facing front. For neck positioning during sleep, it was observed that four cocks and four hens used the three neck positions while three hens and one cock had their neck placed normally more often than stretching their neck. Observation on the sleeping posture showed that a hen was consistently (100%) standing while sleeping for the 12 days. Five hens and two cocks slept in a standing position (41.7-83.3%). Three cocks crouched (91.7%) during sleeping. Throughout this study, a cock was very consistent in sleeping behaviour, 100% use of upper tier of the perch, 91.7% facing forward, 83.3% sleeping in crouching position and 75% sleeping with head in normal position. This study showed that sleeping behaviour of Nigerian indigenous chickens differs with personality.

## Behaviour of white Fulani calves as affected by plant species, age and spacing

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Forages are vital in livestock production hence the most important feed resource in ruminant animal feeding with concentrates diet being fed as supplements. Forages were certainly known as a cost effective feed rather than commercial concentrate. There are many species of grasses with high production capability which thrives well under local tropical conditions. Some of them are *Megathyrsus maximus* (Guinea grass) and *Cenchrus purpureus* (Elephant grass). Studying animal behaviour on pasture helps in detecting various behaviours exhibited and the reason for the exhibition of such behaviours. The objective of the study is to determine the effect of plant age (3 and 6 week after cutback), plant species (*M. maximus* and *C. purpureus*), and plant spacing (0.5m x 1m and 1m x 1m) on the grazing and other behaviours (walking, standing, collision, grooming and foot stamping) of 12 white Fulani calves. The experimental design was a split-split plot with three replicates. The calves were allowed to graze for 2 hours per day for 3 days per week and exhibited behaviours were recorded with the aid of digital video recorder mounted at each experimental unit. The playbacks of the recorded behaviours were performed using Behavioural Observation Research Interactive Software (BORIS) and the time budget analysis of each behaviour was extracted. Data extracted was subjected to analysis of variance (ANOVA) using General Linear Model. The time spent on grazing, standing, grooming, and foot stamping were significantly ( $p < 0.05$ ) affected by the plant age. Time spent on grazing was higher at 6 weeks after cutback (6769.00 sec) than at 3 weeks after cutback. At 3 week after cutback, time spent by the calves on standing (638.20 sec), grooming (113.83 sec) and foot stamping (102.50 sec) were higher than that at 6 weeks after cutback. However, time spent on walking and collisions were not affected by plant age. Plant species influenced ( $p < 0.05$ ) all the behaviours except foot stamping. Grazing time was higher on *M. maximus* (6765.10 sec) plot compared to time spent on *C. purpureus* plot. Walking, standing, collision and grooming were performed more by calves on *C. purpureus* paddock. Time spent on grazing, walking and standing by the calves were significantly ( $p < 0.05$ ) affected by plant spacing, while collision, grooming and foot stamping were not affected ( $p > 0.05$ ) by plant spacing. On the plot of 0.5m x 1m plant spacing, grazing time was relatively higher (6591.60 sec) than 1m x 1m plant spacing plot (6311.30 sec). The time spent on walking and standing were lesser on 0.5m x 1m plant spacing plot compared to 1m x 1m plant spacing plot. From these results, it could be concluded that grazing behaviours of White Fulani calves were influenced by plant species, age and spacing.

**Improving pig welfare, production and farms' economic, by avoiding husbandry mutilations in piglets and providing environmental enrichment**

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Piglets around the world undergo a set of invasive procedures during the first days of their lives, which commonly includes surgical castration and tail docking and in many countries, even teeth clipping. Those procedures are a top welfare concern since it potentially resulting in pain and stress, which may have long term effects on the animals' health, welfare and production. Our objective was to examine the welfare and production parameters of pigs from birth to slaughter. Litters (n=32 sows; 329 piglets; 3 days after farrowing) were allocated randomly into 4 groups; G1: Surgical castration, tail docking and teeth clipping, without environmental enrichment; G2: same as G1, but meaningful environmental enrichment was provided; G3: Non-surgical sterilization with anti-GnRH vaccine (Improvac®), tail docking, teeth clipping, with environmental enrichment; G4: none of the invasive procedures were performed, piglet were vaccinated (Improvac®), and environmental enrichment was provided. Mixed-effects Linear Regression model revealed that standardized slaughter weight significantly increased when invasive procedures were avoided and environmental enrichment was provided (G1: 99.2±1.07, G2: 99.9±1.4, G3: 103.6±1.58, G4: 106.5±1.6 Kg; P<0.05). The odds ratio to be weak, dead in the conventional, non-enriched G1, was 89% higher than in G4 (P<0.05; Chi-square goodness of fit). Hair Cortisol on weaning day, as a marker for chronic stress, decreased as compared to the conventional G1, when the husbandry procedures were gradually avoided (G3: -32.18%, P=0.014; G4: -24.5%, P=0.102). Saliva Cortisol, from weaning to slaughter, as a marker for acute stress, was significantly lower in all enriched groups as compared to the conventional, non-enriched Group1 (Group 2: -0.74±0.37, Group3: -1.1±0.35, Group 4: -0.98±0.36; Linear Regression). Anti-GnRH vaccine was effective in reducing serum and hair testosterone, and hair DHEA, similar to surgical castration. An economic model based on this study suggests an expected combined consumer and producer surplus to increase by 1.48 to 1.92 billion US\$ per year if implemented by the entire USA swine market. In conclusion, replacing surgical castration by anti-GnRH vaccine, avoiding tail docking and teeth clipping, and providing meaningful environmental enrichment, are welfare friendly alternatives that substantially benefit both the animals and farmers.



**Tigernut milk (*Cyperus esculentus*) enhances the welfare of broiler chickens**

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The seeming success in the production efficiency of the modern broiler has come with unwanted consequences, particularly musculoskeletal abnormalities and poor walking ability (commonly referred to as 'leg weakness'). Tigernut has high phosphorus, calcium and magnesium which are necessary minerals for bone development. This study was designed to investigate if tigernut milk (TNM) could alleviate leg weakness in broiler chickens using the latency to lie test. Fifty broiler chicks were purchased for this experiment and brooded for three weeks. At 4 weeks of age, forty-five of the broiler chickens were randomly assigned to three treatments with 15 birds per treatment; T1 (0ml TNM/litre of water), T2 (5ml TNM /litre of water) and T3 (10ml TNM /litre of water) daily for 4 weeks. All the birds were subjected to a latency to lie test at the 6<sup>th</sup>, 7<sup>th</sup> and 8<sup>th</sup> week of age which corresponded to the 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> week of TNM treatment. The time spent standing before making the first attempt to lie down was recorded for each bird. Data (average for the 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> week of treatment administration) was subjected to One-way analysis of variance of SPSS statistical package. Broilers offered 5ml and 10 ml TNM/litre of water stood for a longer time ( $P < 0.05$ ) compared to the control birds (263s and 272s vs 217s respectively). The use of TNM can serve as natural additive to alleviate leg weakness in broiler chickens.



## Impact of epilepsy on judgement bias in a population of companion dogs

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Idiopathic epilepsy (IE) is the most common chronic neurological condition in dogs, characterised by recurrent seizure activity with no identifiable cause. With growing owner-reported evidence for the negative impact of IE upon quality of life, behaviour, and cognitive functioning, this study aimed to investigate the effects of IE on putative canine affective state in a cognitive (judgement) bias task; detecting relative optimism or pessimism to ambiguous stimuli. We hypothesised that dogs with IE would exhibit a more pessimistic judgement bias than healthy controls. Dogs from two breeds (Border Collie n=40, Labrador Retriever n=28) were recruited; n=33 diagnosed with IE (meeting International Veterinary Epilepsy Task Force diagnostic criteria) and n=35 controls. Dogs took part in a previously validated judgement bias task in a standardised testing environment. Food rewards were used to teach a spatial discrimination between rewarded (P) and unrewarded (N) bowl locations. Upon reaching criterion, latencies to approach three ambiguous bowl locations (NN, MID, NP) were recorded. Linear mixed models (LMM) were constructed accounting for study group, bowl position and signalment (fixed effects), and the repeated effect of dog ID (random effect). Sixty-eight dogs were tested (IE=33, Control=35); study groups did not differ by age, sex, breed or neuter status ( $p>0.05$ ). Dogs with IE were significantly more likely to fail to learn the spatial discrimination after 50 trials than controls (22% IE vs. 4% Control;  $p=0.019$ ). Thirty-seven dogs completed the training task, acquired the discrimination and were subsequently tested for their responses to probe locations (IE=19, Control=18). Dogs responded differently to the three ambiguous locations (LMM,  $F_{2,102}=11.8$ ,  $p<0.001$ ). There was no main effect of study group (LMM,  $F_{1,102}=0.20$ ,  $p=0.658$ ), or interaction between study group and bowl location (LMM,  $F_{2,102}=0.11$ ,  $p=0.900$ ). In contrast with our hypothesis, there was no evidence that IE altered judgement bias in this study population; however, dogs with IE were less likely to acquire the spatial discrimination, which may reflect IE-related cognitive deficits. In addition, dogs exhibiting signs of anxiety during testing were less likely to complete the task, resulting in missing data from more severely affected dogs. Developing methods to test affective state without excluding cognitively impaired or more anxious individuals is a future challenge for applied ethology.

## Exposure to controlled challenges increases stress resilience in dog puppies

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The socialisation period in dog puppies is one of the most important periods in determining later behaviour, as indicated by retrospective studies. However, only few studies tested interventions to improve stress resilience in dog puppies, and these focused largely on exposure to different social and non-social stimuli. Here we aimed to test the effect of providing early stimulation beyond mere stimulus presentation on stress resilience in dog puppies. We tested 83 dog puppies from 12 litters of eight breeds. Half of each litter was randomly assigned to the treatment group, the other half to the control group. Puppies in the treatment group received age-appropriate ‘challenge’ exercises four times per week (total 12 times) between the ages of 3-5 weeks. Each training day, treatment puppies were exposed to (1) sudden real-life noises, with increased intensity upon repetition, (2) novel objects, many of which were moving, and (3) a problem solving task (e.g. overcoming small obstacles or a detour task). The control group did not receive any training, but spent the same amount of time with the trainer, being cuddled and played with, and received the same amount of food during sessions. Between 40 and 52 days of age, all puppies were tested in a behaviour test. A nonlinear Principal Component Analysis over coded behaviours yielded four principal components, two of which differed significantly between treatment groups. Firstly, “Response to Novelty” ( $F_{1,70}=8.75$ ,  $p=0.0042$ ; Cohen's  $D=0.71$ ), meaning that the puppies in the treatment group solved the problem solving task faster and showed more exploration of a novel object as well as less seeking of humans and whimpering during the novel object test. Secondly “Social-Startle” ( $F_{1,70}=8.93$ ,  $p=0.0039$ ; Cohen's  $D=-0.71$ ), indicating that the treatment puppies showed a reduced startle reaction in response to a loud noise; however, the control group showed a higher interest in a friendly stranger. It is a possibility that the increased handling experienced by the control group had beneficial effects on their sociability. The presentation of a diversity of challenges, surprises and novel objects at an early age seemed to enable the treatment puppies to cope better when confronted with novel stimuli and to react less fearfully and recover faster after a loud noise. To conclude, providing small challenges to puppies between 3-5 weeks of age is effective in increasing their stress resilience.

## Slow and steady wins the race: a comparison of standard commercial broilers against a slower growing alternative

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Standard commercial broilers have been bred to grow quickly but this has implications on behaviour, such as reduced activity, and welfare, such as high mortality and lameness. It has been proposed that slower growing broiler breeds (reaching market weight about 14 days later than standard broilers) be used as a higher welfare alternative. To investigate this, behaviour, welfare, production and slaughter measures were collected from three commonly used standard broiler breeds, one from each of the three main broiler companies (breeds FA, FB and FC) and compared to a commercially available slower growing breed (S). A total of 1600 birds (400 of each breed) were housed in litter floored pens in groups of 50 birds with 8 pens per breed for a total of 32 pens. Welfare measures were assessed when the average bird weight per breed was 2.2 and 2.5kg and home pen behaviour was recorded weekly with hourly scan samples to estimate behavioural time budgets. Birds were given *ad libitum* feed and water and Feed Conversion Ratio (FCR) and Average Daily Gain (ADG) were calculated per breed at 2.2kg average bird weight. After the 2<sup>nd</sup> welfare assessment, birds were slaughtered and meat quality measures were recorded. Overall, breeds S and FC had lower mortality levels and fewer culls due to lameness ( $P < 0.05$  for both). S birds performed better in the welfare assessments and were more active than the faster growing birds ( $P < 0.05$  for all). Faster growing breeds had better FCR and ADG, ate less feed overall and had more breast meat on average than S birds ( $P < 0.05$  for all); although S birds had fewer meat quality issues, such as white striping, and had heavier leg weights ( $P < 0.05$  for all). Therefore, S birds have improved welfare and a more active behavioural repertoire but aren't as efficient in production as standard commercial broilers. However, if the reduction in mortality levels and meat quality issues, as well as the higher price paid for the brands these birds are marketed under, are considered, the use of slower growing breeds may be a feasible commercial alternative.

## Effects of temporary-crating systems on the welfare of lactating sows

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The aim of the study was to assess lactating sow welfare in three farrowing systems. The farrowing systems included one conventional permanent crating (CON) and two temporary-crating systems (SWAP and JLF15, produced by Jyden, Denmark). Four batches of crossbred Duroc sows were studied: 18 sows in CON, 24 sows in SWAP and 23 sows in JLF15. The day of farrowing was considered as Day (D) 0. Crating period in CON was from entry to weaning, while in SWAP and JLF15, sows were crated from one day before expected farrowing day to three days postpartum (D3). Saliva was sampled on 1 day pre- and post-expected farrowing day to evaluate the crating stress, and 1 day pre- (-1D), 1 day post- (+1D) and 2 days post-weaning (+2D) to evaluate the weaning stress. Salivary cortisol (CORT) and chromogranin A (CgA) were determined. Mother-young social behaviors were scan-sampled on D2, D4, mid-lactation and late-lactation. Sow vocalizations were scan-sampled and counted on the day of weaning, +1D and +2D. CORT and CgA were log-transformed to fit into a linear mixed model (LM). Behaviors were analyzed with Kruskal-Wallis test and Dunn's test. Vocalization counts were square-root transformed to fit into an LM. CORT and CgA did not differ between treatments in terms of crating stress ( $P > 0.05$ ). After weaning, the increase of CORT lasted for two days in CON ( $P < 0.05$ ), whereas in SWAP, CgA increased on +2D, compared to -1D and +1D ( $P < 0.05$ ). On D2, piglet-initiated naso-naso contacts towards sow(s) (NNC) ( $P < 0.05$ ) and piglet-initiated sow contacts were higher in SWAP than in CON ( $P = 0.07$ ). On D4, sows in SWAP showed more exploration than in CON ( $P = 0.03$ ). In mid-lactation, more NNC and sow-initiated mother-young interactions (MYI) were observed in SWAP and JLF15 than in CON (NNC: CON vs. SWAP,  $P = 0.04$ , CON vs. JLF15,  $P = 0.01$ ; MYI: CON vs. SWAP,  $P = 0.001$ , CON vs. JLF15,  $P = 0.003$ ). In late-lactation, there were more MYI in SWAP than in CON ( $P = 0.02$ ). Sows' vocalizations did not differ between treatments, but it largely decreased from the day of weaning to +1D in all treatments (CON:  $P = 0.009$ ; SWAP:  $P = 0.0008$ ; JLF15:  $P = 0.05$ ). Our results suggested that temporary-crating did not cause stress to the sows, and compared to CON, SWAP facilitated the social interactions between sows and piglets throughout the lactation period. In conclusion, temporary-crating systems encouraged the expression of mother-young social behaviors and mitigated the stress of sows at weaning.

## Detection of dairy cows' hind-legs activity during milking using a 3D-accelerometer attached to the milking cluster

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Animal behaviours can be used as reliable welfare indicators. Though daily milking is a preponderant routine in dairy cows, evidences were found for increased hind-legs' activity during milking being an indicator of stress, discomfort and pain. Such behaviors can point out to deficiencies in the milking machine and thus to welfare issues. These behaviors should thus be objectively and easily detected. Assuming that the milking cluster follows cows' hind-legs movements during milking, the aim of our study was to validate the use of a 3D-accelerometer attached to the milking cluster to detect cows' hind-legs' activity. The behavior of 45 dairy cows during one morning and one evening milking in a 2x3 ATM has been monitored using direct observations and a 3D-accelerometer (MSR logger; 50Hz) attached directly to the claw's hook of each milking cluster. The experiment took place at the Agroscope Research Station in Tänikon, CH, and consistent with the ISAE Ethical Guidelines. We recorded when hind-legs' foot-lifting, stepping and kicking behaviors occurred and defined a period of activity each time such behaviors occurred within 3s of each other. Detected activities were defined using a standard deviation filter of the x-lateral acceleration axis. We set the detection threshold at 0.13g ( $=1.27\text{m/s}^2$ ), based on the visual evaluation of the graphs of five cows. Matching observed and detected periods of activity were considered when they occurred simultaneously (with a 2s delay acceptance). We calculated accuracy indices and the average number of periods of activity/cow/milking/min, and performed a Pearson's correlation between the observations and detections. 484 hind-legs' periods of activity were observed. The detection of the cows' hind-legs activity using the accelerometer was found to have a good sensitivity (70% of the observed activities were detected), and a high specificity (99%), positive predicted value (76%), and negative predictive value (98%). Cows were observed being active 0.94 times/min while being detected active 0.87 times/min. The observed and detected periods of activity were found to be correlated ( $R=0.657$ ,  $p<0.001$ ). The use of an accelerometer attached to the milking cluster seems therefore promising to reliably estimate the hind-legs' activity of dairy cows during milking. Nevertheless, further investigations remain necessary to clarify whether other factors can cause milking cluster movement variations regardless of cows' hind-legs activity.

## **Social behaviour in pigs: are littermates nicer to each other than to strangers?**

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The importance of social stability and its influence on social interactions in domestic animals remains poorly understood, especially for affiliative behaviours and other putative socio-positive behaviours. This study investigated the occurrence and type of social behaviour within groups of littermates vs. groups composed of pigs unfamiliar to each other. We hypothesised that pigs kept with littermates would show more positive social interactions than pigs mixed after weaning, and that social interactions would change over time as mixed groups progressively regained social stability. The behaviour of 14 groups of indoor-housed weaned pigs (7 groups / treatment; 8-14 pigs / group) was observed after weaning (4 wk of age) and thereafter once per week until the seventh week using scan sampling (48 scans / individual / d) and continuous observations (150 min / group / d). Pigs spent most time lying together (on average 50% of the scans) or performing non-social behaviours (31% of scans). Exploring together was the most common social behaviour, observed in 7.32% of scans. Other social behaviours occurred infrequently and each accounted for only 0.04% to 1.09% of scans. Treatment significantly influenced mounting behaviour, with more mounting between pigs in the mixed groups (0.53% of scans) as compared to littermate groups (0.24% of scans;  $P=0.01$ ). Other behaviours did not differ between treatment or the treatment $\times$ time point interaction. Nose-nose contact peaked at three days after weaning ( $P=0.04$ ), and most oral manipulation and play peaked in week 7 of life (both  $P=0.001$ ). Pigs in littermate groups showed a better growth rate after weaning (littermates:  $1.03\pm 0.11$  kg; mixed:  $0.78\pm 0.11$  kg;  $P=0.01$ ), despite no difference in body weight at the start of the trial. In conclusion, presumed socio-positive behaviours were more prevalent than socio-negative behaviours overall, supporting that positive social behaviour may be an important but overlooked aspect of social life. Groups of littermates did not show more positive social behaviour than mixed groups, but they did show less mounting behaviour and seemed less affected by the stress caused by weaning.

## **Stockperson attitudes and on-farm handling of Swedish finishing pigs, and influence on animal welfare**

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Rearing of pigs involves environmental stressors that can cause both animal welfare, health and productivity issues. Previous research shows that the quality of interactions between stockpersons and pigs on farm influences pigs' behavioural response when interacting with unfamiliar humans, possibly also during pre-slaughter handling. There is evidence of a relationship between stockperson attitudes and behaviours and the pigs' behavioural response to humans, and both negative handling behaviours and minimal tactile contact with stockpersons are associated with increased fear of humans. Low fear facilitates handling and is beneficial to both pigs and stockpeople. We investigated stockperson attitudes, working routines and handling of pigs, and their relationship with pigs' fear of an unfamiliar human at 9 commercial pig farms located in the south, middle and north of Sweden. One section per farm with between 162 to 510 pigs (total of 2795 pigs in 308 pens), aged between 20-25 weeks, and 1 stockperson/farm were studied. Measurements included a questionnaire on stockpersons beliefs about pigs, observations of stockpersons behaviour during one routinely management sequence such as time spent with the pigs and tactile and auditory interactions, and if it was gentle (positive/ neutral) or rough (negative). Pigs' reactions to an unfamiliar human were assessed in 10 randomly selected pens in each section by recording proportion of pigs fleeing, facing away or huddling in a corner while an unfamiliar human slowly walked inside the pen. In general, stockpersons displayed positive normative beliefs about pigs, spent an average of  $49.5 \pm 48.7$  (mean  $\pm$  SD) s/pen, and performed more rough handling behaviours (mean 0.14 interactions/pig) than gentle (mean 0.036 interactions/pig). There were no significant correlations between pig behaviour and either stockperson attitudes or stockperson behaviour. This is not unexpected with the small sample size ( $n=9$ ) however, the stockperson who spent most time in the study section of the farm (174.5 seconds) and performed the most number of positive interactions had the least proportion of pigs displaying a fear response. These limited data indicates that Swedish stockpeople working with finishing pigs spend little time with the pigs and use mainly negative handling behaviours. Training of stockpersons and refined handling routines may improve animal welfare and facilitate handling of pigs' both on-farm and during pre-slaughter handling.



## Electronic sow feeder (ESF) use patterns are associated with locomotory ability and stereotypic behaviour in pregnant gilts

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Variations in feeding behaviour patterns can act as an early warning sign of health issues, and can thus provide insights into the welfare status of animals. Therefore, the aim of this study was to investigate associations between ESF use patterns by pregnant gilts and their likelihood of having poor locomotory ability or performing stereotypic behaviour during pregnancy. Every 3 weeks, gilts (n=51; 8 batches) were introduced to a dynamic group with ESF, on d30 post-insemination. Sixteen aspects of locomotory ability were scored at service, mid-pregnancy (MID), entry to farrowing crates and at weaning using a visual analogue scale. Stereotypic behaviour (sham chewing, mouth stretching, palate grinding, sucking, tongue flicking, licking) was recorded on days 33, 58 and 107 of gestation using instantaneous scan sampling at 5min intervals for 3hrs (n=36 scans/day), and the percentage frequency was calculated for each gilt throughout gestation. We used data recorded automatically by the ESF to determine the order of entry to the ESF at mixing and in mid-pregnancy (d57), and to calculate the average number of daily visits to the ESF per gilt. Associations between ESF use patterns, locomotory ability and stereotypic behaviour were determined using linear mixed models (PROC MIXED; SASv9.4). Later time of ESF entry at mixing and in mid-pregnancy, and fewer daily ESF visits were associated with higher frequencies of stereotypic behaviour ( $F_{1,49}=5.2$ ;  $P<0.05$ ;  $F_{1,49}=7.6$ ;  $P<0.01$ ;  $F_{1,49}=8.7$ ;  $P<0.01$  respectively), and higher MID locomotion scores ( $F_{1,49}=7.9$ ;  $P<0.01$ ;  $F_{1,49}=8.4$ ;  $P<0.01$ ;  $F_{1,49}=9.1$ ;  $P<0.01$  respectively). It is possible that poor locomotory ability impeded the gilts' ability to compete for entry to the ESF. Frustration associated with the inability to attempt to feed by entering the ESF more often could explain the higher frequencies of stereotypic behaviour of gilts with lower locomotory ability. In conclusion, ESF use patterns could help to identify sows with locomotion problems, helping to prioritise such individuals for treatment in order to improve their welfare status.



**Cognition in the barn: range use and its relation to cognitive performance in free-range broiler chickens (*Gallus gallus domesticus*)**

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Free-range chickens are not all the same. Within the same group, under the same conditions, some individuals explore the range more than others. In many animal species, including chickens, individuals differ in behavior and cognition, i.e., how they perceive, process, and memorize information from their environment. We aimed to further understand the relationship between range use, spatial and non-spatial memory, and inhibitory control in two groups of free-range broiler chickens: those who frequently explore the range ('high rangers, HR') and those who prefer to stay in or near the barn ('low rangers, LR'). Experiments were conducted on two flocks of broiler chickens (n=200 per flock) reared in similar conditions. To test spatial and non-spatial memory, an arena with 8 cups was used. During the training phase, individuals (n<sub>LR</sub> = 20, n<sub>HR</sub> = 21) learned that only a white cup among seven black cups was baited. To find the target cup, individuals could rely either on their spatial memory (the position of the cup in relation to the cues on the arena's walls), or rely on their non-spatial memory (the color of the cup). Two probe tests, on two different days, for spatial memory (all cups are black) and non-spatial memory (the white cup changed its previous position) revealed that LR were quicker to visit the target cup during the spatial memory compared to HR (LR: 60.47 ± 44.58 s, HR: 80.60 ± 47.15 s; ranging level: t = - 2.21, p = 0.03; observation day: t = 0.36, p = 0.71). No differences between groups were found for non-spatial memory (Day 1: U = 195, p = 0.70, Day 2: U = 199, p = 0.77). To test inhibitory control, individuals (n<sub>LR</sub> = 7, n<sub>HR</sub> = 8) learned to detour either side of an opaque cylinder to gain a food reward. In the test condition, chickens were presented with a transparent cylinder in ten trials. As a sign of inhibition, subjects needed to refrain from trying to reach the food reward through the walls of the cylinder and detour to its open sides. LR were significantly more successful than HR (LR: 40 ± 12.90%, HR: 23.75 ± 9.16%, p = 0.028). Overall, our results show that LR chickens perform better in cognitive tasks compared to HR chickens. These results contribute to the growing body of research into the behavior and cognition of free-range chickens. This knowledge is essential for, ultimately, applying husbandry procedures that increase range use for all animals.

### Maternal behaviour of buffaloes and effect on calves' vitality

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Although buffalo dairy farming represents a small niche in animal production, it is an important resource in some countries, but little information is available about the behaviour and welfare of this species. One of the critical moments in buffalo dairy farming for both the dam and the calf is peripartum, therefore this study aimed to investigate the dam's behaviour during pre- and post-partum and its relationship with calves' behaviour and vitality. Thirty primiparous Mediterranean buffalo (18 months) were monitored. Animals were kept in an experimental farm in group housing and 15 days before calving were moved in individual boxes with social and visual contact allowed. Behaviour has been recorded in the following intervals: from 72h to 48h before calving, 12h before calving. Behaviour of calves and dams has been recorded for 48h after calving. Data have been analysed with parametric and non-parametric methods after controlling their distribution. Calves were clinically evaluated using the APGAR score and the transrectal temperature at hours 0 (calving), 1, 12, 24, and 72 after calving. Weight was measured at calving, at day 7, 14, 21, and 28. Preliminary results indicated that dams spent more time standing, and transitioning from Standing to Lying in the last two hours before calving ( $p < 0.01$ ) and that could indicate a restless attitude. The APGAR score was lower at hour 0 and hour 1 compared with hours 12, 24, and 72 after calving, but in all the cases was within the normal range. The duration of calving did not affect the behaviour of the calves. A negative correlation between calves' temperature at T0 and the latency time to stand-up ( $r = -0.53$ ;  $p < 0.05$ ) and a positive correlation between the temperature at 12 hours and the number of suckling in the first 48 hours ( $r = 0.59$ ;  $p < 0.05$ ) were found. The maternal grooming time was correlated positively with the standing time ( $r = 0.5$ ;  $p < 0.05$ ) and suckling time of the calf ( $r = 0.67$ ;  $p < 0.001$ ). Dams that displayed negative maternal behaviours (circling, pawing, moving forward) were correlated with lighter calves at d 21 ( $r = 0.51$ ;  $p < 0.05$ ). So, calves receiving more attention from the dams spent more time standing in the 48h while calves that received less attention were lighter at d 21. These preliminary results suggested that stimulation of the calf from the dam is relevant for his vitality and future growth. Furthermore, the temperature at birth and at 12 hours seems to be a good indicator of vitality in buffalo calves.

## Dairy cow motivation for access to open lying space

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With dairy herds being housed for longer periods throughout the year, it is important to ensure cow lying comfort, a behaviour contributing to their health and welfare, is not compromised when housed. A previous study, conducted by the authors, identified space and surface type as two qualities of a lying area that appear to be important to cows. A trade-off study found that cows value lying space over surface. The current study measures the motivation of cow preference for open lying space, using walking distance as a motivation indicator, for two different lying surfaces. Twenty five Holstein-Friesian dairy cows were allocated to one of five experimental periods and housed in a robotic milking unit, given free access to a milking robot, ad libitum feed and water, and access to six mattress cubicles bedded with sawdust. After a 3 day familiarization period in the trial area, cows were given access to a one way indoor raceway at a short distance (S; ~30m), leading to an open lying area (9m x 5m) of either deep-bedded straw or a mattress bedded with sawdust. Cows were trained to use the raceway to access the open lying area for 3 days, followed by 3 days of choice. The raceway distance was changed to a medium distance (M; ~67m) and cows given 1 day training followed by 3 days of choice. The raceway distance was changed once more to a long distance (L; ~112m) and cows given another 1 day training followed by 3 days of choice. The raceway was changed back to S distance and the above protocol repeated for the second open lying surface. Order of surface type was alternated for each experimental period. This allowed cow motivation for open lying space to be measured in terms of walking distance for two different surface types, mattress and straw, when given free access to mattress cubicles. Linear mixed effects modelling was used to assess the significance of raceway distance and open lying surface type in explaining variations in lying time on the open lying area. Cows spent more time lying down when the raceway was S distance (11.8hr/d;  $p < 0.001$ ) and M distance (10.1hr/d;  $p < 0.001$ ) in contrast to L (7.7hr/d). Open lying surface type had an effect on lying time, with cows lying down longer on the straw yard (10.0hr/d) as opposed to a large open mattress (9.1hr/d;  $p < 0.001$ ). Cows are still motivated at the L distance to lie down on the open lying surfaces for around one third of their day, highlighting the importance of an open lying space to cows.

## **Progesterone, physiological reproductive status, stress response and reactivity in female ruminants**

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The concentrations of sexual steroids do modify the sensitivity to stressors in lab mammals: progesterone (P4) reduces the stress response, whilst P4 withdrawal and/or oestrogens increase it. As the concentration of P4 and oestrogens differs according to the physiological status (pregnant, anestrus or cycling females), it may modify the sensitivity to stressors. Therefore, this information may be useful in relation to farmed cattle and sheep, when subjected to stressful situations (eg. artificial weaning, shearing, social isolation). This would also be considered in selecting the appropriate calm animal by way of temperament assessment tests. A series of studies was performed to determine if P4 concentration and the physiological reproductive status of female ruminants affect their stress response or reactivity. After shearing, the increase in cortisol concentration is smaller in pregnant ewes than in non-pregnant ewes. The former was also observed more frequently standing and grazing but less frequently walking than non-pregnant ewes after shearing. Additionally, on the day of weaning, ewes that received an intravaginal P4 releasing device (CIDR) for 32 days previous to artificial weaning paced and vocalized less than untreated controls. Moreover, four days after artificial weaning, globulin concentration was greater in treated ewes than in controls. Contrastingly, anoestrous ewes treated with a CIDR during a 13 day trial period, responded with higher cortisol concentration, if they were socially isolated 24 h after the device was removed, than untreated ewes. Although P4 concentration affects the stress response, the phase of ewes' oestrous cycle had minor effects. In effect, oestrous ewes only tended to vocalize and remain immobile more times during social isolation than dioestrous ewes. Besides, physiological reproductive status and P4 concentration affected the response to temperament assessment tests in female beef cattle. Heifers' flight distance was shorter by one day after inserting a CIDR, than in controls. The exit velocity was greater in previously treated than control heifers 16 h after removing the CIDRs. The exit velocity was slower and the flight distance shorter in pregnant cows than in dioestrous cows. In conclusion, both P4 concentration and gestation influence the response to stressors and to human handling. Furthermore, P4 withdrawal increases the sensitivity to social isolation in ewes as well as, the oestrous to a small degree.

**Lambing season affects ewe milk production, lamb growth and its behavioural response to weaning**

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Out-of-season lambing systems require an integral view of lambs' welfare and production, including lambs growth and their responses to weaning. Therefore, the ewes' milk production, and lambs' weight and behaviours at weaning were compared in single lambs born from multiparous Corriedale ewes that lambed in spring (natural season: September in south hemisphere, n=20) or in autumn (April, n=20). The milk yield measured 77 days after lambing was greater in spring than in autumn lambing ewes ( $552 \pm 116$  g/d vs  $293 \pm 114$  g/d,  $p < 0.001$ ). At lambing, lamb weight was similar between groups (spring:  $4.5 \pm 1.0$  kg vs autumn:  $4.7 \pm 1.0$  kg); but the average daily gain until weaning (80 days of age) was greater in spring-born lambs than in autumn-born lambs ( $200 \pm 40$  g/d vs  $130 \pm 50$  g/d,  $p < 0.001$ ). The day before weaning, the number of suckling events recorded in two 3-h sessions was similar between groups (spring:  $2.9 \pm 1.3$  vs autumn:  $3.4 \pm 2.1$ ). The duration of each suckling event was shorter in spring-born lambs than in autumn-born lambs ( $11.0 \pm 2.9$  s vs  $12.4 \pm 4.1$  s,  $p = 0.03$ ). The ewe-lamb distance was estimated the same day using lambs' body lengths ( $BL \leq 1$  or  $BL \geq 3$ ) every 10 min during the same periods. Spring-born lambs were more frequently closer to their mother than autumn-born lambs ( $BL \leq 1$ :  $54.1 \pm 15.1\%$  vs  $38.7 \pm 16.5\%$ ,  $p = 0.01$ ). The frequency at which each lamb was observed standing and grazing were recorded the day before weaning and the day of weaning using 10 min scan sampling. The standing posture did not differ between groups the day before weaning (spring:  $66.4 \pm 14.7\%$  vs autumn  $68.1 \pm 8.7\%$ ). However, the day of weaning, spring-born lambs increased significantly more their frequency standing than autumn-born lambs compared with the previous day ( $132.9 \pm 27.4\%$  vs  $105.9 \pm 22.4\%$ ,  $p < 0.001$ ). Spring-born lambs grazed more frequently the day before weaning than autumn-born lambs ( $55.8 \pm 18.0\%$  vs  $45.5 \pm 17.4\%$ ,  $P = 0.002$ ). However while spring-born lambs decreased their grazing frequency compared with the previous day, autumn-born lambs increased it ( $71.9 \pm 64.4\%$  vs  $114.1 \pm 73.1\%$ ,  $p = 0.002$ ). Probably due to differences in pasture quantity and quality, ewes that lambed in spring produced more milk and thus, lambs gain more weight and remained with a stronger bond with their mother before weaning. In consequence, they responded more intensively to weaning, affecting more their time allocated to graze.

**Behavior by individual sows at the entrance of an electronic sow feeder (ESF) is impacted by feed order**

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Electronic sow feeding (ESF) has emerged as a common feeding system for gestating sows, but can be a focal point for competition over a valued resource, feed. To better understand the social challenges in this system, we quantified social interactions at the ESF entrance and explored potential influences on these behaviors. Forty-eight study animals were selected from a larger group of 60 gilts and first parity sows housed in a single pen and fed via ESF. Over a 4-month period once every 4 weeks, a 5-day average of feed order was determined and used to select a 12-animal cohort comprised of 4 head from the top, middle, and bottom of the feed order. Behaviors at the ESF entrance were recorded by a video camera, and behavioral sampling was triggered by entry of a study animal into the station. The recording was stopped and reversed to a minute before the entry and then behavior was coded as: No agonistic interactions (enter without contact), initiated agonistic interactions (displace, knock, push, a single bite, repeated bites, lunge) and elicited retaliation (nose wrestling, mutual fight). The relationship between feed order and behavior at the feeder entrance was evaluated using linear mixed models. The majority of feeder entrances (73.44%) occurred without agonistic interactions whereas those involving agonistic behaviors initiated by the sow entering the feeder totalled 24.91%. The later an animal first entered the feeder to eat, the more likely she was to enter the feeder without agonistic interactions ( $p < 0.001$ ). Conversely, the earlier a sow fed, the more likely she was to initiate an agonistic interaction ( $p < 0.002$ ). Specific agonistic behaviors initiated by sows entering the feeder that were impacted by feeding order were displacement ( $p < 0.004$ ) and push ( $p < 0.009$ ). While nearly  $\frac{3}{4}$ 's of the feeder entries occurs without agonistic interactions, these findings suggest that animals of different social status utilize different behavioral patterns when entering the feeder. It appears animals that first eat earlier in the day initiate more agonistic behaviors while waiting to enter the feeder, whereas animals that first eat later in the day find ways to enter the feeder without initiating aggressive behaviors.



## Effects of early familiarization of cattle with feedlot diet and social group on feed bunk attendance and postural behavior during the finishing period in feedlot

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The first days in feedlots are challenging for cattle, and this is triggered mainly by the sudden changes in their social group and diet, which are potentially harmful for animal performance and welfare. The aim of this study was to assess the effect of early familiarization of cattle to feedlot diet and social group on feed bunk attendance and postural behavior. Nine hundred Nellore bulls ( $420 \pm 30$  kg of live weight and  $\sim 30$  months old), were divided into six groups (150 animals each). Twenty-one days before entering the feedlot (D-21), the familiarized groups (FA,  $n=3$ ) were formed and housed separately in pastures of 30 ha each, where they have free access to grass (*Brachiaria brizantha*) and water, and received 3 kg of concentrate/animal/day, in a feed trough. In the first day in the feedlot pens (D0), three other groups (non-familiarized, NFA) were formed, and all of them (FA and NFA groups) were housed in the feedlot pens (13m<sup>2</sup>/animal and 33 cm of linear space in the feed bunk). Cattle behavior were recorded using scan sampling and instantaneous records (with a 15-minute sampling interval) methods, quantifying the percentages of animals standing, lying down, and attending the feed bunk. These measurements were recorded every two days for the first 15 days (adaptation phase) and then weekly until the end of the feedlot period (post-adaptation phase). Data analyses were carried out using mixed linear models with repeated measurements (PROC MIXED from SAS) considering the fixed effect of treatment (FA and NFA) and days in the feedlot. Means were compared using the Tukey test. Significant differences between FA and NFA were found during adaptation and post-adaptation phases, with FA showing higher percentage animals lying down (23.72%) and attending the feed bunk (19.27%) than NFA (19.97 and 15.99%, respectively) and lower percentage of animals standing (55.99 and 63.03%, for FA and NFA respectively,  $p<0.05$ ). During the post-adaptation phase, FA groups continued showing higher ( $p<0.05$ ) percentages of animals attending the feed bunk than NFA (14.20 and 9.47%, respectively), but lower ( $p<0.05$ ) percentage of animals lying down (25.38 and 30.80%, respectively). We conclude that previous familiarization with the feedlot diet and social group facilitates the adaptation of cattle to the feedlot environment.

## Does ponceau red artificial dye influence dog behavior?

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Results from previous studies suggested that artificial food dyes cause behavioral changes in rats and children. Would this be also the case with dogs? Aiming to answer this question, we set up an experiment to evaluate if the consumption of pet food with the artificial dye ponceau red 4R (E124) promotes changes in dogs' behavior. The study was approved by the Ethics Committee on the Use of Animals of UNESP/FCAV, Brazil, protocol #007955/18. The experiment was carried out at the Research Laboratory in Nutrition and Nutritional Diseases of Dogs and Cats Professor Dr. Flávio Prada (FCAV-UNESP, Jaboticabal, São Paulo, Brazil), with 12 beagles from three different litters (6 castrated females and 6 whole males), they were around three years old and weighed on average, 12.29 kg. Three treatments were tested (n=4), all presenting the same nutritional formulation, differing only in the dosage of artificial dye E124: C= control, zero dye; T1 = 0.05% of dye and T2 = 0.2% of dye. In the adaptation period, the dogs consumed the control diet for 17 days and the test diets for 21 days. After a month break, the experiment was repeated. During the 21 days the dogs were filmed from 10 am to 5 pm and was registered the frequency of agonistic behaviors (when a dog faces other, threatening it, or barks, bristle hair, or show the teeth, or invest, or fight), and anxious behaviors (pacing and/or hit the front paws on the wall and grid twice in a row or more, or tap dance in place). Linear mixed models were used to evaluate if the percentage of dye on food impacted dogs' behaviors, using treatment, sex and their interaction as fixed effects. We observed a significant effect of treatment on anxious behaviors ( $F=18.56$   $P=0.002$ ), with T1 presenting higher frequency ( $2.69 \pm 3.0$ ) when compared to C ( $1.1 \pm 1.5$ ) and T2 ( $0.30 \pm 0.5$ ), which did not differ from each other. There was a tendency effect ( $P=0.067$ ) of treatment and sex interaction on agonistic behaviors, with males from treatments T2 ( $1.82 \pm 1.2$ ) and C ( $1.10 \pm 1.5$ ) presenting significantly higher frequencies than females from T2 ( $0.40 \pm 0.5$ ). It was also observed in males that the T2 and T1 ( $0.70 \pm 0.7$ ) were statistically different, but C did not differ from T2 and T1. Our results suggest that dog diet with artificial dye E124 negatively affects dogs' behavior.



**Personality in Brazilian show jumping horses (*Equus caballus*): a methodological comparison study**

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Studying individual differences in behavioral profile, or personality, in animals can help to promote a safe handling environment, increase animals' welfare and improve trait selection. However, there are various methods to access personality in horses (*Equus caballus*). We evaluated the reliability of three different methodologies: questionnaire (with 24 adjectives), experimental tests (object neophobia, reactivity to humans, and gregariousness test) and behavioral observations, using a sample of 40 jumping horses (5 stallions, 27 geldings and 6 females, all adults). Inter-Correlation Class analyses were run to check for questionnaire's interobserver reliability and tests' temporal consistency. To define independent axes of individual differences Principal Component Analysis were run using adjectives, behavioral and tests variables separately. To verify if methods agree we run Spearman's Correlation analyses. Of questionnaire's adjectives only 9 had interobserver agreement, and grouped into 2 factors named Sociability and Novelty seeking, together explaining approximately 75% of total variation. Of the 35 variables extracted from experimental tests, 17 showed temporal stability, grouped into 6 factors explaining 74.84% of total variation, named: Fearfulness, Locomotor Activity, Sociability with humans, Neophobia, Aggressiveness towards humans and Feeding motivation. Behavioral observations also grouped in six classes (74%): Locomotor, Feeding Anxiety, Social Anxiety, Threatening, Sensitiveness and Self-Groom. The questionnaire's novelty seeking had a positive correlation with test's locomotor activity and threatening stereotypies. Three positive correlations were found between tests and behaviors (fearfulness and feeding anxiety; neophobia and self-groom; feeding motivation and feeding anxiety) and one negative correlation was found (fearfulness and self-groom). Nevertheless, one correlation was found between factors derived by questionnaire to the factors inferred by tests. The absence of more correlations suggests that questionnaires should not be used as the single methodology and that using more than one method might offer more complete overview of horses' personality.

**Evaluating self-isolation in group-housed dairy calves following disbudding through provision of a shelter**

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Evidence across species points to changes in social behavior as a potential indicator of individual welfare. Our objective was to evaluate the effects of hot iron disbudding, a common painful procedure, on self-isolation behaviors of dairy calves provided a shelter for visual seclusion. We hypothesized that calves may increase use of the shelter following disbudding. Holstein bull and heifer calves ( $n = 24$ ; 4 calves/pen) were group-housed at  $16 \pm 2$  d of age and fed milk replacer via an automated milk feeder. All pens contained an open-top 3-sided shelter (1.2 x 1.2 m), built of corrugated plastic allowing for visual seclusion from the rest of the pen. Within pen, calves were randomly assigned to receive either disbudding (DB;  $n = 12$ ) or handling only (CON;  $n = 12$ ) at week 5 of age. Calves received both local anesthetic and analgesic prior to disbudding. Behavior was recorded continuously from video for 72 h following treatment to characterize time spent in the shelter (defined as  $\geq 50\%$  of the calf's body inside), social use of the shelter, and lying time. Data were analyzed in a repeated measures general linear mixed model with fixed effects of treatment, time, and their interaction. Calves used the shelter more after disbudding (339.82 vs 194.81 min/d; DB vs CON; SE = 98.1;  $P = 0.03$ ). During daylight hours (0700 to 2000 h), disbudded calves used the shelter more (13.6 vs. 6.0 min/h; DB vs. CON; SE = 2.6;  $P < 0.001$ ) whereas shelter use did not differ at night (11.5 vs. 10.6 min/h; DB vs. CON; SE = 4.4;  $P = 0.48$ ). Social use as a percentage of total shelter use did not differ between treatments ( $P = 0.86$ ). The proportion of social use of the shelter with a calf on the same treatment differed; disbudded calves spent more time in the shelter together compared to pairs of control calves, on d 1 (46.44 vs 14.00 %; DB vs CON;  $P = 0.009$ ) and d 2 (39.56 vs 18.71 %; DB vs CON;  $P = 0.04$ ) after treatment. Calves spent less time lying following disbudding (17.0 vs 17.6 hours/d; DB vs CON; SE = 21.94;  $P = 0.038$ ), and a greater percentage of their lying was located in the shelter (25.1 vs 12.8 %; DB vs. CON; SE = 8.1%;  $P = 0.05$ ). Calves made greater use of the shelter after disbudding, but social use of the shelter suggests that social withdrawal may not be the only cause of increased use. These results suggest that calves make use of resources that provide visual seclusion from the rest of the pen and the choice to make use of this feature may improve welfare.

## The impact of providing periodic exercise on behaviour of stall-housed gestating sows

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The Canadian Code of Practice for the Care and Handling of Pigs permits the operation of existing stall barns after July 2024 if bred sows are given opportunities for a greater freedom of movement. The objective of this study was to determine the effect of providing periodic exercise to gestating stall-housed sows on sow behaviour, in comparison to stall-housed sows receiving no exercise, and group-housed sows. Sows were randomly assigned to one of three gestation treatments (n=60 per group): stall-housed (Control: C), stall-housed and exercised (Exercise: E – 10 min of walking once per week), and group-housed (Group: G). Sow behaviour was recorded in early (d 7), mid (d 57) and late (d 107) gestation, on the days of exercise. Sow postures (lying, standing, sitting) were recorded every 10 minutes (scan sampling) for 2 hrs before exercise (AM) and for 1.5 hrs post exercise (PM). Sows were live scored for stereotypies at 2-minute intervals for 1 hr in AM and PM. Data were analyzed by PROC MIXED (SAS 9.4) to determine the effects of treatment and stage of gestation on sow postures and stereotypies in the separate models for AM and PM. In AM, G sows spent more time lying down and sat less, than C and E sows, which did not differ (Lying: C: 0.44±0.03, E: 0.49±0.03, G: 0.59±0.03, mean proportion of time±SEM, P<0.05; Sitting: C: 0.14±0.02, E: 0.16±0.02, G: 0.10±0.02, P<0.05). In late gestation C sows stood more than G sows, with E sows being intermediate (Early: C: 0.46±0.04, E: 0.54±0.04, G: 0.41±0.04; Mid: C: 0.43±0.04, E: 0.30±0.04, G: 0.37±0.04; Late: C: 0.28±0.05, E: 0.23±0.04, G: 0.12±0.05, P<0.05). In PM, E sows sat more than C and G sows, and C sows sat more, than G sows (C: 0.08±0.01, E: 0.12±0.01, G: 0.04±0.01, P<0.05). Stereotypies: in early gestation, G sows performed fewer stereotypies than E sows, with C sows being intermediate. In mid gestation, G sows performed fewer stereotypies than C and E sows, which did not differ (Early: C: 0.56±0.04, E: 0.55±0.04, G: 0.44±0.04; Mid: C: 0.76±0.04, E: 0.73±0.04, G: 0.50±0.04, P<0.05). Results suggest that group housing provides more comfort (as indicated by sow postures) and helps to reduce the stress more efficiently (as indicated by performance of stereotypies) than housing in stalls, and that limited periodic exercise is not sufficient to improve sow comfort and to reduce stress levels.

**Assessing owner's ability to detect fear and aggression-related behaviours in the domestic cat.**

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Research on companion animal behaviour and welfare often relies on owner reports, since owner reports provide a holistic assessment of the animal's responses to a diverse range of scenarios. However, this approach assumes that owners are able to accurately recognize and/or interpret the behaviours on which these reports are based. We assessed whether cat owners are able to accurately identify various behaviours that can be used to differentiate whether cats are relaxed, fearful or aggressive using an online survey that asked owners to report on which behaviours they believed were present or absent in a series of cat videos. Mixed logistic regression models, with a random intercept for participant, were used to estimate owners' sensitivity (ability to correctly identify the presence) and specificity (ability to correctly identify the absence) for identifying each behaviour while examining the effects of owner demographic characteristics and cat experience on their identification skills. Owners (N=2,653) could accurately identify (sensitivity and specificity  $\geq 0.75$ ): dilated pupils, ears positioned back, a neutral and crouched abdomen, lowered/tucked tail, rapid tail movements, swatting, avoiding, hissing and growling. Owners had low success at identifying the presence of many other important behaviours such as a lowered head, neutral and constricted pupils, sideways ears, arched back, upright tail, lip licking, freezing and yowling. Comparatively, owners did better at identifying the absences of behaviours, only reporting the absence poorly for an upright head, neutral pupils, and neutral back. Owner reports of having advanced knowledge in cat behaviour consistently improved the ability of owners to detect the presence and absence of various behaviours, while other factors such as previous cat ownership, cat-related work experience, and demographic variables differentially affected identification abilities for a number of behaviours. Understanding which fear and aggression-related behaviours owners can identify can be used to develop training tools to teach owners what these behaviours mean, giving owners the necessary materials to accurately rate fear and aggression in their companion cats. Overall, this information can improve researchers' interpretations of owner reports on cat behaviour, and can help owners better understand their own cats' behaviours, improving the human-animal bond and the welfare of feline companions.

## Effects of milk- and starter-feeding methods on the performance and behavior of individually and pair-housed calves in outdoor hutches

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For dairy producers, housing infrastructure and undesirable social interactions such as cross sucking are potential barriers to adopting social housing. For farms already housing calves in outdoor hutches, an option is pair raising using adjacent outdoor hutches with a shared fence. Our objective was to investigate milk- or starter-feeding strategies to mitigate undesirable calf behaviors in this pair-housing system. Eighty Holstein heifers were individually (n=16 calves) or pair housed (n=32 pairs). Pairs were divided into 4 feeding treatments (n=8 pairs each) in a 2×2 factorial design: milk fed in an open bucket or slow-flow teat bucket and starter fed in an open bucket or open bucket plus a specialized teat bottle. Body weights (BW) and starter intakes (adjusted for dry matter %) were measured weekly and averaged within pairs for pair-housed calves. Calves were observed during and immediately after the afternoon milk feeding 2×/wk for 30 min using continuous video recording. Linear mixed models were used to evaluate housing differences in performance and feeding-treatment differences in behavior. Overall from wk 2-9 of age, paired calves had higher starter dry matter intake (DMI) than those individually housed ( $0.76 \pm 0.04$  vs.  $0.61 \pm 0.05$  kg/d, mean±SE;  $P=0.04$ ), driven by differences during and in the week following weaning ( $P=0.02$ ). Paired calves tended to have a higher average daily gain (ADG) of BW during weaning ( $0.81 \pm 0.06$  vs.  $0.66 \pm 0.08$  kg/d,  $P=0.10$ ), but there were no housing differences in overall or pre-weaning ADG. Before and during weaning, when paired calves were fed milk in an open bucket but provided starter in a Braden bottle in addition to a bucket, cross sucking was reduced compared to pairs with only open buckets (pre-weaning:  $1.5 \pm 0.3$  vs.  $2.9 \pm 0.3$  min,  $P=0.003$ ; weaning:  $1.8 \pm 0.4$  vs.  $3.9 \pm 0.4$  min,  $P=0.009$ ). Regardless of starter-feeding method, paired calves who had slow-flow teat buckets rather than open buckets for milk spent significantly less time cross sucking before and during weaning (pre-weaning difference:  $\geq 1.1$  min, SE=0.2 min,  $P < 0.02$ ; weaning difference:  $\geq 2.7$  min, SE=0.3 min,  $P < 0.003$ ). In conclusion, paired calves performed better than those housed individually during and immediately following weaning. Providing milk through slow-flow teat buckets had the greatest effect on reducing cross sucking, and providing starter through a specialized teat bottle is another novel option for reducing this unwanted behavior.

**Exploring the Relationship Between Daily Visitor Attendance and Aggression Displayed by Three Zoo Primate Species**

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Aggressive interaction is a fundamental property of nonhuman primate social behavior in wild and professionally managed populations. The form, severity, and direction of aggression can vary depending on context and species. In zoo populations, high rates and severe aggression can cause concern for animal health and welfare. In the present study, we conducted a retrospective analysis to investigate how and if attendance was related to the occurrence of intraspecific and/or human-directed aggression in Geoffroy's spider monkeys (*Ateles geoffroyi*, N=8), white-cheeked gibbons (*Nomascus leucogenys*, N=5), and Western lowland gorillas (*Gorilla gorilla*, N=8) over the course of five years. Utilizing historic animal care logs and zoo attendance records, binomial logistic regressions revealed a single significant relationship. Specifically, daily visitor attendance was related to the presence of human-directed aggression in the gorillas ( $p < 0.05$ ) but not the spider monkeys ( $p = 0.80$ ) or the gibbons ( $p = 0.80$ ). Further, daily visitor attendance was not related to the presence of aggression directed toward conspecifics (spider monkeys,  $p = 0.51$ ; gibbons,  $p = 0.59$ ; gorillas,  $p = 0.67$ ) or all aggressive interactions combined (spider monkeys,  $p = 0.52$ ; gibbons,  $p = 0.73$ ; gorillas,  $p = 0.07$ ). Since the exhibits were relatively large and provided opportunity for the primates to be out of view and/or create a substantial distance between themselves and guests, future research which examines space use could provide deeper insight by revealing if the primates were adjusting their location in accordance with visitor numbers. Furthermore, research which incorporates gorilla personality measures to determine if certain characteristics play a role in addition to attendance on rates of human-directed aggression would be of great interest.

**Human-animal interactions and their effects on the welfare of farm animals**

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Interactions between stockpeople and farm animals have received little attention from the public, despite evidence that these interactions have great impact on the welfare of farm animals. The relationship between humans and animals has been developing since the Neolithic period, with many animal species being domesticated. Despite long-term selection for many characteristics, exposure to humans and sudden changes in their social and physical environments remain challenging events for many domesticated animals. Although farm animals' fear thresholds have been reduced by domestication, fear responses to humans have not been eliminated. It is well documented that fear of humans reduces farm animal productivity, including reductions in fertility, milk production and growth rates. Furthermore, stressful situations increase cortisol concentrations in animals and reduce their welfare. As some routine tasks on farm are considered aversive but are difficult or impossible to replace (e.g., vaccinations, transport, etc.), they should be done in the best possible and least aversive ways, minimizing unnecessary animal suffering. It is noteworthy that, in most cases, the inappropriate or misguided opinions, customs, and behaviors of some stockpeople are not due to intentional cruelty, but rather to a lack of knowledge. Although implications of stockpeople behavior on farm animal welfare and productivity have been well characterized, it has not received extensive recognition. On many farms, turnover rates for stockpeople are high and there is limited investment in training. This leads to a problematic, self-perpetuating cycle, in which employees who are not valued and not adequately trained have short employment intervals, which “justifies” to the producers their non-investment in employee training. Studies in dairy and beef cattle, pig, and broiler production systems demonstrate the high potential of training, involving behavioral and cognitive techniques, designed specifically to improve stockpeople attitudes and behavior toward animals. This type of intervention improves their handling skills and, consequently, decreases animals' fear toward humans. Improving animal handling yields benefits for both animals and workers, improving welfare and productivity for both. Additionally, improvements in the human-animal relationship leads to an increase in stockpeople motivation and, therefore, enhances the performance of their work. In conclusion, specialized training programs targeting changes in negative attitudes and behaviors towards animals offer an excellent opportunity to improve human-animal interactions in livestock industries, with numerous benefits.



**Freedom to move versus piglet crushing: citizens' attitudes towards farrowing housing systems.**

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Most sows in intensive production systems are housed in farrowing crates, which protect the piglets from being crushed by the sow. However, crates also cause extreme restriction to sows' freedom of movement and prevent them to perform many natural behaviors. This study aimed to investigate, firstly, citizens' attitudes towards farrowing housing systems (farrowing crates, loose pens and outdoor farrowing) and, secondly, the influence of information about piglets' death by crushing on the acceptability of loose pens system. Brazilian citizens were recruited via social media to participate in an online questionnaire with closed and open questions. The questionnaire started with a text explaining the 3 systems with a short video exemplifying each, after which participants were equally and randomly assigned to answer about one of the systems. All participants were asked if they supported a move from crates to farrowing pens. Demographics of participants (n=1,172) approximately corresponded to the Brazilian population in age, gender, and region of residence. Attitudes (on a 5-point Likert scale; 1 = totally negative, 5 = totally positive) were lowest towards farrowing crates (1.7±0.08), followed by loose pens (3.1±0.06) and outdoor farrowing (4.5±0.06) (ANOVA, F= 597.9, P<0.0001). Participants' attitudes were positively correlated with their perception of quality of life of sows (Spearman, r=0.78, p<0.0001). Ninety percent of the participants that were provided no information about piglet crushing supported a move from crates to loose pens, compared to 84% of those informed that piglet death by crushing would increase 2.5-5% (Chi=7.1, d.f.=1; p=0.01). Qualitative analysis showed overwhelming objection to restriction of animals' freedom to move and perform natural behaviors. A common voice was that piglets' death by crushing does not justify housing sows in farrowing crates, and that farmers, industry and scientists should develop solutions to prevent piglets' crushing while providing animals freedom to move. Many participants considered that the loose farrowing pens fail to provide for the animals' needs. Changing the housing system for lactating sows is vital to maintain the sustainability of the industry and will require substantial financial investments. Before moving to any alternative system, it is important to investigate if it resonates with public expectations.



## **Application of different loading densities during transport to the slaughter plant and their effects on pigs' behavioral response**

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Transportation has been defined as one of the most stressful events for animals and, among others, factors, such as loading density in the truck has an impact on the welfare of pigs. The objective of this study was to evaluate the effect of three loading densities during transportation from the farm to the slaughter plant on pigs' postural changes in the truck. A total of 288 pigs originating from two similar commercial farms were transported to a commercial slaughter plant, all located in Southern Brazil, through 8 trips ( $8,4 \pm 0,5$  hours) (or replicates) using the same truck type and driver. For each trip, 36 barrows ( $120 \pm 10$  Kg) were randomly distributed into three groups and transported in three different compartments located in the middle deck of the truck at the three loading densities, i.e.,  $200 \text{ kg/m}^2$  (D1),  $235 \text{ kg/m}^2$  (D2) and  $270 \text{ kg/m}^2$  (D3), in terms of one compartment/loading density. During transport, behavioral recordings were made through a video-camera installed in each compartment. Pig behavior was observed from videos using instantaneous recording (with a 5-minute fixed sampling interval) and scan sampling route. The number of animals in standing (ST), sitting (SI), lying (LY) and other postures (OP) were registered. For all variables, generalized mixed models were adjusted using PROC GLIMMIX of SAS. During transport, the proportion of LY pigs was higher ( $P < 0.05$ ) at D1 than at D3 ( $77.2 \pm 20.8$  vs  $63.0 \pm 26.0$ ), while the proportion of SI pigs was higher ( $P < 0.01$ ) at D3 compared with D1 ( $28.9 \pm 19.5$  vs.  $13.54 \pm 11.9$ ). No difference in ST or OP was found between loading densities in this study ( $P > 0.05$ ). We conclude that during transport under tropical ambient conditions the application of lower loading densities in the truck allows pigs to have sufficient space to rest and travel more comfortably.

**The importance of behavioural observations in assessing the welfare of animals under human care: an example with elephants.**

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Systematic and regular assessment of the welfare of wild animals under human care is a central component of compassionate conservation. Observing behaviour provides information about animals' needs, preferences and affective states. The purpose of our research was to objectively assess the welfare of 3 adult female elephants (2 *Loxodonta africana*, 1 *Elephas maximus*) housed in an Argentinian zoo, using a two-step protocol previously validated by our team. We visited the zoo once a week, from 9 am to 7 pm, for 1 month. During the first visit, we collected baseline data on the elephants' life histories through caregiver interviews and document review. Also, we filled the first step form, which included the assessment of resource-based indicators (19 environment & 3 management) and animal-based indicators (13 health & 9 behaviour). The subsequent visits we filled the second step form (follow-up), which included only animal-based indicators. To assess health and resource-based indicators we performed visual inspections following protocol guidelines. Regarding behavioural assessment, protocol included carrying out nude-eye observations 3 times a day, by instantaneous and individual sampling (20-min. blocks & 1-min. intervals), using worksheets to record functional categories and space usage. We also rated response to environmental enrichment and stereotypic behaviours, among other indicators. For both forms, we rated all indicators on a 3-point scale (A- normal/no observable risk; B- mild deviation/risk; C- severe deviation/risk). Total direct observation time was 20 h for each individual. For the three elephants, resource and management based indicators were rated 36% A; 9% B and 55% C. Health indicators were rated: elephant a) 100% A; elephant b) 92% A; 8% B; elephant c) 88% A; 12% B. Behavioural indicators were rated: elephant a) 52% A; 33% B; 15% C; elephant b) 50% A; 20% B; 30% C; elephant c) 51% A; 23% B; 26% C. These results suggest that environment and management were not adequate to promote good elephant welfare. However, health indicators did not show major deviations, but behavioural indicators did. This is consistent with other studies which propose that careful observation and interpretation of behaviour can be used to gain a good overall picture of animal welfare, and should always be included in assessment protocols.