Standard Operating Procedure

Novel Surface Test - Group

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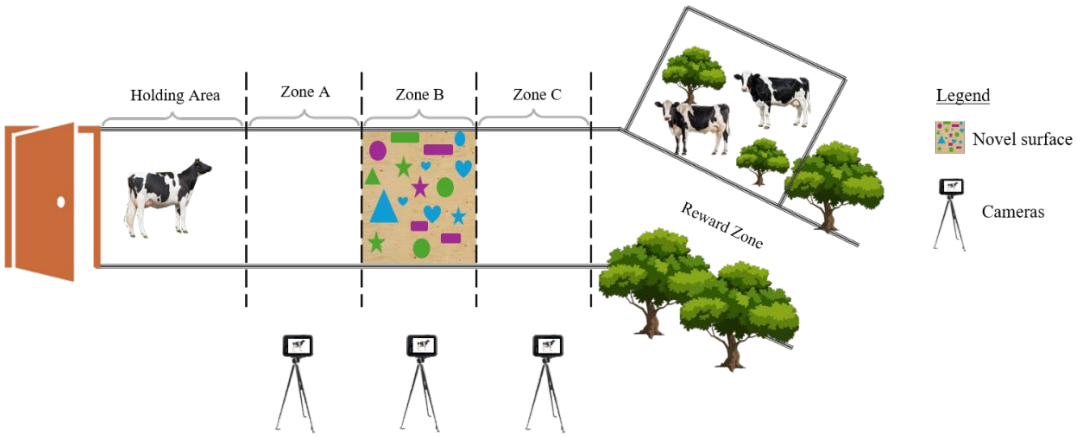
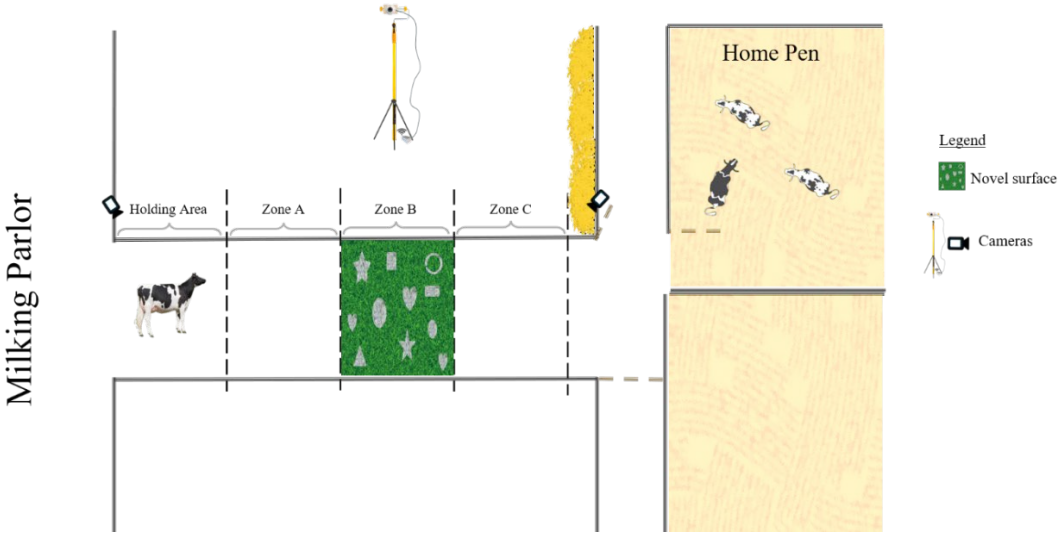
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Description/objective

The Novel Surface Test (NST), also known as the Risk-Taking Test, evaluates cows' willingness to cross an unfamiliar surface placed in their normal walking path. The test measures individual and group-level differences in approach-avoidance, risk-taking tendencies under mild uncertainty, and leadership. This test can be performed in different environments and is designed to test single or multiple animals in one testing session. *The test procedure described in this SOP is performed in two different types of corridors and is designed to test two or more animals.*

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Terminology

Corridor types:	
Outdoor corridor:	The novel surface was installed in an outdoor corridor routinely used by the animal (e.g., a corridor providing access to an exercise yard). Animals were encouraged to progress through the test corridor by the presence of conspecifics housed in a pen located beyond Test Zone C, which served as the reward stimulus.  ***Figure 1.*** *Illustration of the test corridor showing the novel surface (e.g., plywood with yoga mat pieces glued in different shapes), the test zones (A, B, and C), and the reward zone located beyond the test area.*
Indoor corridor:	The novel surface was installed in an indoor corridor routinely used by the animal (e.g., a corridor providing access to the milking parlour). Animals were encouraged to progress through the test corridor by the presence of conspecifics housed in a pen located beyond Test Zone C, which served as the reward stimulus  ***Figure 1***. *Illustration of the test corridor showing the novel surface (e.g., artificial grass with painted shapes), the test zones (A, B, and C), and the reward zone located beyond the test area.*

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Surface:	
Novel surface:	The novel surface must provide secure footing and minimize the risk of injury by being non-slippery. It should cover the full width of the corridor and allow the animal to take a minimum of two steps on it while crossing. The surface must be unfamiliar to the animal and should be of a size, colour, and pattern that ensure clear visibility to both the animals and the video cameras within the corridor. Importantly, it should contrast clearly with the corridor floor so that the animal can visually detect it before stepping onto it, rather than only perceiving it upon contact. Hatch or striped patterns may be used to enhance the perceived risk associated with the surface by creating an illusion of depth or discontinuity, thereby increasing the challenge. This is particularly relevant because the test is intended to assess responses not only to novelty but also to perceived risk. If the test is repeated, the same surface material must not be reused to maintain novelty.

Requirements

✓ **Personnel (n = 4 - 5):**

Personnel	Number	Role
Testing assistant	1	Responsible for wait times and test sequence.
Camera manager	1	Responsible of managing the camera operation (i.e., installing the cameras, validating the views and starting/stopping the recording) and test safety.
Trained handler	2-3	Responsible for handling of animals to and from the test corridor and for handling animals during the test.

✓ **Materials Needed:**

- Novel surface
- Stopwatch
- Whiteboard
- Dry erased marker
- PPE
- Camera(s)
- Power source for camera(s) (i.e., extension cords, power bank with AC outlet)
- Cones or floor markers

General information

- **Location:** The test corridor must be designed to ensure both animal safety and appropriate testing conditions. It should be enclosed by gates of sufficient height to prevent animals from escaping. The flooring or ground surface must provide secure footing and minimize the risk of

injury by being non-slip (e.g., rubber mats, grooved concrete, or sand). The corridor should be wide enough to allow animals to move freely (e.g., a minimum width of 3 m for cattle). A reward should be positioned at the end of the corridor to encourage animals to move through the test area and cross the novel surface.

- **Lighting and noise conditions:** The test environment must be sufficiently illuminated to ensure a clear video recording of the animal. To perform a reliable and reproducible test all potential distractions must be avoided, as such the presence of non-essential personnel and any unnecessary noise during testing is strictly prohibited.

- **Camera positioning:** Camera(s) should be positioned to perfectly cover the animal, the holding area and the 3 zones of the test corridor.

- **Animal preparation**: To avoid confounding effects, test animals should be familiar with the corridor and the route used during testing. If companion animals are used as a reward at the end of the corridor, they should be placed in the designated pen before the test begins. In addition, a reliable identification method should be implemented to ensure that individual cows can be accurately identified from video recordings.

- **Test preparation**: Cones or floor markers should be used to delineate the three zones.

Procedure

1. A camera manager sets up the cameras, initiates recording for all cameras and verifies that the field of view for each camera is correctly aligned to ensure full coverage of all 2 test zones (Figure 1, 2).

2. A camera manager writes on the whiteboard the information that will allow to identify the video (animal ID/markings, test being conducted, date and time) and displays it in all camera views.

3. A trained handler guides the animal into the holding area of the test corridor (Figures 1 and 2) and releases it near the threshold of Zone A (Figures 1 and 2). The handler then exits the corridor, allowing the animal to proceed through the test area without further guidance.

4. After the trained handler exits the corridor, a research assistant initiates a 2-minute observation period during which the animal is allowed to enter Zone A voluntarily.

    - **If the animals do not enter zone A**, after two minutes, the trained handler re-enters the corridor, approaches the animals and starts encouraging them to move forward

into Zone A following the steps below. If the step is unsuccessful after 30 seconds, the handler moves onto the next:

Step 1: Use vocalization (i.e., talking to the animals with regular voice volume) and hand gesture (i.e., soft hand clapping).
Step 2: Use gentle tactile stimulation (i.e., touching the animals on the rump).
Step 3: Select one animal from the group that is still in the handling area and physically prompt the animal to move into Zone A (i.e., putting your hands on the animal's rump and using consistent forward motion to encourage the animal to move forward into Zone A).
Step 4: If the selected animal is still unwilling to enter Zone A, repeated step 3 on another animal that is still in the handling area. If none or certain animals of the group are not willing to enter Zone A they are walked back to their home environment.

- **For the animal entering Zone A**, the testing assistant initiates a 3-minute timer, records the latency to enter Zone A and the time required for each animal to move from Zone A to Zone C (Figures 1 and 2). The testing assistant also records the time at which each animal enters each test zone (A, B, and C).

NOTE: If any animals slip, becomes agitated, or refuses to move for safety reasons, then the camera manager calls "End test" and notes the event as "End test – safety concern."

5. The test is concluded when all the animals have reached Zone C (all 4 foot in Zone C) or when the 3-minute timer has elapse even if Zone C was not reached.

6. After completing the test, animals that have successfully reached Zone C will be rewarded either with a 20-second interaction time with conspecifics in the reward zone (outdoor corridor) or joining its conspecific in their home pen (indoor corridor).

7. Before testing another group, the surface must be cleaned, urine and/or feces must be removed, and the floor must be dried.

Safety Note

All personnel should maintain proper distance from test animals to stay safe and not distract them. All personnel should remain calm and avoid sudden movements to prevent startling the animals. Surface of the enclosure should be safe and the gates sturdy enough to withstand unpredictable behaviour of the animals.

AI ASSISTANCE DISCLOSURE

Microsoft 365 Copilot, an AI-based writing and editing tool was used to support language refinement, structural clarity, and consistency of technical descriptions.

CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

WELL-E Authorship Governance policy, adapted from the Contributor Roles Taxonomy (CRediT) proposed by Brand et al. (2015).

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Analysis	Formal Analysis	N/A
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* WELL-E authors and co-authors, who take accountability for the manuscript, must contribute to each stage of the paper. These stages are conceptualization/design, data analysis, and writing & editing.