

Development of a scale to assess pigs' immediate behavioural reaction when encountering novel odours

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ABSTRACT

It is currently not possible to predict how pigs will perceive novel odours, or whether certain odours might evoke avoidant or exploratory behavioural reactions. This study aimed to develop a reaction scale to assess the immediate behavioural reactions of pigs when encountering an odour for the first time. A secondary aim was to compare if reactions to different odours varied consistently among pigs, which could indicate that the reaction scale could be used to assign odour valence. The experiment included 184 growing-finishing pigs tested in 92 pairs of opposite-sex littermates. All pig pairs were presented with three out of a total of twelve different odours, in a balanced order. The odour samples consisted of six drops of essential oils (undiluted) on filter paper in a plastic container. Each of the three odours was presented along with an odourless control (distilled water). The odour and control samples were available to the pigs during 1-minute trials. The pigs' behavioural reactions were recorded six seconds after each pig had first approached the sample. The reaction was scored on a scale from 1 to 6, where 1 was exploratory behaviour (e.g., sniffing) and 6 was active avoidance behaviour (e.g., leaving, head shaking). An ordinal mixed-effects model was fitted to the data and showed no effect of sex, age, or odour triad on reaction scores, but an effect of treatment with odour eliciting significantly more exploratory behaviour (lower reaction score) than control ($P < 0.001$). Pigs expressed large individual differences in reactions to 10 of the 12 odours, but a consistent pattern of significantly more exploratory reactions to vanilla and blood orange (both odours: $P < 0.05$), which could suggest that pigs may perceive these odours as particularly pleasant. Reaction scores were significantly lower (i.e., pigs performing more explorative behaviour) for the first odour tested compared with subsequent odours ($P \leq 0.04$) irrespective of the specific odour. The 6-point scale effectively described explorative and avoidance behaviours that may reflect the valence of the pigs' reactions. To our knowledge, this is the first scale specifically developed to assess pigs' immediate behavioural reactions to novel olfactory stimuli in a commercial production environment. The development of the reaction scale and the findings provide useful guidance for future research on odour perception. The observed decrease in interest over time should be taken into account when repeated testing is used.

1. Introduction

Olfaction and the role of chemical signals in animal behaviour have been studied in a range of taxa with the main focus on behavioural ecology in birds, e.g., migratory behaviour (Wikelski et al., 2015), olfactory camouflage and communication (Grieves et al., 2022), and rodents as models of human olfaction (Ache and Young, 2005). Chemical signals, including odours, also play a central role in mate selection, development and direction of parental behaviour, food seeking, and group cohesion in many mammalian species (Nielsen, 2018). However,

the role of olfaction is often overlooked in farm animal behaviour and welfare research (Campbell-Palmer and Rosell, 2011; Nielsen et al., 2015). While research on flavour-associated learning processes, e.g., prenatal flavour exposure (Figueroa et al., 2013a) and social learning with flavoured feed (Figueroa et al., 2013b) in livestock species are more established, the study of novel odour perception has yet to be explored.

The domestic pig (*Sus scrofa domestica*) has a highly developed sense of smell, which likely evolved as a result of pigs obtaining useful information from odours in their everyday life (for review see: Schild and Rørvang, 2023). Being omnivorous animals, pigs naturally forage

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across large areas due to the irregular availability of feed resources. During exploration, pigs rely heavily on their sensory apparatus, incorporating sight, hearing, taste, touch, and smell (Stolba and Wood-Gush, 1989). Sniffing, intrinsic to pig exploratory behaviour and evident from birth, is a vital behaviour that allows pigs to detect volatile odourants, especially when exploring new environments or seeking food (Day et al., 1995; Studnitz et al., 2007; Wood-Gush and Vestergaard, 1989). Pigs use their snout to sniff and root, but will also lick and bite, and even express rubbing and rolling behaviours when exposed to certain odours (Rørvang et al., 2023a, 2023b). The underlying mechanisms are not fully understood, and the multifaceted behavioural repertoire of pigs' responses to odours emphasise the necessity for further research into their olfactory perception. One aspect which currently remains unknown is how pigs react to odours when encountering them for the first time: is this response similar for pigs when exposed to the same novel odour, or does perception differ among individuals? This may reflect the aversiveness or pleasantness of odours. In humans, this is relatively easy to study, as the human test subjects can rate the odours on a scale from pleasant to unpleasant verbally (Moskowitz et al., 1976) which may be accompanied by analysis of skin conductance and heart rate as well as behavioural analysis of, e.g., facial expressions (Alaoui-Ismaïli et al., 1997; Bensafi et al., 2002; Brauchli et al., 1995). However, this is not as straightforward when studying animals. Understanding how pigs perceive odours when first encountered may enable us to assess if and how they attribute valence upon first smelling an odour. Valence may, based on human literature, be defined as the intrinsic attractiveness (positive valence) or aversiveness (negative valence) of a stimulus (Vieitas-Gaspar et al., 2025).

This study aimed to develop a reaction scale to assess pigs' immediate behavioural reaction when encountering an odour for the first time (i.e., a novel odour). A secondary aim was to compare if reactions to different odours varied consistently among pigs, which could indicate that the reaction scale could be used to assign odour valence. The study was hypothesis-generating (basic research) as no previous studies have attempted to describe pigs' reactions to novel odours. The null hypothesis was that pigs would react randomly (i.e., no pattern in reaction according to odour) to specific odours.

2. Materials and method

This study relied on data collected as part of a larger project investigating pigs' odour exploration behaviour, and an extensive description of all materials and methods can be found in Rørvang et al. (2023a). Below is a summary of the experimental setup.

2.1. Location and ethical approval

The study was conducted at the Swedish Livestock Research Centre, Lövsta, in Uppsala, Sweden. Prior to the experimental start, the procedures and details of the experiment were evaluated by the Board of Ethical Use of Animals in Teaching and Research, Swedish University of Agricultural Sciences, Uppsala, Sweden, and an ethical permit was obtained from the Swedish Board of Agriculture, Uppsala, Sweden ID number: 5.2.18-02900/2020. All procedures were conducted in accordance with the ethical guidelines proposed by the Ethical Committee of the ISAE (International Society of Applied Ethology; (Tahamtani et al., 2023)) and met the ARRIVE guidelines (Kilkenny et al., 2010).

2.2. Animals and experimental conditions

The study included 184 growing pigs of crossbred commercial breeds (Yorkshire x Hampshire, Duroc x Yorkshire, Landrace x Hampshire). The pigs were tested in littermate pairs ($n = 92$ opposite-sex pairs) to prevent stress associated with social isolation. At the time of testing, the average weight per pig was 64.9 kg (SE = 0.7 kg) and the average age was 86.8 days (range: 32–110 days). All pigs were bred and kept at the research

centre, and housed and managed according to commercial-production-like routines including feeding, housing condition and enrichment (for more information see: The Swedish Livestock Research Centre, 2017). Before the start of testing, all pigs were moved in litter groups from their home pens to an experimental section where they were divided into pairs before being assigned to an experimental pen (Fig. 1). Testing was done during two different time slots, either in the morning or afternoon, ensuring the pigs had the possibility to eat before the tests to minimise the risk of hunger influencing the pigs' responses to the odours. Details of the odour testing design and results about the habituation/dishabituation test can be found in Rørvang et al. (2023a); briefly: The experimental pens each had two circular odour insertion points (Ø: 14 cm; 55 cm apart) drilled through the front wall of the pen (Fig. 1). The insertion points were designed to allow pigs to insert their snouts allowing them to sniff the odour without having physical contact with the odour source (i.e., to not allow pigs to taste the odours). These two insertion points were used to present the pigs with the odour and odourless control (distilled water) samples during the tests, balanced between the sides. The twelve odours selected for the tests were all oil-based and approved for human use. All odours were selected based on the criteria that these were not present in the pig feed or bedding, or in the veterinary remedies at the farm to ensure novelty. Prior to the experiment, both the odour and the odourless control were assessed by a human nose. Eight odours were single-source essential oils (from one origin, i.e., the oils are pure and not mixed), four were mixtures of several essential oils, and all oils were undiluted. The twelve odours were grouped together in four triads, where each of the three odours was of different origin (herb, spice, tree, root, fruit, flower, seed, or a mixture): A) vanilla, *aniseed*, and *blood orange*; B) musk, apple, and *cinnamon bark*; C) *ginger*, *pine*, and jasmine; and D) *cedarwood*, *thyme*, and *lavender* (single-source essential oils are marked in *italic*). Within each triad, the order of presentation was balanced to ensure all order combinations of the three were tested. Each pig pair was randomly assigned to an odour triad (A, B, C, or D) prior to testing. Each pig pair was tested once on three successive odours (i.e., one triad) only (i.e., 24 pairs/odour), to limit the risk of sniffing fatigue or loss of motivation to explore the odours as the test paradigm (Habituation/Dishabituation) relies on voluntary investigation. To preserve novelty, minimize the risk of odour contamination, and avoid prior exposure, all pig pairs tested on a given day were exposed to the same odour sequence, and no pig pair was tested on more than one day (i.e. one sequence). Each odour and control sample were presented simultaneously for 1 min, three times in a row, with 2-minute breaks in-between exposures. After the third presentation of the same odour, a new odour was presented in the same manner resulting in a nine odour and control presentations for each pig pair in total. In this study, we were interested in the immediate reaction to a novel odour and therefore used only the first presentation of each odour and the respective control, i.e., three first presentations of an odour and control pair per pig pair. All tests were video recorded using GoPro Hero 9 (Fig. 1), and the immediate behavioural reactions to the odour and control were subsequently assessed and recorded for each pig in the pair.

2.3. The basis for constructing a behavioural reaction scale

We used the video footage from the Habituation/Dishabituation test to develop a scale to score pigs' immediate behavioural reactions to novel odours. The development of the reaction scale was based on two key premises: 1) to ensure odour novelty to the greatest extent possible, and 2) to register active behavioural reactions when the pigs were within sniffing distance for the first time. Thus, we focused on the pig's very first encounter with each odour as done in human olfactory research (see e.g., Bensafi et al., 2002; Cupchik et al., 2005; Moskowitz et al., 1976). In our study, we had no possibility to include heart rate or skin conductance due to the production setting and social housing of the pigs, and the assessment was thus based purely on the behavioural reaction to the odours, such as sniffing, of the pigs tested.

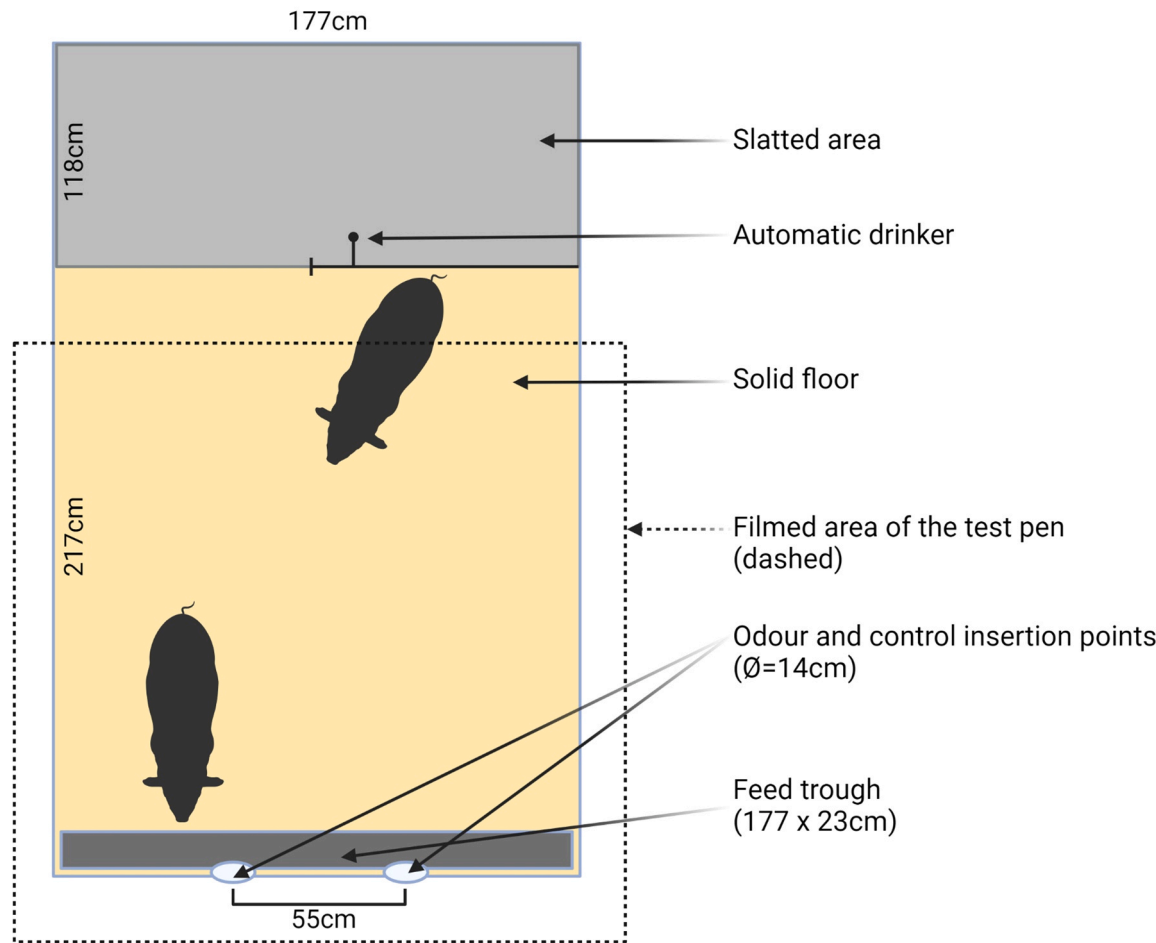


Fig. 1. Top-view diagram of the experimental pen layout, with two pigs. The grey area illustrates the slatted floor with an automatic drinker. The yellow area illustrates the solid floor area with straw provided on the floor and access to a feed trough (23 cm wide). The white circles in the front above the feed trough illustrate the odour and control insertion points, which were spaced 55 cm apart. The dashed square-shaped outline illustrates the area covered and filmed by the GoPro camera.

2.4. Development of the scale

The behavioural reaction scale was developed by first defining a specific point in time and duration (6 s, i.e., 10 % of total presentation time, after fulfilling an approach criterion) to capture the immediate behavioural reaction. This duration was chosen to allow the pigs enough time to sniff the odours and make a decision (e.g., stay and sniff, move away) without becoming habituated to the odour. The pigs were allowed

to approach and interact freely with the odour and control (i.e., pigs could also choose to not approach and/or interact). An ethogram (Table 1) was then constructed to categorize the range of possible behaviours observed at this time. The approach criterion that pigs needed to fulfil in order to have an immediate reaction that could be scored was defined as the snout of the focal pig being positioned within 12 cm from an insertion point (Fig. 2), which was adapted from the sniffing behaviour definition used in Rørvang et al. (2023a). At this distance, it

Table 1
Ethogram used to categorise and score the immediate behavioural reaction of pigs when approaching an odour or an odourless control (either of the two insertion points). Both pigs in a pair were observed (one at a time) during the first 6 s after fulfilling an approach criterion, and the distinct behavioural reactions defined were allocated a score from 1 to 6 (modified from Grut, 2022) and used as the basis for the ordinal scale.

Reaction	Description
1 – Sniff	The pig has their snout through an insertion point.
2 – Oral	The pig is within 12 cm (Fig. 2) of an insertion point and makes direct contact with the insertion point or pen wall (e.g., bites/licks/roots). May include rubbing aimed at insertion point.
3 – Approach	The pig is within 12 cm (Fig. 2) of an insertion point, but does not make direct contact with insertion point or pen wall. May include snout above insertion point, but not above pen bars.
4 – Turn	The pig has approached but turned < 90° away from the insertion point (Fig. 3) and is no longer within 12 cm. May include moving up to 3 steps.
5 – Leave	The pig has approached but turned ≥ 90° away from the insertion point (Fig. 4) or moved more than 3 steps away from the insertion point, and is no longer within 12 cm.
6 – Head shake	The pig has approached the insertion point and shakes their head at least twice from side to side in a sudden movement. May have been preceded by sniff, oral, approach, turn, or leave.
No approach ^a	The pig may approach the front pen wall but does not come within 12 cm of an insertion point. Also includes cases where the pig is out of camera range (i.e., on the slatted area of the pen).

^a Pigs not receiving a score and excluded from the analyses.



Fig. 2. Top-view of the front of the experimental pen as filmed by the camera (see Fig. 1), with two pigs shown. The centre of the feed trough is marked by a green line, which at this view was approx. 12 cm from the insertion points, highlighted by the dashed orange circles. When the snout of the pig was within 12 cm of an insertion point (i.e., on or passed the green line), the timing of the 6 s started, and the pig would receive a score. Pigs who did not fulfil the approach criterion were marked as “no approach”. The pig at the front of the pen has fulfilled the approach criterion and meets the definition of “sniff” (score 1) for the right-hand insertion point. The second pig at the top of the figure did not approach further and was subsequently marked as a “no approach”.

was most likely that the pig would smell and thus detect the odour. The behaviours described in the ethogram were then ranked and used to construct an ordinal scale, developed as an animal analogue to the pleasantness scale from human olfaction studies (e.g., Bensafi et al., 2002; Moskowitz et al., 1976) but based solely on the behavioural reaction of the pigs. The scale ranged from 1 to 6, with 1 being the maximal exploration of an odour (sniffing) and 6 being the maximal expression of aversion (head shake). To categorize attempts of pigs to move away from the odour/control, scores 4 and 5 (Table 1) incorporated degrees to which the pig faced towards/away from the insertion point after approaching and a criterion for how many steps were taken away from the insertion point (Figs. 3 and 4): Step count alone would not provide a fair representation of the pigs’ reaction. Step length can vary between individuals, and factors like another pig blocking the way could influence the focal pig’s movement from the insertion point. To

account for this, the orientation of the pig’s head relative to the insertion point was included. When a focal pig was facing more than 90° away from the insertion point, which they initially approached, they were facing the side and back of the pen and thus most likely actively moving away from the odour/control (Fig. 4). The active movement away from the insertion point was considered an indication of avoidance behaviour or a loss of interest. Using 90° as a cut-off provided a standardised and easily quantifiable criterion for distinguishing a mild avoidance/non-interest from a stronger avoidance/interest reaction, which helped to avoid ambiguity in scoring. Conversely, pigs facing less than 90° away from the insertion point, which they initially approached, were likely moving away from the insertion point (if facing the pen side wall) or towards the other insertion point (Fig. 3). These thresholds were chosen because pigs who turned 90° or more suggested a more definitive movement away from the insertion point compared to turning less than

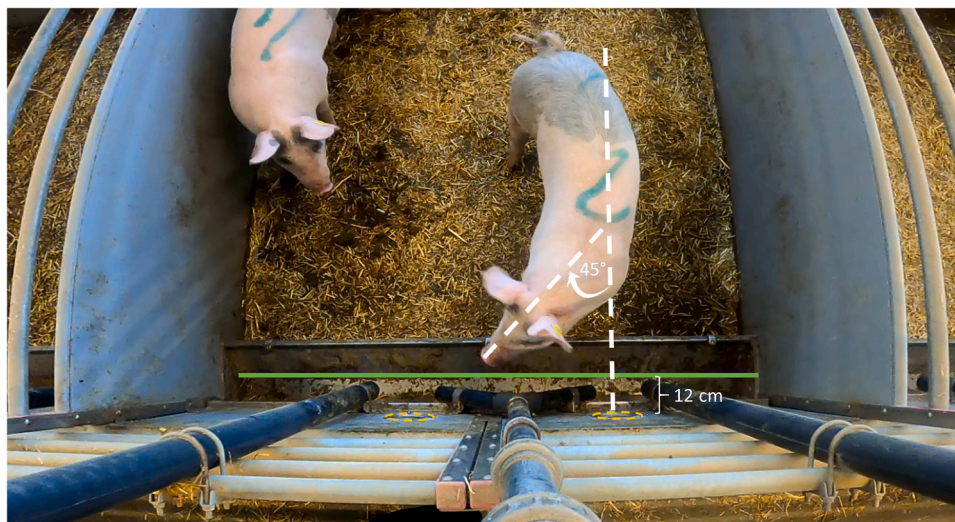


Fig. 3. Top-view of the experimental pen as filmed by the camera, with two pigs shown. The centre of the feed trough is marked by a green line, which at this view was approx. 12 cm from the insertion points, highlighted by the dashed orange circles. The pig at the front of the pen illustrates the position for a reaction score 4 (“turn”), having turned less than 90 degrees away from the right-hand side insertion point within 6 s of fulfilling the approach criterion. The same pig does not yet fulfil the approach criterion for the left insertion point.



Fig. 4. Top-view of the experimental pen as filmed by the camera, with two pigs shown. The centre of the feed trough is marked by a green line, which at this view was approx. 12 cm from the insertion points, highlighted by the dashed orange circles. The pig at the front of the pen illustrates the position for a reaction score 5 (“leave”), having turned more than 90 degrees away from the right-hand side insertion point within 6 s of fulfilling the approach criterion.

90°. Within each pig pair, both pigs’ behavioural reactions to both the control insertion point and the odour insertion point were measured. Pigs not fulfilling the approach criterion within the 1-minute presentation were marked “no approach” (Table 1) and excluded from the analysis on the basis that it was uncertain whether they detected the odour/control or not. Pigs who were not visible in the camera footage (further away than camera coverage, Fig. 1) were also categorized as “no approach” and excluded from the analysis. The scores given to the pigs’ behavioural reactions to the odours ranged from maximal exploration of an odour (sniffing, score 1) to expression of aversion (head shake, score 6) with the scores in between indicating behavioural reactions of less exploration (scores 2 and 3) and increasing aversion/un-interest (scores 4 and 5). The scale was developed as a gradient scale, and did not represent a numerical relationship; a score 6 did not indicate that the reaction was six times more aversive than a score 1, but reflected a relative ordering of the responses based on the degree of exploration or aversion/non-interest.

2.5. Behavioural scoring

Two observers developed the reaction scale using post-hoc video analysis of a subset of 141 behavioural reactions (i.e., approximately 20 % of all reactions). The scale was subsequently validated by the observers scoring the same subset of videos, and calculating an inter-observer reliability score (82 %, kappa: 0.92). The final scores of all pigs were made by an observer who was not involved in developing the reaction scale definitions and who was blind to the specific odours in a given test.

2.6. Data editing and statistical analyses

The total number of reaction scores possible were 1104 (i.e., six reaction scores (three odours and three control reactions) for each of the 184 pigs). Out of these, 34 reactions were lost due to camera error or power failure. From the remaining 1070 reactions, 88 were categorised as “no approach” because the pig was outside the camera view (i.e., 44 for an odour, and 44 for control), and of the remaining 982 reactions scored, 251 reactions (odour and control summed) were “no approach” behaviour and thus also excluded, leaving 731 reactions in total to be further analysed.

As the pigs were all tested in pairs, the statistical unit was the pig pair. Prior to model fitting, a correlation analysis (Spearman rank-order

correlation) was performed to analyse if individual pig reaction within a pair was correlated. The individual reaction within a pig pair was not correlated ($r_s(346) = 0.32$, $P = 0.52$), hence, data were analysed at individual level. An ordinal mixed-effects model was fitted to the data from all pigs (ordinal: all scores 1–6), and the full model included insertion point (categorical: odour, control), odour triad (categorical: A, B, C, D), specific odour (categorical: 1–12), odour number (categorical: 1st, 2nd, 3rd; i.e., all three odours presented to each pair), pig age (numerical: mean \pm SE = 86.8 ± 1.4 days), and sex (categorical: female, male) as fixed effects and pig ID as random effect to control for repeated measures on each pig. The categorisation of the scores being either exploratory (scores 1–3) or avoidant (scores 4–6) was done post hoc.

All analyses were performed using R (R-version: 4.2.2 (2022–10–31 ucrt) – “Innocent and Trusting”; (Computing, 2021) working in the R-interface of R-studio (version: 2023.06.0, build 421). For tidying data prior to modelling, the packages “dplyr” (Wickham et al., 2023), “tidyverse” (Wickham, 2023), and “Rmisc” (Hope, 2013) were used. For building the model, the package “ordinal” (Christensen, 2022) was used, and for comparing factors within categorical fixed effect the package “emmeans” (Lenth, 2021) was used. Graphs were produced using R and the package “ggplot2” (Wickham, 2016), and Microsoft Power Point (version 2017) was used to add the axes of scores, representing either exploratory or avoidant behaviour, and to adjust the resolution.

3. Results

Across both the odour and control reactions, score 1 was most frequently recorded with a total of 327 out of the 731 reactions observed (45 %). The model fitting showed that the fixed effects of odour triad, age, and sex had no significant effect on the reaction scores ($P \geq 0.1$) and hence a reduced model excluding these variables was applied. The full model was checked against the reduced version in a likelihood ratio test, which showed no significant difference between the models and hence these variables were removed from the final model. The final model thus included the fixed effects of insertion point, odour, and odour number and the random effects explained in Section 2.6 above. The model showed a significant effect of the insertion point (odour/control) on the reaction scores, with the odour insertion point receiving significantly more exploration, i.e., lower reaction scores (control – odour: estimate \pm SE = 1.18 ± 0.15 , z-ratio = 7.67, $P < 0.001$) than the control insertion point. The fixed effect of odour significantly affected the reaction score but only for certain odours. Vanilla and blood orange

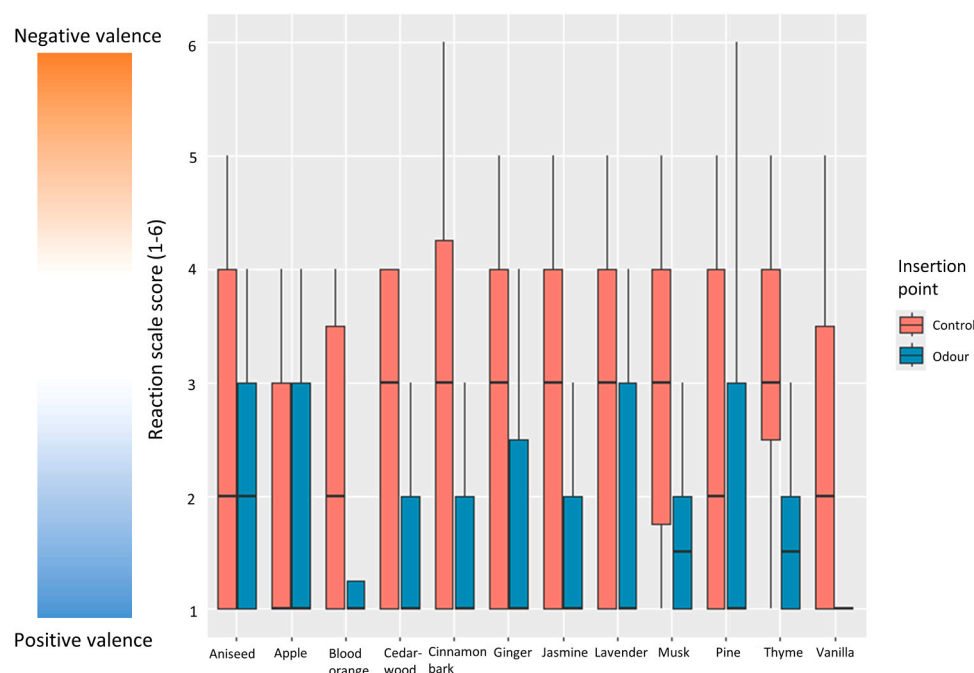


Fig. 5. Boxplot of reaction scores (1–6) for the 12 odours (blue bars) and corresponding control (red bars) for each pig during their first presentation of each odour, shown on the x-axis. The bars span the 25–75 % quartiles, with median shown as a horizontal line, and error bars indicating the maximum and minimum. For vanilla odour insertion point, the median, and 25 %–75 % quartiles were all 1, hence no blue bar is visible.

received significantly lower reaction scores than any of the other odours (pairwise comparisons: vanilla: estimate \pm SE = -1.29 ± 0.54 , z-ratio = -2.40 , $P = 0.017$, blood orange: estimate \pm SE = -1.07 ± 0.52 , z-ratio = -2.06 , $P = 0.04$). The large variation in reaction scores within all 12 odours is illustrated in Fig. 5. The post-hoc categorisation of the reaction scores being either exploratory or avoidant/un-interest, is shown on the left side of Fig. 5 illustrating the patterns for each odour.

A significant effect of odour number was also found, with scores increasing significantly between odours presented first and second (estimate \pm SE = -0.41 ± 0.17 , z-ratio = -2.39 , $P = 0.04$), and odours presented first and third (estimate \pm SE = -0.60 ± 0.19 , z-ratio = -3.24 , $P = 0.003$) but not between the second and third odour ($P > 0.1$).

4. Discussion

The aim of this study was to develop a reaction scale to assess pigs' immediate behavioural reaction when encountering an odour for the first time (i.e., a novel odour). A secondary aim was to compare if reactions to different odours varied consistently among pigs, which could indicate that the reaction scale could be used to assign odour valence. The odours were all assumed to be novel to the pigs, none of the odours were present in their feed, bedding, or veterinary treatments making it unlikely that they had any prior exposure to them. In this study, a 6-point scale scoring pigs' immediate behavioural reactions was developed. We found no effect of sex, age, or odour triad, but a significant effect of odour presentation number and insertion point (odour vs. control), as well as a within-odour effect of specific odours on the reaction score with vanilla and blood orange receiving significantly lower (performing more explorative behaviour) reaction scores. These results show that apart from vanilla and blood orange, pigs' reactions to the odours were not consistent among pigs and thus no general valence could be attributed to these ten odours. Vanilla and blood orange on the other hand received more explorative reactions across pigs, suggesting a consistent interest in these odours, which in turn may indicate a positive valence.

4.1. The reaction scale

The three first reactions of the scale (scores 1–3) included sniffing, oral manipulation and pigs approaching the odour. These behaviours are commonly seen in pigs' normal exploratory behavioural repertoire (Day et al., 1995; Studnitz et al., 2007; Wood-Gush and Vestergaard, 1989) indicating that the pig was motivated to explore the odour, and could be seen as an indicator of positive (or non-aversive) odour valence. Sniffing alone does not necessarily equate to enjoyment, but attempts to lick, root, and remaining close to the odour, were considered supportive indicators of positive valence. Pigs are motivated to explore, even when their needs are met (Studnitz et al., 2007), further supporting the interpretation of exploratory behaviours as a sign of positive valence. Pigs reacted more exploratively during their first encounter with a novel odour, regardless of the specific odour, meaning that pigs reacted more avoidant or with less interest in the subsequent first encounter with the next novel odour. This is likely to indicate a habituation effect of encountering several new odours over the relatively short period of time that was the 25-minute test duration. These findings may aid the understanding of the overall effect of odour number (1st, 2nd, 3rd) found in the results of Rørvang et al. (2023a), which has also been found for horses in the same test paradigm (Rørvang et al., 2022). Previously, this effect has been suggested to be a result of sniffing fatigue, or loss of motivation to explore new odours. This study adds further support to that theory and points to an important drawback when testing repeatedly using the Habituation/Dishabituation test which relies on the animal's motivation to investigate novel odours (Yang and Crawley, 2009). This aspect is important to take into consideration when testing several odours repeatedly over a short time period, when testing similar odours, and in the future development or adaptation of test paradigms to assess animal olfaction.

Scores 4–6 represent pigs choosing to leave the insertion point after first approaching (either leaving to explore the other insertion point, or leaving both insertion points completely), or shaking their heads. These reactions, in particular the head-shaking, may be avoidance reactions as they shared resemblance with pigs' reactions to frightening or unpleasant stimuli (Imfeld-Mueller et al., 2011), or may be indicative of

un-interest in the odour stimuli. These reactions may be indicative of a negative valence of the odour. This highlights an important consideration for the use of odours as enrichment. If not all pigs perceive certain odours as pleasant, the enrichment value may be limited to only some individuals. The results show that pigs reacted significantly less exploratory towards the odourless control than the odours. In this study, no other options were present, and hence the higher reaction scores to the odourless control is likely to reflect a loss of interest due to the absence of odour, rather than avoidance behaviour, but the scale in its current form cannot readily distinguish these, apart from the head-shaking which is a known aversive behaviour in pigs (Norscia et al., 2021). As head shaking was seldom observed in response to the odourless control, the odours eliciting this behaviour were likely more aversive than odours that did not result in head-shaking. The finding that the pigs reacted less explorative and more neutral toward the control supports the validity of the scale, as a somewhat neutral reaction was expected from a neutral odour. In the first presentation (first exposure to the odour and control), scores were generally more evenly distributed between the odour and odourless control, compared with the second and third presentations, indicating that pigs were more reluctant to explore the odourless control once discovering the lack of odour in that insertion point. This finding is also reflected in the sniffing durations measured in Rørvang et al. (2023a) with longer sniffing time for odour compared to odourless control.

4.2. The reaction scale as a predictor of odour valence in pigs

In this study, the 6-point reaction scale based on the immediate behavioural reaction of the pigs to a novel odour, revealed a large individual variation among the 184 pigs tested. This could imply that individual pigs attribute different valence to the majority of the odours tested, but variability could also be influenced by emotional reactivity (Reimert et al., 2013) and individual coping styles (Bolhuis et al., 2005). Differences in personality may therefore have contributed to the variability in the observed responses. Similarly, individual differences in olfactory sensitivity could influence how pigs perceive odours. In this study, we controlled for a potential effect of sex and age, which had no significant effect on the reaction scores. Future studies could test effect on perception of odours in pigs with demonstrated differences in personality.

Although no clear patterns were uncovered with regard to specific reactions to each specific odour, pigs' reactions towards vanilla and blood orange were consistently explorative. This could indicate that for these two odours, we may be able to predict how pigs in general will react to these odours. However, this result appears to contrast with findings on sniffing duration from the same experiment reported in Rørvang et al. (2023a), where vanilla received the lowest total sniffing duration. These contradicting results hint that there may be another facet to measure when investigating pigs' perception of odours, for example attempts of licking and rooting of the odour source. In another study, where the pigs were provided with odours in their home environment, they interacted less with vanilla and orange odours compared with natural, perhaps more biologically relevant odours such as soil and grass (Nowicki et al., 2015). It is important to emphasise, that considerable effort was made in the present study to test odours that the pigs were unlikely to have had prior exposure to, based on the feed, bedding or veterinary treatments not containing these. In this study, the pigs also spent less time close to the odour source when vanilla was tested compared with all other odours, which is in line with the results in Rørvang et al. (2023a). To further investigate the validity of the proposed reaction scale, the immediate behavioural reactions of pigs to more biologically relevant odours may be of interest, but it is difficult to obtain pigs who have not encountered such odours before.

Uccheddu et al. (2018) investigated whether a blend of nine essential oils (including blood orange) influenced the affective states of sheltered dogs, and found that the blend reduced the dogs' latency when

responding to an ambiguous cue suggesting an optimistic bias. Again, these results need to be viewed with caution as the positive effect might be less specific to the particular odour, and rather an effect of the mere presence of an odour compared to no odour. The current findings show that the immediate reaction of pigs to odours is more likely to be exploratory when the odours are vanilla and blood orange, but it remains unknown if these odours are pleasant to the pig, and if they evoke any immediate effects on pigs' emotional state. Vanilla and blood orange might, however, be interesting odours to further assess, both in relation to pigs' perception of odours and their potential use in olfactory enrichment. In this study, the observation period was six seconds, to capture pigs' immediate reactions rather than potential long-term interest. Further studies could investigate whether and how the long-term reactions may differ from the immediate response and if repeated exposure to these odours strengthens positive associations.

5. Conclusion

This is the first olfactory reaction scale designed to record pigs' immediate behavioural reactions when encountering novel odours. The developed 6-point scale showed that pigs reacted most exploratively towards the first novel odour encountered, regardless of the specific odour. The pigs showed less explorative behaviour toward the odourless control compared with any of the odours. The immediate reaction to vanilla and blood orange may be more likely to be exploratory across pigs, but for other odours tested here, the immediate reactions may be associated with odour valence of individual pigs.

CRediT authorship contribution statement

Rebecca Grut: Writing – review & editing, Writing – original draft, Methodology, Investigation, Formal analysis, Conceptualization. **Maria Vilain Rørvang:** Writing – review & editing, Writing – original draft, Supervision, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Birte L. Nielsen:** Writing – review & editing, Investigation. **Johanna Stenfelt:** Writing – review & editing, Writing – original draft, Supervision, Methodology, Investigation, Formal analysis, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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