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Using cognitive paradigms to measure emotion in pigs

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### **Summary**

This work is divided into a number of chapters. The topic is introduced in Chapter 01, where both the importance of emotion with respect to animal welfare and the benefit of cognitive approaches to assess emotion are discussed. In Chapter 02, we reviewed the common behavioural measures used to assess emotion in pigs, for example, behavioural tests such as the Open Field test or the Novel Object test, as well as individual behaviours, such as play and vocalisations. Each behavioural test and measure was evaluated according to a set of criteria, including the ecological validity of the task design and behavioural measures used, as well as the specificity of the measures with respect to emotion. Important aspects of a good task design, such as standardisation of the design and outcome measures, as well as the capacity for repeated testing and automation are discussed. Firstly, we found that it was difficult to compare the common behavioural tests due to the lack of standardisation of task designs and behaviours recorded across studies. For example, the manner in which the object is introduced in a Novel Object test varies in speed from slow to sudden; suddenly introduced objects may create a ceiling avoidance response in all animals, while introducing an object slowly may be more conducive to observing different behavioural responses to novelty. A second finding from this chapter is that many of the tests, having been adapted from rodent research, do not tap into ethologically relevant behaviour for pigs, and rely on quite broad measures which may not specifically and unambiguously reflect emotional responses, for example measures of movement in the Open Field test may represent arousal more than the valence of an emotional state.

Having proposed to study emotion through its influences on cognitive processes, Chapter 03 describes the development of a suitable task design to study emotion-biased judgement of ambiguity. This chapter compares the responses of pigs in two discrimination task designs. First, a Go/No-Go design was used, where a Go tone-cue signalled the availability of reward at the end of a runway while a No-Go tone-cue signalled the absence of reward. Latency to reach the end of the runway in response to each cue type was taken as measure of discrimination learning. A second and third task design used active choice (Go/Go) responses, where pigs had to respond in the correct location (goal-box) for each tone-cue to receive the signalled large or small reward. Choice of goal-box was used to indicate discrimination learning. Pigs in the final active choice design showed clearer and more consistent learning than pigs in the Go/No-Go design. We therefore proposed that active choice task designs are more suitable to study judgement bias in pigs as the task requires an animal to first master and then maintain a discrimination between control ‘positive’ (predicting a large reward) and ‘negative’ (predicting a small reward) cues.

Using the final active choice task design developed in Chapter 03, the learning and responses to ambiguous cues (choice of positive or negative goal-box) in common farm and laboratory breeds of pig were compared in Chapter 04. While Göttingen minipigs were initially faster to learn the discrimination between positive and negative cues, both minipigs and conventional farm pigs maintained the discrimination over time. Furthermore, while minipigs were faster at responding in general, these differences in learning and response speed were not reflected in the proportion of

choices for the positive goal box ('Optimistic' choices) when ambiguous cues were presented, demonstrating that activity, or possibly motivation, did not influence our judgement bias measure. Both groups showed increasing optimistic choices across the ambiguous cues as they neared in frequency to the positive cue. With repeated presentations of the ambiguous cues, both conventional and minipigs reduced their optimistic choices over time, suggesting that both groups quickly learned that ambiguous trials were not rewarded. Any effect of the short-term manipulation of mood state used (restraint treatment) was likely masked by this learning about the outcomes in ambiguous trials. This finding demonstrates that the judgement bias paradigm as is has a limited application for longitudinal measurement of emotional states. Subsequent designs countered this problem by rewarding ambiguous cues 'as expected'.

In Chapter 05 we explored the possibility of using other forms of biases in cognitive processes for measuring emotional valence in pigs. Here we compared decision making under two forms of uncertainty, ambiguity (Judgement Bias Task) and risk (Pig Gambling Task), in pigs. To assess the predictive validity of our tasks, responses of low-birth-weight (LBW) pigs were compared to their normal-birth-weight (NBW) siblings. LBW in humans is associated with later cognitive and emotional problems, such as deficits in learning and attention and increased risk of depression. Increased physiological responses to stress in LBW piglets suggests that they may also be at risk of emotional problems. The Pig Gambling Task was a simple two-choice task where each option differed in the amount of reward and the probability that a trial would yield a reward. While one option, termed the 'advantageous' option yielded fewer rewards per trial, the probability (risk) that a trial was unrewarded was low, resulting in an overall greater gain. LBW pigs made more choices for the advantageous option in later blocks of trials than NBW pigs in the Pig Gambling Task, and, as predicted, made fewer optimistic choices in response to the ambiguous cues in the Judgement Bias Task. No differences in discrimination learning were found. Our findings indicate that LBW pigs use different behavioural strategies when making decisions under conditions of uncertainty, they may be more likely to avoid risk and make more pessimistic judgements of ambiguous stimuli. As advantageous choices in the Pig Gambling Task can also be interpreted within the framework of rational decision making (i.e. advantageous choices lead to a greater gain), we suggest that the judgement bias paradigm may be more suitable for specifically assessing emotional valence.

In Chapter 06, the effect of prenatal treatment with Allopurinol, a potential mediator of some of the causes of long-term effects of LBW, on responses in the Judgement Bias Task was investigated. Physiological measures of acute (e.g. saliva cortisol) and chronic stress (e.g. hair cortisol) were compared between LBW and NBW pigs and with responses in the Judgement Bias Task. Both LBW and NBW pigs showed increasing optimistic choices as the ambiguous cues neared the positive cue frequency, suggesting that they were judging the ambiguous cues within the context of the reference positive and negative cues. Unlike in the previous study, LBW pigs did not make more pessimistic judgements in response to the ambiguous cues, and no effect of allopurinol treatment was observed. However, the LBW pigs also did not differ from NBW pigs in measures of chronic stress as would be expected, possibly explaining the lack of differences found in judgement bias. Moreover, neither acute nor chronic physiological measures of stress were related to responses to ambiguity in the Judgement Bias Task.

To further investigate the independence of responses to ambiguity from cognitive capacities, such as learning and memory, we compared behaviour of pigs in our Judgement Bias Task to working and reference memory measures from a free-choice maze task, the Spatial Holeboard Task, which the pigs

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had performed prior to entering into our studies. Initial comparisons using a small sample of conventional farm pigs suggested a positive correlation between reference memory and optimistic choices contrary to expectation. However, a subsequent study with more animals failed to confirm this result, suggesting that optimistic choices are independent from memory performance.

Chapter 08 sums up the findings from the thesis as a whole and presents the work in the broader context of task design and theoretical expectations. The advantages and disadvantages of the two cognitive paradigms proposed to study emotional valence in pigs are discussed.

Appendices A – D provide further background information from pilot studies on the design of the Judgement Bias and Pig Gambling Tasks, as well as posing some questions for refinement of future research.