

## **The use of sensor technology and genomics to breed for laying hens that show less damaging behaviour**

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### **Abstract**

The European COST Action GroupHouseNet aims to provide synergy for preventing damaging behaviour in group-housed pigs and laying hens. One area of focus of this network is how genetic and genomic tools can be used to breed animals that are less likely to develop damaging behaviour to their pen-mates. When focussing on laying hens, one of the main problems is that feather pecking (FP) occurs in large groups, making it difficult to identify birds performing damaging behaviour. We propose a combination of sensor technology and genomic methods to solve this issue. Research on genetic lines selected divergently on high and low FP as well as on a F2 cross established from these lines has pointed to mechanisms that may underlie this behaviour, revealing relationships between FP, fearfulness and activity levels and locating genomic markers related with FP. Birds selected for high FP were found to be less fearful and highly active in a range of tests and home pen situations. This knowledge may be used to automatically detect high feather-pecking individuals in a group setting. Research on using novel methods such as ultra-wideband tracking to detect phenotypic differences between individuals in a group is ongoing. First results confirm previously found line differences in fearfulness and activity. Future work will focus on exploring potential of other sensor-based methods to accurately measure individual phenotypes, and linking this information to

genomic markers. This should lead to the development of novel breeding methods to select against damaging behaviour in laying hens.

**Keywords:** laying hens, damaging behaviour, sensor technology, genomics, transcriptomics

## **Introduction**

Damaging behaviour, like feather pecking (FP) in laying hens, is a welfare and economic problem in commercial livestock production. There is an urgent need to reduce FP, especially due to the ban on conventional cages in the European Union and the expected future ban on beak trimming in many European countries. Several studies showed that FP can be reduced using genetic selection (Muir, 1996; Kjaer et al., 2001). FP depends on the behaviour of the individual itself and the behaviour of its group members. Associations between fearfulness and FP or feather damage have been found (Hughes and Duncan, 1972; Rodenburg et al., 2004). Several studies found that fearful chicks in an open field test showed more FP behaviour as adults (Jones et al., 1995; Rodenburg et al., 2004). Furthermore, it was found that activity of birds was related to FP behaviour (Kjaer, 2009). To select against FP, it is important to take both the genotype of the individual itself (direct genetic effect or victim effect; DGE) and the genotype of the group members into account (indirect genetic effect or pecker effect; IGE) (Bijma et al., 2007; Ellen et al., 2007). For plumage condition, it was found that DGE contribute 6-31% of the total heritable variation, while IGE contribute 70-94% of total heritable variation (Brinker et al., 2014). Together they explain 10-54% of total phenotypic variation in plumage condition. Therefore, it is important to use a selection method that takes both DGE and IGE into account (Ellen et al., 2014). Nowadays, there is a tendency to keep commercial laying hens in large groups. To identify peckers and victims in these large groups is a challenge. With the use of sensor technology, like ultra-wideband tracking, video tracking, or radio frequency identification (RFID), there is a possibility to track and trace individuals in large groups. Sensors can be used to track individual interactions in large groups, and to identify peckers and victims. Using genomic approaches, we can link the information from sensor technologies to the individual's genotype. This could also help us to identify regions or gene expression patterns and to further understand FP behaviour. Here, we propose a combination of sensor technology and genomic methods to select against FP in large groups of laying hens. In this review, we will give an overview of sensor technologies that can be used for breeding, present the first results of experiments performed in PhenoLab, describe the use of omics approaches to understand FP, and discuss the

identification of indicator traits from both technologies. In this review the focus is on FP behaviour in laying hens.

### **Use of sensor technology to inform breeding**

Researchers have developed a variety of technologies to monitor the activity and behavioural patterns of laying hens in an automated way. For instance sound analysis, image analysis and IR thermography are used to continuously monitor the activity and presence of laying hens (Lee et al., 2010; Quwaider et al., 2010; Mench and Blatchford, 2014). The major advantage of these technologies is that they are cheap (one sensor for the entire house) and non-invasive. However, they do not allow us to monitor the activity of individual birds unless individual markings are applied on the birds, which is a time consuming process that must be repeated regularly since the markings fade away after some time. In recent years, researchers have also started using accelerometers as well as RFID, ultra-wideband and geographic information system (GIS) technology where birds are equipped with a body worn sensor to monitor their individual behaviour patterns. Although these systems make it possible to monitor individual birds, they are difficult and expensive to upscale to farm size (one sensor per bird + transmitters) and often require a lot of battery power for the transmission of data (Banerjee et al., 2014; Nakarmi et al., 2014; Rodenburg and Naguib, 2014; Zaninelli et al., 2016). Furthermore, it is not certain that these body worn sensors can survive the harsh environment of the bird house during the entire life span of the laying hens.

In all this, it is important to take into account that when birds receive markings or are equipped with a sensor, which is often placed on their back, their physical appearance is changed which could make them more susceptible to receive pecking by other birds, enhancing the problem rather than solving it (Daigle et al., 2012). To date, there is no system available that can accurately monitor individual behaviour of laying hens in a real farm setting. Nevertheless, the currently existing technology can be used in research settings where the number of laying hens is small and where time efficiency is less important than in commercial conditions. According to literature, body worn sensors can reach an agreement of up to 95% between automated measurement and labelling by human observers to monitor the behaviour of individual laying hens (Nakarmi et al., 2014). This makes that this technology has potential to automatically phenotype FP behaviour in laying hens, either by recording the behaviour itself or by recording related traits. Such technology could potentially also be used by breeding companies to phenotype their selection candidates, as this is a much smaller number of birds than their crossbred offspring and generally the selection candidates are housed in smaller groups than birds in commercial flocks.

### **PhenoLab: automatic tracking of laying hens**

One possibility for automatic tracking of laying hens is by using ultra-wideband tracking. Here, hens are outfitted with an active tag in a small backpack and the location of each bird is detected by sensing beacons based on time and angle of arrival of the signal. In the PhenoLab project, we compared ultra-wideband tracking using TrackLab (Noldus Information Technology, Wageningen, The Netherlands) with video tracking of individual hens using Ethovision from the same company. As can be seen from Figure 1, both systems yielded very similar data and the ultra-wideband system was able to detect the location of the bird with an 85% accuracy.

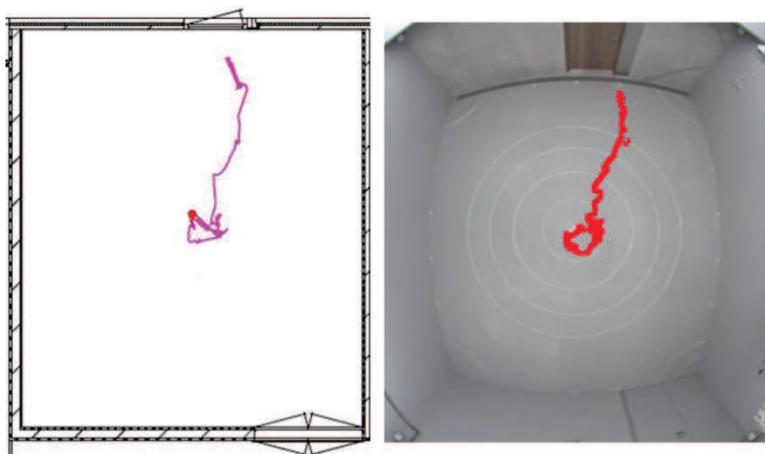


Figure 1. Tracking an individual laying hen in the PhenoLab using ultra-wideband tracking (left panel) and video tracking (right panel).

One of the major benefits of ultra-wideband tracking over video tracking is that tracking is based on a unique tag that is fixed to the individual animal. With current video tracking technology, tracking individuals in groups still seems challenging. However, recent developments in the field of video tracking seem promising (Perez-Escudero et al., 2014). We used PhenoLab to explore behavioural differences between lines selected for high and low FP. We were interested to see if we could record previously found differences in activity between the lines automatically, with the high FP line being hyperactive. This was indeed the case, with the High FP line moving almost twice the distance as the Low FP and unselected control line. Furthermore, based on the tracking data also individual differences within lines could be investigated. This showed by birds characterised as feather peckers (based on video observations) were much more active than birds characterised as victims, an observation that was very much in agreement with the line differences found. Next steps in the PhenoLab

project include measuring distances between individual animals and linking this information to relevant phenotypes (sociality, damaging behaviour). Furthermore, we would like to compare the ultra-wideband tracking method with RFID tracking, as RFID tracking might have potential as a cheaper and more robust technology that could be applied on commercial farms (Richards et al., 2011; Gebhardt-Henrich et al., 2014; Rodenburg et al., 2015).

### **Understanding feather pecking through ‘omics’ approaches**

As compared to other traits, the number of available genome-wide mapping studies for FP is sparse. Buitenhuis et al. (2003) conducted a microsatellite-based study in an F2 population established from the cross of commercial lines differing in their propensity for FP. They reported three suggestive QTL for gentle and one significant QTL for severe FP. Another group only identified a single QTL in an F2 cross of Red Jungle Fowl and White Leghorn (Jensen et al., 2005). Later, studies were conducted based on SNP genotypes. In an across-line association study using a low-density panel of approximately 1000 SNPs, Biscarini et al. (2010) analysed a direct as well as an associative effect of cage mates on feather damage, where the latter can be interpreted as the propensity to perform FP. A major finding was the implication of a serotonin receptor in FP supporting earlier evidence for a prominent role of monoamine signaling. The total number of QTL identified in this study, however, was very large. The most recent mapping studies applied the Illumina 60k SNP chip in lines divergently selected for FP (the same lines as used in the PhenoLab project) and an F2 cross of these lines. Mapping results based on selection signatures between the lines (Grams et al., 2015b) and association results from the F2 cross were jointly analyzed in a meta-study (Lutz et al., 2017) revealing 13 clusters of significantly associated markers and pointing to a candidate gene that might also be related to monoamine signaling. Available large-scale transcriptomic studies have so far been performed in chicken lines divergently selected for FP propensity applying microarray technology. In the high FP selection line, Labouriau et al. (2009) found significant gene expression differences between extreme feather peckers and other birds performing FP at a lower level. These authors proposed the presence of a single allele affecting severe pecking behaviour. Later, Hughes and Buitenhuis (2010) reported a globally reduced variance of gene expression in high FP animals and found distinct expression patterns associated with gentle and severe FP, which supports the hypothesis mentioned above. Brunberg et al. (2011) studied differential hypothalamic gene expression in FP hens, victims and controls. Their findings fitted with the hypothesis that FP is redirected foraging behavior. Wysocki et al. (2013) identified a number of candidate genes related to neurotransmission and psychopathological disorders including monoamine signaling. The available genomic and transcriptomic studies so far point to a

major role of monoamine signaling in FP, which fits well with other available data (Kops et al., 2013a; Kops et al., 2013b; Kops et al., 2014; Kops et al., 2017). Otherwise, however, little congruence is found between studies. This can be expected given the fact that FP is a complex trait. It has a heritability ranging from 0.1 to 0.4 (Kjaer et al., 2001; Rodenburg et al., 2003; Bennewitz et al., 2014; Grams et al., 2015a) and a large number of genes can be supposed to contribute to this phenotype. With respect to breeding for low FP propensity, however, a major outcome of ‘-omics’ studies would be biomarkers and indicator traits that can be applied in selection. Future research in this area will likely also focus on the metabolome and microbiome.

## **Conclusions**

We are now at a point where both sensor technology and omics approaches have the potential to provide a large amount of data at the level of the individual. If we take the example of the selection lines, selected on high and low FP: these lines have now been characterised in genomic and transcriptomic studies. These studies have added to our knowledge of the mechanism underlying FP behaviour, but can also be used to record genomic profiles of individual birds. Similarly, using sensor technology we can record an individual behavioural profile, describing activity, location and distance to other individuals. If we can combine both approaches in a breeding population, we can link the genomic data to the sensor data, and define the genomic profile of individuals that show the desired behaviour (e.g. low or no FP). This approach may be feasible, because breeding companies have begun to genotype their breeding stock routinely. Once the desired genomic profile has been defined, the method should be put to the test by breeding a next generation based on genomic selection and then phenotype this generation with the same tools that were used to phenotype the parent stock. We feel this approach has great promise to select against complex behavioural traits that involve multiple individual animals in a group, such as FP in laying hens.

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