Effects of peat as a litter material on ammonia emissions from experimental broiler houses

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REPORT 1528

WAGENINGEN
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Effects of peat as a litter material on ammonia emissions from experimental broiler houses

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Samenvatting NL
In dit rapport worden de resultaten beschreven van een onderzoek dat is uitgevoerd door Wageningen Livestock Research en Proefbedrijf Pluimveehouderij te Geel, België, in samenwerking met het Poultry Expertise Centre waarbij beoogd werd het perspectief vast te stellen van turf voor het verminderen van de ammoniakemissie uit experimentele vleeskuikenstallen, waarbij houtkrullen diende als referentie strooiselmateriaal. Andere gemeten variabelen waren onder meer persoonlijke fijnstof blootstelling, zoötechnische prestaties, strooiselsamenstelling en dierenwelzijn. Afdelingen met turf hadden een ammoniakemissie van 12,81 g dierplaats^{-1} jaar^{-1}, wat significant hoger was dan die met houtkrullen (4,75 g dierplaats^{-1} jaar^{-1}). Het strooiseltype had geen invloed op de zoötechnische prestaties (gewicht, groeisnelheid, voerconversie, uitval) en de uiteindelijke strooiselsamenstelling (totaal N, ammonium N, P, K, as, drogestofgehalte, pH). Vleeskuikens die op turf werden gehouden hadden echter minder voetzoollaesies en hakdermatitis dan vleeskuikens die op houtkrullen werden gehouden. De resultaten uit deze studie geven aan dat turf als strooiselmateriaal de ammoniakemissie uit experimentele vleeskuikenstallen niet verminderd in vergelijking met houtkrullen, waarschijnlijk omdat het minder nat en meer rul was, maar dat het mogelijk het dierenwelzijn verbeterd.

Summary UK
This report describes the results of a study conducted by Wageningen Livestock Research and Proefbedrijf Pluimveehouderij in Geel, Belgium, in collaboration with the Poultry Expertise Centre, to ascertain the potential of peat as litter material in reducing ammonia emissions from experimental broiler houses in comparison to white wood shavings (reference litter material). Other variables measured included personal dust exposure, zootechnical performance, litter quality, litter composition and animal welfare. Rooms with peat litter had an ammonia emission of (12.81 g animal place^{-1} year^{-1}), which was significantly higher than those with wood shavings (4.75 g animal place^{-1} year^{-1}). The litter material had no influence on zootechnical performance (body weight, growth rate, feed conversion ratio, mortality) and the final litter composition (total N, ammonium N, P, K, ash, dry matter content, pH). However, broilers kept on peat litter had fewer footpad lesions and hock dermatitis than broilers kept on wood shavings. The results from this work indicated that peat litter does not reduce ammonia emissions from experimental broiler houses compared with wood shavings, probably due to being less wet and more friable, but it could improve animal welfare.

This report can be downloaded for free at https://doi.org/10.18174/679212 or at www.wur.nl/livestock-research (under Wageningen Livestock Research publications).

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Foreword

The project Region Deal Foodvalley (RDFv) project works to "*accelerate the transition towards a sustainable and healthy food production system*". In this project, work package 1 aims to contribute to the development and scientific assessment of low-emission housing systems, emission-mitigating techniques, and feed and management measures that can reduce emissions of airborne pollutants from livestock barns.

This report contains the results of a semi-practical study of the potential of peat litter in reducing ammonia emissions in experimental broiler houses, with attention to personal dust exposure, zootechnical performance of the broilers, footpad lesions, hock dermatitis, litter quality and composition, in comparison to wood shavings, the mixture of wood shavings and peat, and straw pellets litter.

I would like to thank Kris De Baere and Ine Kempen from Proefbedrijf Pluimveehouderij (PP) for carrying out the experiments together and for their critical reviews of the report. Additionally, I thank Anne-Jo Smits and Jan Workamp from the Poultry Expertise Centre for the coordination and providing ideas for this study.

Albert Winkel – Project leader at Wageningen Livestock Research

Summary

In the Netherlands, peat is used regularly as litter material in broiler houses because of its alleged positive effects on animal welfare. Additionally, peat could also have an ammonia-reducing effect, given the fact that it has a relatively low pH due to humid compounds/acids produced by soil bacteria under low oxygen levels, but scientific evidence from empirical data is lacking. The aim of this study was to ascertain the effect of peat litter (in comparison with wood shavings) on ammonia emission, and on personal dust exposure, zootechnical performance, litter quality, litter composition and animal welfare. In this study, peat litter was compared with three other litter materials (wood shavings, straw pellets, and a mixture of wood shavings and peat), looking at the effects on ammonia emission, litter quality (friability and wetness), litter composition, production performance, and animal welfare (footpad lesions and hock dermatitis) of regular fast-growing broilers (Ross 308) under semi-practical conditions inside. The study comprised two 40-day production rounds with 42,880 broiler chickens from September 2022 to February 2023. Results showed that peat had ammonia emissions of 12.81 g animal place^{-1} year^{-1}, which was significantly ($P < 0.001$) higher than wood shaving (4.75 g animal place^{-1} year^{-1}). The ammonia emissions of straw pellets (9.95 g animal place^{-1} year^{-1}) and the mixture of wood shavings and peat (8.81 g animal place^{-1} year^{-1}) did not differ from peat and wood shavings. The litter material had no influence on the production performance (body weight, growth rate, feed conversion ratio, and mortality) of broilers and the final litter composition (total N, ammonium N, P, K, ash, dry matter content, and pH). However, broilers kept on peat litter had fewer footpad lesions and hock dermatitis than broilers kept on wood shavings. It can be concluded that peat as a litter material cannot reduce ammonia emissions from broiler houses compared with wood shavings, probably due to the drier and more friable litter, but it can improve animal welfare.

The ammonia emissions found in this study were much lower than common broiler houses, which may be due to the poor quality of the litter (friability and wetness). More insight is needed to investigate how the litter variables interact with each other, how they influence ammonia emissions, and how the litter should be managed.

Keywords: broiler, peat, litter materials, ammonia emission, CO_2 mass balance method, PM_{10} exposure, zootechnical performance, footpad lesions, hock dermatitis

1 Introduction

1.1 Dutch livestock context

The Netherlands traditionally has a large, knowledge-intensive and innovative agricultural sector. This sector underwent rapid development mostly from the nineteen fifties to the nineteen nineties. During this period, initially small-scaled and mixed-family farms developed along the lines of specialisation, land reparcelling, intensification, scaling up, mechanisation, automation, and robotisation. Today, the Dutch agricultural sector is the world's second biggest exporter of agricultural goods with a total value estimated at € 104.7 billion in 2021, of which 72% was Dutch produce and 28% re-export of foreign goods (Jukema et al., 2022). In 2020, the Dutch livestock sector consisted of (WUR, 2022):

- 1.6 million dairy cows at 15,700 farms;
- 476,000 dairy goats at 569 farms;
- 996,000 veal calve places at 1620 farms;
- 32 million laying hen places at 736 farms;
- 49 million broiler places at 637 farms, and;
- 12 million pig places (sows and piglets + fattening pigs) at 3557 farms.

This primary livestock sector is surrounded by an extensive periphery of schools for agricultural education, universities, veterinary practices, animal feed-producing companies, and companies active in developing housing equipment, feeding systems, ventilation systems, milking systems, air cleaners, and so on. Since the nineteen eighties, the focus on highly efficient and high-quality food production has been substantially broadened due to societal and political debate and altered visions on how to produce agricultural goods sustainably. As a result of EU and national regulations, as well as ambitions formulated by the livestock sector itself, great strides have been made in for example improving animal health and well-being, lowering antibiotic use, reducing emissions of airborne pollutants (ammonia, malodorous molecules, greenhouse gasses, dust/bioaerosols), and reducing leakage flows from nutrient cycles (e.g. nitrogen, phosphorus) to soils and waters. At the same time, however, financial margins in the livestock sector are constantly under pressure, partly as a result of the cost-increasing effects of this transition against limited possibilities of livestock farmers to pass on their additional price to the food industry and customers. The transition towards a more sustainable food production system has partly been driven by scientific research, innovation and product development, as well as the resilience and entrepreneurship of forerunner farmers in response to changing political and societal demands.

1.2 The Region Deal Foodvalley (RDFv) project

In the 'Region Deal Foodvalley' project (RDFv; 2020-2025), a broad array of organisations located in the so-called Foodvalley region[1] in the Netherlands work together to "*accelerate the transition towards a sustainable and healthy food production system*". The Foodvalley region lies roughly between the cities of Utrecht and Arnhem. It is a typical livestock and food production region, characterized by many farms with laying hens and veal calves (to a lesser extent also broilers, pigs, dairy goats, dairy cows), as well as agricultural schools, universities, animal feed producers, food industry, hospitals, et cetera. Region Deals are a type of projects launched by the third cabinet under prime minister Mark Rutte (2017-2022) in which a region in the Netherlands receives co-financing from the national government to make substantial progress on problems and challenges typical for that region. Given the presence of the livestock sector as well as related organisations, the Foodvalley region is considered an ideal 'field lab' or 'living lab' to work on the aforementioned transition of the food production system.

[1] The Foodvalley region is a framework of cooperation involving eight municipalities (Barneveld, Ede, Nijkerk, Rhenen, Renswoude, Scherpenzeel, Veenendaal and Wageningen) with altogether 350,000 residents, and many educational/scientific institutions and businesses related to agriculture and food production.

The project is divided into three major themes, of which theme 1 focusses on the transition of the primary agricultural sector. Theme 1 can be further divided into sub-projects I through IV and herein: work packages 1 through 12 (Figure 1.1).

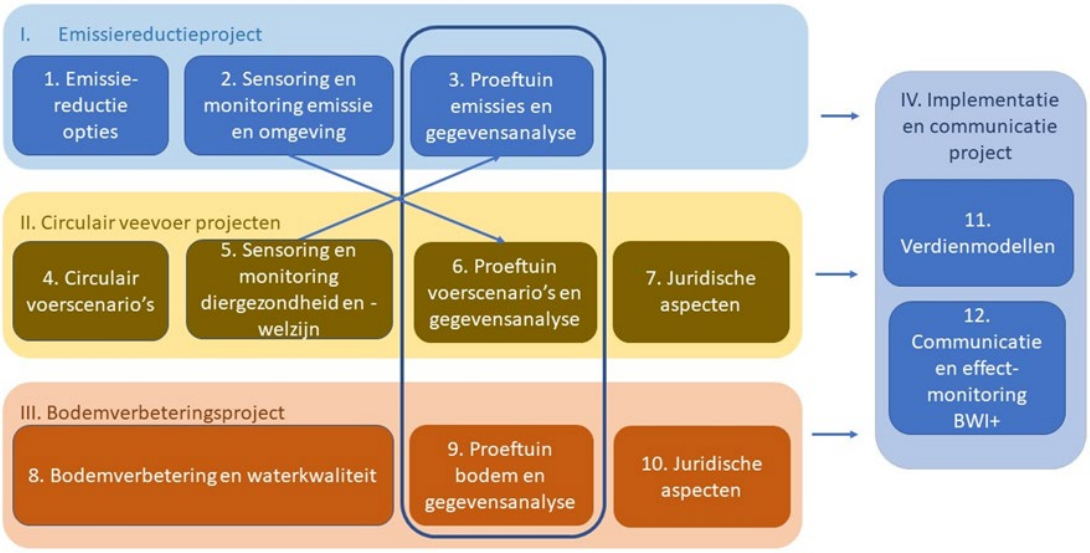


Figure 1.1 *Structure within theme 1 of the Region Deal Foodvalley project. English translation: I: Emission reduction project, II: Circular feed project, III: Soil improvement project, IV: Implementation and communication project, 1: Emission reduction options, 2: Sensoring and monitoring emissions and environment, 3/6/9: Field labs and data analysis, 4: circular feed scenario's, 5: Sensoring and monitoring animal health and welfare, 7/10: Legal aspects, 8: Soil improvement and water quality, 11: Business models, 12: Communications and effect monitoring.*

Work package (WP) 1, within project I of theme 1, aims to contribute to the development and scientific assessment of low-emission housing systems, emission mitigating techniques, and feed and management measures that can reduce emissions of airborne pollutants from livestock barns. The organisation of WP1 is carried out by the Poultry Expertise Centre (PEC, part of the agricultural school community Aeres Group; Barneveld, the Netherlands) whereas Wageningen Livestock Research (WLR; Wageningen, the Netherlands; part of Wageningen University and Research) performs the scientific work in this WP. The farmers union 'LTO Noord' sets up the field lab with affiliated livestock farms.

The assessment of emission reduction options takes place at 'field lab' (or 'living lab'; 'proeftuin' in Dutch), which is the term for all facilities (commercial farms as well as labs and experimental facilities) within the Foodvalley region, available to the project. Options for emission reduction are assessed from an integral view: options should not only reduce emissions, but also have no side effects, or even have beneficial effects, on aspects like animal health and wellbeing, closing nutrient cycles, and the earning potential of farmers. Furthermore, WP1 focuses on emission reduction solutions for the following livestock sectors: 1) poultry, 2) veal calves, 3) dairy goats, 4) pigs, and 5) dairy cows.

Companies with interesting options for emission reduction that are close to the market were asked to submit their option to the project. Subsequently, PEC carried out intakes with each applicant and produced a dossier for each option. These dossiers were then assessed by an independent Expert Team (ET), whose members are active as livestock farmers, livestock consultants, lecturers, environmental scientists, animal feed scientists, or veterinary scientists. Upon discussing the details of the dossiers, the ET issued advice to the project team on the general perspective of the submitted option. Based on this advice, the project team decided on which options are granted access to the project and its field lab.

1.3 Peat as litter substrate for broilers

One of the options submitted to the RDFv project under dossier number 28 is to use peat litter for ammonia reduction in broilers. Peat is regularly used as litter substrate in broiler houses because of its positive effects on animal welfare (less skin dermatitis, e.g. footpad lesions). Estimations are that peat (usually as a mixture of peat and wood shavings) is used as litter material in 20-30% of winter flocks in Dutch broiler farms (Jan van Harn, 2022, personal communication). Peat has a high absorbent capacity of water and a relatively low pH of 3.5 to 4.2 due to humid compounds/acids produced by soil bacteria under low oxygen levels (Agroveen, 2022a; 2002b). The latter may shift the ammonia – ammonium (NH_3 <> NH_4^+) equilibrium towards ammonium, which is not volatile, potentially reducing the evaporation of ammonia from the litter material and thus the ammonia emission from the broiler barn.

Such a working mechanism is plausible since a study on emissions from broiler barns with fresh maize/corn silage (1.75 kg/m^2), dried maize/corn silage (0.8 kg/m^2), and white wood shavings (control; 0.8 kg/m^2) as litter materials showed a statistically significant reduction of the ammonia emission of 37% for both fresh and dried corn silage in comparison to the control (Van Harn et al., 2015). Subsequently, corn silage as litter material was included as ammonia mitigation measure for broiler farms without plans to proceed their farms in the long run in the Dutch 'Programma Aanpak Stikstof' regulation (2015-2019). The working mechanism of corn silage litter is also believed to be the low pH (<4.5) as a result of organic acids such as lactic acid produced by anaerobic bacteria during the silage-making process. However, a recent study by the ILVO Institute in Merelbeke, Belgium, on peat did not show lower ammonia concentrations for peat litter in comparison to wood shavings (Vanheerenthals, 2020).

Claims made about peat litter include (Forfamers, 2021; Agroveen, 2022a, 2022b):
- Inhibition of pathogenic/zoonotic bacteria (e.g., *Salmonella spp.*, *Campylobacter spp.*, *Klebsiella spp.*, *E. coli*), a lower infectious pressure, a higher level of hygiene;
- High absorbent capacity of water;
- A lower incidence of foot pad lesions;
- Comfort for the broilers due to the softness of the material;
- Less ammonia volatilization;
- A better indoor air quality.

1.4 Aim

The Expert Team (ET) of the RDFv WP1 has judged that peat may show perspective for ammonia reduction in broiler production. However, scientific evidence from empirical data is lacking. The ET also expressed that peat is a fossil and finite product, often harvested in the Balkan states and transported to the Netherlands, which are unsustainable peat properties for the long term. Ideally, for the long term, artificial acidification of conventional litter materials could be a sustainable alternative for peat. The ET advised gaining evidence for the relationship between the pH of acidic litter materials (peat, corn silage) and ammonia emission, as well as exploring whether artificially acidified litter material could be a sustainable substitute for peat (and corn silage). The latter, however, is not part of this work. The aim of this study was to ascertain the effect of peat litter (in comparison with wood shavings) on ammonia emission, and on personal dust exposure, zootechnical performance, litter quality, litter composition and animal welfare.

2 Methodology

2.1 General study design

The study was conducted for two production rounds of 40 days in two experimental broiler barns of the PP Geel in Belgium from September 2022 to February 2023. In each round, 42,880 day-old fast-growing broilers (Ross 308) were housed with a density of 20.4 animals/m^2. Four types of litter materials were applied across the rooms (randomly assigned): wood shavings (WS, control, 1.0 kg m^{-2}), peat (PE, 1.5 kg m^{-2}), straw pellets (SP, 1.5 kg m^{-2}), and the mixture of wood shavings and peat (WS+PE, 0.5 kg m^{-2} + 0.75 kg m^{-2}). One barn (hereinafter referred to as barn ABCD) consisted of four mechanically ventilated rooms (A, B, C and D), whereas the other barn (hereinafter referred to as barn E) consisted of eight mechanically ventilated rooms (E1 to E8). All rooms are climate-separated. A detailed description of the two barns and their rooms can be found in section 2.2. In barn ABCD, only peat and wood shavings were compared (two rooms * two rounds = four replicates per litter type). In barn E, all four litter materials were compared (two rooms * two rounds = four replicates per litter type). Therefore, wood shavings and peat were replicated eight times in total, while straw pellets and the mixture of wood shavings and peat were replicated four times. The allocation of litter types across the rooms is shown in Table 2.1. A summary of all variables measured in these rooms is given in Table 2.2.

Table 2.1 *Allocation of litter types across the rooms.*

Room / Round	A	B	C	D	E1	E2	E3	E4	E5	E6	E7	E8
1	WS	PE	WS	PE	SP	PE	WS	WS+PE	SP	WS	WS+PE	PE
2	PE	WS	PE	WS	WS+PE	WS	PE	SP	PE	WS+PE	SP	WS

Table 2.2 *Summary of measured variables.*

Variable	Measurement method	Unit	Day[1]
Aerial concentrations and emission			
Ammonia (NH_3)	Wet chemical method; Dräger Polytron P8100 sensor; Fourier Transform Infrared Spectroscopy (FTIR) analyser (Gasmet CX4000)	ppm	Continuously[2] 25, 31 (1st round); 11, 18, 25, 34 (2nd round)[3]
CO_2	Lung method; Vaisala GMP252 (0-5000 ppm) sensor; FTIR analyser (Gasmet CX4000)	ppm	Continuously[4] 25, 31 (1st round); 11, 18, 25, 34 (2nd round)[5]
Personal PM_{10} exposure	DustTrak™ II Aerosol Monitor	mg m^{-3}	21, 25, 28, 32, 35 (1st round); 11, 18, 22, 25, 34, 36 (2nd round)
Environmental condition			
Air temperature	Vaisala HMP60 sensor for barn ABCD; Fancom temperature sensor standard for barn E	°C	Continuously
Relative humidity	Vaisala HMP60 sensor for barn ABCD; Fancom RV sensor RHM.2 / RHO.2 for barn E	%	Continuously
Ventilation rate	Fan-wheel anemometers (Fancom ATM35 and ATM80), CO_2 mass balance method	m^3 h^{-1}	Continuously
Bird performance			

Mortality	Visual inspection, counting	%	Daily
Body weight	Automatic weighing device	g	Daily
Body weight (weighing of all birds delivered to slaughterhouse)	Manual weighing	g	40
Feed consumption	Feed weighing system	kg d^{-1} $room^{-1}$	Daily
Water consumption	Water meter	L d^{-1} $room^{-1}$	Daily
Water: feed ratio	Calculation	-	-
Growth rate	Calculation	g d^{-1}	-
Feed conversion ratio	Calculation	-	-
Footpad lesions	Visual observation, scoring	Score	31, 39 (1st round); 32, 39 (2nd round)
Hock dermatitis	Visual observation, scoring	Score	31, 39 (1st round); 32, 39 (2nd round)
Litter characteristic			
Dry matter content	Oven-drying, weighing	g kg^{-1}	Weekly
Total nitrogen content	NEN-7434	g kg^{-1}	40
Ammoniacal content	NEN-7438	g kg^{-1}	40
P content	NEN-7435	g kg^{-1}	40
K content	NEN-7436	g kg^{-1}	40
Ash content	NEN-7432	-	40
pH	pH meter	-	Weekly
Thickness	Meter	cm	Weekly
Floor and litter temperature	Infrared thermometer	°C	Weekly
Friability	Visual observation, scoring	Score	6, 13, 20, 27, 34, 38 (1st round); 14, 21, 28, 35, 39 (2nd round)
Wetness	Visual observation, scoring	Score	6, 13, 20, 27, 34, 38 (1st round); 14, 21, 28, 35, 39 (2nd round)

[1] Day: numbers refer to measurement days for two production rounds, otherwise: measurement frequency.

[2] This frequency applies to the Dräger Polytron P8100 sensor and the FTIR Gasmet CX4000 analyser.

[3] Measurement days apply for the wet chemical method.

[4] This frequency applies to the Vaisala GMP252 (0-5000 ppm) sensor; and FTIR Gasmet CX4000 analyser.

[5] Measurement days apply for the lung method.

2.2 Description of the experimental broiler barns

Figure 2.1 shows the location of the two broiler barns. Both barns were heated via central heating with delta tubes. The overall number of climate-separated rooms was 12.

Barn ABCD is indicated by the red frame in Figure 2.1 and consists of two buildings, each with two climate-separated rooms (four rooms in total), equipped with roof ventilators. Each room measured 300 m^2 (length 18.75 m, width 16 m) and was divided into four pens of 75 m^2 separated by fences. Each room has in total 32 air inlets (2*16) in the side walls. Eighteen oval feed pans (Haikoo™; Roxell; Maldegem, Belgium) and 96 drinking nipples (I-flex 15TM; Impex; Barneveld, the Netherlands) per pen were equipped in barn ABCD. The picture of barn ABCD is shown in Figure 2.2 and the layout is shown in Figure 2.3.

Barn E was located in the building indicated by the yellow frame in Figure 2.1, which consists of eight climate-separated rooms from E1 to E8. Each room measured 112.5 m^2 (length 18.75 m, width 6 m) and is divided into 2 pens of 56.25 m^2, separated by fences. Each room had in total 11 air inlets in the side walls.

Fourteen oval feed pans (Haikoo™; Roxell; Maldegem, Belgium) and 78 drinking nipples (Swii'Flo™; Roxell; Maldegem, Belgium) per pen were equipped in barn E. Barn E was equipped with longitudinal ventilation with the option to precondition the incoming air. There are two exhaust towers on the other side, which are used to extract the air using fans (Fancom, ATM35 and ATM80). Pictures of barn E are shown in Figure 2.4 (inside the rooms, working passage, outside the barn). An FTIR measurement system is installed in barn E as shown in the layout of barn E in Figure 2.5.

Figure 2.1 *Barn ABCD consists of two buildings with two climate-separated rooms; four rooms in total (A, B, C, D, indicated by red frame). Barn E consists of one building with eight climate-separated rooms (E1 through E8, indicated by a yellow frame). The overall number of climate-separated rooms therefore is 12.*

Figure 2.2 *Photos of barn ABCD (inside the climate-separated rooms).*

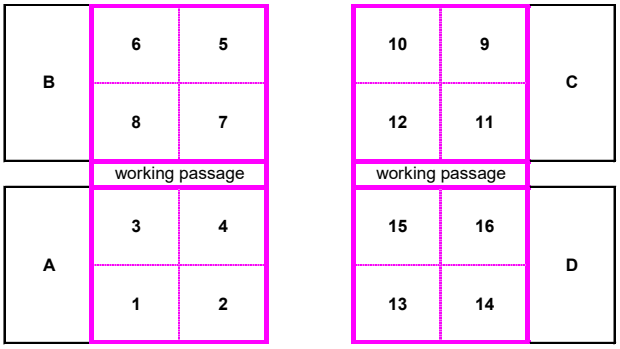


Figure 2.3 *Layout of barn ABCD. Serial numbers of pens are marked. Pink solid lines represent the border of the climate-separated rooms and pink dashed lines represent the separation of the pens in each climate-separated room.*

Figure 2.4 *Photos of barn E (inside the climate-separated rooms, outside the barn, and working passage).*

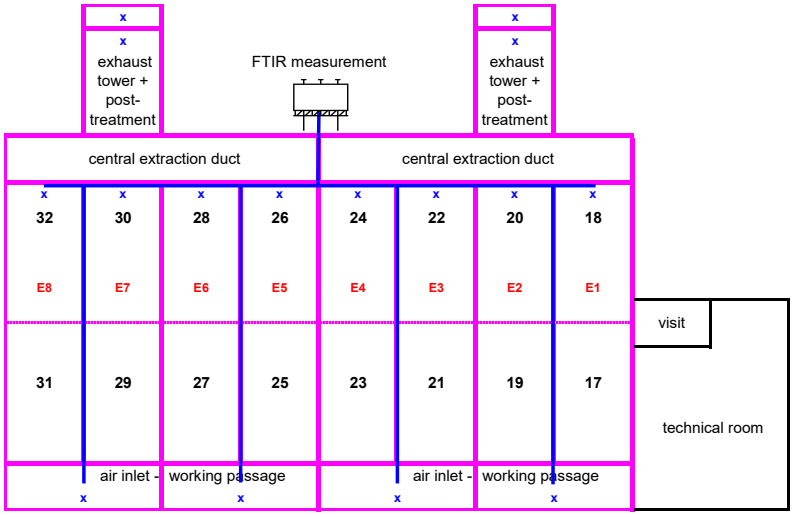


Figure 2.5 *Layout of barn E. Serial numbers of the pens are marked. Pink solid lines represent the border of the climate-separated rooms and pink dashed lines represent the separation of the pens in each climate-separated room. Blue x represents the sampling points of the FTIR analyser. Blue lines show the FTIR connections.*

2.3 Animals and management

The overall management followed the brochure by Proefbedrijf Pluimveehouderij (2016). In barn ABCD, 6,120 one-day-old broilers (Ross 308, as hatched) were placed per room (24,480 broilers in total) and in each room of barn E, 2,300 were placed (18,400 in total). The broilers originated from a parent stock of 48 (round 1) and 42 (round 2) weeks of age and were obtained from a commercial hatchery (Belgabroed, Merksplas, Belgium).

Broilers were delivered to the slaughterhouse at 40 days of age, with a live end weight of approximately 2.6 to 2.8 kg. Broilers had *ad libitum* access to drinking water and feed. Thinning was done on day 33 by taking out 1,280 (20.9%) broilers from each room of barn ABCD and 560 (24.3%) from each room of barn E for round 1 (estimated body weight around 2.1 kg; male/female were taken randomly), and 1,280 (20.9%) from each room of ABCD and 576 (25.0%) from each room of barn E for round 2.

A four-phase diet program was provided: starter diet (crumbled feed) from day 0 to day 9, growing diet A (crumbled feed for round 1 and pelleted feed for round 2) from day 10 to day 17, growing diet B (pellets) from day 18 to day 30, and finisher diet (pellets) from day 31 to the end. Growing and finisher diets were complement diets based on feeding 10%, 25%, and 35% whole wheat in growing A, growing B and finisher phase, respectively. From day 8, whole wheat was added to the feed according to the following schedule: 5% wheat from day 8 to day 9; 10% wheat from day 10 and gradually increased to 20% on day 17; 25% wheat from day 18 and gradually increased to 34% on day 29; 35% wheat from day 30 and increased gradually to 39% on day 38. A percentage of 25% of wheat was maintained on days before thinning and slaughter.

The broilers were vaccinated against Newcastle Disease and Infectious Bronchitis first at one day of age at the hatchery via spray; then again on day 15 via drinking water; and against Gumboro Disease on day 21 via drinking water. No other medications were used in either round.

The light regime was 24L:0D on the first day upon arrival, 8L:3D:10L:3D from day 1 to day 5, 1D:3L:4D:3L:1D:12L from day 6 to day 36, and 23L:1D from day 37 to day 40. The light intensity was 20 lux on the first 2 days, 15 lux on day 3-4 and 36-40, and 10 lux from day 5 to day 35.

All rooms were pre-heated to 32 ^{o}C three days before the broilers arrived to ensure sufficient dryness of the litter materials (around 85%), especially for peat. Ambient temperature was decreased gradually from 34 ^{o}C upon arrival to 20 ^{o}C until day 40. The minimum ventilation rate was set at 1.5 $m^3\ h^{-1}$ per kg live weight on day 0 and gradually decreased to 0.7 $m^3\ h^{-1}$ per kg live weight on day 20, until the end of the trials. The ventilation rates increased automatically by a climate computer based on the expected weight development/gain and climate inside. When a production round ended, litter was removed and barns were fully cleaned (wet) and disinfected. No additional cleaning or refill of the litter materials was performed during the trials.

2.4 Measurements of aerial variables

2.4.1 Ammonia (NH_3) concentration

Ammonia concentrations were measured in three ways:

- Wet chemical method (= Standard Reference Method, SRM)
  The ammonia concentration of the incoming air (background) and the outgoing/exhaust air of rooms E3 and E4 of barn E were measured using the wet chemical method. The wet chemical method with acid traps was used as reference, following the protocol described by Mosquera et al. (2019). A series of impingers (three sets, all in duplicate) were connected with pumps (model 617CD32, Thomas Industries Inc., Wabasha, Minnesota, USA). The first two impingers contained 100 mL 0.05 M nitric acid (HNO_3) solution. The flow rate was limited to 1 L min^{-1} by a critical orifice and was checked before and after the 24-hour sampling period by a calibrated flow meter (DryCal® Defender 510-m, Bios Int. Corp, Lakewood, USA). Impingers were transported to the laboratory for ammonium nitrogen content analysis using spectrophotometry. Sampling points included outside (background air of both barns), E3, and E4. Sampling took place on day 25 and 31 in round 1, and day 11, 18, 25, and 34 in round 2 (Table 2.2).
- Ammonia concentrations (ppm) at the exhaust were measured continuously throughout both rounds by electrochemical sensors (Dräger Polytron P8100, Lübeck, Germany) in rooms A, B, C, D, E3, E4. Prior to the measurements, sensors were calibrated in the Air Quality Lab of WLR by offering a dilution series of ammonia gas to the sensors. Regression lines were used to convert signals to the actual ammonia concentrations (ppm). During measurements, sensors were wired to Koenders dataloggers (CR1000X; Campbell Scientific Inc.; Logan UT, USA) and data was stored as minute-averages.

- The ammonia concentrations (ppm) in barn E (rooms E1-E8) were measured by an FTIR analyser (Gasmet CX4000, Gasmet Technologies, Vantaa, Finland).
- Daily zero-point calibration was performed and the analyser was regularly checked with calibration gases. Each channel carried out three measurements (60 s) and a sufficient rinsing time of 90 s was reserved between channels (4.5 minutes per measurement). Data of the measurements with the FTIR analyser was stored in a txt file with 1 line per measurement per measuring channel.

To express variables in a uniform standard, the ammonia concentrations of the Dräger sensors and the FTIR analyser were calibrated to the wet chemical method values. The application of the three measurement methods for ammonia across the rooms are summarized in Table 2.3.

Table 2.3 *Application of the three measurement methods for ammonia across the rooms.*

Method \ Room	A	B	C	D	E1	E2	E3	E4	E5	E6	E7	E8	outside
Wet chemical							Yes	Yes					Yes
Dräger	Yes	Yes	Yes	Yes			Yes	Yes					
FTIR					Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

2.4.2 Carbon dioxide (CO_2) concentration

Carbon dioxide (CO_2) concentrations (ppm) were measured in three ways:
- The lung method was used as standard reference method (Mosquera et al., 2020). The air samples were analysed in the WLR lab by gas chromatograph (GC 8000, Interscience/Carlo Erba Instruments Inc., Breda, the Netherlands). Sampling points included outside (background air), and outgoing/exhaust air of rooms E3 and E4. The sampling took place on day 25, 31 in round 1, and day 11, 18, 25, 34 in round 2 (Table 2).
- The carbon dioxide (CO_2) concentrations in barn ABCD, E3, E4 were measured continuously by a CARBOCAP® Carbon Dioxide Probe with a measurement range of 0 – 5000 ppm (Vaisala GMP252, Vaisala GmbH, Vantaa, Finland). Sensors were calibrated in the same way as described in section 2.4.1 (using carbon dioxide as calibration gas) and connected to the same datalogger in the same way mentioned above. Outside barn ABCD and barn E, background concentrations of carbon dioxide were measured by the same sensor mentioned previously but with a measurement range of 0 – 2000 ppm.
- The carbon dioxide (CO_2) concentrations in barn E (rooms E1 - E8) were measured by the FTIR analyser as well. Details of measurements are the same as described in section 2.4.1.

To express variables in a uniform standard, the CO_2 concentrations of the Vaisala sensors and the FTIR analyser were calibrated to the lung method values.

2.4.3 Air temperature and relative humidity

The air temperature (°C) and relative humidity (%) of barn ABCD were monitored constantly by combined sensors (Vaisala HMP60; Vaisala GmbH, Vantaa, Finland). Data was stored in Koenders datalogger every minute (CR1000X, Campbell Scientific, Logan UT, USA), and daily mean values were calculated. The air temperature (°C) and relative humidity (%) of barn ABCD and E were monitored by Fancom temperature sensor standard every 5 minutes. In barn ABCD, sensors were hung around 3.5 m high next to the ventilator outlet, while in barn E it was hung around 1.0 m at the outlet.

2.4.4 Personal dust (PM_{10}) exposure

The exposure of workers to PM_{10} was determined in round 1 on day 21, 25, 28, 32, 35, and in round 2 on day 11, 18, 22, 25, 34, 36 for all rooms (Table 2). A DustTrak™ II Aerosol Monitor (model 8530, TSI Inc., Shoreview MN, USA) was suspended on the shoulder of a researcher at a height of approximately 1.5 m, who then mimicked a 10-minute standard inspection through each room. When carrying out measurements, the researcher was wearing a mask to prevent breathing into the inlet. Concentrations of PM_{10} were measured every second, and minute averages were logged in the memory of DustTrak.

2.5 Litter variables

2.5.1 Visual friability and visual wetness

Litter quality in each room was assessed weekly by a panel of two persons. Friability and wetness were scored visually based on Table 2.4, on a 1 - 10 point scale.

Table 2.4 *Score table of litter quality assessment and the description of each score (van Harn et al., 2017)*

Score	Friability description	Wetness description
1	Complete caked litter	Wet litter, by pressure on the litter water is appearing in the total area
2	80-90% of the area is caked	Wet litter, by pressure on the litter water is appearing beneath the drinking line
3	70-80% of the area is caked	Wet litter, by pressure on the litter no water is appearing beneath the drinking line
4	60-70% of the area is caked	Wet litter, dark-coloured, litter can be pressed ball-shaped
5	50-60% of the area is caked	Wet litter, dark-coloured, ridges beneath the drinking line
6	40% of the area is caked	Almost dry litter, small ridges beneath drinking line. Litter between drinking line and feeders is still friable
7	30% of the area is caked	Almost dry litter, dark-coloured beneath drinking line and in other areas light-coloured, ridge formation beneath drinking lines just started
8	10% of the area is caked	Almost dry litter, light-coloured, no ridges beneath drinking line
9	Friable litter, some litter particles are caked	Dry litter, light-coloured
10	Friable litter, no caked litter particles	Very dry litter

2.5.2 Litter sampling and composition analysis

Litter samples were taken weekly (100 g litter per sample for day 6-7, 13-14 and 250 g for day 20-21, 27-28, 34, 38-39) according to the standard protocol (Appendix 1) of PP Geel for dry matter determination. Three samples per pen (near a drinking line, near a feeding line and in-between the two) were taken in eight out of 16 pens in barn ABCD and 16 pens in barn E (24+48=72 samples per measurement day). Samples were oven-dried at 105 °C for 24 h to determine the dry matter content of the litter (weighed before and after oven-drying). At the end of each round, representative samples of the litter were taken according to the protocol of PP Geel (Appendix 1). In contrast to the weekly dry matter samples, the 6 - 8 samples per room were pooled into one representative sample per room (= treatment). These pooled samples were frozen at -20 °C and transported to the lab for analysis on:

- Dry matter content (g/kg)
- Ash content
- Total N-content (g/kg)
- Ammonium-N (g/kg)
- P-content (g/kg)
- K-content (g/kg)
- pH

The samples were treated and tested following the standards provided by the Royal Netherlands Standardization Institute (NEN). Figure 2.6 shows the treatments in turn and the protocols followed. Each sample was divided into a few parts in order to finish all the analyses. To measure pH value, 50 g of the samples was treated following NEN 6411 and then measured by a pH meter. Samples were then pre-treated following NEN 7431. Dry matter and ash were measured following NEN 7432.

The rest of the samples underwent a destruction process with acid following NEN 7433. Total N, ammonium N, P, K were determined based on the processes of NEN 7434, NEN 7438, NEN 7435, NEN 7436, respectively.

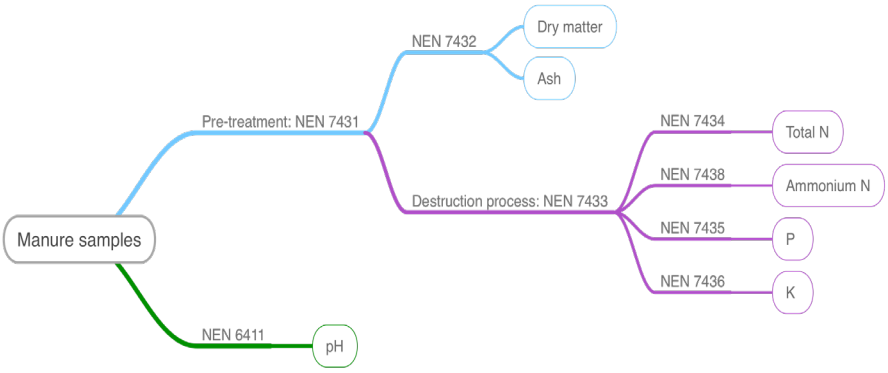


Figure 2.6 *Treatments of litter samples and the corresponding standards.*

2.6 Animal variables

2.6.1 Zootechnical performance

Mortality, feed consumption and water consumption were recorded daily. The body weight of the broilers was measured automatically via a weighing device/system. This measurement was checked by the result of manual weighing every week (50 birds were randomly chosen per pen on day 7, 21 and 75 birds on day 32, 39). The following variables were determined per pen (see Table 2.2):

I total feed consumption (g);
II total water consumption (mL);
III water: feed ratio (II divided by I);
IV average end weight (g);
V total growth (g, IV minus the start weight of one-day-old chicks);
VI growth rate (g/d, V divided by the number of days in the production round);
VII feed conversion ratio (I divided by V).

2.6.2 Footpad lesions and hock dermatitis

Occurrence and severity of footpad lesions and hock dermatitis were determined before thinning (day 31 of the first round and day 32 of the second round) and slaughter (day 39) by an experienced assessor according to the Welfare Quality (WQ) assessment protocol for poultry (see Appendix 2, Welfare Quality®, 2009). Footpad lesions and hock dermatitis were scored on a scale from 0 (no lesions) to 4 (severe lesions on the foot or hock). Fourty animals (20 males/20 females) per pen (in total 24 pens, 8 pens from barn ABCD and 16 pens from barn E) were randomly chosen for assessment. For footpad lesions, the footpad score (FPS) was also calculated according to the following formula: 100% × ((0.5 × the total number of birds with score 1+2) + (2 × the total number of birds with score 3+4))/the total number of scored birds. The footpad score (FPS) can range from 0 (all birds having no lesions) to 200 (all birds having score 3 or 4) per flock.

2.7 Data processing and statistical analysis

2.7.1 Calculation of the ventilation rate

The ventilation rates in barn E were determined directly by fan-wheel anemometers (ATM35 and ATM80, Fancom BV, Panningen, the Netherlands) as the standard reference method.

The signals were logged in a data storage box as five-minute averages. The anemometers were calibrated by a calibrated impeller. The rotations per minute (RPM) of the two impellers were recorded at different ventilation rates (>10) where calibrated lines were derived from. The calibration curves of measurement units were drawn up according to international standards.

The ventilation rates in barn ABCD as well as in barn E were calculated by the CO_2 mass balance method, which uses CO_2 produced by the animals and their manure as a natural tracer gas. The CO_2 production (PCO_2; $m^3\ h^{-1}$ animal^{-1}) is calculated based on the total heat production φ_{tot} (W) of the animals. For broilers, PCO_2 (barn level, 24-hour average) can be estimated by the following equations (Pedersen et al., 2008; CIGR, 2002):

$$PCO_2 = 0.18 * 10.62 * m^{0.75} * 0.001, m < 0.5 \quad (1)$$
$$PCO_2 = 0.185 * 10.62 * m^{0.75} * 0.001, m > 0.5 \quad (2)$$

where:

- m = the body mass of a broiler (kg).

The ventilation rate V ($m^3\ h^{-1}$; 24-hour average; on the level of a room) is further calculated as shown in Equation 5 (Pedersen et al., 2008; CIGR, 2002):

$$V = \frac{PCO_2 * number\ of\ animal}{([CO_2]_{outlet} - [CO_2]_{inlet}) * 10^{-6}} \quad (3)$$

where:

- PCO_2 = CO_2 production ($m^3\ h^{-1}$ animal^{-1}) at the barn level;
- $[CO_2]_{outlet}$ = CO_2 concentration at the outlet of the barn (ppm);
- $[CO_2]_{inlet}$ = CO_2 concentration at the inlet of the barn (ppm).

Ventilation rates in barn ABCD were calibrated to fan-wheel anemometer values.

2.7.2 Calculation of ammonia emission rates

The ammonia emission rate E (g per animal place per year) was calculated by the following equation:

$$E = ([NH_3]_{outlet} - [NH_3]_{inlet}) * \beta * V' * \frac{24\ hours}{1\ day} * \frac{365\ days}{1\ year} * \frac{1g}{1000mg} * 0.82 \quad (4)$$

where:

- $[NH_3]_{outlet}$ = ammonia concentration at the outlet of the barn (ppm)
- $[NH_3]_{inlet}$ = ammonia concentration at the inlet of the barn (ppm)
- β = coefficient of conversion from ppm to mg m^{-3} for ammonia gas at a certain temperature
- V' = ventilation rate ($m^3\ h^{-1}$ animal place^{-1})
- 0.82 (40/49) = vacancy factor

2.7.3 Statistical analysis

The experimental unit was per room. All measurements were considered statistically independent. The statistical analyses were performed using R (version 4.4.1; R Core Team, 2021) and Genstat (VSN, 2022). Litter material types were identified as the main effect and the barn types as the random effect. The interactions between treatments (litter materials) and barns were tested for significant differences as well. The normality of the data was checked with the Shapiro-Wilk test. When the data was normally distributed, unbalanced one-way ANOVA was performed to test for significant differences. When a variable was not normally distributed, the data was transformed to the natural log scale with base *e*, and then analysed by unbalanced one-way ANOVA. In case of a significant effect, the Bonferroni post-hoc test was used to determine the significant differences between groups. Results were shown as MEAN (SE) without transforming to log scale. *P*-values for effects on not normally distributed variables were based on the analyses on log-transformed data.

Data of the following variables were tested:

- Zootechnical performance as listed in section 2.6.1, variables I through VII;
- air temperature;
- relative humidity;
- CO_2 concentration;
- ventilation rate;
- ammonia concentration;
- ammonia emission;
- litter quality and composition mentioned in section 2.5.1 and section 2.5.2;
- PM_{10} concentration;
- footpad lesion score;
- hock dermatitis score.

For the measurement methods of ventilation rate (fan wheel anemometry (SRM), CO_2 mass balance method), CO_2 concentration (lung method and GC analysis (SRM), Vaisala sensor, FTIR analyser) and ammonia concentration (wet chemical method (SRM), Dräger sensor, FTIR analyser), linear regression analysis was performed with the SRMs on the Y-axis (dependent) variable and the candidate method on the x-axis. The functions were used to calibrate values measured by the alternative method to the SRM values. Two-tailed paired samples *t*-tests were used to test the intercept of the regression lines against zero, and the slopes against one to check the differences between the CO_2 concentrations measured by the lung method, Vaisala sensor, FTIR analyser; the ventilation rates measured by the CO_2 mass balance method and fan anemometers; and ammonia concentrations measured by the wet chemical method, Dräger sensor, FTIR analyser. All differences were declared statistically significant at a *P*-value < 0.05.

3 Results

3.1 Environmental conditions

Table 3.1 shows the air temperature and relative humidity (RH) of the indoor and outdoor environment of the two rounds. The outdoor air temperature ranged from - 0.9 °C to 17.2 °C and the outdoor RH ranged from 47.9% to 94%. The outside temperature and RH of the first (13.0 °C and 77.8%) and second round (4.9 °C and 82.0%) match the long-term (2022-2023) average weather data during the study: 11.8 °C and 80% for the period of round 1, and 3.6 °C and 83% for the period of round 2. For both rounds, there were no significant differences in room temperature and RH between litter materials. However, barn ABCD had higher temperatures than barn E in both rounds., while barn E had higher RH than barn ABCD in round 1. One possible reason could be the different heights of the locations where the sensors were hung.

Table 3.1 *Mean (SE) temperature and relative humidity of the outdoor air and the indoor; the latter per barn and treatment.*

Variable	**Round**	**Outside**	**Barn**		**Treatment**				***P*-value**		
			ABCD (n=8)	E (n=16)	WS (n=4)	PE (n=4)	SP (n=2)	WS+PE (n=2)	Litter material	Barn	Litter material *barn
Air temperature (°C)	1	13.0 (0.4)	26.7 (0.1)[a]	25.9 (0.04)[b]	26.1 (0.2)	26.2 (0.3)	25.9 (0.2)	25.9 (0.07)	0.013[1]	<0.001	0.276
	2	4.9 (0.5)	26.2 (0.1)[a]	24.9 (0.07)[b]	25.4 (0.4)	25.3 (0.4)	24.8 (0.1)	24.9 (0.2)	0.033[1]	<0.001	0.803
Relative humidity (%)	1	77.8 (1.5)	57.3 (1.1)[b]	60.7 (0.8)[a]	60.5 (1.2)	59.5 (1.2)	59.3 (1.6)	59.7 (1.6)	0.942	0.043	0.876
	2	82.0 (1.0)	50.5 (1.2)	53.7 (0.9)	52.8 (1.3)	53.3 (1.3)	53.4 (1.7)	51.4 (1.7)	0.846	0.064	0.889

[a-b] Values between barns and litter materials with different superscripts have significant differences ($P<0.05$).

[1] Bonferroni test did not show any significant differences.

3.2 Ventilation rate

Figure 3.1 (upper) shows the linear regression fit for the CO_2 concentrations in rooms E3 and E4, measured by the lung method (x, reference), FTIR analyser (y_1, in blue), and Vaisala sensor (y_2, in red). The two-tailed *t*-test showed that there were significant differences between both FTIR analyser ($P_{intercept} < 0.05$, $P_{slope} < 0.05$), Vaisala sensor ($P_{intercept} < 0.05$, $P_{slope} < 0.05$) and the lung method. The CO_2 concentrations of all rooms in further context (measured by the FTIR analyser and Vaisala sensors) were corrected to the lung method values based on the regression lines shown in Figure 3.1 (lower).

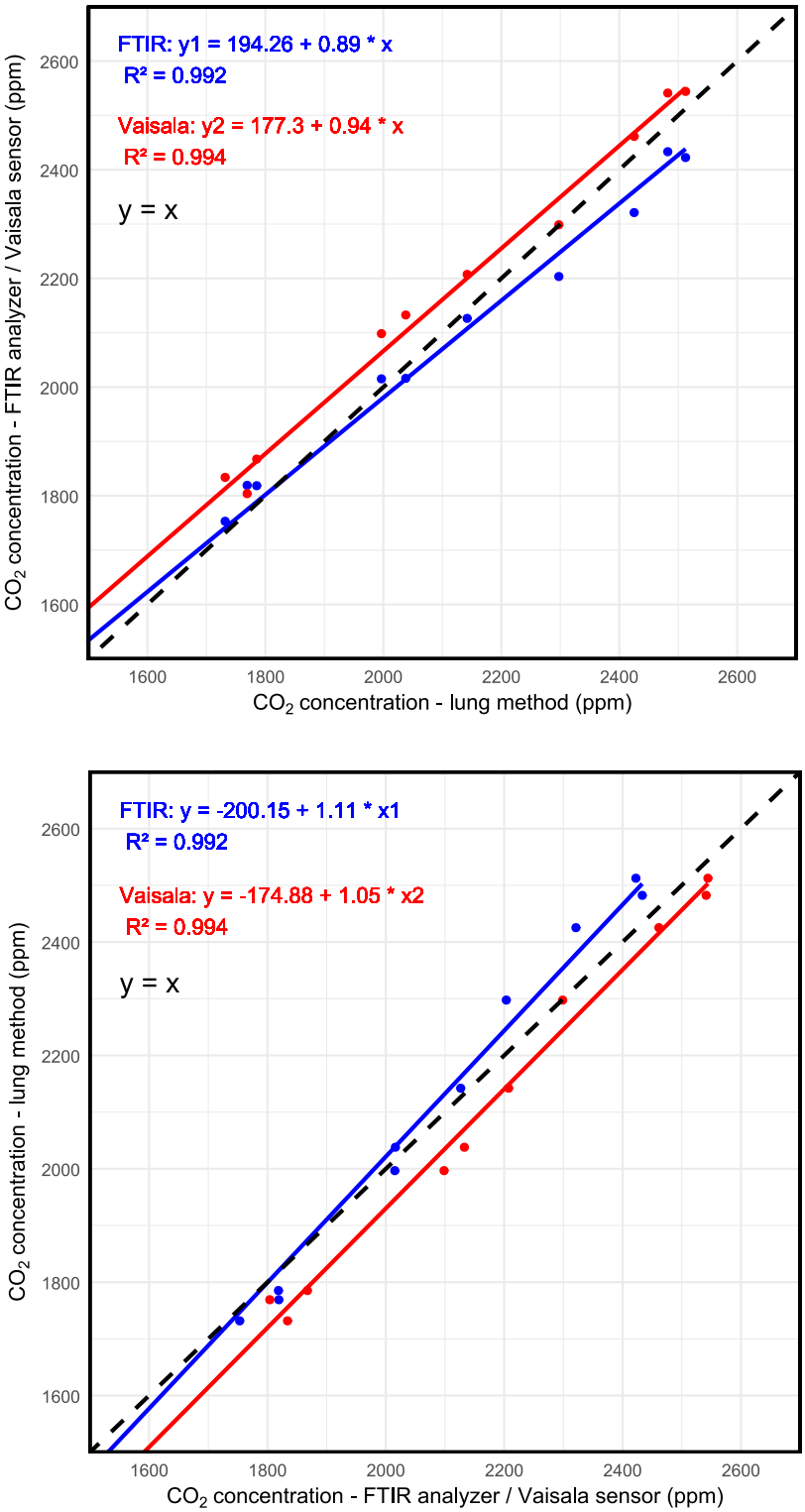


Figure 3.1 *Upper: Linear regression fit for CO_2 concentrations (ppm) measured by the lung method (x), FTIR analyser (y_1, in blue), Vaisala sensor (y_2, in red). Lower: Linear regression fit for CO_2 concentrations (ppm) measured by FTIR analyser (x_1, in blue), Vaisala sensor (x_2, in red) and the lung method (y).*

Figure 3.2 (upper) shows the linear regression fit for ventilation rates in rooms E3 and E4, measured by fan-wheel anemometer (x, reference) and the CO_2 mass balance method (y). The two-tailed *t*-test showed that the ventilation rates calculated by the CO_2 mass balance method were significantly higher than the reference ($P_{intercept} < 0.05$, $P_{slope} < 0.05$). The regression model can describe 97% of the variation between the two methods. In further calculations, ventilation rates in barn ABCD were corrected by the anemometer values based on the regression line shown in Figure 3.2 (lower).

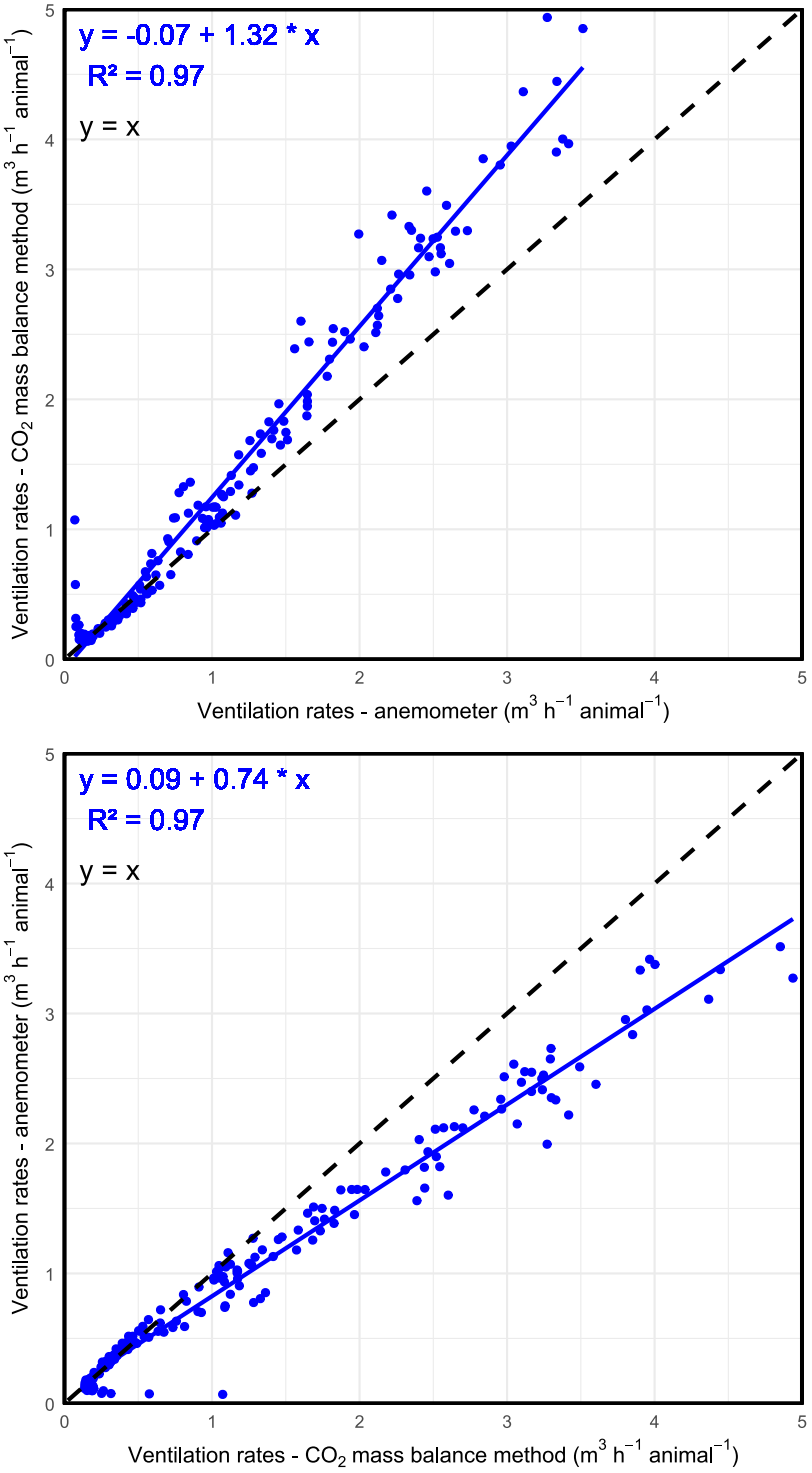


Figure 3.2 *Upper: Linear regression fit for ventilation rates (m^3 h^{-1} $animal^{-1}$) measured by the anemometer (x) and the CO_2 mass balance method (y). Lower: Linear regression fit for ventilation rates (m^3 h^{-1} $animal^{-1}$) measured by the CO_2 mass balance method (x) and the anemometer (y).*

Figure 3.3 shows the ventilation rates (m^3 $hour^{-1}$ $animal^{-1}$) per treatment and room for both rounds. The ventilation rates went up gradually over the course of a round as body weights and heat production increased. From day zero to two, barn ABCD had unusually high ventilation rates, mostly likely caused by the sensitivity of the CO_2 mass balance method at the low delta CO_2 concentrations (inside minus outside) at the beginning of the round. Therefore, the values of day zero to two were replaced by values obtained from extrapolation using regression lines based on the data points of day three to ten. All rooms showed similar trends. The different trends of rounds 1 and 2 could be related to differences in outside temperature between those periods.

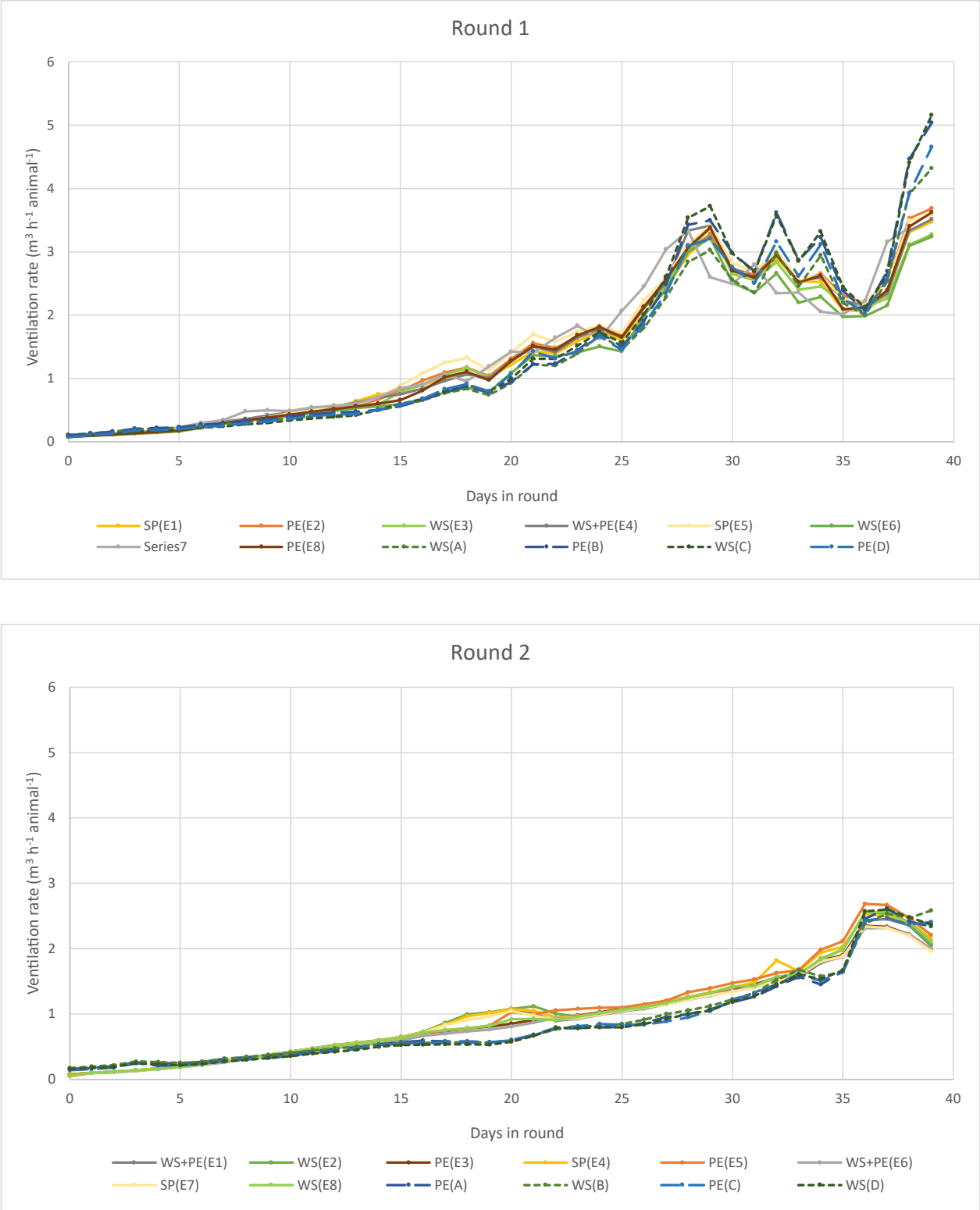


Figure 3.3 *Ventilation rates (m^3 $hour^{-1}$ $animal^{-1}$) of round 1 (upper picture) and round 2 (lower picture).*

Figure 3.4 shows the ventilation rates (m^3 $hour^{-1}$ kg^{-1} live weight) per treatment and room compared with the minimum ventilation requirements from the guidelines of the Klimaatplatform Pluimveehouderij (2015). The minimum required ventilation rates are presented in red lines. The ventilation rates in round 1 fluctuated without an obvious trend and in round 2 there was a rough trend going down. At certain periods in both rounds, the minimum ventilation requirements were not met in some of the rooms.

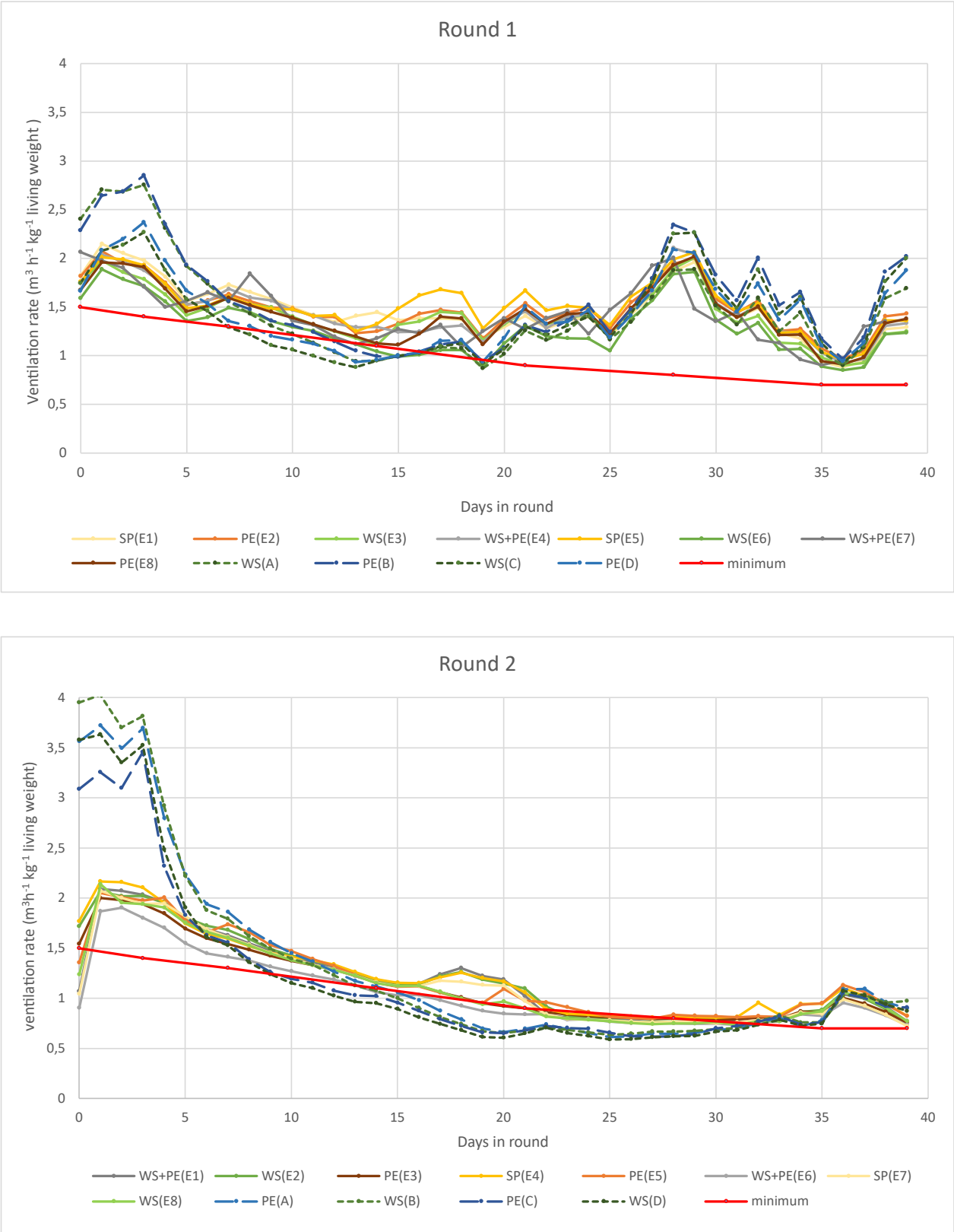


Figure 3.4 *Ventilation rates (m^3 $hour^{-1}$ kg^{-1} living weight) of round 1 (upper picture) and round 2 (lower picture).*

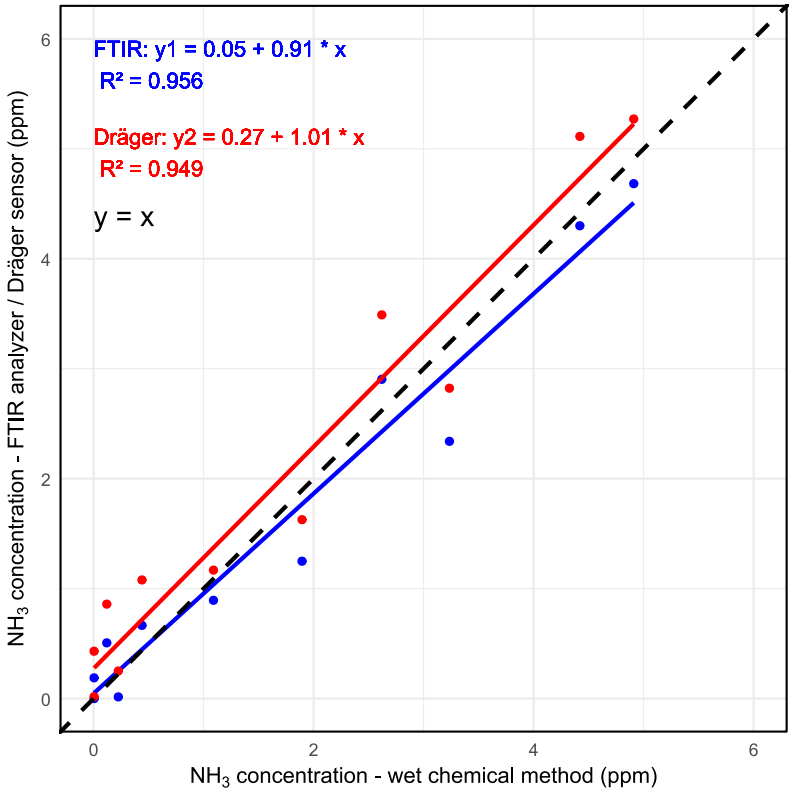
Table 3.2 shows the average ventilation rates of each treatment. There were no significant differences or interactions found in the ventilation rates between different litter materials and barns.

Table 3.2 *Mean (SE) ventilation rates per barn and treatment.*

Variable	Barn		Treatment				*P*-value		
	ABCD (n=8)	E (n=16)	WS (n=8)	PE (n=8)	SP (n=4)	WS+PE (n=4)	Litter material	Barn	Litter material *barn
Ventilation rate ($m^3\ h^{-1}$ $animal^{-1}$)	1.23 ± 0.12	1.27 ± 0.06	1.23 ± 0.10	1.26 ± 0.11	1.29 ± 0.15	1.25 ± 0.15	0.991	0.730	0.893

3.3 Ammonia (NH_3) emission

Figure 3.5 (upper) shows the linear regression fit for the ammonia concentrations measured by the wet chemical method (x, reference), FTIR analyser (y_1, in blue) and Dräger sensor (y_2, in red). One data point in round 1 was discarded since condensations were observed in the tubes of the wet chemical method, which led to an unrealistic lower ammonia concentration (outlier). The two-tailed paired *t*-test showed that neither of the two methods had significant differences from the wet chemical method ($P_{intercept} > 0.05$, $P_{slope} > 0.05$). Given that Dräger gave constant higher values than FTIR, the ammonia concentrations of all rooms were still calibrated to the values of the wet chemical method by linear regression lines shown in Figure 3.5 (lower). It is important to mention that noticeable differences were observed in replicates (max/min ratio = from 1 to 10) of the wet chemical method due to unknown reasons.



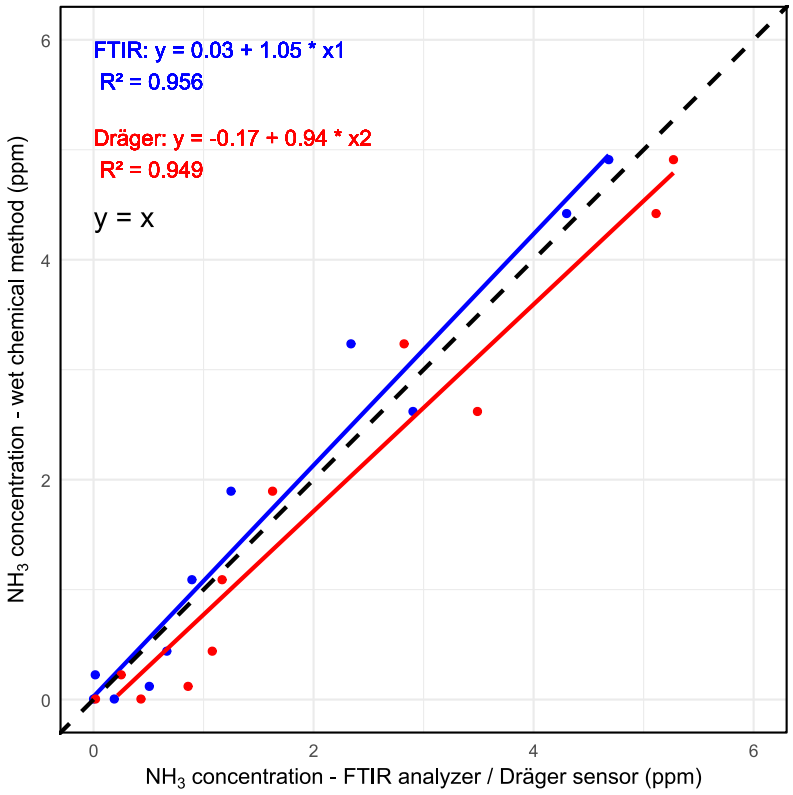
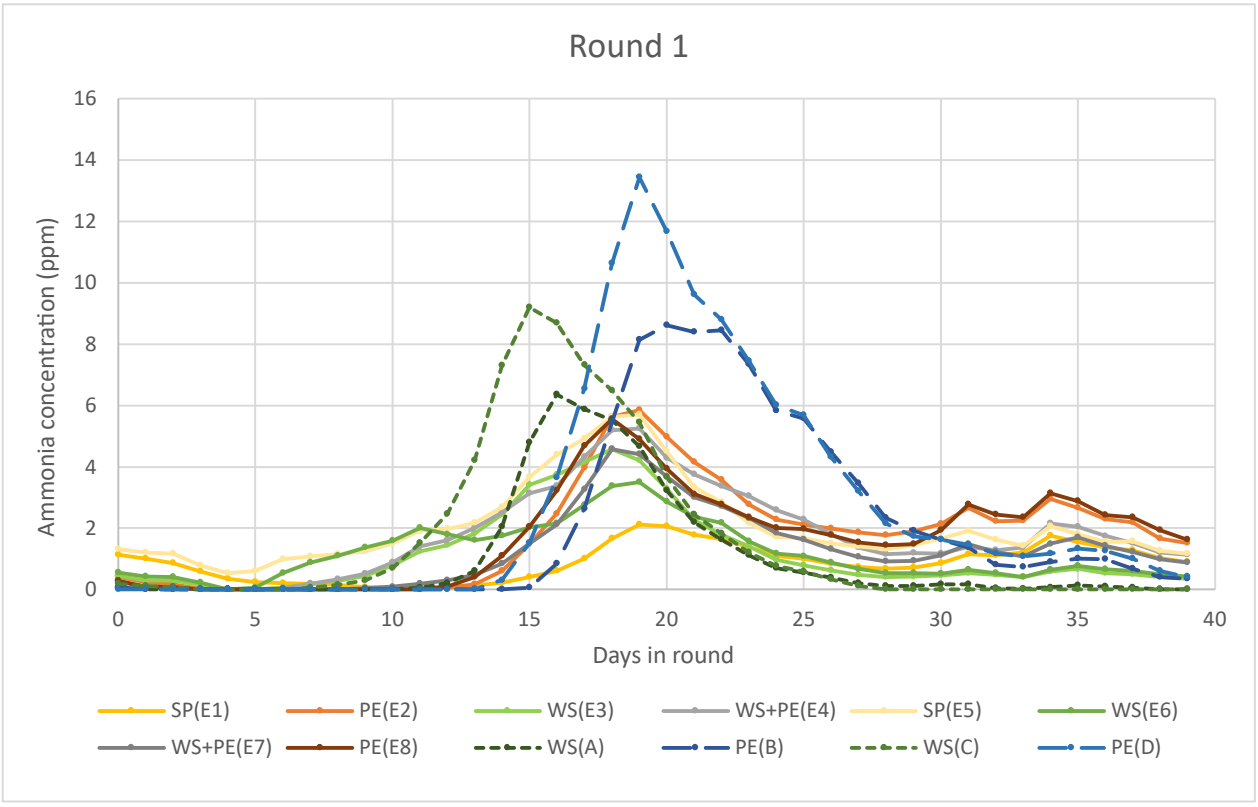


Figure 3.5 *Upper: Linear regression fit for the ammonia concentrations (ppm) measured by the wet chemical method (x), FTIR analyser (y_1, in blue), Dräger sensor (y_2, in red). Lower: Linear regression fit for ammonia concentration (ppm) measured by FTIR analyser (x_1, in blue), Dräger sensor (x_2, in red) and the wet chemical method (y).*

The ammonia concentrations and emission rates are shown in Figure 3.6 and Table 3.3, respectively. Differences were observed between round 1 and round 2 and the ammonia concentration in barn ABCD was higher than in barn E for both rounds. In round 1, ammonia concentrations remained at a low level (0-2 ppm) from day 0 to day 10. After rising until day 15-20, concentrations dropped until the end of the trial. In round 2, concentrations remained low from day 1 to day 15 (0-2 ppm). Then the concentrations increased and had some fluctuations. Peak concentrations in round 1 (2 - 14 ppm) were higher than the values of round 2 (1 - 6.5 ppm). Peat had ammonia concentrations significantly higher than wood shaving. No interactions between barns and litter materials were found.



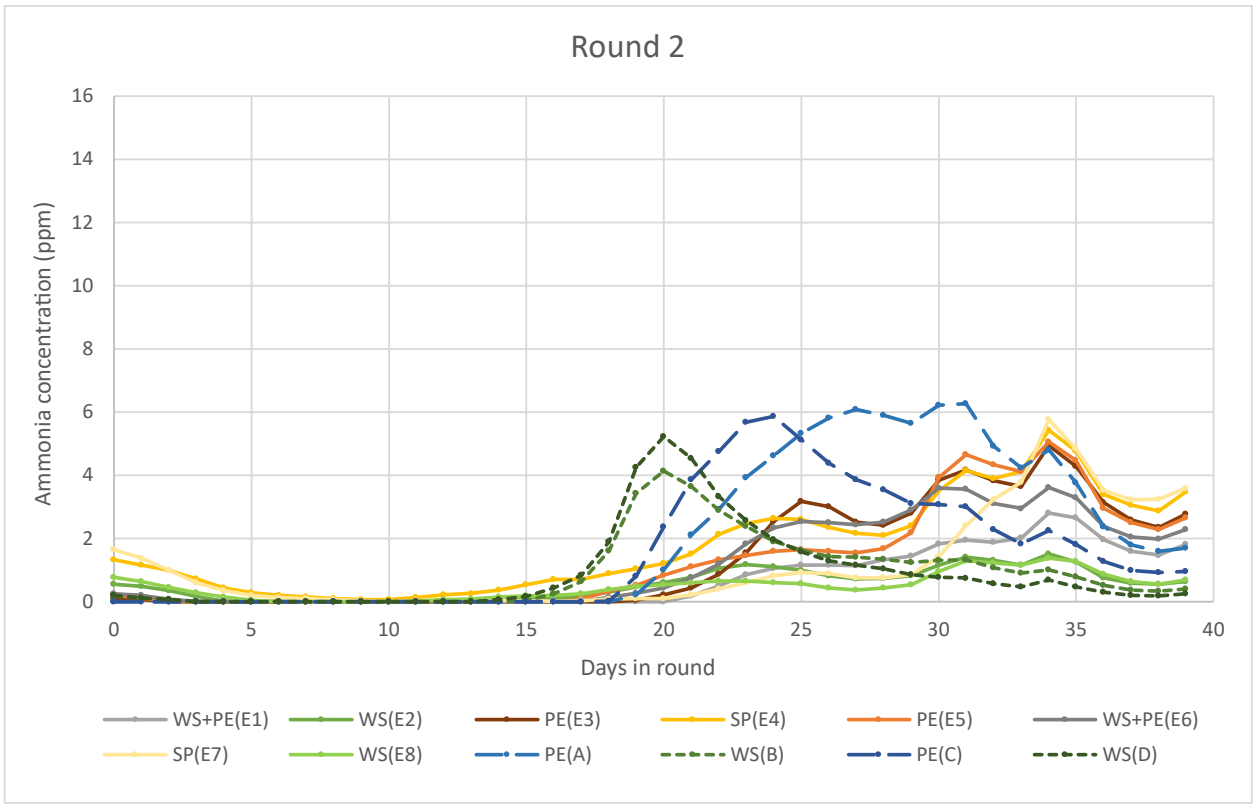


Figure 3.6 *Ammonia concentration (ppm) of round 1 (upper picture) and round 2 (lower picture).*

Table 3.3 *Mean (SE) ammonia concentrations and emissions per treatment and room.*

Variable	Barn		Treatment				*P*-value		
	ABCD (n=8)	E (n=16)	WS (n=8)	PE (n=8)	SP (n=4)	WS+PE (n=4)	Litter material	Barn	Litter material *barn
Ammonia concentration (ppm)	1.69 ± 0.15	1.30 ± 0.11	0.97 ± 0.16^{b}	1.83 ± 0.16^{a}	1.49 ± 0.22^{ab}	1.32 ± 0.22^{ab}	0.005	0.058	0.547
Ammonia emission rate (g animal place^{-1} year^{-1})	9.10 ± 1.12	8.88 ± 0.83	4.75 ± 1.18^{b}	12.81 ± 1.18^{a}	9.95 ± 1.58^{ab}	8.81 ± 1.58^{ab}	<0.001	0.768	0.646

$^{a-b}$ Values between barns and litter materials with different superscripts have significant differences (P<0.05).

3.4 Personal dust (PM_{10}) exposure

Figure 3.7 and Table 3.4 show the PM_{10} concentrations. PM_{10} concentrations went up from day 11 until day 25 and then dropped slightly. This could be caused by the poor litter quality (less friable litter) in the late days of the period and therefore less (fine) dust (see section 3.5). The average PM_{10} concentrations (Table 3.4) were below the values of common Dutch broiler barns (1.931 mg m^{-3}, Winkel et al., 2015). The PM_{10} concentrations in rooms with peat (0.996 mg m^{-3}) and straw pellets (1.017 mg m^{-3}) were 32% and 35% higher than in those with wood shaving (0.753 mg m^{-3}). No interactions between barns and litter materials were found.

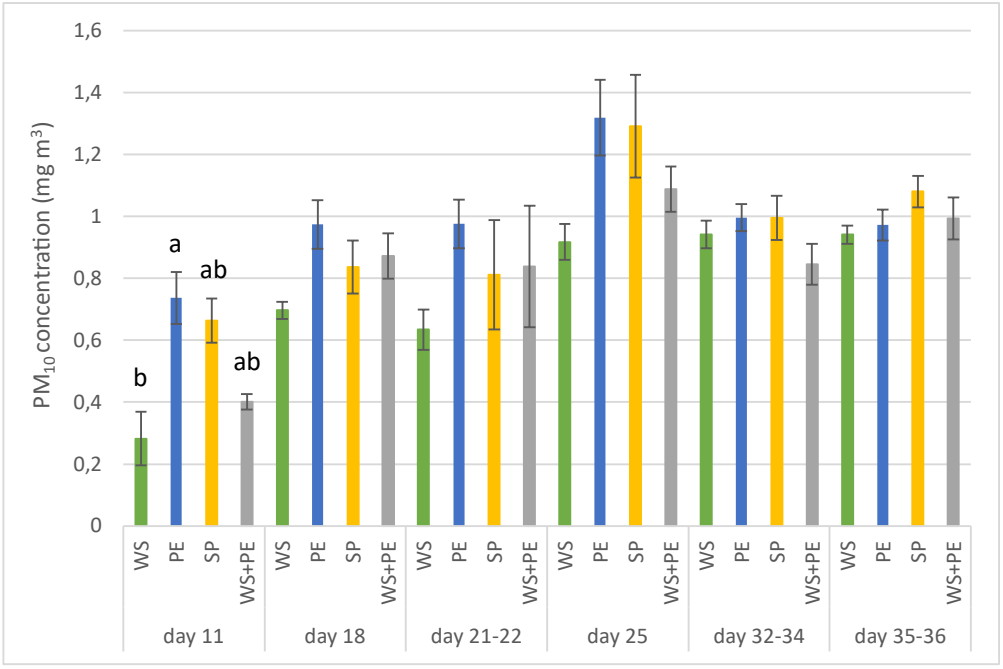


Figure 3.7 *PM_{10} concentration (mg m^{-3}). Error bars represent standard errors.*

Table 3.4 *Mean (SE) PM_{10} concentration per treatment and room.*

Variable	Barn		Treatment				*P*-value		
	ABCD (n=8)	E (n=16)	WS (n=8)	PE (n=8)	SP (n=4)	WS+PE (n=4)	Litter material	Barn	Litter material *barn
PM_{10} concentration (mg m^{-3})	0.920 ± 0.053	0.881 ± 0.032	0.753 ± 0.043^{b}	0.996 ± 0.043^{a}	1.017 ± 0.085^{a}	0.863 ± 0.085^{ab}	<0.001	0.837	0.693

$^{a-b}$ Values between barns and litter materials with different superscripts have significant differences (P<0.05).

3.5 Litter quality and litter composition

Figure 3.8 and Table 3.5 present the friability and wetness of the litter. Significant differences in friability and wetness were observed on multiple days between different litter materials and barns. Peat had significantly higher friability scores than wood shavings on day 20-21 and day 27-28, indicating more friable litter (better litter quality). Straw pellets and the mixture of wood shavings did not induce any differences. In contrast, on day 34-35, peat and straw pellets had higher friability scores than wood shavings while the mixture litter did not differ from other litter materials. On day 34-35 and 38-39, the litter in the rooms of barn E was wetter than in barn ABCD. From day 27-28 and on, 70% - 90% of the litter was caked. Moreover, the litter was wet and could be pressed ball-shaped. No interactions between barns and litter materials were found.

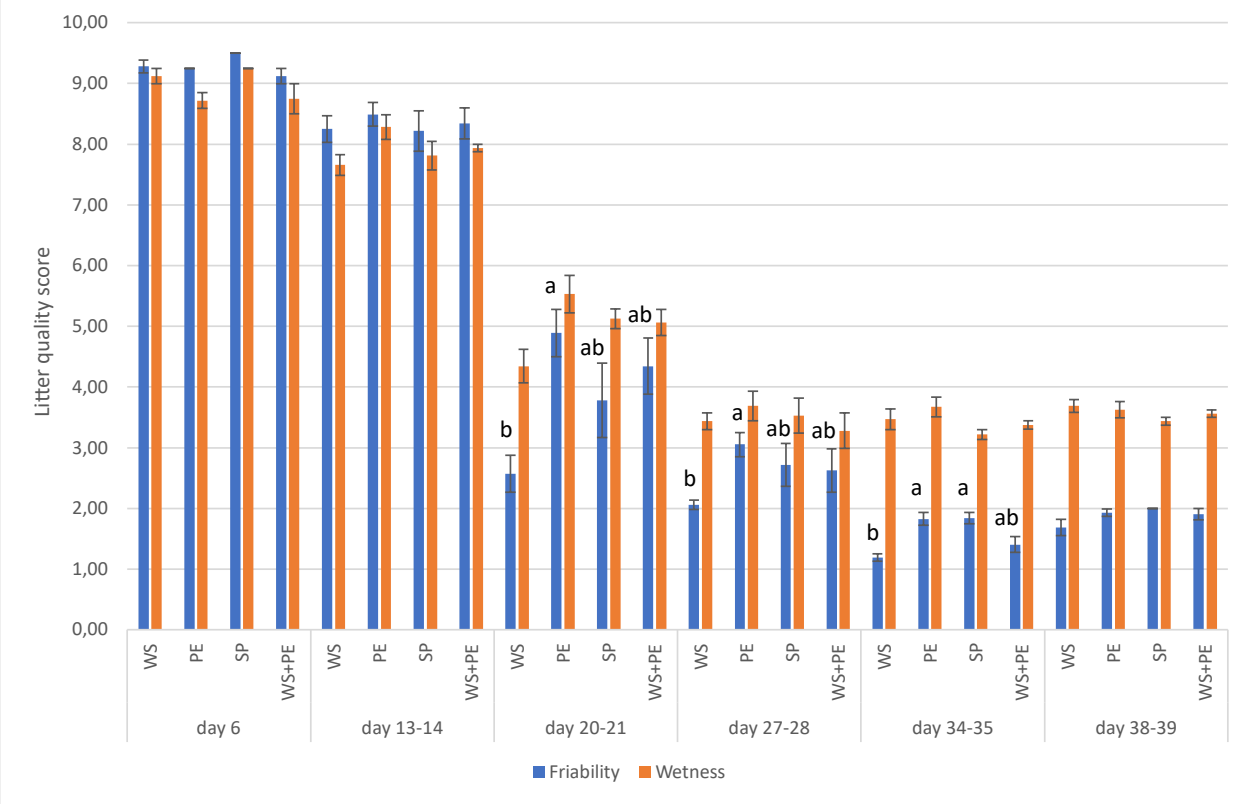


Figure 3.8 *Litter quality (friability in blue, wetness in orange) per treatment and day.*

Table 3.5 *Litter quality (friability[1], wetness[2]) per treatment and room.*

Variable	Barn		Treatment				*P*-value		
	ABCD (n=8)	E (n=16)	WS (n=8)	PE (n=8)	SP (n=4)	WS+PE (n=4)	Litter material	Barn	Litter material *barn
Day 6									
Friability	9.28 ± 0.08	9.27 ± 0.06	9.27 ± 0.09	9.25 ± 0.09	9.50 ± 0.12	9.13 ± 0.12	0.251	0.789	0.789
Wetness	9.09 ± 0.10	8.83 ± 0.08	9.08 ± 0.11	8.65 ± 0.11	9.25 ± 0.15	8.75 ± 0.15	0.053	0.057	0.546
Day 13-14									
Friability	8.60 ± 0.21	8.19 ± 0.15	8.17 ± 0.22	8.42 ± 0.22	8.22 ± 0.29	8.34 ± 0.29	0.824	0.133	0.901
Wetness	8.14 ± 0.16	7.82 ± 0.12	7.66 ± 0.17	8.17 ± 0.17	7.81 ± 0.23	7.94 ± 0.23	0.082	0.150	0.150
Day 20-21									

Friability	3.77 ± 0.36	3.81 ± 0.27	2.68 ± 0.38[b]	4.75 ± 0.38[a]	3.78 ± 0.51[ab]	4.34 ± 0.51[ab]	0.002	0.867	0.152
Wetness	5.17 ± 0.25	4.83 ± 0.19	4.33 ± 0.26	5.39 ± 0.26	5.13 ± 0.35	5.07 ± 0.35	0.027[3]	0.200	0.264
Day 27-28									
Friability	2.66 ± 0.19	2.53 ± 0.14	2.01 ± 0.20[b]	3.04 ± 0.20[a]	2.72 ± 0.27[ab]	2.63 ± 0.27[ab]	0.008	0.307	0.607
Wetness	3.83 ± 0.19	3.33 ± 0.14	3.37 ± 0.20	3.58 ± 0.20	3.53 ± 0.27	3.28 ± 0.27	0.637	0.065	0.732
Day 34-35									
Friability	1.45 ± 0.09	1.59 ± 0.06	1.21 ± 0.09[b]	1.85 ± 0.09[a]	0.84 ± 0.12[a]	1.41 ± 0.12[ab]	<0.001	0.292	0.850
Wetness	3.92 ± 0.09[a]	3.25 ± 0.07[b]	3.35 ± 0.09	3.55 ± 0.09	3.22 ± 0.12	3.38 ± 0.12	0.044[3]	<0.001	0.902
Day 38-39									
Friability	1.73 ± 0.09	1.91 ± 0.07	1.71 ± 0.10	1.96 ± 0.10	2.00 ± 0.13	1.91 ± 0.13	0.158	0.231	0.873
Wetness	3.94 ± 0.05[a]	3.42 ± 0.04[b]	3.60 ± 0.06	3.51 ± 0.06	3.44 ± 0.08	3.56 ± 0.08	0.119	<0.001	0.389

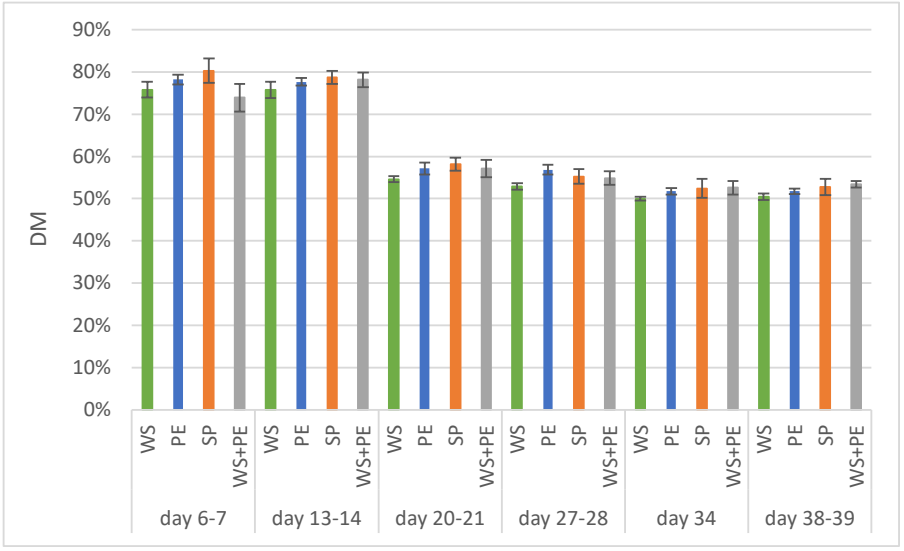
[1] Visual score of the friability of the litter material: 1 (completely caked) – 10 (completely friable).

[2] Visual score of the wetness of the litter material: 1 (very wet) – 10 (very dry).

[3] Bonferroni test did not show any significant differences.

[a-b] Values between barns and litter materials with different superscripts have significant differences ($P<0.05$).

Figure 3.9 shows the pH, thickness, litter and floor temperature on each measurement day. Table 3.6 shows the overall results. More detailed data can be found in Appendix 4. On the day upon arrival, the dry matter content (DM) of litter materials varied. Peat and the mixture litter had a higher DM on day 6-7 compared to day -2 since fresh peat has relatively higher moisture and the room temperature made the moisture evaporate. After that, all groups had a similar trend with no effects observed. The DM remained 75% to 80% from day 6-7 to day 13-14 and then dropped drastically to 55% on day 20-21. After that, it slightly decreased until the end of the round to around 50%. There were no significant differences found between litter materials and barns in the overall DM data. The pH of the litter increased from day 6-7 until day 20-21 (around 7.0) and then kept dropping until the end of the trial. Straw pellet litter had higher pH than wood shavings and peat litter. The litter thickness increased as the broilers aged and excreta was accumulated. The litter layer in barn E was thicker than in barn ABCD but no effects were observed in litter layer thickness between litter materials. The litter and floor temperature rose from day 13-14 until day 27-28 and decreased until day 38-39. The rooms in barn ABCD had higher floor and litter temperatures than the rooms in barn E. No interactions between barns and litter materials were found.



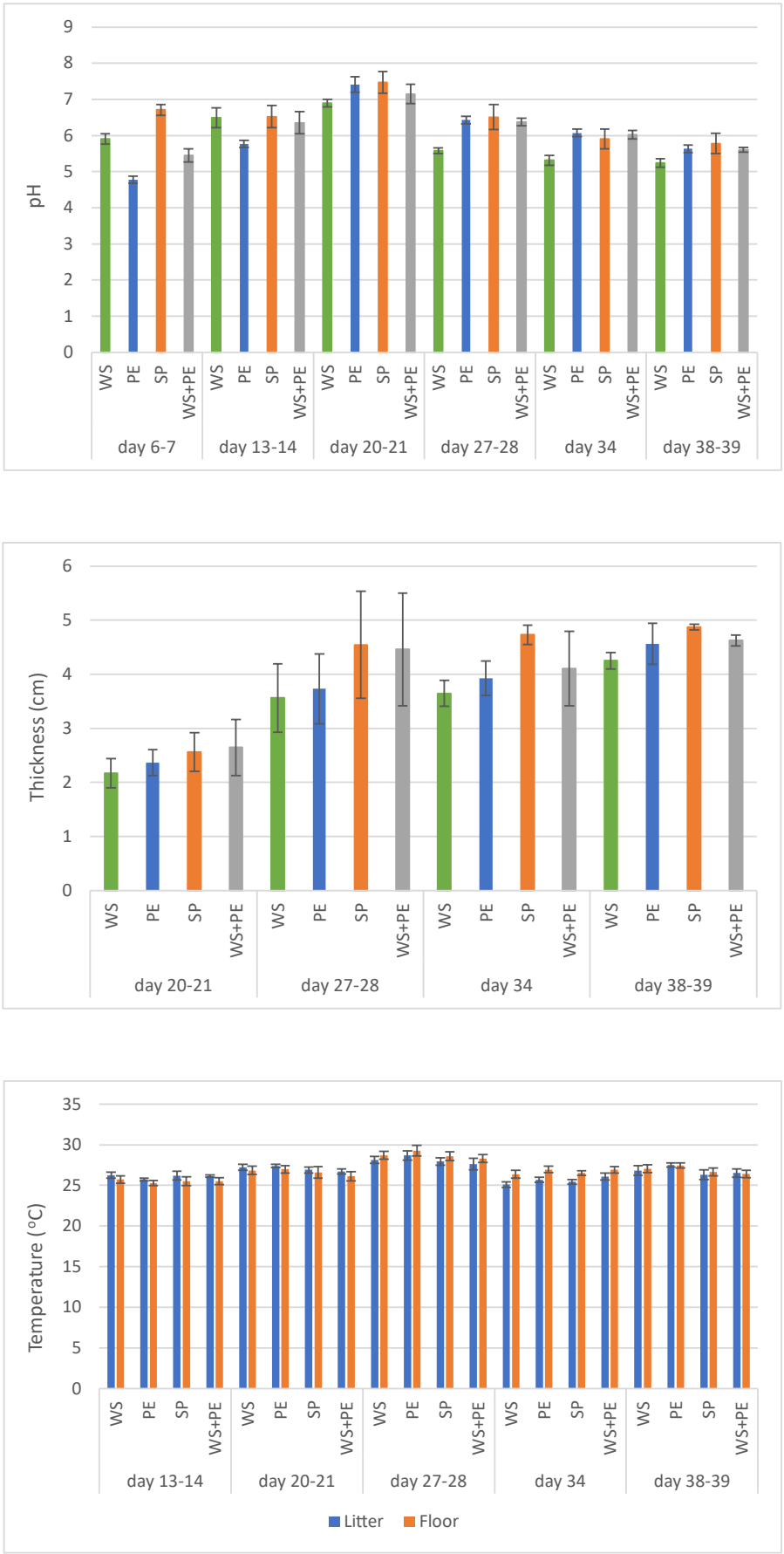


Figure 3.9 *Litter quality (DM, pH, thickness, litter and floor temperature) per treatment and day.*

Table 3.6 *Mean (SE) litter quality (DM, pH, thickness, litter and floor temperature) per treatment and room.*

Variable	Barn		Treatment				*P*-value		
	ABCD (n=8)	E (n=16)	WS (n=8)	PE (n=8)	SP (n=4)	WS+PE (n=4)	Litter material	Barn	Litter material *barn
Dry matter content	61.36% ± 0.82%	61.06% ± 0.61%	59.60% ± 0.87%	61.93% ± 0.87%	62.78% ± 1.16%	61.66% ± 1.16%	0.101	0.449	0.926
pH	5.85 ± 0.08	6.15 ± 0.06	5.96 ± 0.09^{b}	6.03 ± 0.09^{b}	6.48 ± 0.12^{a}	6.16 ± 0.12^{ab}	<0.001	0.122	0.243
Thickness	3.13 ± 0.13^{b}	3.97 ± 0.09^{a}	3.53 ± 0.13^{b}	3.79 ± 0.13^{ab}	4.18 ± 0.18^{a}	3.96 ± 0.18^{ab}	0.002	<0.001	0.781
Temperature (°C)	27.2 ± 0.2^{a}	26.6 ± 0.1^{b}	26.5 ± 0.2	27.0 ± 0.2	26.6 ± 0.2	26.6 ± 0.2	0.301	0.003	0.052
Floor temperature (°C)	27.5 ± 0.2^{a}	26.7 ± 0.1^{b}	26.8 ± 0.2	27.1 ± 0.2	26.8 ± 0.2	26.7 ± 0.2	0.208	<0.001	0.101

$^{a-b}$ Values between barns and litter materials with different superscripts have significant differences (P<0.05).

Table 3.7 shows the litter composition on day 40. Litter materials did not have any significant effects on litter composition. The litter pH in barn E was higher than in barn ABCD. No interactions between barns and litter materials were found.

Table 3.7 *Litter composition on day 40 per treatment and room.*

Variable	Barn		Treatment				*P*-value		
	ABCD (n=8)	E (n=16)	WS (n=8)	PE (n=8)	SP (n=4)	WS+PE (n=4)	Litter material	Barn	Litter material *barn
Total-N (g/kg)	24.08 ± 0.55	22.89 ± 0.41	22.98 ± 0.58	23.25 ± 0.58	23.28 ± 0.78	23.54 ± 0.78	0.896	0.080	0.263
Ammonium-N (g/kg)	2.36 ± 0.12	2.48 ± 0.09	2.23 ± 0.13	2.68 ± 0.13	2.28 ± 0.17	2.58 ± 0.17	0.054	0.446	0.574
P-content (g/kg)	2.81 ± 0.10	2.89 ± 0.07	2.83 ± 0.11	2.90 ± 0.11	2.86 ± 0.14	2.91 ± 0.14	0.895	0.539	0.598
K-content (g/kg)	12.98 ± 0.28	13.08 ± 0.21	12.79 ± 0.30	13.31 ± 0.30	13.10 ± 0.40	13.07 ± 0.40	0.543	0.822	0.602
Dry matter content (g/kg)	488.7 ± 11.2	488.5 ± 8.3	482.6 ± 11.8	488.8 ± 11.8	493.9 ± 15.8	500.2 ± 15.8	0.760	0.775	0.361
Ash content	60.73 ± 2.04	60.93 ± 1.52	60.38 ± 2.15	60.16 ± 2.15	61.25 ± 2.88	64.17 ± 2.88	0.698	0.812	0.329
pH	5.05 ± 0.04^{b}	5.19 ± 0.03^{a}	5.10 ± 0.04	5.21 ± 0.04	5.16 ± 0.06	5.20 ± 0.06	0.143	0.017	0.651

$^{a-b}$ Values between barns and litter materials with different superscripts have significant differences (P<0.05).

3.6 Zootechnical performance

Table 3.8 shows the zootechnical performance of the two production rounds, as described in section 2.6.1. Overall, the litter materials did not affect the zootechnical performance of broilers. The broilers raised in barn ABCD had lower ADWC and WFR than in barn E. Furthermore, broilers raised on the mixture litter had higher WFR than those on wood shavings. No interactions between barns and litter materials were found.

Table 3.8 *Mean (SE) zootechnical performance per treatment and room.*

Variable	Barn		Treatment				*P*-value		
	ABCD (n=8)	E (n=16)	WS (n=8)	PE (n=8)	SP (n=4)	WS+PE (n=4)	Litter material	Barn	Litter material *barn
ADFC[1]	95.38 ± 0.73	94.73 ± 0.54	95.21 ± 0.77	94.69 ± 0.77	93.81 ± 1.03	95.66 ± 1.03	0.535	0.541	0.537
ADWC[2]	164.0 ± 1.2[b]	168.5 ± 0.9[a]	166.2 ± 1.3	167.2 ± 1.3	167.1 ± 1.7	171.6 ± 1.7	0.058	0.027	0.888
WFR[3]	1.720 ± 0.009[b]	1.778 ± 0.006[a]	1.745 ± 0.009[b]	1.767 ± 0.009[ab]	1.781 ± 0.012 [ab]	1.794 ± 0.012[a]	0.004	<0.001	0.475
BW[4]	2521 ± 15	2524 ± 11	2529 ± 16	2521 ± 16	2500 ± 21	2537 ± 21	0.572	0.747	0.400
BWG[5]	2476 ± 15	2481 ± 11	2486 ± 16	2476 ± 16	2457 ± 21	2492 ± 21	0.586	0.714	0.424
dBWG[6]	61.90 ± 0.37	62.02 ± 0.28	62.16 ± 0.39	61.91 ± 0.39	61.43 ± 0.53	62.30 ± 0.53	0.586	0.714	0.424
FCR[7]	1.541 ± 0.012	1.527 ± 0.009	1.532 ± 0.013	1.530 ± 0.013	1.527 ± 0.017	1.536 ± 0.017	0.982	0.377	0.980
MRT[8]	2.218 ± 0.468	3.082 ± 0.349	2.431 ± 0.494	2.634 ± 0.494	4.293 ± 0.662	3.435 ± 0.662	0.228	0.970	0.988

[a-b] Values between barns and litter materials with different superscripts have significant differences ($P<0.05$).

[1] ADFC: average daily feed consumption (g bird^{-1}).

[2] ADWC: average daily water consumption (g bird^{-1}).

[3] WFR: water:feed ratio.

[4] BW: average end body weight (g).

[5] BWG: total body weight growth (g).

[6] dBWG: daily body weight growth rate (g bird^{-1} day^{-1}).

[7] FCR: feed conversion ratio.

[8] MRT: mortality (%).

3.7 Footpad lesions and hock dermatitis

The footpad lesion and hock dermatitis scores on each measurement day are listed in Table 3.9. On day 31-32, broilers kept on wood shavings had a higher incidence and severity of footpad lesions than those kept on peat while on day 39 higher than on all other three litter materials. On day 39, broilers kept on wood shavings had a higher incidence and severity of hock dermatitis than those kept on straw pellets. More hock dermatitis was observed in barn ABCD than in barn E on day 39 of age as well. Overall, wood shaving provided the worst animal welfare. No interactions between barns and litter materials were found.

Table 3.9 *Footpad lesions and hock dermatitis scores pretreatment and day.*

Variable	Day	Barn		Treatment				*P*-value		
		ABCD (n=8)	E (n=16)	WS (n=8)	PE (n=8)	SP (n=4)	WS+PE (n=4)	Litter material	Barn	Litter material *barn
Footpad lesion	31-32	1.08 ± 0.10	1.14 ± 0.08	1.48 ± 0.11[a]	0.86 ± 0.11[b]	1.06 ± 0.15[ab]	0.95 ± 0.15[ab]	0.001	0.379	0.071
	39	2.33 ± 0.12	2.29 ± 0.09	2.81 ± 0.12[a]	2.05 ± 0.12[b]	1.87 ± 0.16[b]	1.96 ± 0.16[b]	<0.001	0.370	0.380
Footpad score	31-32	84.9	47.3	86.8	51.6	43.6	38.9	-	-	-
	39	123.6	113.0	151.8	100.9	96.7	96.9	-	-	-

Hock dermatitis	31-32	1.53 ± 0.09	1.37 ± 0.07	1.62 ± 0.10	1.25 ± 0.10	1.17 ± 0.13	1.48 ± 0.13	0.015[1]	0.305	0.373
	39	2.11 ± 0.05[a]	1.76 ± 0.04[b]	1.94 ± 0.05[a]	1.85 ± 0.05[ab]	1.62 ± 0.07[b]	1.75 ± 0.07[ab]	0.003	<0.001	0.900

[1] Bonferroni test did not show any significant differences.

[a-b] Values in a row with different superscripts have significant differences (P<0.05).

Figure 3.10 presents the average percentage of footpad lesion scores and hock dermatitis scores on day 31-32 and day 39 of two rounds. The incidence and severity of footpad lesions and hock dermatitis increased as the broilers aged. Severe footpad lesions occurred more often when the broilers reached slaughter age.

Figure 3.10 *Average percentage of footpad lesion (upper) and hock dermatitis (lower) scores on day 31-32 and day 39.*

4 Discussion

This study was designed to ascertain the effect of peat litter (in comparison with wood shavings) on ammonia emission, and on personal dust exposure, zootechnical performance, litter quality, litter composition and animal welfare. The temperature (ranging from - 0.9 °C to 17.2 °C) and relative humidity (ranging from 47.9% to 94.0%) outside the barns were consistent with the long-term weather data in Belgium. The study was conducted using a commercial broiler breed (Ross 308). The housing, feeding and management of the broilers resembled that of commercial barns in Belgium and the Netherlands. The study was therefore conducted under conditions representative for real life situation.

Overall, peat litter did not reduce ammonia emission compared with wood shavings but resulted in better animal welfare. No differences were observed in final litter composition and zootechnical performance. Peat had ammonia emissions of 12.81 g per year per animal place, which was 2.7 times as high as wood shavings (4.75 g per year per animal place). Ammonia emissions of straw pellets (9.95 g per year per animal place) and the mixture litter (8.81 g per year per animal place) did not differ from other litter types.

The category of both barn ABCD and E are equipped with delta heating tubes, but not in a way as described in the Dutch Livestock Farming Ammonia Regulations (abbreviated as Rav). The Rav category of both barns ABCD and barn E is between housing system E 5.15 and E 5.100 (E 5 Diercategorie Vleeskuikens, n.d.). However, the ammonia emissions in the present work were substantially lower than the corresponding emission factors of 21 and 68 g per year per animal place for E 5.15 and E 5.100, respectively (E 5 Diercategorie Vleeskuikens, n.d.). This could be related to the poor litter quality and the low dry matter content. From day 28 and on, more than 70% of the litter was caked and the wet litter could be pressed ball-shaped due to the high moisture. It was demonstrated that the crust formed on the top litter layer and a low litter friability (caked litter) can slow down ammonia volatilization (Brink et al., 2022). Furthermore, the dry matter content was below 60% from day 21 until the end of the trials. It was found that the ammonia formation from the litter decreases when the dry matter content is higher than 80% or lower than 60% (Groot Koerkamp et al., 2000). This could also explain why such low amounts of ammonia were observed. Additionally, peat has a rather high moisture content, and it tends to keep the moisture it absorbs (preheating should be carried out thoroughly). At some points, the ventilation rates did not meet the minimum ventilation requirements either, which made the moisture hard to evaporate. Wet conditions can lead to anaerobic decomposition and anaerobic microbial activities, producing more ammonia. Nevertheless, the nitrogen content of the litter could not explain the differences in ammonia emissions. The variables of litter quality in broiler barns are changeable and hard to predict. Little is known about how these variables interact with each other and how they influence ammonia emissions. More focus is required to investigate how the litter variables interact with each other in broiler barns and how they influence ammonia emissions.

Three methods were used in this study to measure ammonia concentration. The Dräger Polytron P8100 sensor is an electrochemical sensor that has been reported with an accuracy of 5% (according to the manufacturer) and the FTIR analyser has an accuracy of 2% - 5% (Schulte et al., 2022; Singh et al., 2019). In this study, both FTIR analyser (y_1=0.91x + 0.05, R^2=0.956) and Dräger sensor (y_2=1.01x + 0.27, R^2=0.949) showed significant differences from the wet chemical method. Although the wet chemical measurements fitted the overall concentrations measured by Dräger and FTIR, it is important to mention that a number of large differences were observed between the data points of duplicate measurements of the wet chemical method (max/min ratio ranged from 1 to 10). The reason for this has not become clear. The wet chemical method was still used as a reference to cancel out the systematic differences between Dräger and FTIR, while the reliability of wet chemical measurements may be challenged.

Another question shown in this study was the validity of the CO_2 mass balance method to calculate ventilation rates. Mosquera et al. (2012) proved that the CO_2 mass balance method was able to provide a reasonably accurate estimate of ventilation rates for broilers.

In this study, however, the values calculated by the CO_2 mass balance method were significantly higher ($P < 0.05$) compared with the ventilation rates measured directly by anemometers. As known, one of the uncertainties of the CO_2 mass balance method is the CO_2 contribution from the litter (mixture of litter materials and excreta). It could be that the litter contribution of 10% in the current equation (CIGR, 2002) overestimated the actual CO_2 emissions from the litter, given that the thickness of the litter was less than 5 cm. Therefore, the litter might contribute to the CO_2 emissions less than proposed in the equation (CIGR, 2002). Further studies need to be conducted to validate the accuracy of the carbon dioxide mass balance method for today's broilers.

Furthermore, the ventilation rates calculated based on the CO_2 mass balance method were unrealistically high during day zero to two. A few possible reasons could be that (1) the ventilation system is not accurate enough to control and measure the ventilation flow correctly at very low ventilation rates (<1% of the installed ventilation capacity) during the first days, resulting in a higher ventilation rate; (2) it was recommended that the difference between indoor and outdoor CO_2 concentration should be more than 250 ppm (Seedorf et al., 1998). Some of the measurements on day 0 did not meet this condition; (3) on day 0, the chickens were in the barn for less than 24 hours, which would lead to inaccuracy of the calculation since the carbon dioxide mass balance method is based on 24-hour averages; (4) the sampling points of CO_2 measurements were fixed in the climate-separated rooms and might not be representative. To provide more accurate results, multiple sampling points over a full barn can help (Edouard et al., 2016).

5 Conclusions

In this study, we investigated the effects of using wood shavings, peat, straw pellets, and the mixture of wood shavings and peat as litter material in broiler houses and their effects on ammonia emissions, personal dust exposure, zootechnical performance, litter quality and animal welfare.

The main conclusions are:

- Peat did not reduce ammonia emissions compared with wood shavings, straw pellets, and the mixture litter. Instead, peat had ammonia emission rates of 12.81 g $year^{-1}$ animal $place^{-1}$, which was 2.7 times as high as wood shavings (4.75 g $year^{-1}$ animal $place^{-1}$).
- Peat and straw pellets caused higher PM_{10} concentrations (personal bound PM_{10} concentrations) than wood shavings (0.996, 1.017 and 0.753 mg m^{-3} respectively). This can be related to the more friable litter.
- Peat had more friable litter (better litter quality) than wood shavings on day 20-21 and day 27-28 of broiler age. Straw pellets and the mixture of wood shavings did not induce any differences. On day 34-35 of broiler age, peat and straw pellets had more friable litter (better litter quality) than wood shavings while the mixture litter did not differ from other litter materials.
- Litter materials did not influence the overall (the average of all measurement days) litter DM content, litter pH, litter layer thickness, litter temperature, floor temperature and final litter composition (total N, ammonium N, P, K, DM, ash, pH).
- On day 31-32 of broiler age, peat had lower incidence and severity of footpad lesions compared with wood shavings, which indicates better animal welfare. Straw pellets and the mixture litter showed no differences from peat and wood shavings. Litter material did not influence the incidence and severity of hock dermatitis. On day 39, peat, straw pellets and the mixture litter had less incidence and severity of footpad lesions than wood shavings. Straw pellets reduced the incidence and severity of hock dermatitis compared with wood shavings. Peat and the mixture litter showed no differences in hock dermatitis from straw pellets and wood shavings.

Some recommendations for research in the future would be:

- More attention should be paid to investigating the litter characteristics and litter composition in broiler barns, how they interact with each other, how they influence ammonia emissions, and how the litter should be managed.
- Further studies need to be carried out to validate and/or improve the accuracy of the carbon dioxide mass balance method for today's broilers.

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Appendix 1 Litter sampling and processing protocol

The sampling location depends on the barn. Figure A1 and A2 show the sampling locations in barn ABCD and barn E, respectively.

Take the samples with a so-called manure auger down to the (concrete) subfloor. Collect the samples per room in a bucket, plastic bag or stainless steel container (mixing sample).

For the litter analyses at the end of the round, the litter samples are pooled per climate-separated section (n=12) and placed in the freezer as soon as possible after sampling, after which WLR will take the samples to the laboratory for analyses.

For the (weekly) dry matter determination, individual samples per climate-separated room (n= 6 x 12) will be dried in a drying oven at 105 °C for 24 hours immediately after sampling.

If direct processing of the (mixing) samples is not possible, the samples are stored in plastic bags in the freezer (-20 °C), after which they are later processed/dried.

Figure A1.1 *Sampling barn ABCD*

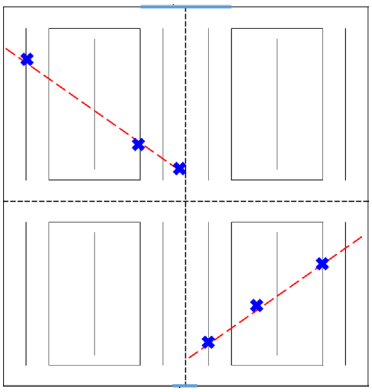
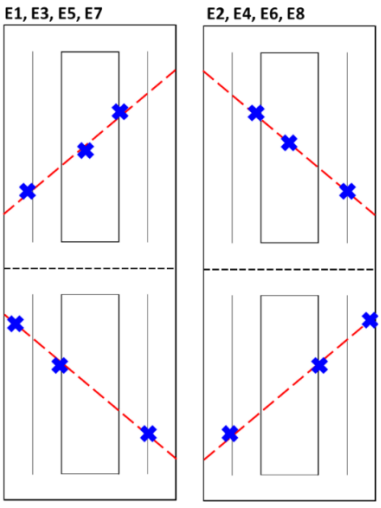


Figure A1.2 *Sampling barn E*



Appendix 2 WQ protocol footpad lesions and hock dermatitis

The presence and severity of foot pad and hock lesion is determined for each department of 2 x 40 chicks (males and females). A score from 0 (no lesion) to 4 (severe lesion) is given in accordance with the WQ protocol.

Foot pad dermatitis farm card

0	1	2	3	4
No lesion	**Very mild lesion:** Very small superficial lesions, slight discolouration on a limited area, mild hyperkeratosis	**Mild lesion:** Discolouration of the foot pad, superficial lesion, superficial dermatitis	**Moderately severe lesion:** Ulcers or scabs	**Very severe lesion:** Ulcers or scabs, signs of haemorrhages or deep dermatitis

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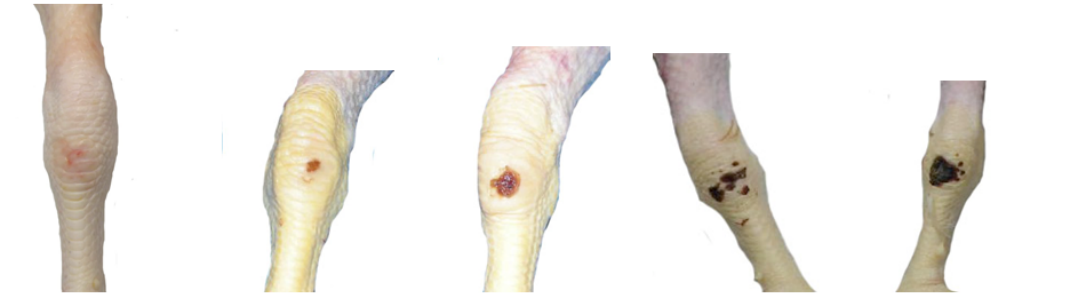
Hock burn farm card



0	1	2	3	4
No lesion	**Very mild lesion:** Small red discolouration (< 0.5 cm^2)	**Lesion:** bigger (> 0.5 cm^2), but still only red discolouration	**Hock burn:** Brown or black discolouration of the hock, small area. More than 1 spot possible. (total <0.5 cm^2).	**Serious hock burn:** Brown or black discolouration of the hock. More than 1 spot possible. (total >0.5 cm^2). Probably with crust. Infected hock is also in this category (no picture).

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Appendix 3 Detailed data on emissions

Table A3.1 *Average ventilation rate, ammonia concentration, ammonia emission corrected for vacancy, room temperature, outside temperature, room RH, and outside RH per round of barn E.*

	Round 1				Round 2			
Variables	**WS**	**PE**	**SP**	**WS+PE**	**WS**	**PE**	**SP**	**WS+PE**
Ventilation rate ($m^3\ h^{-1}$ $animal^{-1}$)	1.44	1.54	1.55	1.51	1.04	1.04	1.04	1.00
Ammonia concentration (ppm)	1.23	1.84	1.48	1.60	0.50	1.43	1.49	1.03
Ammonia emission (g animal $place^{-1}$ $year^{-1}$)	6.58	14.98	10.59	10.99	3.09	9.84	9.31	6.63
Room temperature (°C)	25.86	25.91	25.91	25.86	24.95	24.88	24.84	24.91
Outside temperature (°C)	13.00	13.00	13.00	13.00	4.90	4.90	4.90	4.90
Room RH (%)	61.90	60.78	59.27	59.74	54.16	54.54	53.40	51.42
Outside RH (%)	77.84	77.84	77.84	77.84	82.01	82.01	82.01	82.01

Table A3.2 *Average ventilation rate, ammonia concentration, ammonia emission corrected for vacancy, room temperature, outside temperature, room RH, and outside RH per round of barn ABCD.*

	Round 1		Round 2	
Variables	**WS**	**PE**	**WS**	**PE**
Ventilation rate ($m^3\ h^{-1}$ $animal^{-1}$)	1.53	1.56	0.92	0.91
Ammonia concentration (ppm)	1.39	2.51	0.95	1.91
Ammonia emission (g animal $place^{-1}$ $year^{-1}$)	5.30	17.48	3.84	9.76
Room temperature (°C)	26.56	26.86	26.30	26.12
Outside temperature (°C)	13.00	13.00	12.99	12.99
Room RH (%)	57.65	57.03	50.04	50.92
Outside RH (%)	77.84	77.84	82.01	82.01

Appendix 4 Detailed data on litter quality

Table A4.2 *Average effects of barn and litter material on litter dry matter content, litter pH, litter layer thickness, litter temperature and floor temperature on different days.*

Variable	Barn		Treatment				*P*-value		
	ABCD (n=8)	E (n=16)	WS (n=8)	ABCD (n=8)	E (n=16)	WS (n=8)	ABCD (n=8)	E (n=16)	WS (n=8)
Day -2									
Dry matter content	69.5%	78.0%	89.0%	54.8%	89.0%	74.5%	-	-	-
Day 7									
Dry matter content	76.55% ± 1.63%	77.33% ± 1.21%	76.80% ± 1.72%	77.50% ± 1.72%	80.03% ± 2.30%	73.89% ± 2.30%	0.213	0.703	0.042
pH	5.53 ± 0.11[a]	5.46 ± 0.08[b]	5.83 ± 0.12[b]	4.72 ± 0.12[c]	6.71 ± 0.16[a]	5.45 ± 0.16[b]	<0.001	0.024	0.742
Day 14									
Dry matter content	75.45% ± 0.96%	78.10% ± 0.72%	77.19% ± 1.01%	77.04% ± 1.01%	78.70% ± 1.36%	78.14% ± 1.36%	0.278	0.083	<0.001
pH	6.39 ± 0.15[a]	6.06 ± 0.11[b]	6.27 ± 0.16	5.81 ± 0.16	6.52 ± 0.21	6.35 ± 0.21	0.010[1]	0.030	0.001
Temperature (°C)	26.2 ± 0.3	25.9 ± 0.2	26.1 ± 0.3	25.7 ± 0.3	26.2 ± 0.4	26.1 ± 0.4	0.560	0.419	0.258
Floor temperature (°C)	25.6 ± 0.3	25.4 ± 0.2	25.4 ± 0.3	25.5 ± 0.3	25.5 ± 0.4	25.5 ± 0.4	0.782	0.552	<0.001
Day 21									
Dry matter content	56.74% ± 1.17%	55.89% ± 0.87%	54.51% ± 1.24%	56.67% ± 1.24%	58.13% ± 1.66%	57.11% ± 1.66%	0.299	0.313	0.539
pH	7.15 ± 0.17	7.21 ± 0.13	6.83 ± 0.18	7.49 ± 0.18	7.47 ± 0.25	7.15 ± 0.25	0.163	0.940	0.089
Thickness (cm)	2.23 ± 0.21	2.40 ± 0.16	2.38 ± 0.22	2.18 ± 0.22	2.56 ± 0.30	2.65 ± 0.30	0.548	0.810	<0.001
Temperature (°C)	27.4 ± 0.2	27.1 ± 0.2	27.0 ± 0.3	27.5 ± 0.3	26.9 ± 0.3	26.7 ± 0.3	0.360	0.571	0.015
Floor temperature (°C)	26.9 ± 0.3	26.7 ± 0.2	26.4 ± 0.3	27.3 ± 0.3	26.6 ± 0.4	26.1 ± 0.4	0.481	0.854	<0.001
Day 28									
Dry matter content	55.52% ± 0.95%	54.52% ± 0.71%	53.19% ± 1.01%)	56.17% ± 1.01%	55.26% ± 1.35%	54.85% ± 1.35%	0.066	0.361	0.048
pH	5.98 ± 0.13	6.16 ± 0.10	5.58 ± 0.14[b]	6.44 ± 0.14[a]	6.51 ± 0.19[a]	6.37 ± 0.19[a]	<0.001	0.802	0.801
Thickness (cm)	3.68 ± 0.46	3.91 ± 0.35	4.07 ± 0.49	3.19 ± 0.49	4.54 ± 0.66	4.46 ± 0.66	0.522	0.923	<0.001
Temperature (°C)	28.3 ± 0.3	28.3 ± 0.3	27.8 ± 0.4	29.1 ± 0.4	28.0 ± 0.5	27.6 ± 0.5	0.321	0.700	<0.001

Floor temperature (°C)	28.9 ± 0.4	28.9 ± 0.3	28.3 ± 0.4	29.7 ± 0.4	28.6 ± 0.5	28.3 ± 0.5	0.439	0.774	<0.001
Day 35									
Dry matter content	50.98% ± 0.94%	51.34% ± 0.70%	50.25% ± 1.00%	51.42% ± 1.00%	52.39% ± 1.34%	52.58% ± 1.34%	0.332	0.871	0.215
pH	5.71 ± 0.13	5.77 ± 0.10	5.33 ± 0.14[b]	6.05 ± 0.14[a]	5.91 ± 0.19[ab]	6.02 ± 0.19[ab]	0.004	0.862	0.520
Thickness (cm)	3.73 ± 0.31	4.04 ± 0.23	3.57 ± 0.33	4.04 ± 0.33	4.73 ± 0.44	4.10 ± 0.44	0.274	0.796	0.206
Temperature (°C)	25.6 ± 0.2	25.4 ± 0.2	24.9 ± 0.3	25.8 ± 0.3	25.4 ± 0.3	26.1 ± 0.3	0.146	0.249	0.005
Floor temperature (°C)	26.8 ± 0.3	26.6 ± 0.2	26.0 ± 0.3	27.2 ± 0.3	26.5 ± 0.4	26.9 ± 0.4	0.429	0.439	<0.001
Day 39									
Dry matter content	50.62% ± 0.80%	52.13% ± 0.59%	50.89% ± 0.84%	51.71% ± 0.84%	52.81% ± 1.13%	53.41% ± 1.13%	0.166	0.378	0.188
pH	5.38 ± 0.13	5.56 ± 0.10	5.28 ± 0.14	5.63 ± 0.14	5.78 ± 0.18	5.60 ± 0.18	0.085	0.550	0.441
Thickness	4.21 ± 0.20	4.65 ± 0.15	4.17 ± 0.21	4.78 ± 0.21	4.88 ± 0.28	4.63 ± 0.28	0.336	0.177	0.005
Temperature (°C)	27.3 ± 0.5	26.8 ± 0.3	26.8 ± 0.5	27.4 ± 0.5	26.3 ± 0.7	26.5 ± 0.7	0.426	0.674	0.811
Floor temperature (°C)	27.7 ± 0.4	26.7 ± 0.3	27.0 ± 0.4	27.2 ± 0.4	26.7 ± 0.5	26.4 ± 0.5	0.384	0.138	0.238

[1] Bonferroni test did not show any significant differences.

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