

Factors affecting presence of feed in crops of broilers at slaughter

- With focus on a hygienic slaughter

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Factors affecting presence of feed in crops of broilers at slaughter – With focus on a hygienic slaughter

Faktorer som påverkar förekomsten av foder i krävan hos slaktkycklingar vid slakt – med fokus på en hygienisk slakt

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Abstract

Food security and sustainable consumption and production are parts of the United Nation Sustainable Development Goals 2 and 12. Filled broiler crops at slaughter poses a risk to contaminate carcasses with gastrointestinal contents. Due to the risk of contamination of pathogenic and other bacteria, the carcasses will be rejected for further processing if contaminated. The purpose of this thesis was to investigate which factors that influence the content load of crops at slaughter, and possibilities to reduce the problem. Furthermore, investigation if rinsing can improve the hygienic quality of carcasses. This was done by palpation of the crop in live birds at farms, interviewing of producers, observations of the assessment of filled crops at slaughter, and by analyses of the load of total aerobic bacteria and Enterobacteriaceae before and after rinsing of carcasses at slaughter. The result of the interviews with farmers revealed that on-farm factors had no significant effect on the percentage of filled crops at slaughter. However, a significant difference in the percentage of crops assessed as filled was observed for the two working shifts in the meat inspection. Rinsing of carcasses resulted in a significantly decrease in total aerobic bacteria and Enterobacteriaceae, which is a positive result towards improving the hygienic quality of the carcasses. In conclusion, results from this thesis show that further research within this topic is required to better understand which factors that are linked to broiler crops assessed as filled at the slaughterhouse and to ensure a hygienic slaughter. A better understanding of which factors affecting the occurrence in filled crops at slaughter will be of importance for the continuous work toward the Sustainable Development Goals 2 and 12.

Keywords: Crop, Broiler, Feed Withdrawal, Bacteria, Enterobacteriaceae

Sammanfattning

Delar av Förenta nationernas hållbarhetsmål 2 och 12 syftar till att ha säkra livsmedel för människor och att upprätthålla en hållbar konsumtion och produktion. Fyllda krävor hos slaktkycklingar riskerar att kontaminera slaktkropparna på slakteriet vilka riskerar att kasseras. Syftet med denna uppsats var därför att undersöka vilka faktorer som påverkar förekomsten av fyllda krävor för att på sikt minska problemet vid slakt. Dessutom studerades om duschning av slaktkroppar minskar bakteriemängden på slaktkropparna. För att genomföra detta besöktes fem slaktkyckling besättningar och ett större antal producenter intervjuades. På slakteriet undersöktes hur bedömningen av fyllda krävor gjordes, krävinnehållet granskades visuellt, och slutligen beräknades totalantalet bakterier samt Enterobacteriaceae före och efter in-och utvändig tvätt. När resultaten av intervjuerna kopplades till förekomsten av fyllda krävor vid slakt kunde ingen av inverkan på förekomst av fyllda krävor härledas till någon enskild faktor i gårdens rutiner inför slakt. Däremot fanns det en signifikant skillnad i andelen krävor som bedömts som fyllda mellan de två arbetsskiften på slakteriet. In- och utvändig tvätt visade en signifikant minskning av såväl totalantalet bakterier som bakterier tillhörande familjen Enterobacteriaceae, vilket är ett positiv resultat för de hygieniska kvalitetsaspekterna hos slaktkyckling. Sammanfattningsvis visar resultaten från denna studie att vidare forskning är nödvändig inom detta område för att bättre kunna förstår vilka faktorer som påverkar krävans innehåll vid slakt. Kunskapen skulle kunna bidra till att minimera antalet kontaminerade och kasserade slaktkroppar, samt säkerställa en mer hygienisk slakt. Ökad kunskap inom detta område skulle även vara ett steg i arbetet mot att uppfylla hållbarhetsmålen 2 och 12.

Nyckelord: Kräva, Broiler, Borttagning av foder, Bakterier, Enterobacteriaceae

Preface

"Anyone who has never made a mistake has never tried anything new." Albert Einstein

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Abbreviations

A. caviae	Aeromonas caviae
A. veronii	Aeromonas veronii
E. coli	Escherichia coli
EU	European Union
PCA	Plate count agar
P. mirabilis	Proteus mirabilis
SDGs	Sustainable Development Goals
SLU	Swedish University of Agricultural Sciences
SVA	The Swedish National Veterinary Institute
UN	United Nations
VRBG	Violet red bile glucose agar

1. Introduction

The United Nations (UN) invented the 17 Sustainable Development Goals (SDGs) in 2015 with the purpose of creating peace and prosperity for all humans on the planet. The 2nd SDG target is to end hunger, achieve food security, improve nutrition, and promote sustainable agriculture, meanwhile SDG 12 target is to ensure sustainable consumption and production patterns (United Nations n.d.).

From 2020, there has been a gradual increase in the number of filled crops for Swedish broiler chickens. Broilers uses the crop to temporarily store feed and water before it is passed further in the gastro-intestinal tract. At slaughter, filled crops causes hygienic problems since the crop content contains bacteria that may contaminate the carcasses. Broiler carcasses that are contaminated with crop content may become discarded at the slaughterhouse because of the impaired hygienic quality of the carcass (Almeida et al. 2018). This mode of action leads to both food waste and a financial loss for the slaughterhouse as well as the broiler producers and are neither in line with SDG 2 nor SDG 12.

In Sweden, the consumption of chicken has had an increasing trend over the last two decades with an average consumption of 22.3 kg meat per person in 2019 (Jordbruksverket 2022). Europe has also followed the same trend even though the consumption is predicted to stagnate (European Commission 2022). The EC Agricultural and Rural Development (2021) stated that the consumption of poultry meat within the European Union (EU) has increased with 2 % annually between 2011-2021. The increase is predicted to continue but with 0.5 % per annum until 2031. The EU poultry meat consumption per capita is predicted to increase from 23.5 kg in 2021 to 24.8 kg in 2031. Reasons for a continued demand is that poultry meat is considered as a healthier alternative and less impact of climate changes than red meat, and accepted in all religions (European Commission 2021).

This study was performed to evaluate if there is a correlation between actions, like light programme, feed, and water withdrawal, at farm level prior to transportation to the slaughterhouse and occurrence of filled crops at slaughter. To avoid filled crops at slaughter, feed and water are withdrawn prior the catching starts. The breeding company Aviagen, which supplies Sweden with Ross 308 chickens, has recommendation for feed and water withdrawal times and light programme to broiler producers, to reduce the number of filled crops at slaughter.

To find a best practice to handle the filled crop problem would be finically beneficial for both producers and the slaughterhouse. It would also be a step in the right direction towards SDG 2 and SDG 12 since it would lower the amount of wasted food and increase sustainable production.

1.1 Purpose and objective

The purpose of this project was to study possibilities to reduce the problem with filled crops at slaughter and to investigate if rinsing with water can improve the hygienic quality of carcasses. A lower frequency of filled crops at slaughter and improved hygienic status by rinsing could enable economic profits for both broiler producers and slaughter company. Specific aims were:

- Evaluate how broiler producers apply the recommended crop routine by visiting five broiler producers and interview the producers,
- Observe how the assessment of filled crops is performed at the slaughterhouse and investigate the content in the crops by ocular inspection,
- Quantify the load of total aerobic bacteria and *Enterobacteriaceae* on neck skin before and after rinsing, to evaluate if rinsing of carcasses improves the hygiene.

2. Background

2.1 The broiler chicken

Broiler chickens (Gallus gallus) can be either fast- or slow-growing meat-types birds. In the past, Ross 308 or Cobb were used for both fast- and slow-growing purposes (Rezaei et al. 2018), but are today mainly used for fast-growing purposes. Ross 308 is the dominating hybrid for conventional broiler production in Sweden and it takes approximately 35 days to reach a live weight of 2200 grams (Aviagen 2019). Broilers maintain a body temperature between 40.5 and 42.0°C at rest and are therefore categorised as *homeothermic* animals (Sjaastad et al. 2010). Like dogs, chickens lack sweat glands and therefore panting when overheated.

2.1.1 Gastrointestinal system of avian species

Avian species have a unique digestion system unlike carnivores, ruminants, and single-stomached herbivorous (Sjaastad et al. 2010). To begin with, broiler chickens do not have any teeth and instead of chewing the feed, they churn it with the beak into smaller pieces. The fast movement of the tongue makes the bird swallow the feed immediately after intake. Peristaltic movements helps the feed to reach the crop where the food is stored before further movements in the gastrointestinal system (Sjaastad et al. 2010). The transit time is rapid and it takes on average five to six hours for the feed to transit the whole gastro-intestinal system (Ravindran 2013; Svihus & Itani 2019). Factors influencing the digestion time are feed composition and particle size, water intake, temperature, light, and stress (Aviagen 2018). Apart from the domestic animals, broiler chickens neither suck nor lap up water when they drink. Instead, when the oral cavity is filled, the chicken leans the tip of the beak backwords. By doing so, the water drains to the pharynx and reaches the crop (Sjaastad et al. 2010). The crop is a storage organ and is explained more in detail in sub-chapter 2.1.2. After the crop, the feed is passed onto the glandular stomach (proventriculus) and muscular stomach, (gizzard), where the feed is further digested by chemical digestion, and mechanical grinding. The feed transit through the proventriculus is fast with a transition time of approximately 5-10 minutes. During that time, the enzyme pepsinogen and hydrochloric acid are secreted from the gland cells which lowers the pH from 5.5 to approximately pH 2.5-3.5 (Rose 1997; Sjaastad et al. 2010; Ravindran 2013).

Thereafter, the feed enters the gizzard, where it is mechanically degraded by contractions of the muscle layers, but also with help from the enzymes secreted in the glandular stomach. The contraction process in the gizzard takes between 60 to 90 minutes before the feed continues to the small intestine (Sjaastad et al. 2010). It is in the small intestine the main absorption of nutrients (fat, starch, and protein) takes place. The small intestine can be divided into duodenum, jejunum, and ileum but there are no distinct limit between these segments (Klasing 1998). A pair of caeca are connected to the ileum and one function the caeca are microbial fermentation (S. Adil & S.N. Magray 2012). Finally, the undigested feed, together with uric acid, reaches rectum and cloaca before voided from the intestinal tract as faeces (Rose 1997).

2.1.2 Crop

The crop is a bulging pouch located between the esophagus and the glandular stomach (proventriculus) in birds. It is used for temporarily storage of feed when the gizzard is full. Bolus is passed in smaller portions from the crop to the glandular stomach (Rose 1997; Sjaastad et al. 2010). According to Sturkie & Benzo (1986) when bolus nears the opening of the crop, the state of contraction in the gizzard decides if the bolus will enter the crop or bypass directly to the glandular stomach. Studies by Paştea et al. (1968) showed that there is a correlation between the contractions in the crop and the glandular stomach. Therefore, glandular stomach contractions may influence the crop activity.

The crop has a pH of approximately 5.5. However, Ravindran (2013) found that the pH of the crop could be as low as 4, meanwhile Hinton et al. (2000) concluded that the crop could hold a pH of 6.5 when feed has been withdrawn for 12 hours. The degradation of starch also begins here by the secreted enzyme amylase (Sjaastad et al. 2010; Ravindran 2013). Lactic acid bacteria (LAB) is the most abundant bacterium in the crop and is of great important for the fermentation of the feed (Rose 1997:101; Gussem et al. 2016). Less frequently found bacteria in the crop are coliforms, streptococci, and bifidobacterial (Classen et al. 2016).

The pellets needs water and/or secreted mucus to dissolve in the crop and the feed can therefore stay in the crop between 10 to 60 minutes (Sturkie & Benzo 1986; Dänicke et al. 1999; Aviagen 2018). Dänicke et al. (1999) found that the average retention time for feed in the crop was 50 min. When water is present in the crop, the feed will be moistened, and the crop becomes soft and round. If the broiler chicken has not had enough to drink, it is possible to feel pellets feed in the crop by palpation. The pellet feed will remain solid until the bird drinks or mucus is secreted (Gussem et al. 2016; Aviagen 2018).

To continue, even though water availability is an important factor for the feed transit time in the crop, other factors like the pellet quality, restricted vs. *ad libitum* feeding, lighting programme, temperature, and stress also influence the digestion time (Svihus et al. 2013; Aviagen 2018). Important to highlight is that birds strive to fill the crop prior to resting. During the dark period, the content within the crop changes very slowly (Klasing 1998). Because of that, it is important to make sure that the crop is emptied prior to loading, since it is dark inside the trucks and there is no access to water during transport. Otherwise, the feed in the crop will probably remain when entering the slaughterhouse. Filled crops increases the risk for contaminated carcasses and may cause condemnation, which in turn poses a loss of income for the farmer and the slaughterhouse (May et al. 1990; Buhr et al. 1998; Aviagen 2018).

2.2 Potential bacterial contamination at slaughter

Bacteria belonging to family *Enterobacteriaceae* are gram-negative bacteria and commensals in the intestinal flora of broiler chickens as well as other animals (Hinton et al. 2000). *Enterobacteriaceae* are often used as indicators of faecal contamination, and to evaluate efficacy of cleaning and disinfection to estimate the risk of presence of pathogenic bacteria. Therefore, it can be found on the neck skin that have been contaminated with intestinal content, especially if the slaughter house applies a technology where the broilers are hung up site down with the neck skin facing the floor (Ellerbroek & Lox 2004). Further, it is rod shaped, facultatively anaerobic and oxidase-negative (Adams et al. 2016; Carroll et al. 2019). The optimum condition for growth varies within the *Enterobacteriaceae* family but most bacteria will grow at 37 ± 1 °C and at a pH ranging between 6-8 (VetBact 2022a). The most common genera that belongs to *Enterobacteriaceae* are *Escherichia, Salmonella, Enterobacter, Shigella* and *Klebsiella* (Adeolu et al. 2016).

2.2.2 Escherichia coli

Escherichia coli (*E. coli*) is one of several species belonging to the *Enterobacteriaceae* family. It is a mesophilic bacterium with an optimum growth temperature of $37 \,^{\circ}$ C at a neutral pH, but can grow in the range of 7 to $50 \,^{\circ}$ C (Adams et al. 2016). There are several stereotypes within family Enterobacteriaceae, most of them are non-pathogenic and commensals in the intestines. However, some of them could cause several different kinds of diseases in humans including food borne pathogens that can cause gastroenteritis in all age groups, from infants to elderly people. Common symptoms are diarrhoea, cramps, nausea, and vomiting (Adams

et al. 2016). One of the stereotype of *E.coli* is *Enterohaemorrhagic E.coli* (EHEC) which causes most serve illness (Adams et al. 2016). Within the EHEC, *E. coli* 0157:H7 is the most frequently discussed serotype. Symptoms by *E. coli* 0157:H7 are often related to consumption of undercooked minced beef and green salad contaminated with faeces. Undercooked poultry meat can also lead to illness (Adams et al. 2016).

2.2.3 Salmonella

Poultry meat and eggs are two food categories associated with Salmonella. Salmonella outbreaks in humans often occur after inadequate cooking of contaminated meat or due to cross-contamination in the kitchen. At farm level, Salmonella is spread by poultry consuming feed or water that has been contaminated with infected intestinal content. Rodents and birds can also be a source of infection of Salmonella (Adams et al. 2016). The Swedish Board of Agriculture has established regulations on mandatory Salmonella control of poultry (SJVFS2007:19). According to the regulation (SJVFS2007:19), all broiler producers that have a stock of >500 broilers for slaughter per annum are obliged at, 10 days before thinning and 14 days prior to slaughter, take samples from the chickens which and must be tested free from Salmonella spp. The Salmonella sampling is performed by boot sock and must be performed according to the Appendix in regulations on mandatory Salmonella control of poultry (SJVFS2007:19) by the broiler producer or veterinarian. The analysis must be performed in accordance with the standard method ISO 6579:2022 (SJVFS2007:19). If Salmonella spp. is present, the flock is culled at the farm and destroyed to ensure that no birds reach the slaughterhouse or the market (SFS1999:658). Worth mentioning is that Norway, Finland, and Denmark have also implemented the same Salmonella routine as Sweden (LIVSFS2018:9).

2.3 Swedish broiler production

2.3.2 Housing and management

The concept "all in all out" is applied for conventional broiler chicken management in Sweden. This means that all new hatched chicks are placed in the house at the same time, but also sent to slaughter together (Aviagen 2018). However, the practice "thinning" or "partial depletion" is also applied in broiler chicken management to maximize the production volume (Alfifi et al. 2020). Thinning occurs approximately 28 days after hatching and 20% of flock is sent to the slaughterhouse. At this time, the broiler chickens have reached a live weight of about 1600 grams (Aviagen 2018, 2019).

A vast majority of the chicken houses in Sweden are divided into two compartments. Each compartment has an area which varies from 750 m² to 3000 m². Before chick placement takes place, all areas must be cleaned, and new bedding material (litter) should be placed on the floor. The chicken house must be preheated minimum 24 hours prior to the arrival of the chicks since chicks cannot regulate their body temperature by themselves before day 12-14 (Aviagen 2018). The temperature in the compartment on day 1 should be between 30-35 °C and hold a relative humidity of 60-70 %. During the rearing period, the temperature should be decreased and at the day of slaughter, the compartment should hold a temperature of approximately 20 °C (Aviagen 2018). Whole house brooding is used to keep a uniform temperature in the chicken house. Heat exchanger is gaining more popularity in whole house brooding since they are efficient and creates a controlled environment by heating the ingoing air with the outgoing air (Aviagen 2018).

Nipple lines for access of water are the most frequently used in Swedish chicken houses. Chicks should have access to water from the minute they are placed in the house (Aviagen 2018). According to 8§ of the Swedish Board of Agriculture's regulations and general guidelines on poultry farming in agriculture (SJVFS2019:23), there should be enough nipples so not more than 20 chickens need to share the same nipple during week 0-7 and 15 chickens during week 8-10. Broilers are in general fed with manufactured pellets including all nutrients, but whole grains (mainly wheat, but also barley, and oats) can be used together with a protein concentrate (Aviagen 2018). Feed can be placed on paper or directly on the litter during the first three days before the chick has adapted to use feeders (Aviagen 2018). If the farmer uses round feeders where the chickens have free access to feed, the minimum space requirement for each chick is 8 and 20 mm per chicken during week 0-7 and 8-10, respectively (SJVFS2019:23). The hight of the nipple lines and feeders should be monitored during the growing period to suit the size of the chicken.

2.3.3 Legislation and recommendations

The Swedish Poultry Meat Association, also known as Svensk Fågel in Swedish, is representing the whole production chain for Swedish meat-type chickens and turkey, from breeding companies and hatcheries to producers and slaughterhouses (Svensk fågel 2017). The Swedish Poultry Meat Association have come up with a quality and control programme besides Swedish legislation, which will be explained later in this chapter. By fulfilling the quality and control programme, chicken and turkey products will be labelled with the Swedish Poultry Meat Association own logotype, called "Gula pippin".

Production of Swedish broiler chickens are regulated by the EU and more specific the council of the European Union (2007/43/EC) regulates the minimum standards for the protection of chickens kept for meat production. In addition to the EU directive, Swedish broiler producers have to comply with additional requirements stipulated by the Swedish Board of Agriculture's regulations and general advice on poultry farming in agriculture (SJVFS2019:23). However, beyond the council of the European Union (2007/43/EC) and regulations and general advice on poultry farming (SJVFS2019:23). Swedish conventional broiler producers must follow obligations specified in the Swedish Poultry Meat Associations quality and control programme to be able to deliver chickens to Kronfågel, or any other slaughterhouse connected to the Swedish Poultry Meat Association.

Within the EU, a conventional broiler chicken producer is allowed to have between 33 to 42 kg chicken live weight/m² depending on if the producer fulfils specific requirements in the Annex I, II and/or V in the council of the European Union (2007/43/EC), whereas the Swedish Board of Agricultural has set the stocking density to 20 kg/m². Broiler producers that are members of the Swedish Poultry Meat Association are allowed to have up to 36 kg/m² or a maximum of 25 broiler chickens per m² (SJVFS2019:23), if the producer fulfils the requirements in the Swedish Poultry Meat Associations quality and control programme, which includes animal welfare and feet welfare programmes, and *Salmonella* and *Campylobacter* controls.

According to the regulation of the European parliament and the council of the European Union (1831/2003) antibiotics, except for coccidiostat, is not allowed to give preventively to commercially reared broilers. Since 2011, the Swedish Poultry Meat Association collects information about how many broilers, parents, and grandparents that have undergone treatments each year. In the report Swedres-Svarm (2020), the Swedish National Veterinary Institute (SVA) summarized the number of broiler flocks treated with antibiotics from 2011 to 2020. The number of flocks treated varies a lot. When the compilation started in 2011, six broiler flocks out of 3185 slaughtered were treated with antibiotics. The number of antibiotic treatments peaked in 2019 when 54 flocks, out of 3368 flocks, were treated. Other years' that are worth to highlight are 2012 and 2017. Both years, only one flock out of 2853 and 3300, respectively, had undergone treatment (National Veterinary Institute & Public Health Agency of Sweden 2020).

2.4 Recommended routine to avoid filled crops at slaughter

Adaption to avoid filled crops and to minimize an excessive weight loss for broilers who are sent to slaughter starts three days prior to catching. To achieve this, feed and water withdrawal time and light programme are adapted. When deciding the feed withdrawal time, following parameter must be considered: time in house with feed, catching time, transport time, lairage, and local legislation (Aviagen 2018). According to the council of the European Union (2007/43/EC), and 6§ of the Swedish Board of Agriculture's regulations and general guidelines on poultry farming in agriculture (SJVFS2019:23), feed must not be withdrawn more than 12 hours before planned slaughter time. For farmers using whole wheat, Aviagen (2018) recommends to stop feeding wheat two days prior to catching, to avoid kernels in the intestinal tract.

Broiler producers are recommended to withdrawal the feed 8-12 hours before processing starts to avoid a filled crop and intestines at slaughter. At processing, feed in the crop may be an indication that feed has been withdrawn less than 8 hours prior to slaughter, meanwhile litter in the crop can be an indication that the chicken has been without feed for more than 12 hours. Leaving the feed cups on the floor until catching starts perhaps lowers the risk for chickens to peck in the litter (Aviagen 2018). Northcutt et al. (1997) reported that broilers that had been without feed less than nine hours prior to slaughter had feed in the crop, meanwhile broilers slaughtered nine hours after feed withdrawal had a watery content in the crop. Their study also showed that broilers slaughtered 12 hours after feed withdrawal had empty crops. To continue, neither the council of the European Union (2007/43/EC) nor the regulations and general guidelines on poultry farming in agriculture (SJVFS2019:23), describe when water earliest can be withdrawn prior to slaughter but Aviagen (2018) recommends free access to water until catching starts to avoid dehydrated broilers.

According to the council of the European Union (2007/43/EC) and 9§ of the Swedish Board of Agriculture's regulations and general advice on poultry farming (SJVFS2019:23), broiler chickens shall have a dark period of at least six hours per day, of which four hours are coherent. However, this directive does not include the first production week and the three last days before slaughter. The Ross Broiler Management Handbook (2018) recommends, but emphasize that local legislation applies, that from day seven, a minimum of four-hour darkness is required to avoid disrupted feeding and water behaviour due to lack of sleep and to not reduce the animal welfare. Three days prior to slaughter, broilers are recommended to be provided with 1 hour of dark and 23 hours of light to remain calm during catching and to stabilize their eating behaviour after feed withdrawal. The light intensity should be reduced to an extent so catching can occur safely (Aviagen 2018).

Research of how light programmes affects crop content has also been performed. Duve et al. (2011) investigated how feeding behaviour and intestinal content were affected of dividing the dark period. The results showed that broilers exposed to an eight-hour dark period used their crop in a greater extent as a storage organ compared to those broilers that had had a dark period of four+ four hours.

When catching starts, chickens can be caught either by hand or by a chicken harvester (Aviagen 2018). Research by Wolff et al. (2019) has shown that catching chicken mechanically can reduce stress and increase animal welfare for the chickens. Examples of chicken harvesters are Chicken Cat Harvester (JTT Conveying A/S, Bredsten, Denmark) and Apollo Generation 2 (CMC Industries – Ciemmecalabria S.r.l., Cazzago, Italy) (Mönch et al. 2020). Caught chickens are loaded to trucks and transported to the slaughterhouse. According to the Swedish Board of Agriculture's regulations and general guidelines on the transport of live animals (SJVFS2019:7), broilers must be slaughtered within eight hours after catching, but the time can be extended with four hours if transportation occurs during the dark hours. Broilers are left in a heated and ventilated hall when they arrive to the slaughterhouse. The lairage time for broilers that are slaughtered in the beginning of the day is 180 minutes. Subsequently during the day, the lairage time will be lowered and can be as short as seven minutes (Unpublished material).

The stunning does not start until a veterinarian has approved the chickens to be in good condition (The European parliament and the council of the European Union 853/2004). When approval is given, broilers are stunned according to the Swedish Board of Agriculture's regulation and general advice on slaughter and other killing of animals (SJVFS2020:22). After stunning, broilers are bled to ensure death (The council of the European Union 1099/2009). It takes approximately 10 minutes from when the chickens are stunned until the chicken carcass reaches the meat inspection. Meat inspection can be performed by trained employees at the slaughterhouse under supervision of an official veterinarian if approved by the national authorized authority, which in Sweden is the Swedish National Food Agency (The European parliament and the council of the European Union 853/2004).

3. Materials and methods

This project contains four different parts:

- Telephone interview where broiler chicken producers were asked about feed and water routines, lightning programme, and temperature prior to slaughter.
- Evaluation of possible links between routines on farms prior to slaughter and percentage of crops assesses as filled in the meat inspection. Including considering the impact of working shift in the meat inspection.
- Investigation of crop fill in chickens at farms prior to catching for slaughter, followed by dissections of a sample of filled crops from the same flocks at the slaughterhouse.
- Microbiological analysis regarding the amount of *Enterobacteriaceae* and total aerobic bacteria on the neck skin from carcasses at the slaughterhouse, before and after rinsing the carcasses with water.

3.1 Collaboration

The project was conducted in collaboration with the company Kronfågel, which is a subsidiary to Scandi Standard, and their Swedish conventional broiler chicken producers (Scandi Standard 2022). Out of 39 producers, 22 chose to participate in the project.

Kronfågels role in this project was to delegate the contact between me as an investigator and the broiler producers. Kronfågel also provided access to the slaughterhouse and shared data regarding percent of chickens assessed as having filled crops in the meat inspection. Staff at the slaughterhouse also assisted in collecting samples for crop dissection and the microbiological analysis.

3.2 Farm interview

A description of the project and a registration of interest were emailed to all producers contracted by Kronfågel on the 15^{th} of February 2022. The producers had seven days, whereof five working days, to reply if they were interested in participating in the project. Those who participated could choose to conduct the interview during a farm visit, a telephone interview, or a Teams/Zoom meeting. The registration of interest closed on the 22^{nd} of February 2022. The survey was emailed to the registered producers on beforehand, so they had time to prepare their answers before the interview was conducted.

Microsoft Word was used to create the survey. The survey had 24 questions written in Swedish since it was easier to communicate with the producers in their native language. The survey had two sections. Section 1 had questions related to thinning meanwhile Section 2 was focused on the main slaughter (Appendix I). The telephone interviews and Teams/Zoom meetings took between 20 to 60 minutes per participant.

3.3 Farm visits

Five broiler farms, named A, B, C, D and E in this report, were selected for farm visits. The farms were selected so there would be maximum one visit per week, and the slaughter should occur in the months of March or April to suit the time framework. All farm visits started with practicing hygiene routines before entering the production plant, which includes hand washing and changing to clean clothes and shoes. Safety clothes was also worn (disposable coverall DC 10 Blue from L.Brador, plastic boot covers by Granberg, and a 3M 9322+ Fine Dust mask with Valve FFP2).

3.3.2 On farm crop investigation

At each farm, the compartment was visually divided into six rows across the long sides. A number of 60 broiler chickens per compartment, ten per row, were randomly selected to investigate if the crop was filled by palpation of the crop while holding the bird. Each crop was categorised at site as filled, semi-filled or empty depending on the texture of the crop. If the crop was categorised filled or semi-filled, it was further categorised into pellets, hard, soft, or watery (Fig.1). Crops

categorised as hard had a texture like playdough. The texture of soft crops reminded of a slurry, meanwhile crops with water felt washy.

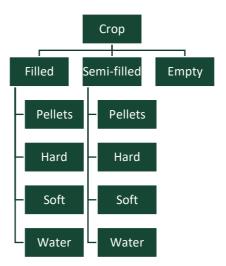


Figure 1. Schematic illustration of crop categorization at farms based on palpation. Crops were categorised as filled, semi-filled, or empty. Filled and semi-filled crops were further sub-categorised into pellets, hard, soft, or water.

At all farm visits, crop investigation was performed on birds in one compartment, and the palpation of crops started approximately 60 minutes before planned catching time. The compartments were chosen to correspond the answers in the survey questions as different routines were sometimes applied in different compartments. The compartment should preferably also be the first one slaughtered from that specific farm. However, this was not possible in farm E, where compartment 4 was chosen since it was correlated to the answered question in the survey but was not the first compartment to be slaughtered from that farm. Planned catching time differed between the farms. At farm A, the catching time was planned to start at 21.02. Farm B had planned catching time at 02.44, farm C 21.42, farm D 02.13, and farm E approximately 02.00 (Table 2).

3.4 Slaughterhouse visits

3.4.2 Crop control routine at slaughter

At the slaughterhouse, two to three meat inspectors per shift, took turns in performing the crop control. The control was performed once every 30 minutes for all slaughtered flocks during main slaughter. Therefore, the number of controls could vary depending on how many chickens there were within one flock.

For five minutes, the meat inspector stood still, looking at the slaughter line and counted the number of carcasses assessed as having filled crops, using a tally counter. Approximately 1000 chickens were controlled during the 5 minutes. If more than 2% of the carcasses had filled crops during the control, the employees were allowed to discard carcasses with filled crops until the next control was performed. However, if less than 2% had filled crops, the employees were obliged to manually cut off the filled crops and re-hang the carcasses to the slaughter line. When the controls for a flock were completed, crop max (%) and crop mean (%) were calculated. A made-up, but plausible, example has been illustrated below (Table 1, Fig. 2 and 3). A farm had five crop controls for one slaughtered flock. Control 1 had five filled crops, control 2 three filled, control 3 had no filled crops, control 4 had nine, and control 5 had seven filled crops with 1000. In this case, control 4 had most filled crops (Table 1).

Table 1. An example of possible outcomes in the crop controls performed by the meat inspector. Each crop control was conducted during five minutes under which 1000 crops were inspected. Filled crops were the number of crops assessed as being filled with content (n).

Crop control	Filled crops (n)	Controlled crops
1	5	1000
2	3	1000
3	0	1000
4	9	1000
5	7	1000

The calculation below was performed to calculate and producers crop max (%) value for the slaughtered flock. The crop max value became of 0.9% (Fig. 2):

$$\frac{9}{1000}$$
 * (100) = 0.9%

Figure 2. The calculation illustrates how crop max (0.9%) was calculated after the crop controls for a specific compartment with broilers.

To calculate crop mean (%), the number of filled crops for each control were add up and divided with the total number of controlled carcasses. The crop mean for the producer became 0.48% (Table 1, Fig. 3).

$$\frac{(5+3+0+9+7)}{5000} * 100 = 0.48\%$$

Figure 3. The calculation illustrates how crop mean (0.48%) was calculated after the crop controls for a specific compartment with broilers.

3.4.3 Investigation of crop content at slaughter

At the slaughterhouse, staff working in the meat inspection selected 60 broiler chickens with filled crops, from the same compartment as the palpation had been performed during the farm visit. The staff cut off the crops by hand and left them on a chopping board for further investigation of the crop content. All 60 crops were dissected and photographed.

Based on the photos, the crop content was visually categorised into the following twelve categories, water, water with kernels, water with feathers, water with kernels and feathers, feed, feed with kernels, feed with feathers, feed with kernels and feathers, litter, litter with kernels, litter with feathers, or litter with kernels and feathers (Table 2, Appendix III).

3.5 Statistical analyses - Farm interview and impact of working shift on crop fill

Flock mean percentages of carcasses categorised as having content in their crops at slaughter were received from the slaughter company. Flock was defined as birds arriving at the same time to the farm, being housed together in the same compartment. The data file comprised all flocks slaughtered in the period 2021-11-01—2022-03-11. The file included data from slaughter at thinning of flocks and from main slaughter, but only data from the main slaughter were included in the analyses. In total, data from 449 flocks reared on 41 different farms were included in the analyses. The number of flocks from the same farm varied between 2 and 28. Of the 449 flocks, 292 had been inspected by shift 1 and 257 by shift 2. There were totally 48 employees working at the meat inspection at the slaughterhouse. Shift 1 and shift 2 had 24 employees each, whereof 19 employees from each shift work morning and afternoon shifts every second week. Five employees from each shift (1 and 2) worked only mornings or afternoons

Data from 22 different farms were gathered in the interviews. In the statistical analyses of results in the survey 20 farms were included, 2 were excluded due to missing values. Four factors from the interviews were included in the statistical evaluation. These were whole wheat kernel, feed withdrawal, water withdrawal, and light. In the category whole wheat kernels, producers were categorised as Yes or No - Yes for producers using whole wheat kernels and No for the ones not using it in their feed. For feed withdrawal, producers were categorised as feed withdrawal 0-5 hours or feed withdrawal 6-9 hours prior to planned catching time. Similar group categorisation was performed for water withdrawal. Producers were either categorised as water withdrawal 0-15 minutes or water withdrawal 16-60 minutes prior to planned catching time. The light category stands for the number of days prior to slaughter that chickens have 24 hours. Producers were categorised as either 24-48 hours or 49-72 hours, depending on how many days the chickens had full light in the compartment.

All statistical analyses were performed with Proc mixed in SAS statistical software (release 9.4, SAS Institute Inc., Cary, NC). According to diagnostic plots of residuals the percentages of carcasses with filled crops deviated from normality and homoscedasticity, and a logit transformation of data was therefore conducted prior to statistical analyses. The significance level was set to p < 0.05.

The effect of work shift was analyzed using data from all farms in the dataset with the following model:

Proc mixed; CLASS shift farm; MODEL crop = shift; RANDOM farm farm*shift;

Effects of routines applied at the farm prior to slaughter, according to the interviews, were analyzed using the following model:

Proc mixed;

CLASS whole wheat kernels, feed withdrawal, water withdrawal, light; MODEL crop = whole wheat kernels, feed withdrawal, water withdrawal, light;

3.6 Bacteriological analysis

The aim with the bacteriological analysis was to evaluate the differences in *Enterobacteriaceae* and total aerobic bacteria before and after carcasses had been rinsed inside and outside along the slaughter line. The sampling was split into three sampling occasions over a two-week period. Ten broiler chickens from different producers were randomly selected from the slaughter line at each visit, which resulted in 60 pieces of neck skin were sampled 30 before and 30 after washing.

3.6.2 Inside/Outside washer

After the meat inspection, before entering the cooling line, the carcasses entered a mechanical washer (Inside/outside washer RW-16 RS, Marel Garðabær, Iceland). The washer was equipped with two spray pipes. One spray pipe cleaned the inside of the carcass meanwhile the other pipe cleaned the breast and back of the carcass. A row of static sprayers washes the opposite side of the carcass. Every minute, 220 carcasses passed through the washer.

3.6.3 Sample collection

At all three visits, neck skin from ten carcasses were sampled and labelled before (A) and after (B) the washer. To collect sample A, the carcass was taken down from the slaughter line by Kronfågel staff and neck skin samples of approximately 10 g was aseptically collected with a sterile pair of scissors. The samples were placed in Stomacher bags labelled with an A and a number (1-30).

Before the carcass was re-hanged on the line, a red plastic ribbon was tied around the wing to recognise the same carcass after the washer. After the carcass had passed through the neck skin chopper and the washer it was taken down a second time to collect sample B. Again, approximately 10 g of neck skin was cut off with sterile pair of scissors and the samples were placed in a new Stomacher bag labelled with B and a corresponding number (1-30).

All Stomacher bags (A1-30 and B1-30) were placed in an esky right after sampling and transported to the lab at the Swedish University of Agricultural Sciences (SLU) in Uppsala the same day of sampling.

3.6.4 Bacteriological analysis

The bacteriological analysis started the same day as the sampling. All samples were kept in an esky below 10°C until analysis. No samples were frozen. Every neck skin sample was weighted and nine times the volume (approximately 90 ml) of buffered peptone water (CM0509, Oxoid, Basingstoke, UK) was added to each sample. The stomacher bag with neck skin sample and buffered peptone water was homogenized for 2 min at 240 rpm (easyMIX Lab Blender, AES-Chemunex, Weber Scientific, Hamilton, NJ). After homogenization, a 10-fold dilution was prepared.

3.6.5 Enumeration of Enterobacteriaceae

For quantification of bacteria belonging to family *Enterobacteriaceae* NMKL 144, 3rd ed. *(22)* was used. Briefly, from each dilution, 1.0-mL from the 10-fold dilution described in sub-chapter 3.6.3 was mixed in a petri dish (9 cm in diameter) with 10 to 15 mL of violet red bile glucose agar (VRBG) (BD, Sparks, MD) (Fig.4). After

the agar had solidify, an overlayer of approximately 5 mL of VRGB agar was added.

Accidently, plate 1-10 A and B were incubated in 30 ± 1 °C for 16 hours instead of 37 ± 1 °C. Hence, the plates were transferred to 37 ± 1 °C after 16 hours and incubated for another 8 ± 2 hours before bacterial count was performed. Plates A and B 11-30 were incubated at 37 ± 1 °C for 15 ± 2 hours. Bacterial count was performed on plates with 2 to 150 colonies.

To identify *Enterobacteriaceae*, five colonies were re-cultured on horse blood agar (SVA, Uppsala, Sweden) and incubated at 37 ± 1 °C for 24 ± 2 hours and then tested for the production of oxidase (<u>www.vetbact.org.se</u>), assessed May 2022). Oxidase-negative colonies were identified to species level using matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) (Brucker Daltonics, Bremen). The number of micro-organisms were compiled in Microsoft Excel and calculated by using the standard formula ISO 7218.2007/A1:2013 (Fig. 5). The number of *Enterobacteriaceae* bacteria was further expressed as log CFU per gram, with a detection limit of 1.0 log CFU/g.

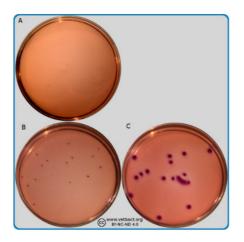


Figure 4. Members of the family Enterobacteriaceae cultured on Violet red bile glucose (VRBG) agar. Bacteria have not been cultured on the plate image A, on the plates B and C bacteria belonging to family Enterobacteriaceae could be identified.

Standard Formula (ISO 7218:2007/A1:2013)

N = Σ C / (V × 1.1 × d)

- N no. micro-organisms
- ΣC sum of the colonies on 2 plates from successive dilutions
- V volume of the inoculum/plate, in ml
- d 1st countable dilution retained
- 1.1 a factor used when the so-called weighed mean is calculated from two plates (if only one plate is used, the factor should be 1.0 and for three plates 1.11)

Figure 5. The standard formula ISO 7218.2007/A1:2013 was used to calculate the number of bacteria.

3.6.6 Identification of bacteria by MALDI-TOF

Sixty re-cultured colonies from horse blood agar were randomly selected to be confirmed and identified to species level by MALDI-TOF MS. The re-cultured colonies were smeared on a MALDI target-plate and 1 µl of matrix (a mixture of: α-cyano-4-hydroxycinnamic acid (HCCA), 250 µl 50% acetonitrile, and 2.5% trifluoroacetic acid) was added on top of the colonies. The MALDI target-plate was irradiated with laser UV light. Moreover, the UV light caused breakage of the molecules in the bacteria into a fragment, which was hurled towards a detector. The time it took for the fragments to reach the detector (time of flight) was measured. The time it takes for the bacterium to reach the detector is dependent on the size and the charge of the molecule. The broad characteristic mass spectrum and fragments the molecules gave rise to was compared to a database with stored mass spectra of well-known bacteria. Bacteria can be identified in different levels depending on the score value. A score value between 0 and 1.699 indicates that the mass spectra do not match any of the bacteria in the database and can therefore not be identified. Hence, if the mass spectra get a score value between 1.700 and 1.999, the unknown isolates genus can be identified but not the species level. For a bacterium to be identified on both genus and species level, a score value between 2.0 and 3.0 is (Bruker Daltonics, Bremen)(Carolis et al. 2014; VetBact 2022a). The identifications of the bacteria were summarised in Table 4.

3.6.7 Enumeration of total aerobic bacteria

Total number of aerobic bacteria were enumerated according to NMKL 86, 5th ed. (24), using the same 10-fold dilution as described in sub-chapter 3.6.3. From each dilution 1.0 mL was mixed in separate petri dishes (9 cm in diameter) with 10 to 15 mL of plate count agar (PCA) (Oxoid, Basingstoke, UK) (Fig.6). After the agar had solidify, an overlayer of approximately 5 mL PCA was added. Samples 1-10 A and B were accidently incubated at 37 ± 1 °C for 16 hours but were transferred to 30 ± 1 °C for incubation another 56 ± 2 hour. Samples 11-30 A and B were incubated at 30 ± 1 °C for 72 ± 2 hours. Bacterial counts were performed on plates with 2 to 250 colonies. The number of micro-organisms was calculated by using the standard formula ISO 7218.2007/A1:2013 (Fig. 5). The total aerobic bacteria count was expressed as log CFU per gram and with a detection limit of 3.0 log CFU/g.

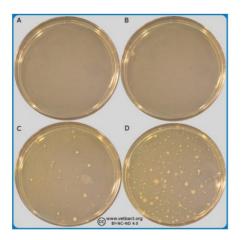


Figure 6. Plate Count Agar (PCA), used for quantification of total aerobic bacteria. Bacteria have not been cultured on the plate in image A. On plate in image B, four colonies could be calculated, on plate C 45 colonies, and plate D > 250 colonies.

3.6.8 Statistical analysis of bacteria

The results were compiled and analysed using Microsoft Office Excel and the website based statistical programme "Social Science Statistic" (https://www.socscistatistics.com), accesses in May 2022). Bacterial count (CFU per gram) was log transformed. The statistical significance was determined by a paired dependent means *t*-test. The *t*-test was performed on both *Enterobacteriaceae* and total aerobic bacteria before and after treatment in the washer. The significance level was set to p < 0.05.

4. Results

4.1 On-farm crop investigation

During visiting the farms (A, B, C, D, and E), palpation was performed on totally 420 broiler chickens and the results were summarised in Table 2. At farm A, two filled crops were found at palpation of 60 broiler chickens. The two crops were categorised as filled and semi-filled, and both were further sub-categorised into soft, according to the schematic illustration (Fig. 1). Meanwhile the other 58 broiler chickens were categorised as empty. At farm B, seven filled crops were found and three of these were categorised as filled and another three as semi-filled. Both the filled and semi-filled were sub-categorised as soft. One crop was categorised as semi-filled watery, and the rest, 53 crops, were categorised as empty. Farm C had most filled crops at site compared to the other farms. At palpation 14 filled crops found in the compartment. Two crops were categorised as filled soft, seven crops were filled watery, and five crops were categorised as semi-filled watery. The rest, 46 broiler chickens, were categorised as empty. At farm D, ten out of 60 broiler chickens had filled crops. Five crops were categorised as filled soft, meanwhile the other five crops were categorised as filled watery. The rest, 50 broiler chickens, had empty crops. At farm E, palpation was performed in compartment 4 since the survey questions were corresponding to that compartment. After palpation, one crop was categorised as semi-filled soft, two were semi-filled watery, and 57 crops were categorised as empty. In conclusion, none of the filled crops were categorised as pellets in combination with hard, or as semi-filled pellets and hard. Among the filled crops 20 were categorised as soft and ten watery. In the group of semi-filled crops seven were soft and 13 watery. There were 370 crops categorised as empty.

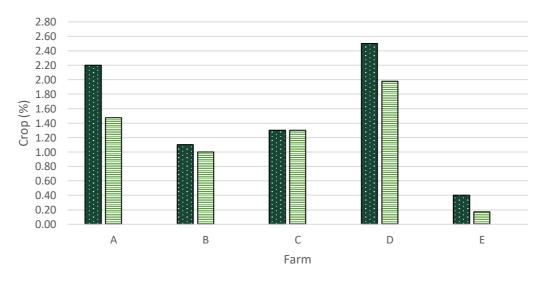
Further, Fig. 7 shows the outcome after meat inspection of crop max and crop mean for each compartment where on-farm crop investigation was performed. Farm D had the highest crop max and mean value with 2.50 and 1.98%, respectively. The feed had been withdrawn for elven hours and 20 minutes prior to slaughter. Furthermore, farm E had the lowest crop max and mean value with 0.40 and 0.17%, respectively, and the feed had been withdrawn for approximately ten hours (Table 2, Fig. 7).

Table 2. A compilation of the used categories, filled and semi-filled with their sub-categories' pellets, hard, soft, and watery, and empty, during the palpation of broiler chickens prior to catching for slaughter at farm A, B, C, D and E. The approximately time when feed was withdrawn, the number of minutes before planned catching time water was lifted, and the planned catching and slaughter time are also expressed. The total numbers of categorised crops are also recalculated into percent (%).

Farm & compartment	Total	Filled Pellets	Filled Hard	Filled Soft	Filled Watery	Semi- filled Pellets	Semi- filled Hard	Semi- filled Soft	Semi- filled Watery	Empty	Feed withdrawal*	Water withdrawal** (minutes)	Planned catching time	Planned slaughter time
A -C1	60	0	0	1	0	0	0	1	0	58	20.32	30	21.02	04.27
B -C1	60	0	0	3	0	0	0	3	1	53	21.15	10	02.44	07.44
C -C1	60	0	0	2	7	0	0	0	5	46	13.42	10	21.42	04.27
D -C1	60	0	0	5	3	0	0	0	2	50	20.15	15	02.13	07.35
E -C4	60	0	0	0	0	0	0	1	2	57	Approx. 21.00	30	Approx. 02.00	Approx. 07.00
Total	420	0	0	20	10	0	0	7	13	370				
Percent (%)	100	0	0	4.8	2.4	0	0	1.7	3	88.1				

* The approximately time the feed line was raised before catching started

** The number of minutes the water line was raised before catching started



Crop max ECrop mean

Figure 7. Crop max and crop mean after meat inspection, expressed in percent (%), for the compartments at farm A, B, C, D, and E where the on-farm crop investigation was performed. The data was achieved from the slaughterhouse.

4.2 Investigation of crop content at slaughter

At the slaughterhouse, the first 60 filled crops from each farm were supposed to be dissected. However, the number of dissected crops varied from 44 to 65. Crop content at slaughter was categorised into twelve categories and the values were recalculated to percent (%) with raw data from Table I (Appendix IV) and summarised in Table 3 and Fig. 8. Most crops (26.1%) at farm A had a content of feed with kernels, meanwhile no crops (0%) had water with feathers. Farm A also had the highest percentage of crops with litter, pure or in combination with, feathers or both feathers and kernels (6.1, 13.8, 4.6, and 7.7%, respectively). However, farm B had the highest percentages of crops with water and feed, 31.8 and 47.7 %, compared to farm A, C, D and E. At farm B no crops were categorised as water with feathers, litter, litter with kernels, litter with feathers, or litter with kernels and feathers. The category feed with feathers was prominent at farm C, just over 50 % of the crops had a content of feed with feathers. The second most abundant category for farm C was water with feathers, with nearly 22% of the crops having a content of water with feathers. Furthermore, the two most abundant crop contents from farm D were feed and feed with feathers, 25 and 30 %, respectively. Most crops at farm E were filled with feed (24.6%). The second most frequently occurring content in the crops were water and feed with kernels and feathers. Both with a percentage of 14.7. Crops with a water content constituted to approximately 28% of the dissected crops, whereof crop content with feed or litter constituted to approximately 61 and 11%, respectively (Fig.9).

Table 3. Categorisation of crop contents after dissection of crops from five farms at the
slaughterhouse. Crop contents were categorised into twelve categories and data was recalculated
into percent (%). The number of dissected crops (n) from each farm is also showed in the table.

Farm	Dissected crops (n)	Water (%)	Water with kernels (%)	Water with feathers (%)	Water with kernels & feathers (%)	Feed (%)	Feed with kernels (%)	Feed with feathers (%)	Feed with kernels & feathers (%)	Litter (%)	Litter with kernels (%)	Litter with feathers (%)	Litter with kernels & feathers (%)
А	65	1.5	7.7	0	3.0	12.3	26.1	6.1	10.7	6.1	13.8	4.6	7.7
В	44	31.8	6.8	0	2.3	47.7	4.5	4.5	2.3	0	0	0	0
С	64	9.4	0	21.9	0	12.5	1.6	51.6	0	1.6	0	1.6	0
D	60	8.3	1.7	10	3.3	25	5	30	8.3	0	0	6.7	1.7
Е	61	14.7	1.6	11.5	8.2	24.6	13.1	4.9	14.7	0	4.9	0	1.6
Total	294	11.9	3.4	9.2	3.4	22.8	10.5	20.4	7.5	1.7	4	2.7	2.4

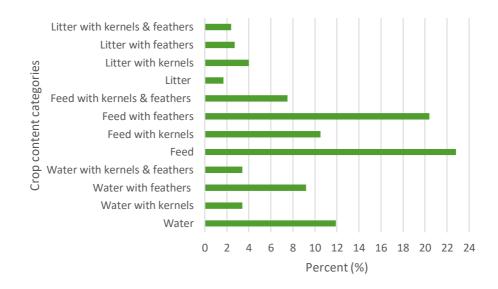


Figure 8. Percentage of investigated filled crops at broiler slaughter categorised in different categories based on visual assessment of their content. Total number of investigated crops from five farms was 294.

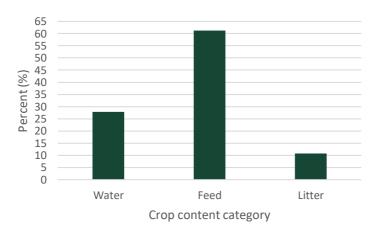


Figure 9. Percentage of filled crops at broiler slaughter compiled to three large categories, water, feed, and litter. Total number of investigated crops was 294.

4.3 Farm interview

The analyses of the impact of whole wheat kernels, feed withdrawal, water withdrawal, and light on filled crops at slaughter, based on the interviews, did not reveal any statistical differences (whole wheat kernels p<0.38; feed withdrawal p<0.54; water withdrawal p<0.69; light p<0.87) (Fig. 10).

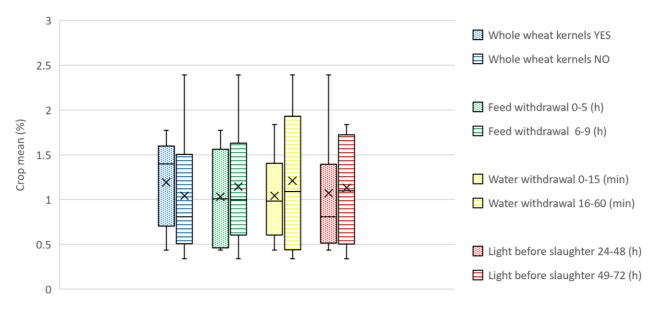


Figure 10. Effects of different on-farm routines prior to slaughter of broilers on crop fill at slaughter, based on interviews of 20 farmers and data from crop fill assessment at slaughter. Boxes shows values between the 25th and 75th percentiles.

4.4 Impact of working shift on crop fill

Work shift performing the inspection at slaughter significantly affected the percentage of carcasses judged as filled in the meat inspection (p < 0.002). Flocks inspected by shift 2 had a higher percentage of carcasses assessed as filled (mean ± sd 1.11± 1.13) compared to shift 1 (mean ± sd 0.74± 0.57) (Fig. 11). It has to be mentioned that this study does not tell which shift that has judged filled crops correctly.

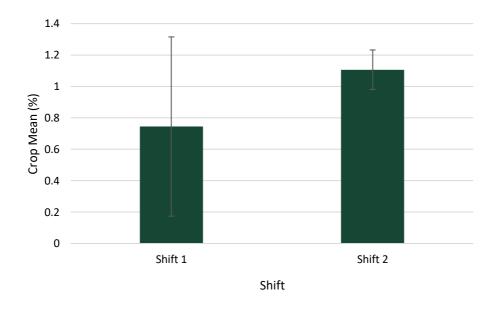


Figure 11. Assessment of filled crops at broiler slaughter performed by shift 1 and 2. The mean percentage of filled crops was higher for carcasses inspected by shift 2 compared to shift 1.

4.5 Enumeration of Enterobacteriaceae

The number of bacteria belonging to family *Enterobacteriaceae* varied from 3.2 to 5.0 log CFU/g before washing, and 3.0 to 5.7 log CFU/g after washing. There was a significant (p<0.01) difference before and after treatment in the washer. One sample (A + B) was removed from the analysis, because the B sample (after washing) was not prepared in accordance with the lab manual. *Enterobacteriaceae* were reduced in 18 (62%) of 29 samples, meanwhile seven (24%) of the samples had an increase. The average number of *Enterobacteriaceae* was before treatment 4.1 log CFU/g and after 3.8 log CFU/g. Four (14%) of the samples did not show either a decrease or increase in the number of *Enterobacteriaceae* after the wash (Fig. 12).

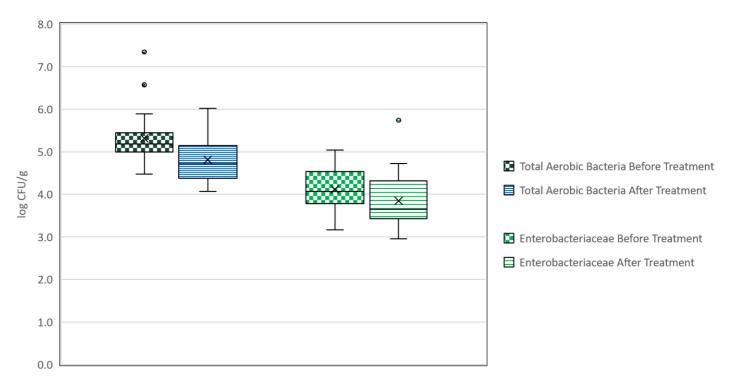


Figure 12. Number of aerobic bacteria and Enterobacteriaceae from neck skins sampled chicken before and after treatment by the washer. Boxes shows values between the 25th and 75th percentiles and outliers as circles.

4.6 Identification of bacteria by MALDI-TOF MS

In the first MALDI-TOF MS analysis, duplicates of samples 1-10 A and B were performed. All analysed isolates got a score value between 2.06 and 2.58, respectively and 8 of 10 isolates were identified as *E. coli* whereas one isolate was identified as *Proteus mirabilis (P. mirabilis)* and one as *Aeromonas veronii (A. veronii)* (Table 4). In the second occasion, sample 11-30 A and B were analysed.

This time, one isolate from each sample was analysed. All isolates got a score value above 2.0 and ranged from 2.18 to 2.58. All isolates were identified as *E. coli*, except one, which was identified as *Aeromonas caviae (A. caviae)* (Table 4).

Table 4. Bacteria isolated from VRBG samples 1-10 A and B, and 11-30 A and B and identified by using MALDI-TOF MS. The total number (n) of each identified bacterium is also expressed in percent (%).

Bacterium	1-10 A and B	11-30 A and B	Total	Total		
	(<i>n</i> =20)	(<i>n</i> =40)	(<i>n</i> =60)	(%)		
E. coli	18	39	57	95		
P. mirabilis	1	0	1	1.67		
A. veronii	1	0	1	1.67		
A. caviae	0	1	1	1.67		

4.7 Enumeration of total aerobic bacteria

The highest number of aerobic bacteria observed before and after treatment were 7.3 and 6.0 log CFU/g respectively, and the lowest observed values were 4.5 and 4.1 log CFU/g, respectively. This result clearly shows that total aerobic bacteria are significantly reduced (p<0.001) during the washing (Fig. 12). The average reduction in total aerobic bacteria by washing was 0.5 CFU/g. The same samples as mentioned in sub-chapter 4.5, was removed from the data compilation. There was a reduction of total aerobic bacteria in 23 (80%) of 29 samples, and an increase in five (17%) samples. In one neck skin no differences in the number of bacteria could be found before and after washing.

5. Discussion

5.1 On-farm crop investigation

On-farm crop investigations showed that the majority (88.1%) of the total palpated crops were empty, meanwhile the rest (11.9%) were either categorised as filled or semi-filled with a soft or watery content. Even though most crops were empty prior to slaughter, it is desired to decrease the number of filled crops further to minimize the risk for crop content to contaminate broiler carcasses at slaughter. Research performed on palpation of broiler crops prior to slaughter is limited and it is therefore difficult compare results from different studies.

Catching time was between 21.00 and 02.00 for all producers and palpation started approximately 45 to 90 minutes before catching. Farm A had most crops, 58 of 60, categorised as empty meanwhile farm C had least number of crops categorised as empty, 46 of 60. With these on-farm results, one would have expected farm A to have the lowest percentage of filled crops in the meat inspection and that farm C the highest. Instead, farm D with 50 of 60 crops categorised as empty had the highest (1.98%) and E had the lowest percentage of filled crops (0.17%). These findings show how hard it is to predict the outcome of crops assessed as filled. Based on these findings, the relevance for the on-farm crop investigation can be questioned since there is limited scientific research to compare with and the palpation is based on a subjective judgement. Other limiting factors in this study are the number of palpated crops on the farms, and pre-slaughter factors like transportation of broilers from the farm to the slaughterhouse, and how the lairage time effects the content in the broiler crop. Only 60 crops per farm were palpated, a greater number of palpated crops would make the result more reliable. Important to highlight is that the palpation should preferably occur as close as possible prior to planned catching time to get a representative result of broilers that have a filled crop. The transportation and lairage time are two factor that has not been studied in this thesis. Mönch et al. (2020) looked at how pre-slaughter factors like catching, transport, and lairage, effect the animal welfare of broilers and concluded that not only the loading method (manual or mechanical) impact the welfare of broiler chickens. Other factors e.g. physical condition of personnel and

intensity of loading are important factors to consider. Similar studies could be performed but with the aim to see how the crop content is affected of these factors.

5.2 Investigation of crop content at slaughterhouse

After compilation of the dissected crops at the slaughterhouse, it was found that feed in the crop represent the largest category, with 61.2%, followed by a watery crop content of 27.9%, and litter content of 10.8%. According to (Aviagen 2018), feed in the crop is an indication of that the feed withdrawal time has been insufficient, whereas litter in the crop is a sign of that the chicken may have been without feed too long and therefore has started to peck in the litter. To avoid broilers pecking in the litter, Aviagen (2018) suggests that feeders shall remain in bird height until catching starts, even if they are empty. Important to highlight is that a crop content with water, feed and/or litter can occur even if broiler producers follow local legislation (SJVFS2019:23) and do their best to optimise the feed and water withdrawal time, and light programme. The investigation of the crop content from farm A showed that the content was mainly categorised as crops with feed and crops with litter. As aforementioned, Aviagen (2018) explains that feed withdrawal time has been insufficient if feed remains in the crop at slaughter meanwhile litter in the crop indicates that feed withdrawal time has been too extensive. According to this statement, crop content results obtained from farm A would then indicate that feed withdrawal time has mainly been insufficient but also too extensive. This result may be interpreted as that feed withdrawal time is not the only factor affecting the crop content. Unfortunately, a combination of on-farm factors was not investigated in this study due to time limitations.

At the meat inspection farm D had the highest percentage of filled crops (1.98 %), meanwhile farm E in contrast had the lowest percentage of filled crops of 0.17%. Both farms had withdrawn the feed between ten to eleven hours prior to slaughter. In a study of broiler crops dissected at different feed withdrawal times, Northcutt et al. (1997) found that the crops were empty after twelve hours of feed withdrawal and had a water content after nine hours. Broilers that had not had access to feed three hours prior to slaughter had feed remaining in the crop. Broilers from farm D and E had a crop content consisting mainly of feed at the dissection, which according to Northcutt et al. (1997) should not be the case, although a watery content could occur after nine hours. The findings in this study therefore contrast with Northcutt et al. (1997) findings since feed was found in crops from both farm D and E.

There were some drawbacks with the dissection of crops at slaughter. The number of dissected crops varied from 44 to 65 crops per farm. For further studies, dissection on a greater number of crops is recommended to obtain even more

reliable results since 44 to 65 crops are not that many crops in relation to the total number of reared broilers within a flock. The categorisation of crops based on documentation by photos was difficult to perform because some crops had contents that was in between two categories, although the number of categories were well-sized. An example of when it was hard to distinguish two categories was when there was feed present in the crop but also water.

5.3 Farm interview

The statistical analysis of the answers in the survey did not show any significant effect on mean percentage of filled crops at slaughter. To be able to conduct the statistical analysis the answers in minutes, hours, and days had to be categorised into two intervals such as e.g. short period of feed withdrawal or long. This way of categorization created categories with large intervals e.g. feed withdrawal 0-5h and 6-9h prior to catching. The large intervals may influence the result. If the categories had been narrower, for example 5-6h and 7-8h, the outcome might have been different. One limitation is that only answers from 20 producers were used in the statistical analysis. The result could had been more reliable if several producers had participated in the project, but 20 of 39 possible answers are still more than expected from the start. Even though individual farm factors did not show a significant difference on the mean percentage of filled crops, it was not possible to include the different farm factors in the same statistical model due to limited data available. For further studies, another approach needs to be considered in order to combine several factors in the same analyses. Northcutt et al. (1997) found that a combination of feed withdrawal time and the farm broilers had been reared on had a significant effect on crop fill. The specific content in crops, e.g. feed, litter etc. was not investigated in the study by Northcutt et al. (1997), but the results contrast with the findings in this study since no effect of on-farm factors and crop fill was found in the present study.

It is beneficial for the animal welfare that water withdrawal time did not show any correlation with mean percentage of filled crops after slaughter. The producers can then let the broiler chickens have access to the water until a few minutes prior to catching, this will also minimize the risk for broilers to be dehydrated (Aviagen 2018). Light programme can have effect on the feed behaviour of broilers (Duve et al. 2011), but in this study no significant difference was found on crop fill between farms having continuous light for 24-48, or 49-72 hours prior to slaughter. However, Duve et al. (2011) looked at how light programmes with eight hours versus four + four hours of darkness, affected the feed intake of broiler chickens. They found that broilers used the crop as a storage organ to a larger extent when given eight hours of darkness compared to broilers reared with four + four hours of darkness, because broilers given eight hours of darkness prepared themselves to be without feed for a longer period. For further studies within this topic, it is suggested to deeper analyse how the light programme at farm level effect the crop content. However, as aforementioned it cannot be excluded that other factors like stress, temperature, transportation, or a combination of farm factors which are not investigated in this study, may have an impact on the crop content.

5.4 Impact of working shift on crop fill

The statistical analysis of impact of work shift on percentage of crops assessed as filled at slaughter showed that flocks inspected by shift 2 had a significantly higher percentage of carcasses assessed as filled compared to shift 1. In a study by Törmä et al. (2022), broilers with feed left in their crops/pendulous crop, were evaluated at four slaughterhouses in Finland. The study showed that slaughterhouses categorised pendulous crops into different condemnation causes (body cavity disorder, and other reasons), although staff at all four slaughterhouses had the same education. Similar findings have also been reported in other studies. St-Hilaire & Sears 2003; Lupo et al. 2008, and Buzdugan et al. 2020 identified variations between slaughterhouses in condemnation causes, and the percentage of condemned carcasses. The difference between shift 1 and 2 in this study regarding assessment shows how hard it is to accomplish a uniformly assessments of carcasses, although meat inspectors have been trained in the same way. The data obtained in the study was from November 2021 to March 2022 which might be a limited period but if a longer period had been implemented, the variation in crops assessed as filled might had varied even more due to e.g. seasonal workers and new employees. However, it must be mentioned that this study does not tell which shift that were most correct in the assessment of filled crops.

5.5 Enumeration of Enterobacteriaceae

Treatment with the inside/outside washer showed a significant (p<0.01) decrease in the number of *Enterobacteriaceae* after washing. This interpretation differs from that of Geornaras & von Holy (2000) who observed no significant effect of reduction in *Enterobacteriaceae* before and after inside/outside wash. In contrast, similar result was found by Moazzami et al. (2021), where the number of *Enterobacteriaceae* decreased significantly after treatment of broiler carcasses in an ultrasound-steamer. Worth mentioning is that heat can reduce the number of *Enterobacteriaceae* and is applied in the ultrasound-steamer but not in the washer. A study that supports the results in this study is the research performed by Althaus et al. (2017). They collected many neck skin samples (n=450) from a slaughterhouse and analysed the number of *Enterobacteriaceae* before and after washing. Their results showed a decrease in *Enterobacteriaceae* after washing. Even though the result in this study showed a significant decrease in Enterobacteriaceae, the neck skin from seven carcasses had a higher number of bacteria after washing, and a difference was found on four neck skins. One reason why some carcasses have a higher number of bacteria after washing, could be that faeces may have been located on the outside or inside of the carcass, and during washing, the faeces was flushed and contaminated the neck skin. Althaus et al. (2017) suggested that bacteria could be redistributed instead of washed off, which also could be a reason to why some carcasses had increased values or showed no effect after washing. A decrease in Enterobacteriaceae is beneficial for the final product to avoid foodborne infections in humans, nevertheless, raw chicken should always be heated before consumption. Anyway, before the chicken reaching the several processing steps will occur where the number of market, Enterobacteriaceae bacteria is expected decrease further. However, further studies are necessary to evaluate how effective the washer is, even though the results in this study showed a significantly decrease in number of Enterobacteriaceae. Since the sample collection was performed randomly with a limited number of carcasses, the author suggests that this can be performed again with an increased number of samples.

Bacteria belonging to the Enterobacteriaceae family were identified by MALDI-TOF analysis and 95% of the identified bacteria were E. coli, while 5% were identified as P. mirabilis, A. veronii, and A. caviae. Finding E. coli in the analysed neck skin samples was excepted since E. coli is commonly present in the intestinal tract of broiler chickens from an early age (Kemmett et al. 2014). In a similar study, Moazzami et al. (2021) also identified E. coli from neck skin samples of broilers using MALDI-TOF MS. P. mirabilis, A. veronii, and A. caviae are well commensals and expected to be found at carcasses. Sanches et al. (2020) explained that both E. coli and P. mirabilis are associated with cellulitis which occur when broiler chickens have scratches in the skin, which favours the entrance of bacteria. Since cellulitis is one of the most common reasons for condemnation of carcasses, it is not unlikely that the broiler chicken had a scratch in the skin where *P. mirabilis* had entered and was rinsed to the neck skin during washing. P. mirabilis is a commensal in the intestinal tract of poultry which can have contaminated the broiler carcass after evisceration (Sanches et al. 2020; VetBact 2022c). The finding of A. veronii, and A. caviae can be explained by the way chicken is slaughtered. It is a wet process compared to slaughter of other livestock, such as beef and pork. Since both A. veronii, and A. caviae have their natural habitat in water, it was not surprisingly to identify the bacteria (Neyts et al. 2000; Benagli et al. 2012), even if A. veronii, and A. caviae are more frequently associated with fish (Benagli et al. 2012). The identification of these bacteria is reliable since the MALDI-TOF score values gave a clear indication that the bacteria were identified on both genus and species level. To confirm the findings of bacteria even more, a higher number of bacteria could be identified by MALDI-TOF, it was not possible due to the limitation of time and money.

5.6 Enumeration of total aerobic bacteria

The total aerobic bacteria showed a highly significant (p < 0.001) reduction after washing. The total number of aerobic bacteria have been analysed in different stations along the slaughter line in several studies. Zweifel et al. (2015) analysed the total aerobic bacteria at three different slaughterhouses and concluded that washing of the carcass reduced the total number of aerobic bacteria between 0.2- $0.4 \log \text{ CFU/g}$, which is in line with the findings in this study. However, in the study by Zweifel et al. (2015) the main reduction occurred after plucking, with a mean reduction of 1.2-1.7 log CFU/g. Further, Moazzami et al. (2021) investigated the difference in total aerobic bacteria before and after ultrasound-steam treatment. The findings revealed that a significant (p<0.001) reduction in total aerobic bacteria occurred after treatment, even though some values increased instead of decreased. An increase occurred in 17% of the samples after washing meanwhile treatment in the ultrasound-steam increased 23% of the analysed samples (Moazzami et al. 2021). This could be due to that it was different parts of the neck skin were analysed and the number of bacteria might not be evenly distributed on the whole carcass. A significant reduction in total aerobic bacteria is a beneficial result because a reduction in bacteria is wanted. The findings also shows that the inside/outside washer helps to reduce the number of bacteria, even though the major reduction perhaps occur in the earlier stages along the slaughter line, as described by Zweifel et al. (2015). To strengthen these findings, more samples could be collected and analysed before and after the washing station, but also from several other stations along the slaughter line.

6. Conclusion

Findings from on-farm crop palpation conclude that it is difficult to predict the outcome regarding crop fill at slaughter based on the proportion of filled versus empty crops estimated on-farm prior to transportation to the slaughterhouse. Based on the investigations of crop contents at the slaughterhouse, it can be concluded that most crops had a content of the main category feed (61.2%), followed by water (27.9%), and lastly litter (10.8%), indicating that feed withdrawal time has been insufficient for some broiler while others have had a too extensive feed withdrawal period. The percentage of filled crops at slaughter was not affected by farm factors such as feeding whole wheat kernels, feed withdrawal time, water withdrawal time, and light three days prior to slaughter. However, the statistical analysis has limitations such as few numbers of observations and a need to categorise of answers into two outcomes per investigated factor. Therefore, more extensive studies are needed to analyse how on-farm factors impact the percentage of filled crops at slaughter. It was revealed that work shifts performing the inspection at slaughter significantly affected the percentage of carcasses judged as having filled crops in the meat inspection. However, the analysis only shows that there is a difference between the shifts and not which shift that performs the assessment of filled crops correctly. Rinsing carcasses at the slaughterhouse decreased both total aerobic bacteria and *Enterobacteriaceae*. This is an expected and positive result because the hygienic quality of the carcass is improved, and it is a step in the right direction to achieve SDG 2 and 12. To summarise this thesis, further research is needed to evaluate which factors that affects crop content.

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Popular science summary

You have probably heard about crops in a field, but have you ever heard about a filled crops in a broiler? A broiler is a bird that is extensively bred for its meat and most birds have a crop. The crop can be equalised with a pouch that is used for temporarily storing feed and water before it is passed to the birds' stomach. Filled crops are not desired during slaughter of broiler chickens because they pose a risk for contamination of the carcasses. The hygienic quality of the carcass will be impaired if crop content is present on the carcass, which means that the shelf life of the chicken product decreases, and it could also increase the risk of food borne illness among humans because more bacteria are present on the carcass than usually.

The purpose of this thesis was to look at which factors that affect the crop content in broiler chickens before slaughter and what actions that can to improve the hygienic quality of the broiler carcasses at slaughter. Less filled crops at slaughter and improved hygienic quality of the carcass could enable economic profits for the producers and the slaughterhouse but are also a step towards the goals the United Nations invented in 2015 to improve the sustainability for all humans around the globe. To find out which factors that affect the crop content in broilers, farm visits, interviews with broiler producers, and visits at the slaughterhouse were performed. Laboratory analyses on bacteria from neck skin samples of broiler chickens were performed to understand how the hygienic quality of carcasses could be improved by rinsing carcasses with water at the slaughterhouse.

Answers from the interviews with the broiler producers could not prove that any of the farm management routines investigated affected crop content at slaughter. However, working shift in the meat inspection at the slaughterhouse affected the percentage of crops assessed as filled but the analysis did not show which shift that performed the correct judgement of filled crops. The rinsing of carcasses with water at the slaughterhouse improved the hygienic quality by reducing the number of bacteria present on the carcass. In summary, results from this study shows that more research is needed within this topic to better understand which factors that affect the crop content in broiler chickens and to further improve the hygienic quality of the broiler carcasses. A deeper understanding of these factors will be a help to work towards a sustainable consumption and production.

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Appendix I Farm interview questions

The interview questions that were sent out to the broiler producers are presented below.

Hej uppfödare!

Denna enkät är en del av min Masteruppsats som jag skriver inom Agronomprogrammet – livsmedelsvetenskap i samarbete med Kronfågel.

I mitt arbete undersöker jag krävaproblematiken hos slaktkycklingar och hur en förbättring av rutinen skulle kunna minska förekomsten av fyllda krävor på slakteriet samt minska risken för ett krävaavdrag.

Enkäten är uppdelad i två delar, där **Del 1** handlar om *delslakten* medan **Del 2** handlar om *huvudslakten*. Du kommer att besvara 24 frågor via telefon och dina svar kommer att vara anonyma.

För att projektet ska kunna genomföras med god vetenskaplig kvalitet behöver enkätsvaren kopplas till gårdens statistik, **per avdelning,** över förekomsten av fyllda krävor för de tre senaste slakttillfällena.

Din medverkan kommer att göra skillnad för mitt arbete och förhoppningsvis på sikt leda till en minskning av fyllda krävor vid slakt.

Tack på förhand! Har du frågor kontakta mig på: Med vänlig hälsning, Ylva Eriksson

DEL 1 – Delslakt

Om du INTE använder samma rutin i alla stallar eller avdelningar. Utgå från den rutin som du använder i störst utsträckning alt. den rutin som fungerar bäst på din gård.

Följande frågor kommer att handla om när du stänger av fodret samt hissar foder och vattenlinjer, i samband med delslakt under vinter respektive sommartid.

Fråga 1

Vid delslakt, har du möjligheten att dela av avdelningen för de kycklingar som ska
lastas ut?

Svar: JA		NEJ	
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Fråga 2

Under	tider	n utla	stningen	till	delslakt	pågår,	har	de	kycklingar	som	ska	till
huvuds	lakt t	illgång	g till fode	r oc	h vatten?							
Svar: JA	\square	NEJ										

Svar: JA		NEJ	
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Fråga 3

Använde	r du	hela	vet	ekärnor	i fodret?
Svar: JA		NEJ			

Fråga 4

- a) Hur lång tid innan utlastning brukar du stänga av fodret vintertid (oktober - april)?
 - Svar: (Ange svaret i hela timmar)
- b) Hur lång tid innan *utlastning* brukar du stänga av fodret sommartid, (maj - september)?
 - Svar: (Ange svaret i hela timmar)
- c) Jag brukar inte <u>stänga av</u> fodret innan utlastning

Fråga 5

- a) Hur lång tid innan *slakt* brukar du stänga av fodret vintertid (oktober april)?
 - Svar: (Ange svaret i hela timmar)
- b) Hur lång tid innan slakt brukar du stänga av fodret sommartid (maj september)?

Svar: (Ange svaret i hela timmar)

c) Jag brukar inte <u>stänga av</u> fodret innan delslakt

Fråga 6

- a) Hur lång tid innan *utlastning* brukar du <u>hissa</u> foderlinjerna vintertid (oktober - april)?
 - Svar: (Ange svaret i hela timmar)
- b) Hur lång tid innan *utlastning* brukar du <u>hissa</u> foderlinjerna sommartid (maj - september)?
 Svar: (Ange svaret i hele timmer)

Svar: (Ange svaret i hela timmar)

Fråga 7

a) Hur lång tid innan *utlastning* brukar du <u>hissa</u> vattenlinjerna vintertid (oktober - april)?

Svar: (Ange svaret "precis innan plockarna sätter i gång" eller i minuter)

 b) Hur lång tid innan *utlastning* brukar du <u>hissa</u> vattenlinjerna sommartid (maj – september)?

Svar: (Ange svaret "precis innan plockarna sätter i gång" eller i minuter)

Nu kommer du att få besvara frågor om ljusschema, stalltemperatur samt krävans innehåll innan utlastning. *Observera att frågorna fortfarande handlar om delslakt!*

Fråga 8

Vilken tid på dygnet är stallarna mörklagda, sju dagar efter insättning fram tills de tre sista dagarna innan slakt? Svar:

Fråga 9

Hur ser ljusschemat ut för respektive dag, tre dagar innan utlastning? Svar:

Fråga 10

- a) Hur lång tid innan *utlastning* brukar du stänga av belysningen (vintertid)?
 Svar: (Ange svaret i minuter)
- b) Hur lång tid innan *utlastning* brukar du stänga av belysningen (sommartid)?
 Svar: (Ange svaret i minuter)

Fråga 11

- a) Vilken temperatur önskar du att ha vid utlastning (vintertid)?
 Svar: (Ange svaret i °C)
- b) Lyckas du ha önskad temperatur längst bort från utlastningsporten (vintertid)?

Svar:

- c) Vilken temperatur önskar du att ha vid utlastning (sommartid)?
 Svar: (Ange svaret i °C)
- d) Under dagar med hög utetemperatur, hur många grader högre är det i hallen längst bort från utlastningsporten?
 Svar: (Ange svaret i °C)

Fråga 12

Brukar du känna på krävan innan utlastning?

- a) JA 🗌 NEJ 🗌
- b) Vid svar JA: ungefär hur många kycklingar brukar du känna på?
 Svar: (Ange ungefärligt antal).
- c) Beskriv krävans innehåll så detaljerat du kan (*T.ex. pellets, mjuk, vattnig eller tom*).

Svar:

DEL 2 – Huvudslakt

Om du INTE använder samma rutin i alla stallar eller avdelningar. Utgå från den rutin som du använder i störst utsträckning alt. den rutin som fungerar bäst på din gård.

Följande frågor kommer att handla om när du stänger av fodret samt hissar foder och vattenlinjer, i samband med huvudslakt under vinter respektive sommartid.

Fråga 13 Denna fråga gäller endast de uppfödare som använder <u>hela vetekärnor</u> i fodret

- a) Om du använder hela vetekärnor i ditt foder, hur lång tid innan utlastning brukar du stänga av fodret vintertid (oktober - april)? Svar: (Ange svaret i hela timmar)
- b) Om du använder hela vetekärnor i ditt foder, hur lång tid innan utlastning brukar du stänga av fodret sommartid (maj - september)? Svar: (Ange svaret i hela timmar)

Fråga 14

a) Hur lång tid innan utlastning brukar du stänga av fodret vintertid (oktober - april)?

Svar: (Ange svaret i hela timmar)

b) Hur lång tid innan utlastning brukar du stänga av fodret sommartid (maj september)?

Svar: (Ange svaret i hela timmar)

Fråga 15

a) Hur lång tid innan slakt brukar du stänga av fodret vintertid (oktober april)?

Svar: (Ange svaret i hela timmar)

b) Hur lång tid innan *slakt* brukar du stänga av fodret sommartid (maj september)?

Svar: (Ange svaret i hela timmar)

Fråga 16

Hur många timmar innan slakt vill du att kycklingarna ska ha ätit rent i foderkopparna?

Svar: (Ange svaret i hela timmar)

Fråga 17

- a) Hur lång tid innan *utlastning* brukar du <u>hissa</u> foderlinjerna vintertid (oktober - april)?
 - Svar: (Ange svaret i hela timmar)
- b) Hur lång tid innan *utlastning* brukar du <u>hissa</u> foderlinjerna sommartid (maj - september)?

Svar: (Ange svaret i hela timmar)

Fråga 18

a) Hur lång tid innan *utlastning* brukar du <u>hissa</u> vattenlinjerna vintertid (oktober - april)?

Svar: (Ange svaret "precis innan plockarna sätter i gång" eller i minuter)

 b) Hur lång tid innan *utlastning* brukar du <u>hissa</u> vattenlinjerna sommartid (maj – september)?

Svar: (Ange svaret "precis innan plockarna sätter i gång" eller i minuter)

Fråga 19 Denna fråga gäller endast uppfödare som har delade foder- och vattenlinjer

Om du som uppfödare har delade foder och vattenlinjer, brukarna du ha olika tider för avstängning och hissning för den främre respektive bakre delen av stallet? Svar:

Nu kommer du att få besvara frågor om ljusschema, stalltemperatur samt krävans innehåll innan utlastning.

Fråga 20

Vilken tid på dygnet är stallarna mörklagda, sju dagar efter insättning fram tills de tre sista dagarna innan slakt? Svar:

Fråga 21

Hur ser ljusschemat ut för respektive dag, tre dagar innan utlastning? Svar:

Fråga 22

- a) Hur lång tid innan *utlastning* brukar du stänga av belysningen (vintertid)?
 Svar: (Ange svaret i minuter)
- b) Hur lång tid innan *utlastning* brukar du stänga av belysningen (sommartid)?
 Svar: (Ange svaret i minuter)

Fråga 23

- a) Vilken temperatur önskar du att ha vid utlastning (vintertid)?
 Svar: (Ange svaret i °C)
- b) Lyckas du att ha önskad temperatur längst bort från utlastningsporten (vintertid)?

Svar:

- c) Vilken temperatur önskar du att ha vid utlastning (sommartid)?
 Svar: (Ange svaret i °C)
- d) Under dagar med hög utetemperatur, hur många grader högre är det i hallen längst bort från utlastningsporten?
 Svar: (Ange svaret i °C)

Fråga 24

Brukar du känna på krävan innan utlastning?

- a) JA 🗌 NEJ 🗌
- b) Vid svar JA: ungefär hur många kycklingar brukar du känna på?
 Svar: (Ange ungefärligt antal).
- c) Beskriv krävans innehåll så detaljerat du kan (*T.ex. pellets, mjukt, vattning eller tom*). Brukar krävfyllnaden skilja sig från delslakten?
 Svar:

Tack för din medverkan!

Om du har några frågor går det bra att kontakta mig på:

Appendix II Carcasses with crops assessed as filled after meat inspection



Figure I. Carcasses assessed as having a filled crop after the meat inspection.



Figure II. Carcasses assessed as having a filled crop after the meat inspection



Figure III. Carcasses assessed as having a filled crop after the meat inspection. The neck skin has been removed to easier visually recognise the crop content in the crop.

Appendix III Photographs: crop content categorisation after dissection



Figure IV. Sixty dissected crops at the slaughterhouse from farm E, assessed as filled after the meat inspection.

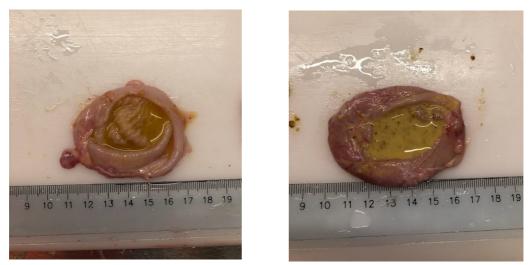


Figure V. Crop content categorised as water after dissection at the slaughterhouse.

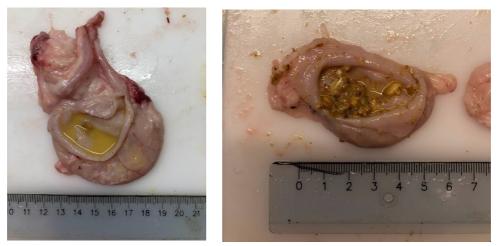


Figure VI. Crop content categorised as water with kernels after dissection at the slaughterhouse.

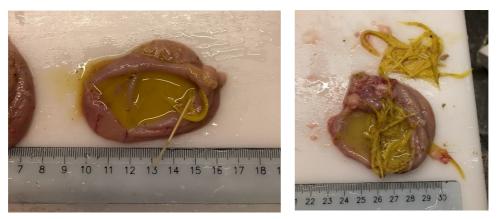


Figure VII. Crop content categorised as water with feathers after dissection at the slaughterhouse.

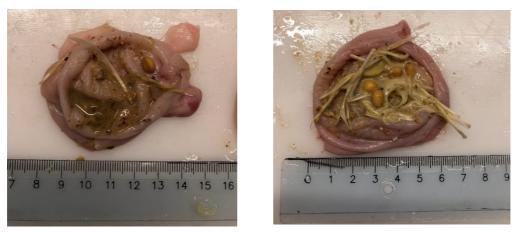


Figure VIII. Crop content categorised as water with kernels and feathers after dissection at the slaughterhouse.

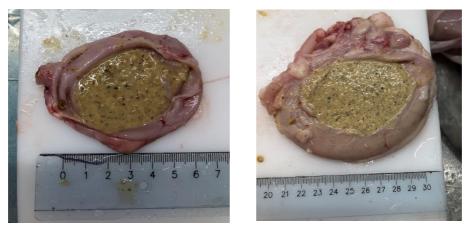


Figure IX. Crop content categorised as feed after dissection at the slaughterhouse.

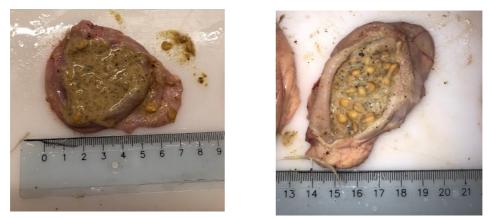


Figure X. Crop content categorised as feed with kernels after dissection at the slaughterhouse.

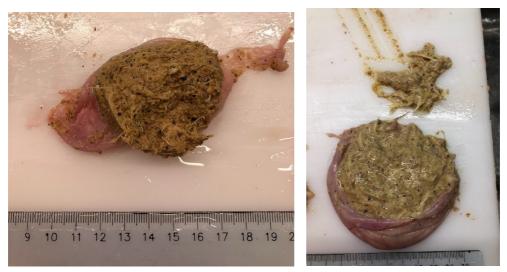


Figure XI. Crop content categorised as feed with feathers after dissection at the slaughterhouse.

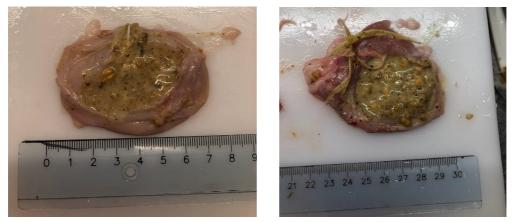


Figure XII. Crop content categorised as feed with kernels and feathers after dissection at the slaughterhouse.



Figure XIII. Crop content categorised litter after dissection at the slaughterhouse.



Figure XIV. Crop content categorised litter with kernels after dissection at the slaughterhouse.



Figure XV. Crop content categorised litter with feathers after dissection at the slaughterhouse.

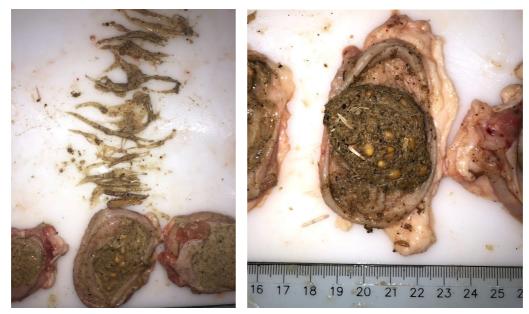


Figure XVI. Crop content categorised litter with kernels and feathers after dissection at the slaughterhouse.

Appendix IV Raw data: crop content categorisation after dissection

Farm	Dissected crops	Water	Water with kernels	Water with feathers	Water with kernels & feathers	Feed	Feed with kernels	Feed with feathers	Feed with kernels & feathers	Litter	Litter with kernels	Litter with feathers	Litter with kernels & feathers
А	65	1	5	0	2	8	17	4	7	4	9	3	5
В	44	14	3	0	1	21	2	2	1	0	0	0	0
С	64	6	0	14	0	8	1	33	0	1	0	1	0
D	60	5	1	6	2	15	3	18	5	0	0	4	1
Е	61	9	1	7	5	15	8	3	9	0	3	0	1
Total	294	35	10	27	10	67	31	60	22	5	12	8	7

Table I. Categorisation of crop content after dissection at the slaughterhouse from farm A, B, C, D, and E. The number of crops dissected from each farm is also present in the table.

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