Towards a safe and sustainable poultry production chain

Jacqueline Berghout, Wibke Roland, Martijntje Vollebregt, Miriam Koene, Ingrid de Jong

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Towards a safe and sustainable poultry production chain

Jacqueline Berghout[1], Wibke Roland[1], Martijntje Vollebregt[2], Miriam Koene[2], Ingrid de Jong[3]

[1] Wageningen Food & Biobased Research
[2] Wageningen Bioveterinary Research
[3] Wageningen Livestock Research

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P.O. Box 338, 6700 AH Wageningen, The Netherlands, T +31 (0)317 48 39 53,
E info.livestockresearch@wur.nl, www.wur.nl/livestock-research. Wageningen Livestock Research is part of Wageningen University & Research.

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Preface

Wageningen Food & Biobased Research, HAS universityUniversity of applied sciencesApplied Sciences, Marks & Spencer, Food Animal Initiative, and Heijs Food Products collaborated in this research project on a safe and sustainable poultry production chain. This is the end report with summaries of our findings. Heijs Food Products likes to share a short explanation for the collaboration and research topic in the Preface.

We as Heijs Food Products, a middle class Poultry Slaughter plant and Meat Producer, supply the high-end market with chicken products in the European market. The development of the demand of customers in these markets goes in the direction of products that offer high transparency, a production based on high animal welfare, products that find their origin in healthy and robust animals and they ask every year for higher valorization and development of high added value products. To find the right answers in the future on all these four customer demands we decided to investigate these important issues for our customers in a project with a scientific independent research party (Wageningen University & Research), an important commercial relation in the European market (Marks & Spencer) and their British Research and Development Institute Food Animal Initiative and, for the operational research in the field, the HAS University of Applied Sciences. In the project we performed research on poultry farms, in slaughterhouses, at the University and in the processing plant(s).

The project was financially supported in the years 2015 - 2018 by Samenwerkingsverband Noord-Nederland (SNN) and the Province of Groningen and the Province of Drenthe.

Heijs Food Products thanks all the people involved over the years in this project:
Wageningen University & Research; project coordinators and researchers,
Marks & Spencer (M & S); technical department of the supply chain,
Food Animal Initiative; project coordinator of the institute working for M & S,
HAS University of Applied Sciences; project coordinators, supervisors and a lot of graduate students on the different projects in the years,
Poultry farmers supplying the Heijs chain,
Heijs Food Products; the departments NPD, Micro lab, Production, Administration, Technical supply chain.

The support from Samenwerkingsverband Noord-Nederland (SNN) and the Provinces in the North of The Netherlands was very welcome and we thank them for making it all possible'.

Sincerely,
Henk Benedictus

Summary

The aim of this project was to develop an innovative poultry production chain that offers high quality and safe products as required by the consumer ('farming for the future') through a guaranteed traceability and product safety; and valorisation and development of distinctive poultry products. The project was divided into four work packages; product transparency and recognisability (WP1), animal welfare (WP2), healthy and robust chickens (WP3), and valorisation and development of added-value products (WP4). The results of the performed projects will contribute to an innovative poultry production chain, through the developed welfare and sustainability model for more transparency and recognisability, through the novel, high-quality product concepts developed, the insights in end-of-life welfare, the demonstrated potential for *Campylobacter* reduction in the slaughter process, and the improved understanding of effective environmental enrichment that can be applied on-farm.

Heijs Food Products, a poultry slaughter plant and meat producer, is a leading company and important party in the poultry production chain in the Northern part of The Netherlands. Heijs Food Products successfully translates new developments in the consumer market into products and processes that are implemented in the poultry production chain. Increasing volumes of poultry meat are produced in countries such as Brazil and Thailand. The production of these 'anonymous' bulky products has stimulated Heijs Food Products to distinguish their products from these bulk products by investing in quality and traceability of their poultry products.

To produce an undisputed and high quality product, the poultry sector needs to be transparent including full traceability from farm to fork, guarantee sustainable production of poultry meat (including animal welfare) and produce safe poultry meat with added value. With this project Heijs Food Products (HFP), Marks & Spencer (M & S), the British Research and Development Institute Food Animal Initiative (FAI), HAS University of Applied Sciences (HAS) and Wageningen University & Research (WUR) collaborate to enable the development of a poultry production chain that meets these requirements. The project is supported by Samenwerkingsverband Noord-Nederland (SNN, the 'North Netherlands Cooperation Agreement'), Koers Noord, the province of Groningen, and the province of Drenthe. It was executed in the years 2015-2018.

The aim of the project was to develop an innovative poultry production chain that offers high quality and safe products as required by the consumer ('farming for the future') through a guaranteed traceability and product safety; and valorisation and development of distinctive poultry products.

The project was divided into four research topics (work packages (WP)); product transparency and recognisability (WP1), animal welfare (WP2), healthy and robust chickens (WP3), and valorisation and development of added-value products (WP4). Within these research topics, 32 projects in total were defined, of which 30 projects were executed by HAS University of Applied Sciences and Wageningen Research, with collaboration of HFP, M & S, and FAI. The remaining 2 projects were carried out by Wageningen Food & Biobased Research. This report is a summary of all the results generated within these 32 projects.

In WP1 the focus was on improving transparency and recognisability of the poultry production chain. HFP performed activities that focused on transparency and recognisability, whilst the student projects focused on designing a sustainability model. Together with WP2, working on animal welfare, the broiler production chain was mapped out. From this production chain three chain links were selected for further study; the broiler farmer, the slaughter house, and the processing plant. For these chain links, the level of sustainability was studied. Sustainability in this project was defined as consisting of indicators for People, Planet, and Profit based on the CSR performance ladder (MVO prestatieladder), and extended with another P for Poultry (welfare and health). For these 4 Ps, indicators, rankings, and scores were developed based on literature study and expert interviews. Subsequently, the indicators were validated with chain data of HFP.

Next to the validation, confrontation matrices were prepared to identify the effect of certain measures on the sustainability indicators (positive or negative). With this information, broiler farmers as well as the processing industry obtained an insight into improvements they can apply with relatively low effort to increase their sustainability level.

The sustainability level of the chain was transparently visualized as a total score per P and the individual scores of the underlying indicators of that P. The sustainability model is ready to be implemented, but it is recommended to gather even more data to improve the sensitivity of the model. To improve the model even further it is recommended to develop a software tool that allows the chain links to enter their data and to benchmark themselves against the average scores of the chain links.

In WP2 and 3 the focus was on animal welfare and healthy and robust chickens. The experiments performed for the development of effective environmental enrichment showed that broiler chickens prefer some enrichments over others. E.g., for resting on elevated structures they preferred platforms over perches and for exploration and foraging they preferred wood shavings bales over lucerne bales and pecking stones. Furthermore, they made better use of enrichments when the stocking density was reduced from 35 kg/m^2 to 25 kg/m^2. Finally, we showed that wood shavings bales only stimulated activity of the broilers better as compared to the combination of wood shavings bales and platforms. None of the experiments indicated that environmental enrichment negatively influenced the technical performance of the broilers. During this project indications were found on how many enrichments should be provided, but more research is needed to determine the optimal number of enrichments in a broiler house.

The most important outcome of the data evaluation on the end-of-life phase is that the percentage of wing fractures increases between lairage, post-shackling, post-stunning, and post-plucking, with an average increase of 4.0% between lairage and post-plucking. Additionally, most injuries and damage occur during the slaughter process and only a small percentage occurs during the pre-slaughter process (catching and transport). To develop preventive measures for injuries and damage, it is recommended to determine where exactly in the process injuries and damage occur. In addition, a scoring system to accurately determine the age of bruises needs to be developed, in order to be able to determine where in the process the bruises occurred. During transport of broiler chickens for thinning under moderate climatic conditions (neither very hot nor very cold) it was shown that there were large differences in the temperature (> 10°C) in the transport containers according to the location of the containers in the truck. During transport, the temperature remained stable regardless of the duration of the transport. However, during stops, the temperatures in the transport containers raised considerably. This implicates that especially during stops there is a welfare risk for broilers in transport containers.

From the study on indicators of the effect of thinning we found that the majority of broiler farms apply thinning as a routine practice, of which 30% thins multiple times during a production cycle. Feed deprivation of the whole flock prior to thinning may negatively affect welfare of the remaining chickens. Flock uniformity and the number of broilers per feeder after thinning could be suitable indicators to determine the effect of thinning.

A welfare model was developed that includes 14 indicators relevant for broiler welfare on-farm and during the end-of-life stage, a scoring system based on risk assessment principles, and limit values for each welfare indicator. The model is sensitive to variation in welfare performance between broiler flocks. Further, indicators for the assessment of flock behaviour on-farm were developed that can be measured by the farmer itself. It is recommended to communicate the individual variable scores to the broiler farmers to allow them to improve their performance.

From the studies on the measures to reduce *Campylobacter* contamination on-farm and during the slaughter process it was concluded that there are multiple introduction routes of *Campylobacter* on farm. Feed additives did not significantly reduce *Campylobacter* in broiler chickens. It is possible that thinning might be an introduction route for *Campylobacter* as contamination was detected on containers. A further study to the risk of contamination in containers confirmed that even after cleaning, containers were contaminated with *Campylobacter*. Due to the container design, it is very difficult to remove all contamination from transport containers. Three different intervention methods were tested in a slaughter house in order to reduce *Campylobacter* spp. and *Escherichia coli* (*E. coli*) contamination during the slaughter process. The results showed that the tested intervention methods are all separately successful in reducing bacterial counts compared to no intervention. All three methods combined led to reductions of *E. coli* by 1.4 log and of *Campylobacter* spp. by 1.3 log. Experiments in laboratory settings, with a fourth method, namely cold atmospheric plasma (CAP), led to *Campylobacter jejuni* reduction by 1.5 log. However, the meat quality was not acceptable anymore after the CAP treatment due to dehydration. Industrial applicability and suitable settings of this pilot technique to maintain meat quality would have to be developed before usage is possible.

WP4 focused on value-added products by the reduction of salt and E-numbers, on gluten-free products and on the valorisation of chicken leg meat. The salt content of spicy chicken wings (Hotwings) could be reduced with 25% (20% in the marinade and 5% in the breading) without noticeable difference in saltiness, tenderness, juiciness, and colour compared with the reference, as evaluated by a sensory panel (n=31). Also, the sensory panel evaluated these aspects for a combination of salt replacer and NaCl to a maximal salt content of 2 grams in one hot wing portion (in line with UK guidelines) to be similar to the reference.

The experiments on allergen-free Hotwings revealed that a combination of chickpea, rice, and tapioca flour resulted in the best gluten-free breading mixture with similar or better properties compared to the reference product with a wheat flour breading.

Another project led to the development of four concepts for world marinades to be used on chicken thighs. All marinades are free from allergens, low in salt, and low in E-numbers.

For the valorisation of leg meat, several product concepts were developed. One of the concepts is a “non-sticky fingers” concept, which consists of drumsticks with a clean bone resulting in clean hands after consumption. Another concept focused on the development of an E-book containing recipes to prepare chicken thigh meat. The next projects focused on the application of enzymatic re-structuring of leg meat. Formed leg meat, cut into slices, and prepared in different ways, was successfully developed by applying the enzyme transglutaminase and the (allergenic) milk protein sodium caseinate. It was investigated whether sodium caseinate could be replaced by non-allergenic plant proteins or chicken proteins, but the stability of those products was insufficient. Another binding mechanism using Fibrimex (fibrinogen and thrombin from bovine plasma) resulted in a very good and stable allergen-free product, implying labelling of beef protein as minor disadvantage. The milk protein containing product was scaled up to industrial scale production.

In general, it is recommended to perform sensorial research by a large consumer panel (n = 60 – 100) to validate appreciation of the newly developed concepts for the specified target groups. Additionally, it is recommended to perform shelf-life tests for the reduced salt products.

With these work packages, we covered a large part of the poultry production chain; starting on the farm to improve welfare, health and sustainability, to the processing industry where sustainability, product safety and health were addressed, to the production of products with lower salt and E-number contents, novel concepts for valorisation of the broilers, up to the customer (wishes). The results of the performed projects contribute to an innovative poultry production chain, through the developed welfare and sustainability model for more transparency and recognisability, through the novel, high-quality product concepts developed, the insights in end-of-life welfare, the demonstrated potential for *Campylobacter* reduction in the slaughter process, and the improved understanding of effective environmental enrichment that can be applied on-farm.

Lastly, to make these findings a success in the poultry production chain, the communication within the poultry production chain is very important; by exchanging more information between the different chain links, the chain as a whole can improve on e.g. sustainability, welfare, and product safety. For the sustainability and welfare model to be implemented, we recommend to invest in a software tool that allows the broiler farmers and the processing industry to digitally record their data and allows them to create outputs with which they can benchmark themselves against the average and get insight into aspects they can improve on. For the developed product concepts, we recommend to strengthen the collaboration with Heijs' customers and perform sensorial tests with large(r) consumer panels.

1 Introduction

1.1 Background

Heijs Food Products (HFP), a poultry slaughter plant and meat producer, is a leading company and important party in the poultry production chain in the Northern part of The Netherlands. HFP successfully translates new developments in the consumer market into products and processes that are implemented in the poultry production chain. Increasing volumes of poultry meat are produced in countries such as Brazil and Thailand. The production of these ‘anonymous’ bulky products has stimulated HFP to distinguish their products from these bulk products by investing in quality and traceability of their poultry products.

Despite the efforts of HFP to distinguish themselves from the bulky market and the appreciation of these efforts by their international customers, such as retailers and fast food producers, HFP needs more instruments to claim the quality and the distinctive character of their products to the consumers and clients. Incidents with meat traceability or quality, but also the discussion on the welfare of fast growing broiler chickens had and will have their negative impact on the meat sector and may as such also negatively affect HFP, despite their efforts to constantly produce high quality poultry meat.

To produce an undisputed and high quality product, the poultry sector needs to be transparent including to have full traceability from farm to fork, to guarantee sustainable production of poultry meat (including animal welfare) and to produce safe poultry meat with added value. The project consortium will demonstrate the development of a poultry production chain that meets these requirements.

The project consortium consists of Heijs Food Products, Marks & Spencer, Food Animal Initiative via Marks & Spencer, HAS University of Applied Sciences, and Wageningen University & Research (WUR). This project was supported by Samenwerkingsverband Noord-Nederland (SNN, the 'North Netherlands Cooperation Agreement'), Koers Noord, the province of Groningen, and the province of Drenthe. It was executed in the years 2015-2018.

Also other chain partners (hatcheries, broiler farms and slaughter plant) were involved in the execution of the project as many of the project goals are a chain responsibility. The chain partners were not part of the consortium.

1.2 Project aim

The aim of the current project is to develop an innovative poultry production chain that offers high quality and safe products as required by the consumer (‘farming for the future’) through a guaranteed traceability and product safety; and valorisation and development of distinctive poultry products.

1.3 Aim and structure of the report

This report is a summary of all research projects performed by HAS University of Applied Sciences and WUR from 2015 until 2018. The project was divided into four research topics, also called the work packages (Figure 1). The four work packages focus on relevant aspects to develop an innovative poultry production chain; work package 1 focuses on product transparency and recognisability, work package 2 on animal welfare, work package 3 on healthy and robust broiler chickens, and work package 4 on the valorisation and development of allergen-free and low-in-E-numbers products.

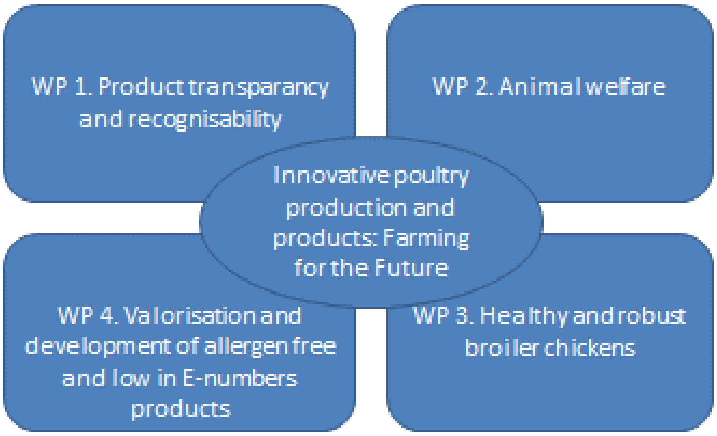

Figure 1 *Illustration of project activities and how these contribute to the overall aim.*

Chapter 2 summarises the way of working in this project and the roles of the project partners. In Chapter 3 the results for each of the work packages are presented. The conclusions and recommendations are given in Chapter 4.

2 Way of working

Each research topic was assigned its own work package, with an aim, activities, time table, intended results and deliverables. The starting point for each work package was a discussion with project and supply chain partners, where we defined research questions, deliverables, major activities and the research approach. This information was translated into an overall work plan, including lead time and planning.

Within each work package, WUR prepared proposals for assignments to be carried out by HAS University of Applied Sciences. Each proposal was matched to either a group of students (2-4) or to a junior researcher. The students were supervised by HAS KnowledgeTransfer and on headlines by three research institutes from WUR; Wageningen Food & Biobased Research (WFBR), Wageningen Bioveterinary Research (WBVR) and Wageningen Livestock Research (WLR). Overall, 32 proposals were defined, of which 2 assignments were performed by WFBR, and 30 assignments were executed by HAS in 2015-2018. Each of the assignments at HAS had a duration of approximately 4 months, starting in February or September. A list of involved people can be found in the Acknowledgments appendix.

Each February or September multiple assignments started at once. For each round of assignments, a kick-off meeting was held to introduce the students to Heijs Food Products, to inform the students about the work performed by their predecessors and the current status of the work packages, and to have a first discussion with the contact persons from Heijs. Next, the students wrote a plan of action including research questions and a time line for their topic/assignment. Activities included on-farm observations, slaughter plant observations, experiments, data feeding, checking and analyses, preparation of presentations and reports in Dutch and English, and joining meetings with all project partners. During the mid-term meeting, students presented their plan of action and possible first results. During the end meeting, students presented their findings, conclusions and recommendations. Each group of students delivered a plan of action, a report in Dutch with summary in English and two presentations in English.

Next to providing scientific knowledge and advice to the student groups, WUR participated in execution of part of the projects. WFBR assisted students with the experimental part, investigated the use of Cold Atmospheric Plasma in reduction of *Campylobacter*, and performed a product search in the Innova database. Genotyping of *Campylobacter* strains was performed by WBVR. WLR trained the students in performing the behavioural observations in the enrichment studies and did the Welfare Quality scoring of other welfare indicators on the farms. Further, WLR trained and assisted the students in the measures performed at the slaughter plant (welfare improvement during end-of-life stage).

Heijs Food Products made many of the on-farm observations possible, as well as slaughter plant observations and even experimental work on farm and in the slaughter plant. Additionally, they provided data for the modelling parts both from the broiler farmers and from their own activities and gathered data for the *Campylobacter* projects, and provided meat and several other ingredients for development of innovative product concepts. The Track & Trace part of WP1 was also performed by Heijs themselves, in collaboration with Van Wouw Engineering B.V. Microbiological analyses were performed by AgroFood Lab and WEK laboratory (Germany).

M & S has a collaboration with the British Research and Development Institute Food Animal Initiative (FAI) for input on welfare and health of broiler chickens. Both parties attended most of the meetings in person or through teleconference, where they gave valuable input to the students' projects.

3 Results and discussion

3.1 Product transparency and recognisability (WP1)

Heijs group wants to distinguish itself from the bulk market by investing in more instruments to ensure the quality and traceability of its products for the consumer. The project partners aim to improve the customer perception of poultry products, especially on sustainable production. The ongoing and planned innovations in production and processing need to be made transparent for customers and consumers.

Previously, efforts on increasing sustainability are aimed at specific parts of the chain, for example on reduction of antibiotics, animal friendly stunning methods, or minimal use of additives in products. To obtain an overview of the sustainability level of an entire chain on different end-user levels, several methods of sustainability assessment could be combined[1]. In this project, the aim is to map the relevant information for sustainable production throughout the poultry production chain and to set up an information system to inform chain partners, both suppliers and customers of Heijs, and consumers on sustainability aspects of their activities or their purchases.

The goal of this project is to develop a sustainability system that HFP can use to give its customers insight in the sustainability level of the chain, and make a visualisation of that system. At the start of the project, the chain links in the poultry production process were mapped out. The following links were included in the sustainability model; the broiler farm, slaughterhouse and deboning plant (or together: processing industry). Table 1 gives an overview of the different projects performed within WP1. The projects are described in more detail in sections 3.1.1 to 3.1.5. The codes within the section titles refer to the thesis project numbers used by HAS.

Table 1 *Overview of projects executed within WP1 Product transparency and recognisability.*

Code	Project title	Main activities	Execution period
7525HFP4	Mapping the broiler chain and identifying sustainability indicators	Description of the activities from breeding farms to customers, this report was shared with all HAS project teams as first introduction to the chain. Sustainability indicators for People, Profit and Planet: description, characteristics and key figures, measurability and ranking. First visualisation developed.	Feb – June 2015
7623LW4	Sustainability indicators applied	Addition of 4th P: Poultry, with focus on animal health. Update of the rankings and targets of the chain. Factors (knobs) influencing one or more indicators were identified and scenario studies to illustrate the effects of turning the knows were presented. Update of visualisation to spider webs.	Feb – June 2016
7664LRW4	Sustainability model I	Selection of indicators by comparing indicators with customer demands and wishes. Integration of animal welfare indicators within the P for poultry. Compiling sustainability level per P. Visualisation including the level per P and selected underlying indicators.	Sept 2016 – Feb 2017
7704LRW, 7705LRW	Sustainability model II	Further development of indicators relevant for broiler farms by discussions with farmers. Selection of feasible indicators for which data is available or can be collected nowadays. The knobs were analysed on level of impact for prioritisation.	Feb – June 2017
18200144	Extension and validation of the sustainability model	Development of forms to be filled in by farmers and the processing industry. Integration of the latest welfare model into the Excel tool. Validation of the sensitivity and efficacy of the model with data from farmers and Heijs.	Mar – June 2018

[1] Van Passel, S., & Meul, M. (2012). Multilevel and multi-user sustainability assessment of farming systems. *Environmental Impact Assessment Review*, *32*(1), 170–180. http://doi.org/10.1016/j.eiar.2011.08.005.

3.1.1 Mapping the broiler chicken chain and identifying sustainability indicators (7525HFP4)

The goal of this project was to determine the level of sustainability in the broiler chicken chain and to identify improvement opportunities related to sustainability for the broiler farm, slaughterhouse and deboning plant.

The project started by gathering information on the broiler chicken chain in collaboration with the team working on the welfare model in WP2 (7526WUR4 see section 3.2.3.1). The chain is displayed in Figure 2. The next step was to interview different companies in the broiler chain and experts on sustainability to identify the appropriate sustainability indicators. Customers of HFP were not included in this research. However, code of practices of the customers such as Marks & Spencer were used as source on sustainability efforts. Based on the CSR Performance Ladder (MVO Prestatie Ladder) and in consultation with experts in the field of sustainability, relevant aspects were pinpointed for each link in the broiler chicken chain within the main themes People, Planet and Profit. Subsequently, these aspects were split up into indicators. For these indicators, a description, characteristics and key figures, measurability and a ranking was given, together with a target value if available, and the information was based on scientific reports, the MVO Performance Ladder and interviews with experts in the field. The ranking was based on the average with two levels below and two levels above the average. This ranking was used to determine the sustainability level of HFP, and is visualized with a colour scheme.

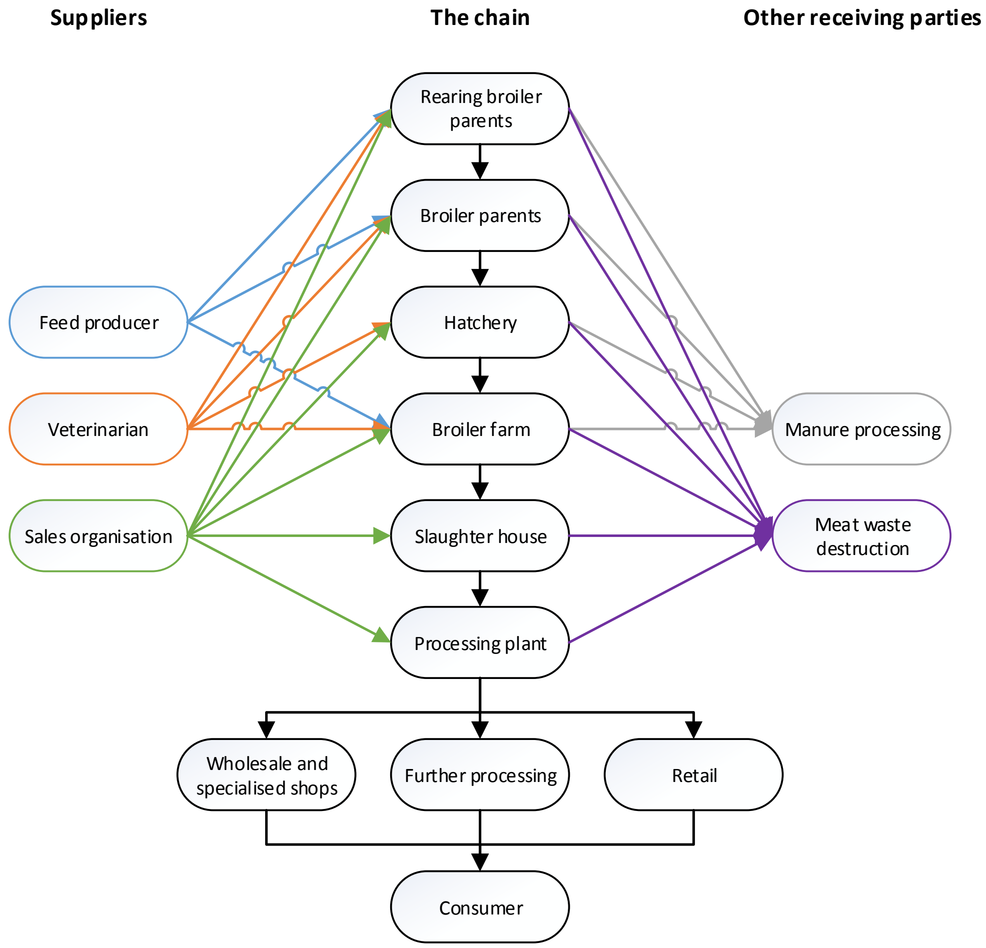

Figure 2 *Scheme of the broiler chicken chain, chain links in red were taken into account in the sustainability analysis.*

Four broiler farmers supplying to HFP were interviewed to determine their sustainability level (June 2015). The average sustainability is displayed in Figure 3. In general the broiler farms performed well on 9/14 indicators (green), and performed not so well (red) on water consumption. Additionally, they should take into account the new ammonia emissions rules. This method was also applied to the slaughterhouse and the deboning plant of HFP. The slaughterhouse scored worse in terms of sustainability compared to the broiler farms. The slaughterhouse scored particularly low for the indicators of green energy and water consumption, which is mainly due to the fact that the slaughterhouse does not use green energy and the high water consumption due to the strict cleaning demands. The deboning plant scored particularly low on the theme Profit. This showed that a high total turnover does not imply that the profit rate will also be high for a profitable business. In addition, more green energy could be used.

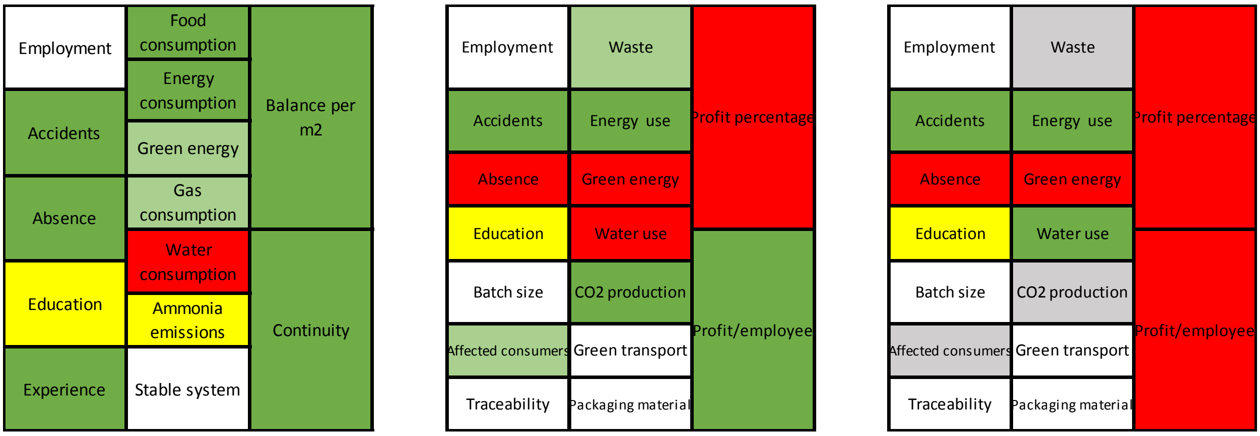

Figure 3 *Sustainability colour scheme for broiler farm average, slaughterhouse, and deboning plant in June 2015.*

Overall, if the slaughterhouse and deboning plant partly use green energy, the chain will score better on the theme Planet. Further scientific research is necessary to create a list of concrete improvements on the broiler farm, slaughterhouse and deboning plant. In addition, there is a lack of information about sustainability throughout the supply chain and more research is necessary on the impact of different farming systems related to sustainability. The recommendation to HFP is to maintain the suggested target values as a guide to obtain a sustainable chain and to inform broiler farmers about sustainability by adding several target values and data flows about sustainability in the manual for the broiler farmer of HFP.

Additionally, it is recommended to make the chain links aware of the importance of information sharing throughout the chain to improve the sustainability of the chain and to improve transparency towards Heijs' customers. The information is registered per chain link, but not shared with all other chain links as some chain partner see the risk of information abuse by competitors.

3.1.2 Sustainability indicators applied (7623LRW4)

The aim of this study was to provide HFP insight into which sustainability issues must be addressed and improved in the chain to meet the requirements and targets of their customers. Another aim was to show the effect of turning knobs on sustainability indicators. The chain parties that were included in the analysis are the broiler farm(er), slaughterhouse and deboning/cutting plant.

The following chain parties were included in the analysis: broiler farm, slaughterhouse and processing plant. The study consisted of desk and field research; desk research included literature review, and internal sources, field research included qualitative research by conducting in-depth interviews with HFP, suppliers and customers of HFP and poultry chain experts.

The determination of the requirements and targets of the customers of HFP focused on four major customers; Marks & Spencer (M & S), Kentucky Fried Chicken (KFC), Tesco and J. Sainsbury's. These customers account for approximately 75% of total sales of HFP.

Next to People, Planet, and Profit, a fourth P was included for Poultry. The sustainability indicators for Poultry all relate to animal health. The level of sustainability of the broiler farms was determined based on 22 sustainability indicators, focusing on 11 sustainability aspects. While mapping the sustainability levels of the chain links slaughterhouse and deboning/cutting plant, it was decided to merge them into a processing industry chain link. The level of sustainability of the processing industry was determined based on 13 sustainability indicators, focusing on 10 sustainability aspects, which are based on the three Ps; People, Planet, and Profit. The slaughter data used to determine the sustainability levels stems from Heijs' database over the years 2013 – 2015.

The analysis showed that action is required to be able to meet the sustainability requirements and targets from the customers. The actions identified that influence the sustainability indicators are called "knobs"; for the broiler farms 25 knobs were described and for the processing industry 10 knobs. Every knob can affect multiple sustainability indicators. The relationships between the knobs and the sustainability indicators are mapped in a confrontation matrix (Figure 4). Knobs affected the sustainability indicators directly and indirectly. The effect of turning the knob was, where possible, supported by (scientific) literature and information from in-depth interviews.

Themes ↓	Indicators ↓ / Buttons →	Communication on the work floor (improve)	Number of training days (increase)	Batch size (reduce)	Number of times cleaning slaughter line (improve)	Sustainable transport (improve)	Way of anesthesia (adapt)	Isolation (improve)	Purchase recycled packacing (increase)	Green energy from energy supplier (increase)	Self-generating green energy (increase)
People	1. Staff turnover	+	+								
	2. Number of absenteeism	+ +	+				-				
	3. Number of workaccidents	+	+								
	4. Food safety		+	+	+ +						
	5. Traceability of products			+							
Planet	6. Square valorisation		+								
	7. Energy consumption per 1000 kg of product				-		-	+ +			
	8. Use of renewable energy									+ +	+ +
	9. Water consumption per slaughtered chicken				- -		-				
	10. CO_2 emmision per kg delivered product				-	+ +	+	+ +	+	+ +	+ +
	11. Percentage use of recycled packaging								+ +		
Profit	12. Net profit margin per kg product	+ +	+	-	-	+/-	+ +	+	+/-	+/-	+
	13. ROI	+ +	+	-	-	+/-	+ +	+	+/-	+/-	+

Figure 4 *Confrontation matrix for the processing industry.*

To visualize the effects of the knobs on the sustainability indicators, the sustainability circle was developed, which is based on a spider web model. The sustainability circle consists of the themes of People, Planet, Profit and *Poultry* (broiler farm) and the corresponding sustainability indicators. A greater surface area in the circle indicates a higher sustainability level.

Figure 5 shows the current sustainability level of HFP, and the requirements and targets of the customers of HFP. Customer requirements often focus on the short term and the targets mainly relate to the long term. Based on the results of a very limited group of broiler farms it can be stated that the broiler farms often comply with the requirements on the sustainability indicators specified by the customers of HFP for the themes People, Planet, and Profit. Looking at the long term targets, the broiler farms should improve on the Poultry theme.

The customers of HFP put fewer demands on the processing industry in the field of sustainability. For the sustainability indicators with requirements from customers, the processing industry often meets the requirements. When specifically looked at the sustainability themes it can be concluded that the levels of sustainability of the themes of People and Planet are at a moderate level and the theme Profit is still at an unsatisfactory level. All three themes still need attention.

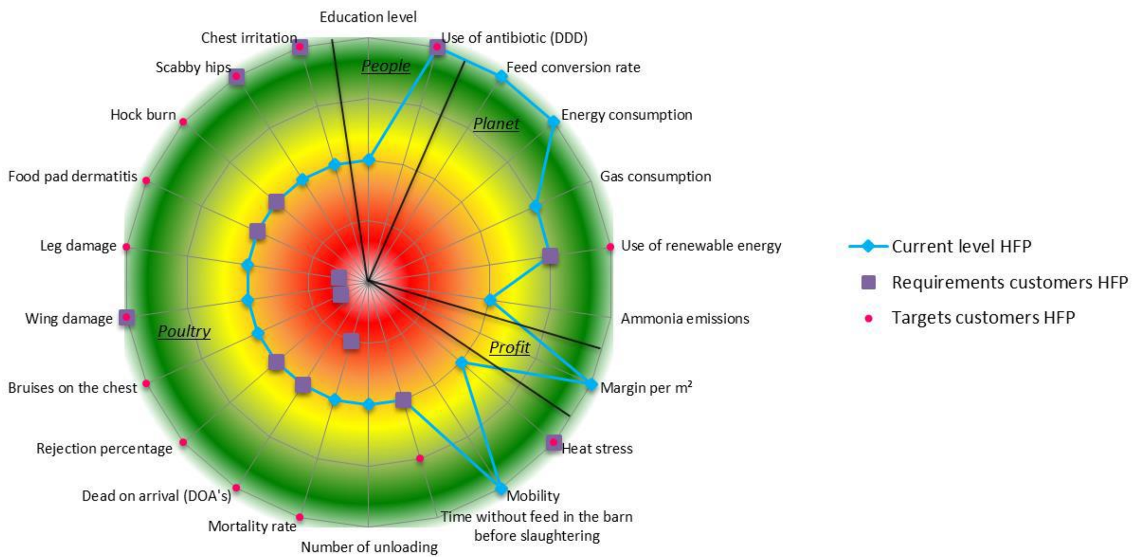

Figure 5 *Sustainability circle with current level of sustainability of HFP over 2013 – 2015 (light blue), requirements customers HFP (purple), and targets customers HFP (pink).*

The sustainability circle also shows the effect of turning knobs on the sustainability indicators, one example being the positive effect of turning the knob 'light scheme' to intermittent, where the indicators of chest irritation, scabby hips, hock burn, foot pad dermatitis, mortality rate, heat stress and ammonia emissions all improved to at least the light green area.

With the mapped effects of the knobs and the identified areas of improvement, it is recommended that a next project translates it into practice. Focus can be on determining which of the described knobs, on the short term or on the long term, are best applicable into practice to achieve the desired level of sustainability. It is advisable to look whether a sufficient enough change can be expected to actually achieve the targets of the customers with these knobs. In addition, HFP is advised to carry out research into possible methodologies, systems or models to monitor and visualise sustainability levels.

3.1.3 Sustainability model I (7664LRW4)

This project focused on optimizing the sustainability circle and on determining how HFP can communicate the required sustainability level to its customers. The required sustainability level was defined as the sustainability level that meets the wishes and needs of the customers of HFP.

Next to reading the reports of previous projects, seven product specification reports of the customers of HFP and literature were studied. Additionally, interviews with HFP, suppliers of HFP, a customer of HFP, chain experts and WUR were held. The data used for the model dates from September 2015 – August 2016.

An important customer of HFP in the top segment is Marks and Spencer (M & S). Another important customer of HFP is Tesco. Tesco operates in the middle segment on the British retail market. The most important customers of HFP on the fast food market are KFC and McDonalds.

Research into the wishes and needs of the customers shows that animal welfare, animal health, traceability and food safety are important issues. The increased consumer attention to animal welfare causes an increasing demand for animal friendly production. On the basis of these results indicators were merged with the indicators from former projects. With that, also the P from Poultry – animal welfare is added to the four other Ps (People, Planet, Profit, Poultry – animal health). Indicators regarding Poultry – animal welfare are for example mortality of the broilers and enrichment, and for Planet for example energy consumption and gas consumption. Eventually, 42 indicators within the 5Ps were used to determine the sustainability level on the broiler farm and in the processing industry.

The sustainability level will be communicated to customers through a dashboard. The dashboard indicators were selected in consultation with experts in the field and chain partners. The indicators can be measured and a formula is used to determine the value. A five-point scale ranking (A till E) was used, where A was the best score and E the worst score. Boundary values for the ranking were determined by requirements of Heijs' customers, legal requirements, national averages, and average and standard deviations in Heijs' data.

The dashboard has four displays; per slaughtered flock, per shed, per year and per category (P). Some indicators can be measured per slaughtered flock, some per shed and some only per year. The indicators that can be measured per slaughtered flock and per shed always relate to the broiler farm. To eventually come to a score per category (P) it is desired to have one score for each indicator.

Eventually the scores per indicator form the score for a category P. These scores are given on the dashboard and with the dashboard the sustainability level can be communicated to the customers. An example of the dashboard is displayed in Figure 6. The figure shows us that a lot of indicators are on average. There is one indicator that scores below average and that is 'green energy vs. fossil energy'. From the four categories, People has the best score, followed by Poultry – Animal health and Planet. Some indicators on the dashboard do not have a score yet, which is due to a lack of data for the indicators. The category Poultry – Animal welfare does not have a score yet because none of its indicators was measured at this point (no data).

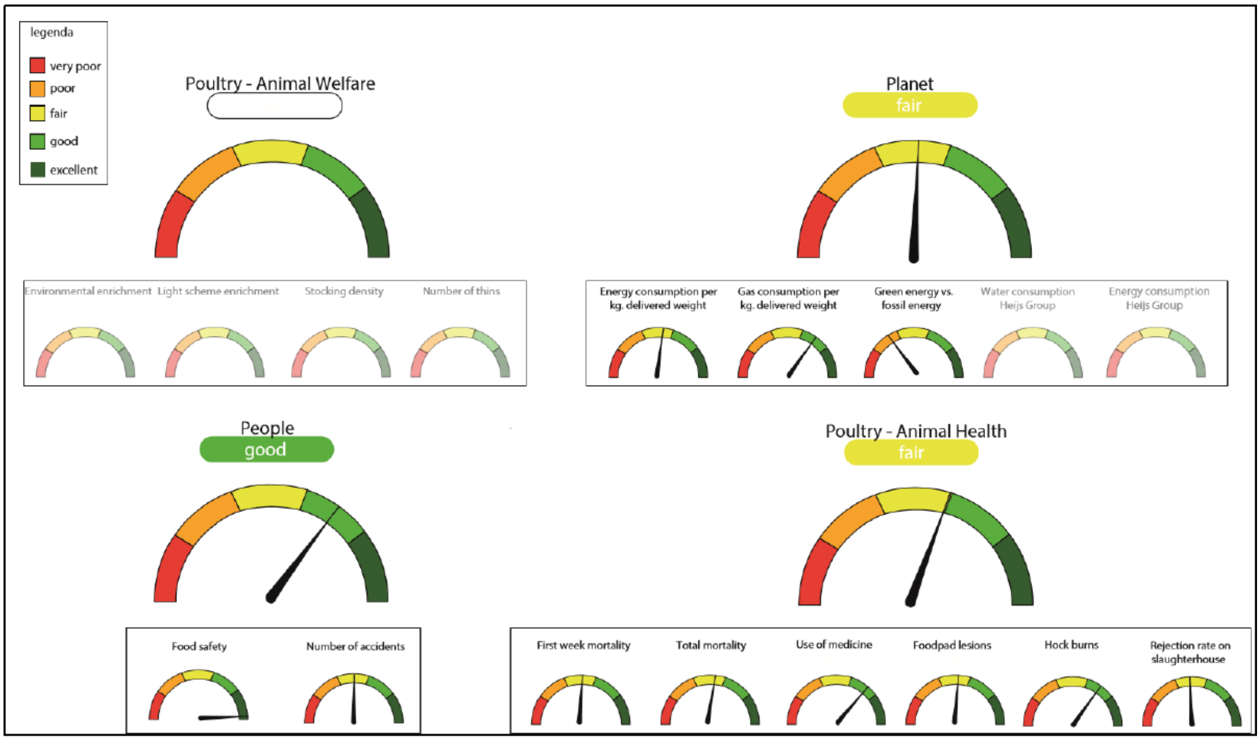

Figure 6 *Sustainability dashboard September 2015 – August 2016; people, planet, poultry animal health and poultry animal welfare.*

The most important recommendations are to obtain the missing data on the dashboard visible for HFP. For some indicators this is possible with the flock card that is delivered from the broiler farm to HFP at the end of every cycle. Next to that, HFP can set up a form for the yearly data and could send that to the broiler farms on the end of every year. The broiler farms then can fill out the form and return it to HFP at the end of every year.

3.1.4 Sustainability model II (7704LRW4 & 7705LRW4)

The aim of this research project was to take the existing sustainability model from project 7664LRW4 one step further by integrating the models of the three chain links in the production of poultry meat, enabling HFP to get an overview of their sustainability level and to pinpoint where in the chain improvements can be made. The integrated model should make monitoring easier and more effective.

Research methods used were desk research; company reports on previous research, scientific papers and various online sources. In addition, in-depth interviews were conducted and discussions were held with 13 broiler farmers (suppliers and non-suppliers to HFP). Data used in the model dates from 2016. Besides, several broiler and sustainability experts were interviewed.

First of all a list of practical measurable indicators was prepared (Figure 7). The indicators with regard to the 'P' of Poultry (animal welfare and health) were not part of the sustainability model yet, these were being developed in report 7676LRW4 in section 3.2.3.3. The indicator traceability was removed due to a lack of reliable data. As recommended by the previous report, two indicators were added, these indicators are: Food safety - *Campylobacter* and the usage of water per kilograms delivered weight.

	Dashboard indicators	System indicators
People	D1. Number of accidents (PI) D2. Food safety – *Salmonella* (BF) D3. Food safety - *Campylobacter* (BF)	S1. Level of education (BF) S2. Staff turnover (PI) S3. Absence (PI) S4. Courses and training (PI)
Planet	D4. Electricity consumption (kWh) per kg delivered weight (BF) D5. Gas consumption (m^3) per kg delivered weight (BF) D6. Green vs. grey electricity (BF) D7. Electricity consumption (kWh) per 10^3 kg delivered weight (PI) D8. Water consumption per 10^3 kg delivered weight (PI)	S5. Feed consumption per kg delivered weight (BF) S6. Reduction of NH_3/particulate matter (BF) S7. Valorisation of the broiler (PI) S8. Green vs. grey electricity (PI) S9. CO_2 emission per kg delivered product (PI) S10. Recycled packaging materials (PI) S11. Water consumption per kg delivered weight (BF)
Profit	D9. Gross feed profit (BF)	S12. Net profit margin (PI) S13. Return On Investment (PI)
Poultry		

Figure 7 *System and dashboard indicators for People, Planet and Profit.*

The indicators for the broiler farmers were validated, the classification of all indicators were corresponding with the rankings, with exception of 'gross feed profit per chicken'. The classification of this indicator was not changed because the validation is based on an insufficient amount of data. For the broiler farmers the standardisation is made per kg delivered weight and for the processing industry this is per ton delivered product. The dashboard showing the average of the broiler farmers interviewed in this project (suppliers and non-suppliers to HFP) is displayed in Figure 8. The average of all broiler farmers (benchmark) scored excellent on Profit, good on People (no data for *Campylobacter* and *Salmonella* though), and fair on Planet.

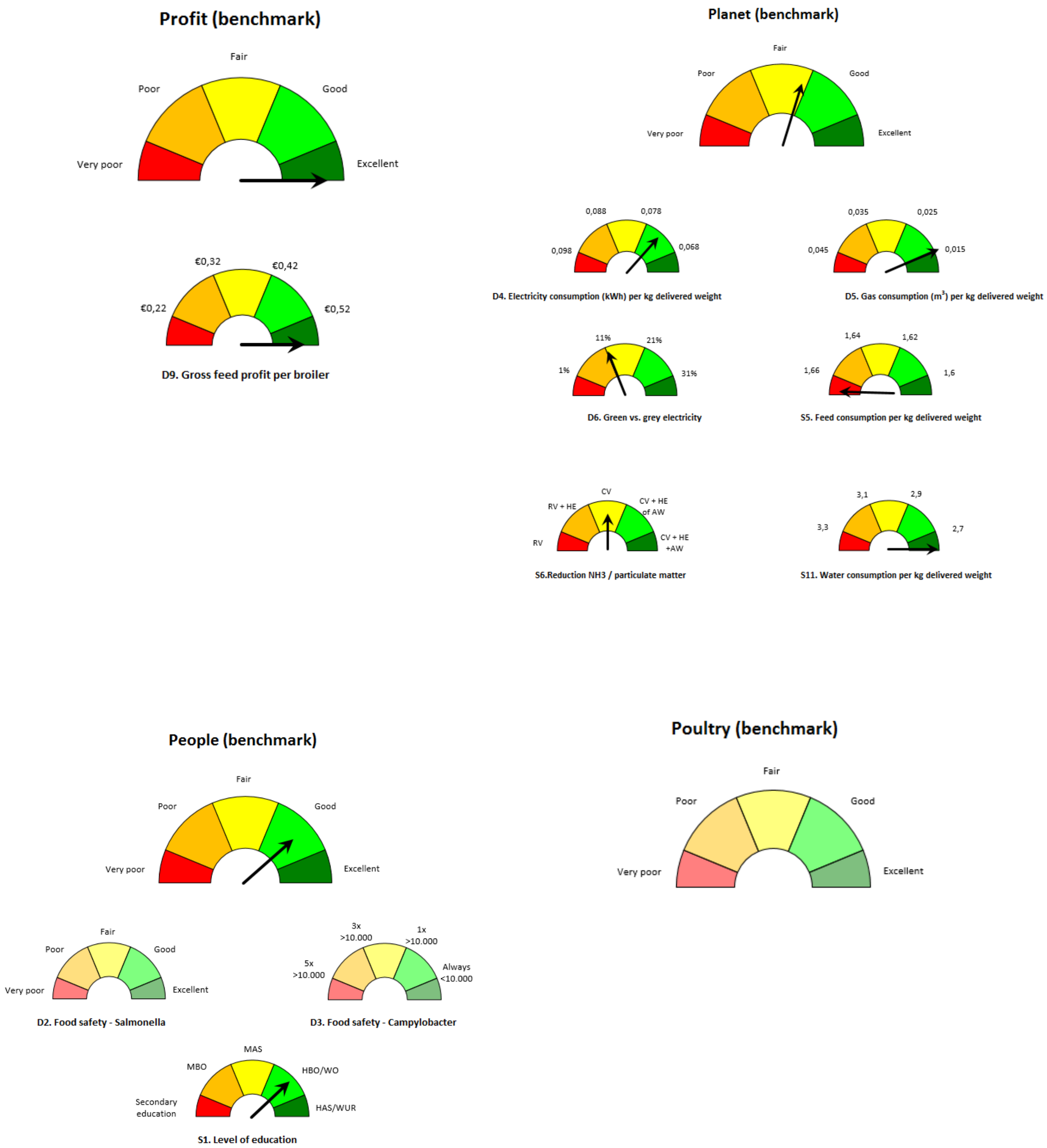

Figure 8 *Sustainability dashboard of the average of the broiler farmers interviewed in this project (suppliers and non-suppliers to HFP).*

A confrontation matrix was prepared to prioritize the 'knobs' that can be influenced by the broiler farms and processing industry. Turning of the knobs influences the sustainability level. The confrontation matrix shows that the broiler farms could start by turning the knobs 'lowering the water pressure' and/or 'reducing the starving period before pre-loading' to improve the sustainability level. For the processing industry the most important knobs to improve their sustainability level are: 'providing language courses', 'taking more green electricity from the electricity supplier', 'better insulation' and 'changing to gas numbing' (Figure 9).

Knobs	People	Number of accidents	Staff turnover	Absence	Courses and training	Planet	Electricity consumption (kWh) per 10^3 kg delivered product	Water consumption per 10^3 kg delivered product	Valorisation of the broiler	Green vs. grey electricity	CO_2-emmission per kg delivered product	Recycled packaging materials	Profit	Net profit margin	Return On Investment
Proving language courses		+	+	+										+	
Increasing amount of course days					+										
Changing to gas numbing		+		+			-	+						-	-
Applying better Insulating							+				+			+/-	
Using more recycled packaging materialsgebruiken												+		+/-	
Using more green electricity from electricity supplier										+	+			-	
Lowering batch size															
Increasing intensity of cleaning the slaughter line															

Figure 9 *Confrontation matrix for the processing industry.*

Transparency in data collection, and data security as well as guarantees of anonymity of data providers are required to ensure the collaboration of the broiler farmers. These broiler farmers are not looking forward to the extra paperwork, therefore the added value of the sustainability model requires further attention. A possible way to convince them of the need for such model is to show them the benefits in terms of profit.

The model output shows the indicators at just one level, namely on a yearly basis. At cycle and flock level, the practical indicators are not measurable at this stage yet. Several kinds of model outputs can be used. A dashboard for the individual company (broiler farm and processing industry) and a dashboard for the average company were developed, so they can benchmark themselves. And lastly, a dashboard for the clients of HFP was developed. On this dashboard four big speedometers are shown, which indicate the four Ps (Profit, People, Planet and Poultry). Below the big speedometers there are several smaller speedometers, which show the dashboard indicators and gives insight in the score of the P speedometer.

Figure 10 shows the dashboard for Heijs' clients, and the indicators are filled with all available data. This picture shows that Profit scores 'very good', Planet scores 'average' and People is scoring 'good'. Currently Poultry is empty because this is still under construction in report 7676LRW4. What draws attention in Figure 10 is that some indicators do not have an arrow, these could not be calculated due to a lack of available data for the processing industry. For further development and validation, data should be provided by HFP for their own as well as other food processing industries. Our main recommendation for HFP is to start an investigation into what incentives broiler farmers need to motivate them to use the sustainability model. For a follow-up study, the indicators Food safety - *Campylobacter* and Food safety - *Salmonella* are important to be included. Currently food safety concerns only broiler farmers, but this may also apply to the processing industry. Moreover, the usage of antibiotics should be included in the 'P' of People, for it is possible to investigate whether residues are measured by default and what are legal boundaries.

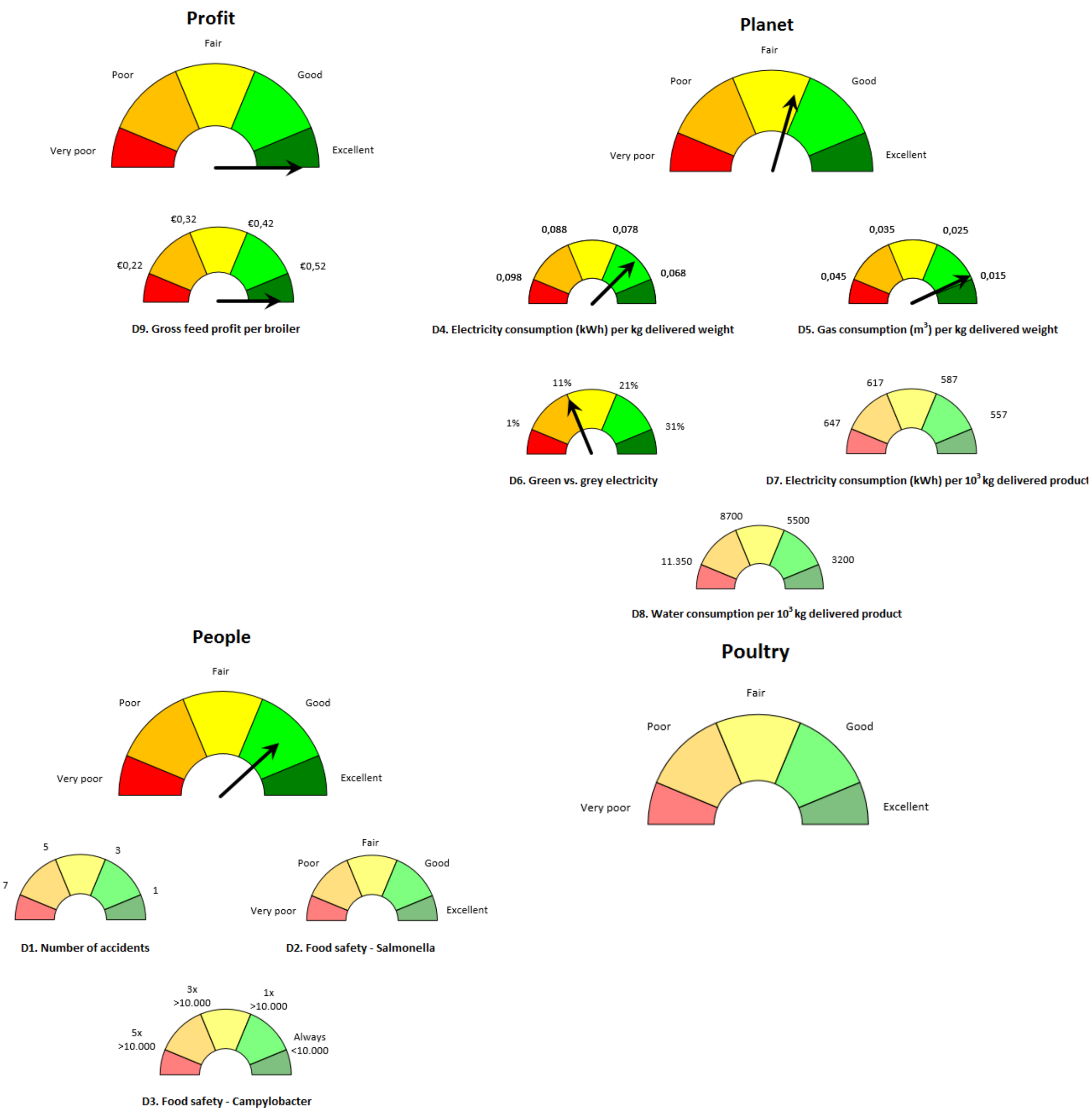

Figure 10 *Sustainability dashboard over 2016 for Heijs' customers.*

The current sustainability model provides sustainability levels per year, a follow-up model could be based on cycles. In addition, unknown data can be classified in class E. Also, it would be useful if a dashboard could be developed of the moving average of the broiler farms and processing industry. Finally, Poultry (animal welfare and health) should be included in the sustainability model once the model is ready and reliable data are available.

3.1.5 Extension and validation of the sustainability model (18200144)

The aim of this research project was to design a sustainability model for the 4 Ps that gives insight in the sustainability of farming and processing chicken broilers within the HFP chain and to show this information in a clear dashboard. The model was first validated for People, Planet, and Profit on efficacy and sensitivity. The model was then extended by implementing the welfare model 3.0, and validated on efficacy.
This research is based on the developed sustainability model as formulated in the report 7704LRW4 & 7705LRW4 (sustainability model 2.0) and the developments of the welfare model 3.0. The used research methods were desk and field research, by studying company reports of previous projects, by interviewing experts from Wageningen Research and HFP.

The sustainability model consists of four categories; People, Planet, Profit, and Poultry. These categories were subdivided into two chain links; broiler farmers and processing industry. During the completion and validation of the sustainability model a total of 36 indicators were included in the model. The indicators for Poultry were determined by the project group that developed welfare model 3.0. An overview of all indicators divided per category and chain link (v for processing industry and p for broiler farmer) is shown in Table 2 Welfare model 3.0 was not based on chain links, but chain links were assigned in this project.

Table 2 *Indicators per category; a (v) refers to the poultry farmer, and a (p) refers to the processing industry.*

People	Planet	Profit	Poultry
Staff turnover (v)	Electricity consumption (v)	Net profit margin (v)	Dead on arrival (v)
Absence (v)	Water consumption (v)	Return on investment (v)	Tilting (v)
Number of accidents (v)	Broiler valorisation(v)	Gross feed profit (p)	End of life (v)
Courses/training (v)	Green vs. grey electricity (v)		Stocking density(p)
Salmonella (p)	CO2 emission(v)		Total mortality(p)
Campylobacter (p)	Recycled packing materials(v)		First week mortality (p)
Level of education (p)	Electricity consumption (p)		Feed-water ratio (p)
	Gas consumption(p)		Enrichment (p)
	Feed consumption(p)		Animal daily dose (p)
	Green vs. grey electricity p)		Light regime (p)
	Reduction NH3 / particulate matter (p)		Hock burn (p)
	Water consumption (p)		Footpad lesions (p)
			Scabby hips (p)
			Behaviour (p)

One of the research questions related to the validation of the information system. The current information system was reviewed with all indicators developed by the previous project (section 3.1.4). The current information had to be adjusted as some indicators required more calculation. For example, the broiler farmer is reluctant to share data on their gross feed profit. Thereto, HFP, WUR and HAS decided that the gross feed profit will be calculated with reference values from the KWIN. In the context of the gross feed profit, the farmer is now asked for the number of placed broilers, and HFP for the yield price per broiler, which is stated on the slaughter report.

During the validation, the calculation and ranking for *Campylobacter* and *Salmonella* were studied. These food safety indicators are important to HFP, but little information could be gathered from the broiler farmer and the processing industry. Thereto, the calculation and ranking for these two indicators was slightly adjusted. For *Salmonella* the ranking is now based on the frequency of *Salmonella* being tested positively, and on the occurrence of SE/ST. For *Campylobacter* the ranking is now based on a frequency of 0 < 10.000 CFU/g, or 1, 2 to 3, 4 to 5, or 5 times > 10000 CFU/gram.

Subsequently, the information gathering system was validated for efficacy. It remains difficult to gather good and useful data within the broiler chain. Not all data was filled in correctly, there were variations between data based on stocking level, house level and company level. For this reason data was corrected into uniform data corrected for house level. There may be several reasons for the incorrect delivery of data. It appears that miscommunication and interests have a major influence on the correctness or completeness of data delivery, and that a number of broiler farmers heat their houses with wood instead of gas. There is no calculation in the current model to translate wood consumption into gas consumption.
During the mid-term presentations, Heijs Food Products indicated that they would like to have a dashboard as the output of the sustainability model, divided in categories and chain links. To achieve this, a number of things were added to the model. All indicators were multiplied with an impact score. In the welfare model, these impact scores were determined on the basis of literature. There was no literature available for the other categories, thereto these impact scores were now assumed to be equal. Next to the impact scores, classifications were re-evaluated and adjusted to five classes with a linear interval for each indicator.

Subsequently, with the obtained and corrected data, the categories People, Planet, and Profit were validated for sensitivity and efficacy. An example of the output of the model is shown in Figure 11. The individual indicators all respond, as do the final classifications. It is only within the Planet category that compensation is higher due to the large number of indicators.

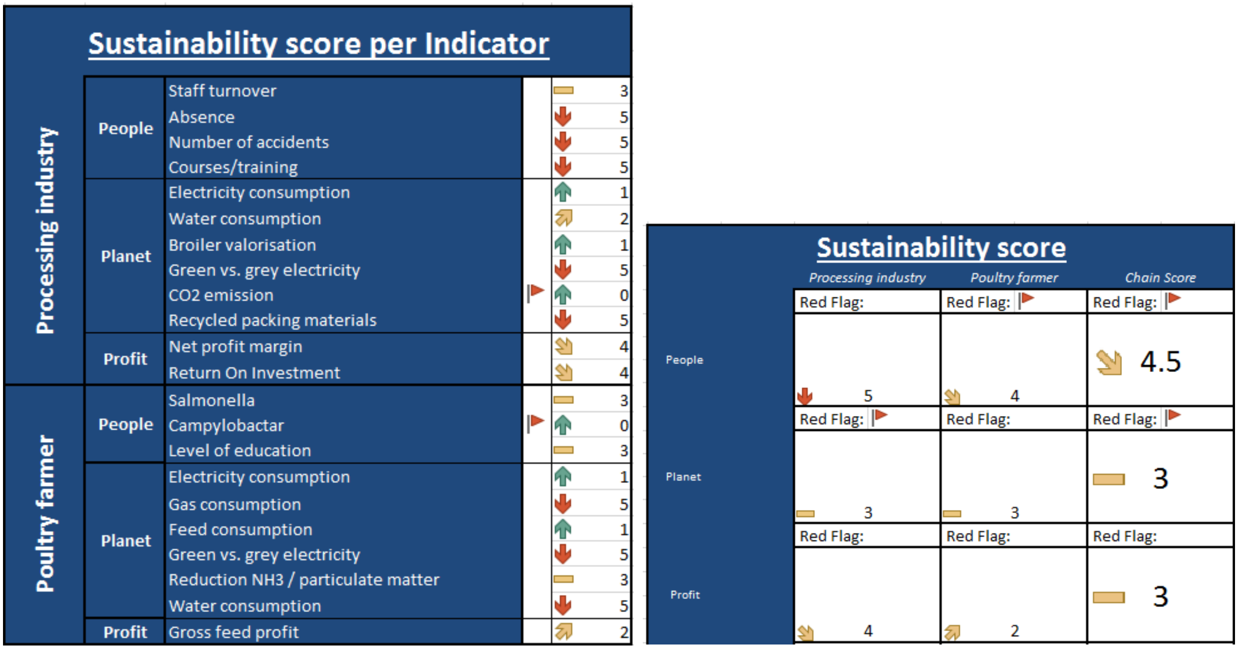

Figure 11 *Left: Sustainability score per indicator, Right: sustainability score per category.*

Finally, the welfare model 3.0 was implemented in the sustainability model and validated for efficacy. An example of the output of the model is shown in Figure 12. Due to a lack of data, it was not possible to validate the model for sensitivity yet, however the welfare model 3.0 was validated in section 3.2.3.4.

Firstly, the most important recommendation is to send an independent person with the model to broiler farmers within different production segments and to validate the model for sensitivity. Secondly, it is recommended to elaborate on the CO_2 indicator with literature study and expert interviews, and to look into translating wood consumption into for example gas (or CO_2) consumption. Thirdly, after proper validation on sensitivity, it is recommended to look into the relationships between the four categories. Lastly, it is recommended to find someone that can maintain the model.

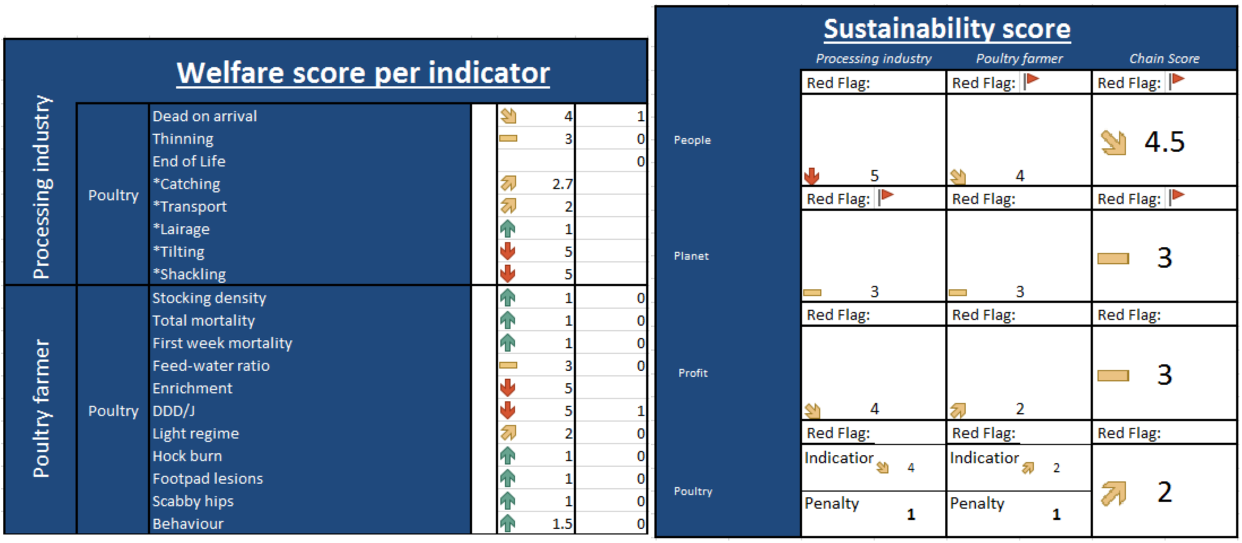

Figure 12 *Welfare score per indicator and total sustainability score per category.*

3.2 Animal welfare (WP2)

The animal welfare topic was divided into three subtopics; enrichment, end-of-life, and welfare model. The section 3.2.1 to 3.2.3 describe the focus of these research topics, contain a table with an overview of the projects performed, and the projects are described in more detail in the subsections.

3.2.1 Enrichment

Five subprojects have been carried out that aimed to develop suitable environmental enrichment for fast growing broiler chickens. Environmental enrichment increases the animal's behavioral possibilities and leads to improvements of the biological function. It is provided with the purpose of: 1) increasing the occurrence and range of the animal's normal or species-specific behavior, 2) preventing the development of abnormal behavior or reducing its extent and complexity, 3) increasing the positive exploitation of the environment (e.g., the use of an outdoor area), and 4) increasing the animal's ability to handle behavioral and physiological challenges[2]. Environmental enrichment must be biologically relevant to be effective. Riber, *et al.* (2018) defined four criteria of success for application of environmental enrichment on-farm: 1) it should increase species-specific behavior, 2) it should maintain or improve levels of health, 3) it should improve the economics of the production system, and 4) it should be practical to employ. With these requirements in mind, the subprojects were designed towards the development of effective enrichment in the HFP production chain, i.e. enrichment that meets the four criteria of success. In the first project, the preferences of broiler chickens were determined with respect to enrichment designed for resting, for foraging and exploration, and for foraging only. The second and third project aimed to determine the effect of stocking density on the use of the enrichment and the effect of different combinations of enrichments on the extent to which natural behaviour was stimulated. The final two experiments were aimed at testing the selected enrichment (wood shavings bales) on different farms (validation) and to determine the effect of the addition of natural light in houses with or without environmental enrichment, respectively. Table 3 gives an overview of the different projects performed within the topic 'Enrichment'. The projects are described in more detail in sections 3.2.1.1 to 3.2.1.5. The codes within the section titles refer to the thesis project numbers used by HAS.

Table 3 *Overview of projects executed within WP2 Enrichment.*

Code	Project title	Main activities	Execution period
Enrichment			
7555LRW4	The preference of broilers for three types of enrichment	Assessment of preference of broilers of for perches / platforms, lucerne / wood shavings bales, and pecking blocks / plastic chains.	Feb – June 2016
7662LRW1	The effect of stocking densities on the broiler behaviour near and use of enrichments	Assessment of the effect of stocking density on the behaviour and use of platforms and wood shavings bales as enrichment in broiler houses.	July – Nov 2016
7734LRW1	Enrichment III	Assessment of the effect of different combinations of enrichments on broiler behaviour.	July – Nov 2017
18200141	Environmental enrichment IV	Assessment of wood shavings bales on welfare of broilers in commercial houses: proof of principle.	Feb – June 2018
18200143	Natural light and environmental enrichment	Assessment of the combination of natural light and enrichment on the welfare of broilers.	Feb – June 2018

3.2.1.1 The preference of broilers for three types of enrichment (7555LRW4)

The aim of this subproject was to investigate the broilers preference for three of the following types of enrichment: enrichments that stimulate pecking, sitting on or in the enrichment, or being close to the enrichment, as well as the behaviour in the area near the enrichments. Two broiler houses, each on a

[2] Riber, A. B., van de Weerd, H. A., de Jong, I. C., & Steenfeldt, S. (2018). Review of environmental enrichment for broiler chickens. *Poultry Science*, *97*(2), 378–396. http://doi.org/10.3382/ps/pex344

different commercial farm, were divided into five sections. Six different types of enrichment were provided in three of these sections, two types per section: perches versus platforms, Lucerne bales versus bales of wood shavings, pecking blocks versus plastic chains (Figure 13). The other two sections in the houses did not have any enrichment and served as control sections. For this trial, platforms were created by placing plastic transport crates upside down in the litter. The preference for enrichments was tested by placing two enrichment types for the same purpose opposite of each other. The preference of the broilers based on the usage of the enrichments and specific relevant behaviours near the enrichment types was tested one day in the second week, two days in the fourth week, one day in the fifth week and one day in the sixth week of the production round.

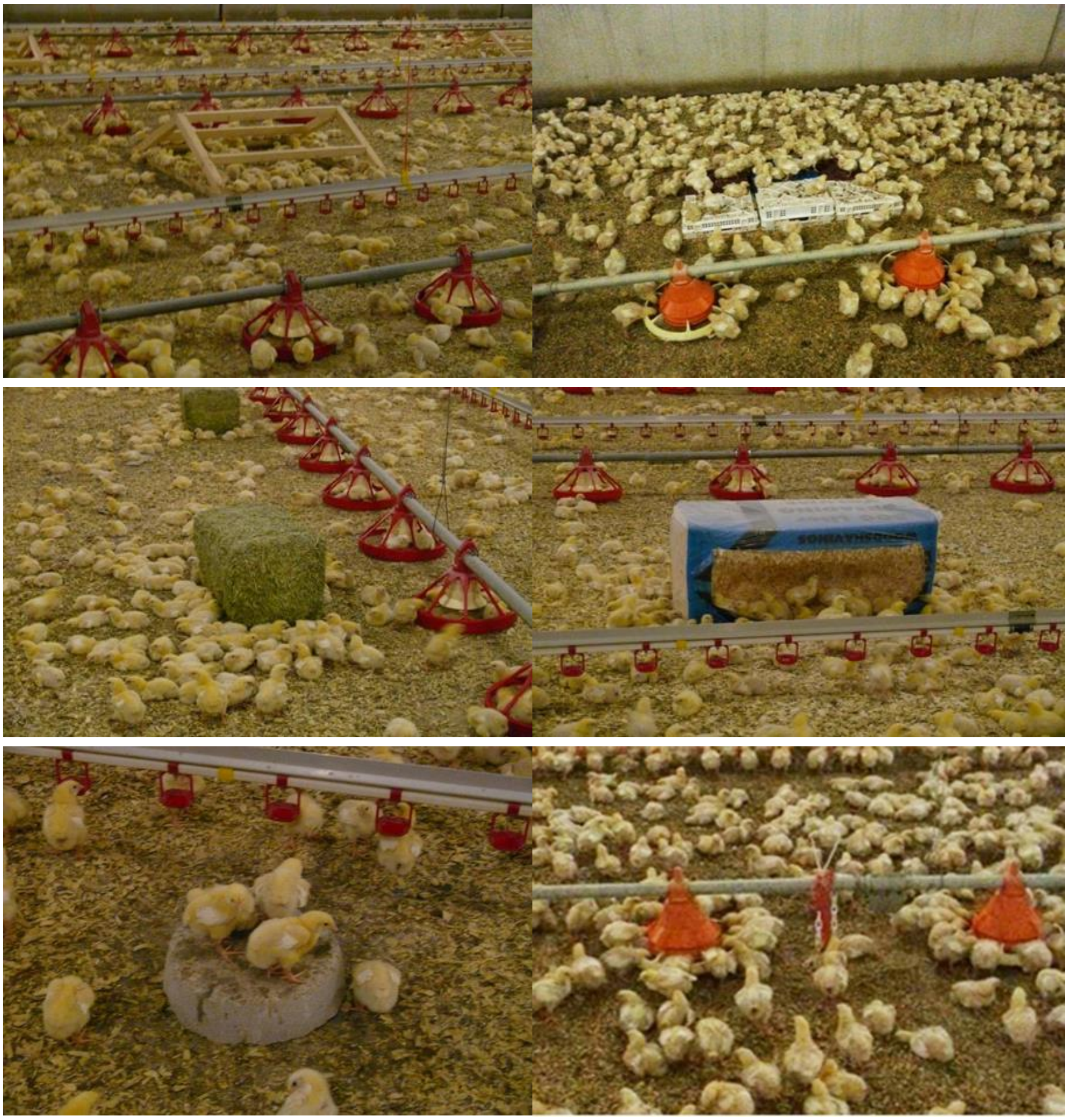

Figure 13 *The various enrichments as used in the preference experiment. Top pictures: perches versus platforms; middle pictures: lucerne bales versus wood shavings bales; bottom pictures: pecking stone versus hanging chains.*

Broilers used the perches more than the platforms, which might be explained by the fact that the broilers could use the perches not only for resting on but also for shelter, i.e. sitting under, whereas this was not possible under the platforms that were used in this trial. However, more broilers were observed sitting on the platforms than on the perches. Observations of the relevant behaviours showed that broilers walked more near the perches than near the platforms, but this was only the case at farm 1. The farmers had no preference for either perches or platforms. The Lucerne bales and bales of wood shavings were evenly used by the broilers, but the observations of the relevant behaviours showed that broilers were foraging, dust bathing and pecking more at the bales of wood shavings than at the Lucerne bales. The loose particles of wood shavings might have attracted the broilers to show these behaviours. The same low amount of aggressive behaviour was shown near these two types of bales which suggested that no competition for these resources. Farmers were however not very positive about the bales of wood shavings due to possible clogging in the intestines of the broilers and a high mortality caused by bales falling on very young chickens. The latter could however be solved by repositioning of the bales. Lastly, broilers used the pecking blocks more than the plastic chains, The farmers indicated that the plastic chains did not have any real added value with respect to broiler behaviour.

In conclusion, not all the enrichments were used as much and were equally good for stimulating the relevant behaviours. For resting, broilers preferred the platforms to sit on, but the perches to sit under (shelter). The bales of wood shavings were the best for stimulating the exploratory and foraging behaviours. Pecking blocks were found to have a more positive effect than the plastic chains. They stimulated the broilers to show more exploratory behaviours and they were used more than the plastic chains when it comes to being in, on, near and active with the enrichment.

3.2.1.2 The effect of stocking densities on broiler behaviour near and use of enrichments (7662LRW1)

Stocking density is one of the factors that has a large effect on broiler welfare. Stocking density directly affects the amount of space the broilers have, but indirectly also may affect other factors such as temperature, humidity and litter quality. Several studies examined the effect of stocking density on the behaviour of broiler chickens; increasing the stocking density was found to increase agonistic behaviour and to reduce the amount of resting behaviour of the broilers. Also, a more crowded house would make the broilers move a shorter distance per hour and to show less pecking, scratching and walking behaviour. A way to stimulate broilers to show more natural behaviour is applying environmental enrichment. In this study, the effect of stocking density on the use of and behaviour near two different types of environmental enrichment were examined: platforms (here provided as plastic transport crates that were placed upside down) and bales of wood shavings. It was also studied whether or not there was an effect of age and time of the day on the behaviour of the broilers and use of the enrichments by the broilers.

The results of this study showed that a low stocking density (25 kg/m^2) compared to a higher stocking density (35 kg/m^2) stimulated behaviours like eating, drinking, dustbathing, foraging, comfort behaviour and object pecking. This is supported by other studies that show that natural behaviour is stimulated by a low stocking density. The broilers showed more dustbathing at a low stocking density, which may be explained by the quality of the litter which was improved at the low stocking density. Eating and drinking behaviour was scored more often in the low stocking density section, which may be related to more space for the broilers to move. Different studies have shown that broilers grow faster at a low stocking density which is in line with the broilers moving more often to the feeders in the present study.

The pilot studies showed that almost all the different ways of using enrichment were higher at a lower stocking density (Figure 14). So more space seems to stimulate broilers to move more toward enrichments, sit on them more and peck more at them, whereas lying and standing were more stimulated at a higher stocking density. Possibly, at a higher stocking density, broilers resting or standing blocked the access to the enrichment for the other chickens.

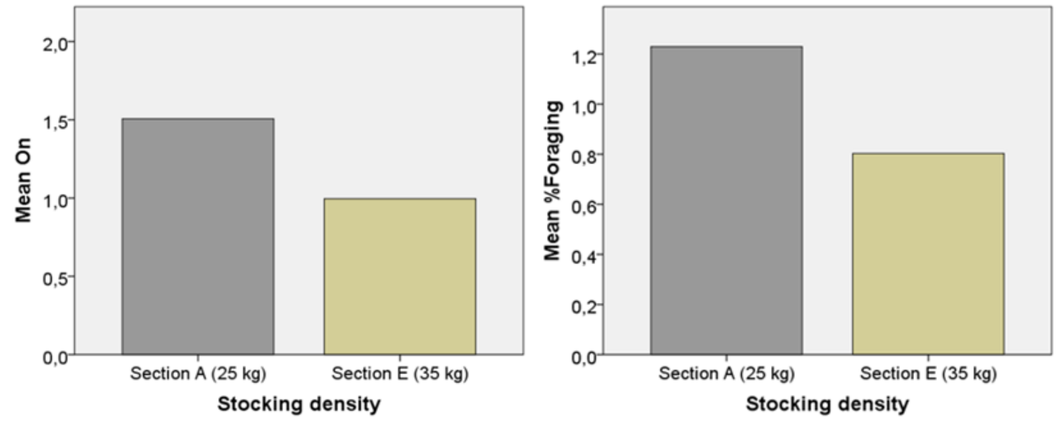

Figure 14 *The mean number of broilers on the enrichments at a an stocking density of 25 kg/m^2 and 35 kg/m^2 (left) and the mean number of broilers showing foraging behaviour near the enrichments at 25 kg/m^2 and 35 kg/m^2 (right panel).*

Broiler welfare was improved at a low stocking density, as it stimulates natural behaviour of the broiler chickens and they suffered less from foot pad dermatitis, hock burns and locomotion problems. High stocking density leads to dirtier litter. This directly affects factors like foot pad dermatitis and cleanliness. Broilers were dustbathing more at a low stocking density, which might have been caused by a better litter quality in the low stocking density sections

The broilers used the enrichments the most at an age of 21 days. This might be caused by the fact that at an age of 10 days the broilers were too small to reach the platform and unable to pull at the bales of wood shavings and at the oldest age, between the thinning's, it possibly became harder for the broilers to get on or near the enrichment because of their body size and weight. Also their growing body size was causing less space for the broilers to use the enrichment, so less broilers could use the enrichments at the same time. The broilers showed more dustbathing and comfort behaviour, but less aggressive behaviour with increasing age.

The number of broilers on or near the enrichments was lower at the final observation day. This result was also found in another study; the use of platforms was lower on the last observation day than during the earlier observation days. The amount of dustbathing and foraging increased over the day and comfort behaviour was most observed around noon.

The present experiment showed that broilers had a preference for platforms, bales of wood shavings and pecking blocks as these were used the most. Next to this a low stocking density seems to stimulate natural behaviour in broiler chickens. It also seems that a low stocking density provides the broilers with more opportunities to use the enrichments.

3.2.1.3 The best combination of environmental enrichments for broilers on commercial farms (7734LRW1)

Broiler's welfare can be improved with environmental enrichment. Different types of enrichments can be applied and already have been tested for the HFP production chain. In this study it was tested which combination of the following enrichments is the best for improving broiler's welfare. The three types of enrichments were; bales of wood shavings, platforms and pecking blocks (See Figure 15 for the platforms used in this experiment). The broiler's behaviour was observed by the scan sampling method. There were always two observation days in a row at three ages. At each observation day there were three time blocks. All the sections were equally treated during each time block, so the number of enrichments by which the broilers were observed were equal for each section. The results showed that wood shavings were the best for stimulating active natural behaviour. This is probably due to the loose particles of wood shavings in which the broilers seem to like for dustbathing. The particles also seem to make it attractive to show more pecking, and comfort behaviour. Next to stimulating behaviours, the bales are also used for shelter (broilers crowding around the bales). The platforms stimulated resting behaviour and the broilers did not get disturbed much by each other on and near the platforms. The platforms were also used for shelter (resting under the platforms). The pecking blocks were not preferred, since only a few broilers can use them at the same time and they stimulated only object pecking. This study showed that bales of wood shavings are advised to stimulate active natural behaviour and that platforms meet the requirements of the birds for resting at an elevated structure.

Figure 15 *Platforms as applied in the 3rd and 5th enrichment study.*

3.2.1.4 The effect of bales of wood shavings on the welfare of broilers (18200141)

The purpose of this study was to investigate whether or not the welfare of broilers can be improved by bales of wood shavings as enrichment. This study built further on previous assignments to determine the most effective enrichment for broiler chickens and involved a validation study on commercial farms. The welfare of broilers was studied by the behaviour and other welfare indicators such as footpad dermatitis, hock burn, injuries and gait score.

The behavioural observations and health parameters were measured on four different farms during one production cycle. Each farm had two identical broiler houses, one with enrichment (wood shavings bales, equally divided over the house, see Figure 16) and one without enrichment. The behavioural observations were performed in week 4, 5 and 6 of the production cycle. The broilers were observed on 6 different locations equally divided over the broiler house. Approximately 40 broilers per observation location were observed. Other welfare parameters were measured in week 6 of the production cycle. These included foot pad dermatitis, hock burns, injuries and plumage cleanliness measured on five different locations with approximately 20 broilers per location. Gait score and litter quality were measured in six different locations with approximately 25 broilers per location.

Figure 16 *Wood shavings bales provided as environmental enrichment.*

Significantly more natural behaviour (dust bathing, comfort behaviour (preening, wing and leg stretching) and foraging) was found in enriched houses than in control houses (Figure 17) and on enriched locations as compared to locations without enrichment. Furthermore, activity was higher in locations with enrichment than in locations without enrichment. Additionally, locomotion activity was lower in enriched houses than in control houses (measured on both enriched and non-enriched locations). This might be explained by the fact that the enrichment might have provided shelter and thereby stimulated resting behaviour close to the enrichment. The other welfare parameters were not affected by the enrichment.

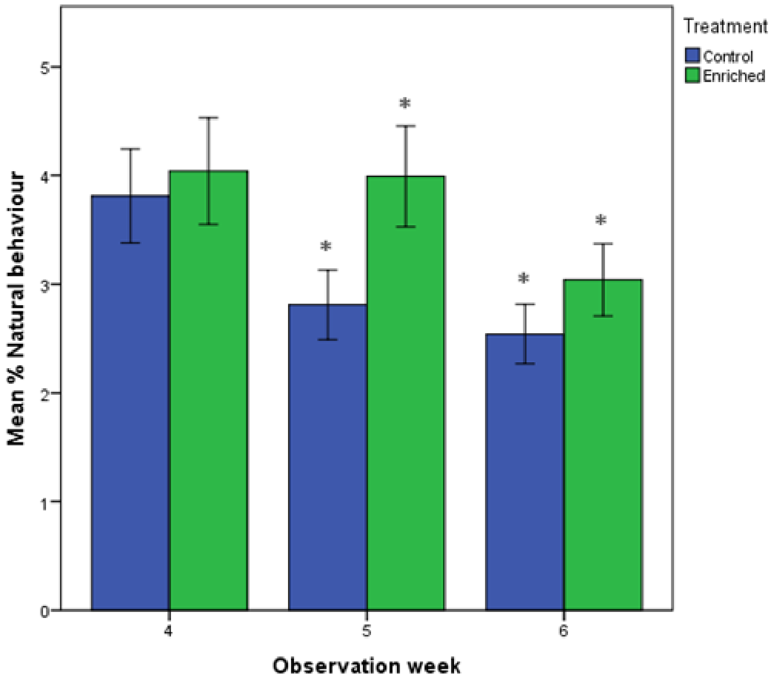

Figure 17 *Proportion of broiler chickens performing natural behaviours in week 4, 5 and 6 of age in enriched versus control houses. An asterisk indicates a significant difference between the treatments (P<0.05).*

It was concluded that environmental enrichment had a positive effect on broiler welfare as it stimulated the performance of natural behaviour, despite the fact that no differences were found in other welfare indicators.

3.2.1.5 Natural light and environmental enrichment (18200143)

Natural light (daylight) can have a stimulating effect on the activity of broiler chickens. Moreover, natural light in combination with environmental enrichment may promote the use of the enrichment objects. The effect of natural light, in combination with environmental enrichment, has been studied in 6 houses on one location. Per house 35,000 broilers were placed. The houses differed by the use of natural light (by windows in the side walls, see Figure 15) with artificial light or only artificial light, in combination with bales of wood shavings and/or platforms or no environmental enrichment. Behaviour was scored weekly from week 2 onwards during two production cycles of 6 weeks. Active behaviour, resting behaviour and foraging behaviour where measured. Gait score, foot pad dermatitis, hock burn, cleanness, and litter quality were scored in week 4 of each production cycle (just before thinning or depopulation).

Broilers where more active in houses with natural light compared with the houses without natural light during cycle 1. This difference was not found in cycle 2, possibly because of the hot weather conditions. Consequently, the temperature inside the houses was higher during cycle 2, which could have caused more resting behaviour of the broilers. Broilers were more active in the houses with only bales of wood shavings (8.5%) in comparison with the houses with bales of wood shavings and platforms (5.9%) (cycle 1). Broilers showed more foraging behaviour in the houses with only bales of wood shavings (2.7%) in comparison with the houses with bales of wood shavings and platforms

(1.5%) and the houses without enrichment (1.4%) (cycle 1). The scores for foot pad dermatitis were lower in the houses without natural light with bales of wood shavings and platforms (1.7%) than in houses with the other enrichment types (1.0% and 1.2%) (cycle 1). Broilers in the houses with natural light, bales of wood shavings and platforms had less foot pad dermatitis than the broilers in the houses with the same type of enrichment without natural light. In production cycle 1 no significant difference was found for foot pad dermatitis between the houses with and the houses without natural light. In cycle 2, more foot pad dermatitis was found in the houses with bales wood shavings and in the houses with natural light. The differences between cycle 1 and 2 can possibly be explained by the difference in breed. In cycle 1 Cobb broilers were used and in cycle 2 Ross broilers.

In conclusion, both natural light without and environmental enrichment had a positive effect on the behaviour of the broilers. There was no additional effect on natural behaviour for the combination of natural light and environmental enrichment. However, it should be taken into account that data of more production cycles should be collected before drawing any firm conclusions.

3.2.2 End-of-life

The process of catching, transport, lairage, stunning and killing of broiler chickens may affect their welfare, and also have a negative effect on product quality[3,4,5]. In the HFP production chain this not only occurs during depopulation, but also at thinning, which is common practice for the majority of farms. The subprojects 'end-of-life' were aimed at identifying possible welfare risk during the end-of-life stage in the HFP production chain, as well as providing solutions for improvement, if necessary. An overview of the projects performed within the topic 'End-of-life' is shown in Table 4. The projects are described in more detail in sections 3.2.2.1 to 3.2.2.3. The codes within the section titles refer to the thesis project numbers used by HAS.

Table 4 *Overview of projects performed within WP2 End-of-life.*

Code	Project title	Main activities	Execution period
7624WUR4	Improvement end-of-life - Thinning	To develop parameters to assess the welfare and health of broilers during and after thinning based on measurement at farm level and analyses of slaughterhouse records.	Feb – June 2016
7677LIV4	End-of-life course II - Occurrence of injuries and damage in the slaughter process	To determine to what extend broiler injuries and damages result from the pre-slaughter phase or from the slaughter process, including at which stage in the slaughter process.	Feb – June 2017
18200142	End-of-life III – Effect of temperature during transport on the prevalence of heat stress in broiler chickens	Measurement of temperature and relative humidity during transport from farm to slaughterhouse. Correlation studies of these measurements with measured heat stress indicators.	Feb – June 2018

3.2.2.1 Improvement end-of-life – Thinning (7624WUR4)

For an optimal use of the floor space in the barn, thinning is a common procedure during the growth period of broilers. Heijs Food Products (HFP) is seeing the development that farmers apply thinning more than once during one production round. Thinning might have a negative effect on broiler health and welfare, which concerns customers of HFP. However, little research has been performed on the effects of thinning on welfare and health. The aim of this project was to find parameters that can be

3 Jacobs, L., Delezie, E., Duchateau, L., Goethals, K., & Tuyttens, F. A. M. (2017a). Broiler chickens dead on arrival: associated risk factors and welfare indicators. *Poultry Science*, 96(2), 259–265. http://doi.org/10.3382/ps/pew353

4 Jacobs, L., Delezie, E., Duchateau, L., Goethals, K., & Tuyttens, F. A. M. (2017b). Impact of the separate pre-slaughter stages on broiler chicken welfare. *Poultry Science*, *96*(2), 266–273. http://doi.org/10.3382/ps/pew361

5 Jacobs, L., Delezie, E., Duchateau, L., Goethals, K., Vermeulen, D., Buyse, J., & Tuyttens, F. (2017). Fit for transport? Broiler chicken fitness assessment for transportation to slaughter. *Animal Welfare*, *26*(3), 335–343. http://doi.org/10.7120/09627286.26.3.335

used to measure the animal welfare and animal health before, during and after thinning. With this knowledge, the thinning process can be optimized in combination with several methods for thinning. Dutch broiler farmers have been asked about the thinning method on their farm with an online survey. In total 29 farmers filled in the questionnaire; 30% of these farmers apply thinning more than once during a production round, of which 90% of the broilers is manually caught. In general, the thinning process does not take longer than 1 hour and usually takes place between 23.00h and 8.00h. About 75% of the farmers apply food deprivation for the thinning process. The duration of the period of feed deprivation before thinning is usually 6 hours.

One of the most important reasons for the use of thinning is the financial benefit. The benefit per square meter has been calculated. For the farms that did not use thinning the feed profit is € 7.64/m^2. When thinning is done once the feed profit is € 9.39/m^2 and when thinning is done twice the feed profit is € 10.06/m^2.

An experiment was designed to determine parameters to measure health and welfare during thinning. Two pilot studies were performed. During these studies, physiological measurements, data from the slaughterhouse, behavioural measurements and technical results were included. With these parameters a follow up study was set up to explore the effect of feed deprivation during thinning relative to no feed deprivation (study A) and another pilot study on the effect of an additive (Hepafit) during the thinning process (study B). The additive (Hepafit) contains a lot of energy. The purpose of the additive is to give the broilers some energy during the feed deprivation.

These studies were conducted at two different farms (1 and 2). At farm 1, the parameters were tested in combination with feed deprivation. In one barn all the broilers were deprived of feed and in another barn only the broilers that were thinned were deprived of feed. At farm 2, the parameters were tested in combination with an additive (Hepafit). In one barn the broilers received the additive (through drinking water) and in another barn the broilers did not receive the additive.

One of the physiological measurements that was scored is the plumage cleanliness. In both studies, no difference between the experimental groups were found. The litter quality was scored before, during and after thinning. In study A, the litter quality became worse in the barn where all broilers were deprived of feed, in comparison with the broilers kept in the barn where only the thinned broilers were feed deprived. In study B, there was no difference in litter quality between the two groups. The dry matter content of the litter increased in both studies. In study A, the dry matter content increased more in the barn where only the thinned broilers were deprived of feed. Possible explanation for this difference is that these broilers produce a better manure quality than in the barn where all broilers were feed deprived. The gait score is also scored, but no differences were found.

To score the behaviour of the broilers after thinning a protocol was set up for study A. This was practically not achievable. For study B was decided to count the number of broilers around the feeder when the farmer started feeding after thinning. The number of broilers was counted every 15 minutes and this was done for 60 minutes. The results showed that the number of broilers around the feeder dropped with increasing time after the start of feeding. There was no difference between the groups. Beside these parameters the walk direction/activity was studied; 25 broilers were marked during thinning. The day after thinning and the day before the next catching moment, the marked broilers were located. Both studies did not show a difference between groups.

Based on the slaughterhouse recordings the following parameters should be scored: hock burn, scabby hips, footpad dermatitis, uniformity and catching damage. In both studies no differences were found between groups. Not all parameters were scored by the slaughterhouse. Catching damage will not be used in the final protocol because the catching damage does not have an effect on the broilers that stay in the barn. Uniformity will be used in the protocol, because uniformity could be an indicator that can be influenced by health issues and maybe also by feed deprivation and thinning.

<u>Technical results</u>
Body weight, growth from first thinning moment until catching and the mortality from first thinning moment until catching were scored. In study A, the broilers of the barn that were partially feed

deprived grew more than the broilers of the barn that received feed deprivation as a whole. The difference was 40 grams. In study B, the broilers that got the additive grew less than the broilers that did not got the additive. Here the difference was 60 grams. No differences in mortality were found.

It can be stated that the parameters for behaviour and catching damage are not interesting for further research. The number of broilers per feeder and uniformity are interesting for further research. For this protocol was made that can be applied in further studies (Figure 18).

Physiological	**Slaughterhouse**
• Plumage cleanliness • Gait score • Manure quality • Dry matter content	• Footpad dermatitis • Hock burn • Scabby hips • Uniformity
Behavioral measurement	**Technical Results**
• Broilers per feedpan	• Final weight • Growth • Mortality

Figure 18 *Indicators that can be used to evaluate the effect of thinning procedures on welfare and health of the broiler chickens.*

3.2.2.2 End-of-life course II - Occurrence of injuries and damage in the slaughter process (7677LIV4)

Previous studies show that it is inconclusive where damage or injuries occur in the pre-slaughter or the slaughter process of broilers. The goal of this study was to determine whether injuries and damage to broiler chickens occur during the slaughter process at HFP processing. It was also examined where in the slaughter process these injuries and damage occur. Furthermore, it was examined if injuries occur during the pre-slaughter process and which factors influence the occurrence of the injuries. For these purposes dislocations, fractured bones and bruises of both legs and wings have been scored. Bruises on the breast have also been included. A distinction was made in size and probable age of found bruises.

To examine which pre-slaughter factors have an influence on carcass damage, historical data of the years 2014 until 2016 from ten different broiler farms, contracted by HFP, were analysed. These ten farms were selected based on their average foot pad dermatitis score (fpd-scores) of 2016. Five farms with a low average fpd-score (<40) for 2016 and five farms with a high average fpd-score (>80) for 2016 were selected. The historical data was tested for correlations between the amount of breast, wing and leg damage and the various pre-slaughter factors. Statistical analysis found positive associations between average live body weight (p=0.000; B=0.001), number of dead broilers on arrival (p=0.001; B=0.004), and wing damage. A negative association was found between wing damage and the number of broilers per container (p=0.037; B=-0.015).

To determine whether or not injuries and damage occur during the slaughter process, the presence of bruises on the wings, legs and breast as well as broken wings and legs were scored in lairage and post-plucking (see Figure 19 for examples). Hanging wings were also scored post-shackling and again post-stunning. An increase in percentage of broken wings between lairage (0.99%), post-shackling (1.67%), post-stunning (2.73%) and post-plucking (5.02%) was found (p=0.000 between all four moments). This is an average increase of 4.03% of broken wings between lairage and post-plucking (Figure 20). The stunner settings (frequency and strength of current) were found to have no correlation with the occurrence of broken wings (p=0.800). The small, medium and large breast bruises all show an increase between lairage and post-plucking (p=0.000). For wing bruises, the small sized bruises showed a decrease between lairage and post-plucking (p=0.047) whereas large wing bruises showed an increase (p=0.033).

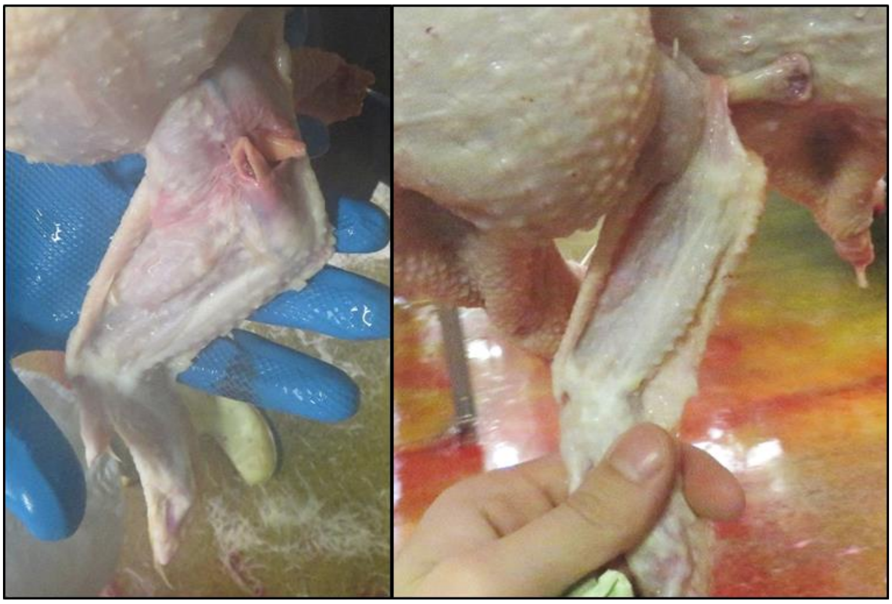

Figure 19 *Left: wing fracture, the bone itself is broken. Right: wing dislocation, the bone is still intact but it is no longer connected to the joint.*

This study shows that injuries and damage do mainly occur during the slaughter process at Heijs and less during the pre-slaughter process. Further research is advised to determine exactly which steps in the slaughter process are the main causes of these injuries and damage. Also, it was very difficult to accurately determine the age of bruises purely based on colour and size on different body parts of the broilers.

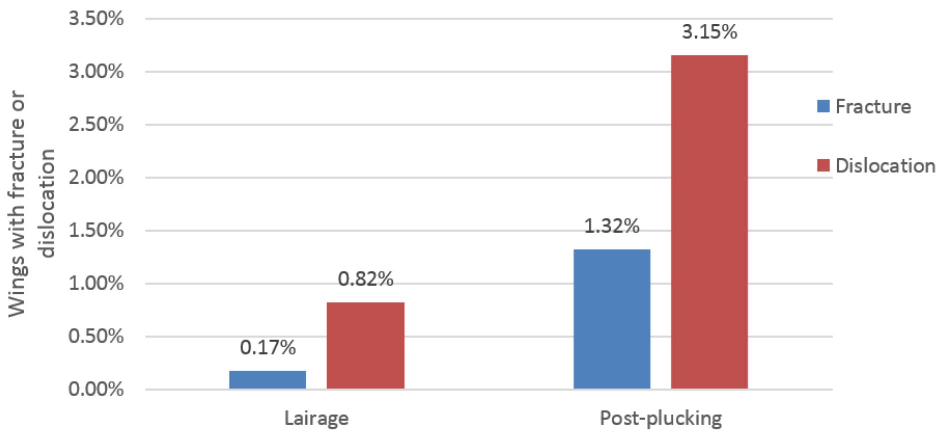

Figure 20 *Mean percentage of wing fractures and dislocations in lairage and post-plucking, based on a sample size of 20 flocks.*

3.2.2.3 End-of-life III – Effect of temperature during transport on the prevalence of heat stress in broiler chickens (18200142)

Broilers are exposed to various stressors during transport, especially heat stress is the main welfare risk during transport. Heat stress is the condition in which the broilers will apply physiological, anatomical and behavioural mechanisms aimed at facilitating heat loss to, or minimising heat gain from the environment. The broiler regulates its body temperature during transport primarily by panting. Therefore, the objective of this study was to assess the effect of transport temperature and relative humidity (RH) on prevalence of panting behaviour in the lairage. Additionally, the effect of transport on the temperature and RH in the trailer, the effect of drawer temperature on weight loss and body temperature and the effect of the health status on the prevalence of panting was studied. It was hypothesized that an increasing transportation duration and temperature results in a higher prevalence of panting and therefore more heat stress in lairage due to longer exposure to high temperatures.

The effect of temperature and RH on the prevalence of panting was studied during 7 transports of thinned broilers, 3 transports of a long duration (> 7 hours) and 4 transports of a short duration (< 4 hours) with 8,000 to 11,000 broilers per transports. The broilers were between 1.2 and 2.1 kg. An overview of the temperature and RH distribution was created by logging at 36 locations in the trailer (Figure 21). In four out of eleven container stacks, in the bottom, middle and top drawer at the driver side, midline and passenger side. Additionally fifteen broilers were measured before and after transport on weight loss and the body temperature during transport. The panting behaviour was observed in the temperature and RH measured containers after transport. The information from the food chain information and slaughter report were used to relate to the prevalence of panting after arrival.

Figure 21 ***Containers with thinned broilers, red circles indicate the place of the loggers at one side of the truck.***

There was no effect found of transport time on the temperature in the trailer, during both long and short transports. The temperature varied in the trailer with higher temperature found in the midline and first stack in contrast to the other locations. In the midline of the last top drawer the temperature was lower than at the other midline top drawers No relation was found between the temperature and weight loss or body temperature during transport. Panting behaviour occurred after 2 transports, with 1% and 6% of panting broilers. No relation was found between the temperature and relative humidity during transport on the prevalence of panting behaviour.

Although the temperatures varied considerably (>10°C) between the different locations on the truck the average temperatures did not rise during the course of the transport. The RH also varied between the locations on the truck and showed more fluctuations over time. The temperature differences found between the midline and driver- and passenger side are in line with results found in other studies. This can be explained by the natural ventilation which was found better at the outside of the trailer. The lower temperature found at the back top, was due to the effect of active ventilation used during the transports, indicating that active ventilation works locally. The transport, with 6% of the broilers panting, took a stationary period of 55 minutes prior to unloading. Limited observations prior to the stationary period showed no sign of panting behaviour while after the period 6% of the broilers were displaying panting behaviour. These broilers seemed to be more distressed, due to higher vocalisation and restlessness, unlike the transport with 1% panting behaviour. Therefore suggested is that heat stress occurs somewhere between 1 and 6% of panting broilers. The health status of the flock did not influence the panting. Due to the increase of panting behaviour during the stationary period, stationary periods are assumed to be the largest influence on panting behaviour.

3.2.3 Welfare model

The assessment of welfare on-farm and during the end-of-life process provides insight in the welfare status of the flocks of the HFP production chain, and provides opportunities for improvement. For broiler chickens, the Welfare Quality® assessment protocol is available[6], but this implicates that an assessor visits and inspects broiler flocks, which is not feasible for HFP. However, routinely collected data on-farm and at the slaughter plant may provide opportunities for a more simple but also more feasible welfare assessment. Such a protocol has been developed for HFP in the following subprojects, with the requirement that the indicators should be valid, reliable and repeatable. An overview of the projects performed within the topic Welfare model is shown in Table 5. The projects are described in more detail in sections 3.2.3.1 to 3.2.3.4. The codes within the section titles refer to the thesis project numbers used by HAS.

Table 5 *Overview of projects executed within WP2 Welfare model.*

Code	Project title	Main activities	Execution period
7526WUR4	Indicators for welfare and health	Welfare measures based on the Welfare Quality assessment protocol were determined for the aspects feeding, housing, health and behaviour. For each aspects indicators were developed, together with characteristics and key figures, absence or presence of scientific evidence, feasibility of measurement and a ranking.	Feb – June 2015
7557LIV4	Welfare model 1.0	Selection of indicators based on scientific evidence and measurability, together with experts. Development of classifications and impact scores. Development of overall welfare score. Testing with first data set.	Feb – June 2016
7676LRW4	Welfare model 2.0	Validation of model 1.0 with a data set and adjustment of classifications in case sensitivity was too low. Heijs acceptability limits formulated, incorporation of penalty system in model.	Feb – June 2017
18200144	Welfare model 3.0	Incorporation of indicators for enrichment, end-of-life and behaviour, including classifications and impact scores. Validation with data set. Development of behaviour assessment forms for farmers. Validation with data on flocks for regular housing as used by Heijs farmers.	Feb – June 2018

3.2.3.1 Indicators for welfare and health (7526WUR4)

The goal of this subproject was to determine the current level of animal welfare and animal health in the Heijs broiler production chain. Another goal of this project is to investigate possible improvements for animal welfare and health during the life of the broiler chicken from the broiler farm until slaughter.

The project started with desk research to gather information about the broiler chicken chain. Few companies, being part of the broiler production chain, were interviewed to get more insight in standard processes in the production chain. Based on the book Welfare Quality, and in consultation with experts in the field of animal welfare and health, important aspects were pinpointed for each phase in the broiler chicken chain (broiler farm, transport, slaughter) within the main themes of feeding, housing, health and behaviour of broilers. The twelve selected aspects were divided and processed into indicators, characteristics and key figures. For each of the indicators, characteristics and key figures a definition, absence or presence of scientific evidence, feasibility of measurement and a ranking was described. This information was collected from scientific reports, the Welfare Quality assessment protocol for poultry and several interviews with experts in the field of animal welfare and health. If possible, a ranking was made for each indicator, characteristic and key figure. This ranking was used to determine the current level of Heijs Group with respect to animal health and welfare.

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[6] Welfare Quality® Consortium. (2009). *Welfare Quality® Assessment protocol for poultry (broilers, laying hens)*. Lelystad.

The level of animal welfare and health was visualized by a colour scheme, with all the feasible indicators, characteristics and key figures. This colour scheme provides a quick overview of the Heijs Group performance in animal welfare and health. Mainly the indicators that have an economic incentive are routinely measured, and these achieved a good score within the Heijs Group chain. The colour scheme also showed a lot of indicators, characteristics and key figures that could not be measured, because for example no valid indicators have been developed or because these are currently not integrated in the Heijs production chain. To determine the influence on animal health and welfare, further scientific research is needed. Especially the broiler chicken farms received a high score for health and welfare, because most of the animal welfare and health themes are covered by the broiler chicken manual from Heijs Group and thus already included in routine monitoring of the production chain.

A lot of information is registered per phase in the production chain, and subsequently fed back to the previous phase. Sending information forward is only on request. Separate phases in the production chain are not aware of the added value of the information for the next phase in the chain, and they are also afraid of the abuse of information by competitors. It is therefore advised to make an application to fill in all the information.

3.2.3.2 Welfare model 1.0 (7557LIV4)

In North West Europe, the attention to animal welfare in broilers is on the rise. In order to respond to these new developments and requirements of customers, poultry processor Heijs Food Products commissioned Wageningen UR and the HAS University of Applied Sciences to develop a new welfare model specific to the needs of Heijs. The new welfare model should be able to measure the broiler welfare and also be able to process the measurement results into a welfare score. Heijs commissioned the development of a new welfare model because the existing welfare models are not feasible for HFP. Also the existing models sometimes lack sensitivity or are not capable of giving an overall welfare score for broilers. The development of the welfare model started with a selection of parameters provided by the previous study (7526WUR4).

First, the non-relevant parameters were discarded. Then a literature study was carried out to give a substantiated motivation to each parameter whether or not to use them in the model. The parameters and motivation were listed together and submitted to several experts who have helped make a further selection. After the meetings with the experts the final selection of parameters was made. For each parameter, a new literature study was carried out in order to compile a classification and impact score. A classification is a measurement value to determine the state of the parameter. Each classification is divided in up to 5 different classification scores. The impact score is a score between 1 and 7, and describes the welfare effects on the broilers. The parameters which both substantiated a classification and impact score have been incorporated into the model. In total there were 22 parameters selected for the welfare model, 11 parameters were not included in the model because more research needs to be done to make the parameter measureable or to develop a classification and impact score. These parameters are described separately in the report with advice on how they could be included in the model. The other 11 parameters: ammonia concentration, stocking density, breast irritation, defined daily dose animal, hock burn, heat stress, mobility, litter quality, mortality and culling, feed-water ratio and footpad dermatitis are included in the first version of the welfare model. For each parameter, the classification score is combined with the impact score. The outcome is the parameter score to each parameter. All parameter scores are added together to define the overall welfare score.

The model has been tested with slaughterhouse data combined with fictitious data for the on-farm measures. The model provides a best possible welfare score of 46 points and the worst score is 229 points. Besides the total welfare score a star graph is given which displays the parameter score of each parameter (see example). The radar chart can be divided into the four criteria of the Welfare Quality Eye: good feeding, housing, health and behaviour. In the present version of the welfare model three of the four criteria are displayed because none of the parameters is related to the behavioural criteria. The star graph can be used by Heijs in order to determine whether the broiler farms meet the reference value, but also to provide feedback.

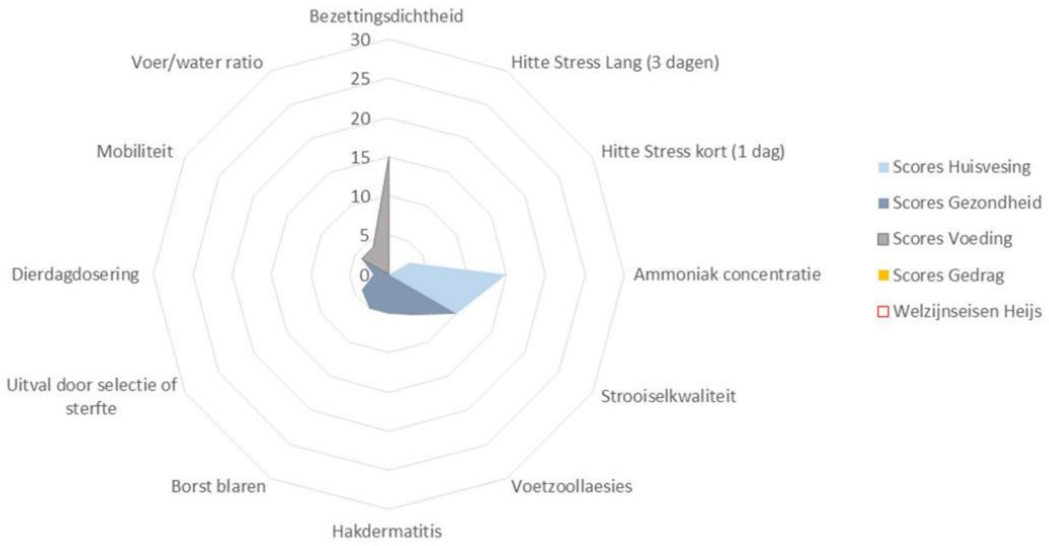

Figure 22 *Example star graph of parameter scores of the welfare model 1.0.*

3.2.3.3 Welfare model 2.0 (7676LRW4)

In this study, welfare model 1.0 (developed in project 7557LIV4) for broiler chickens was validated. HFP intends to use this welfare model to ensure the welfare of broiler chickens towards its customers. The first version of the welfare model gives a total score based on 11 different parameters. These parameters are composed of a classification and impact score. It was expected that in a number of parameters the classification score would have to be adjusted to improve the sensitivity of the welfare model. In order to investigate the sensitivity, 6 regular broiler farms were used to gather the required farm and slaughter data from 2016. These data include a total of 124 flocks. The data has been entered in the first version of the welfare model. The average of the measurements has been calculated per parameter for every farm. The percentage of flocks that achieved a certain final score has been calculated. These calculations were compared to investigate whether the variations in the measurements were also visible in the variation of the final scores. If this variation was not found, the classification score was adjusted. The parameters stocking density, feed- water ratio, defined daily dose for animals, hock burn and footpad dermatitis have been considered sensitive enough and were not adjusted. For the parameter mortality and culling, the classification has been modified because the sensitivity of this parameter was not sufficient. The parameters heat stress, ammonia concentration, litter quality and breast irritations have been removed from the model for a variety of reasons.

Through discussions with experts and literature research, it was decided to include the parameters dead-on-arrival, light regime, scabby hips, thinning, enrichment and 1st week mortality and culling in the welfare model 2.0. The parameters chicken sound, feed conversion ratio, uniformity, damage pre-slaughter procedure and behaviour have been investigated, but cannot be added to the welfare model yet.

Acceptable limit values for HFP have been set per parameter. When a parameter is above or below the limit, one penalty point will be assigned to the flock. These penalty points are set to clearly visualize whether a parameter exceeds the limit. It is important that a large variety of broiler farms will be used in order to validate the welfare model 2.0. This is necessary because if a greater variation in measurements is created, the sensitivity of the model can be tested more reliably. For the execution of the welfare model 2.0, it is important that the broiler chicken farmers keep their data up to date. The same applies to the slaughterhouse. If the administration is not properly maintained, the welfare model 2.0 will no longer be objective.

3.2.3.4 Welfare model 3.0 (18200144)

Animal welfare is globally growing in importance. This is also the case in livestock farming. In order to meet the changing expectations of their customers, companies are starting to improve their animal welfare. One of these companies is Heijs Food Products (HFP). To monitor the animal welfare of companies who supply HFP, HFP has started a collaboration with Wageningen University & Research (WUR) and HAS University of Applied Sciences in 2015 to create a welfare model for broilers. Since that time, the welfare model consisted of twelve parameters, namely: stocking density, loss due to mortality and selection, feed-water ratio, animal daily dose (antibiotic treatments), hock burn, footpad lesions, light regime, first week mortality, enrichment, death on arrival, scabby hips and thinning.

Two important aspects that were missing in this model, were parameters about the end-of-life phase and the behaviour of the broilers. The goal of this project therefore was, to add these two aspects to the welfare model, to validate these separately and to validate the total model.

To be able to establish the parameters, literature research was conducted and consultations with experts in the broiler industry, broiler behaviour and the slaughter industry were conducted. For both the end-of-life as the behaviour parameter, multiple criteria were established. This was done, in a way that multiple components could be added, without the new parameters overpowering the existing parameters. For the end-of-life parameter, the following aspects have been studied: catching, transport, lairage, tilting and shackling. Within the parameter behaviour, the following tests have been established: physiological response, foraging, social and maintenance behaviour.

The end-of-life parameter has been validated, using the data of 155 flocks. By using this data, it has been determined whether the model was sensitive enough and could reflect differences between flocks. When this was not the case, the corresponding classification score or impact score was adjusted. The classification score was adjusted when experts were questioning the range of the classes. The impact score was adjusted when the parameter score did not show any effect on the total welfare score. For validating the behaviour parameter, the behaviour tests were conducted at different companies affiliated by HFP or HAS University. It was being checked whether the tests were practically feasible and whether or not they gave reliable results. When this was not the case, the tests or percentages were adjusted.

Subsequently, the total welfare model was validated, using the obtained data from the 155 flocks (Figure 23). It was researched whether the total welfare score corresponded to the expectations based on the data of the flocks. Besides that, the validation showed if the parameters are correctly balanced and have a correct influence in the total welfare score.

The total welfare score reflects the welfare of the flock. As a result, the welfare of different flocks can be compared. In the model it is also shown for which parameters a flock has a good or bad score. Because of this, the company can make changes in certain aspects for the next flock, when willing to improve the welfare. The total welfare score is divided into eight classes. This way flocks can be assigned to classes, making a clearer overview for HFP of how the welfare scores are divided.

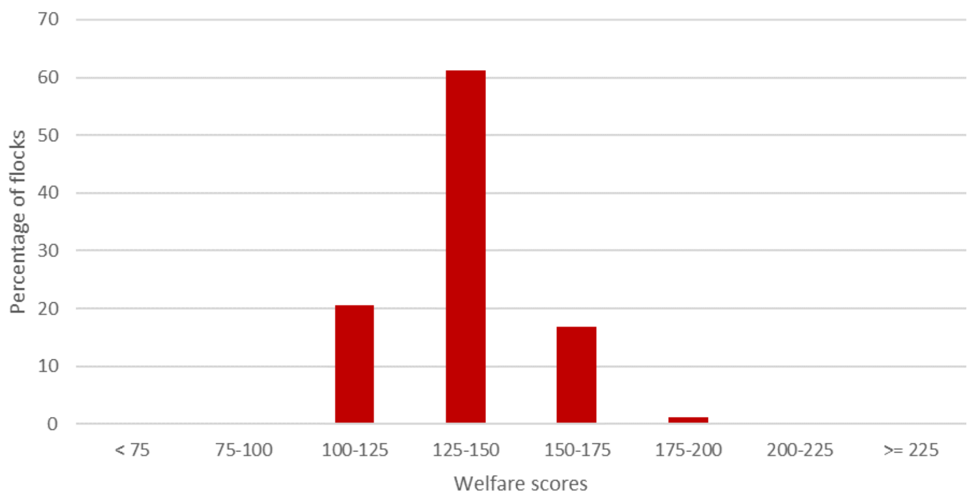

Figure 23 *Percentages of broiler flocks from HFP per welfare score classes from Welfare Model 3.0 (n=155).*

3.3 Healthy and robust broiler chickens (WP3)

Campylobacter is an important food safety problem and a serious threat to public health (EFSA, 2014)[7]. This bacterium is one of the most common zoonosis in Europe and is the biggest cause of bacterial gastroenteritis in humans. According to the European Food Safety Authority (EFSA) 20-30% of the campylobacteriosis cases in humans can be assigned to consumption and/or handling of contaminated broiler meat, while 50-80% of the infections is caused by poultry related *Campylobacter* strains. This implies that transmission routes other than via poultry meat are important (for instance through contaminated (surface) water, air or direct contact with poultry). It also means that preventing *Campylobacter* colonisation in poultry is the most effective way of reducing *Campylobacter* infections in humans.

When *Campylobacter* is introduced in a poultry flock, it rapidly spreads and in a matter of days >95% of all chickens are colonised, shedding the bacterium in huge quantities (±log8 per gram faeces) without showing any signs of disease. There are multiple factors playing a role in the risk of a flock to become colonized. Therefore, reducing *Campylobacter* should aim at the major routes and risks of introduction in broiler flocks.

Reducing *Campylobacter* contamination during the slaughter process involves improving hygiene at plucking and evisceration in the slaughter line, and and interventions aiming to eradicate *Campylobacter* that are present on chicken carcasses and meat. The various subprojects were aimed at identifying the risks for introduction in the HFP chain and to find possible ways to limit these risks.

Table 6 provides an overview of the different projects performed within WP3. The projects are described in more detail in sections 3.3.1 to 3.3.6. The codes within the section titles refer to the thesis project numbers used by HAS.

[7] EFSA. (2014). *EFSA explains zoonotic diseases: Campylobacter.* Italy.

Table 6 *Overview of projects executed within WP3 Healthy and robust broiler chickens.*

Code	Project title	Main activities	Execution period
7625LIV4	*Campylobacter* reduction in the chain	Inventory of possible transmission routes to introduce *Campylobacter* in broiler houses. Analysis of influence of feed additives on prevention. Literature study on reduction in the slaughterhouse.	Feb – June 2016
7678LRW4	*Campylobacter* reduction in the chain II	Further investigation of the effects of feed additives. Influence of dry matter content of the litter on *Campylobacter* investigated through measurements. Possibility of identification of transmission routes via thinning by genotyping explored.	Sept 2016 – Feb 2017
7695LRW4	*Campylobacter* reduction in the slaughter line I	Literature study on possible technologies to reduce *Campylobacter* in the slaughterhouse.	Sept 2016 – Dec 2017
(1)	Inactivation of *Campylobacter* on poultry meat by nitrogen cold atmospheric plasma	Laboratory investigation of the effect of cold atmospheric plasma on *Campylobacter*.	Oct 2017 – Feb 2018
7696LRW4	The role of partial depopulation on the introduction of *Campylobacter* in broiler houses	Assessment of the effect of thinning in the transmission of *Campylobacter* to houses by genotype analyses of farm samples from faeces, courtyard and people, and from transport materials, and slaughterhouse samples from cleaned containers.	Feb – June 2018
18200157	Reduction of *Campylobacter* spp. in the poultry slaughter line	Investigate if *Campylobacter* in the slaughter line can be reduced by use of (a combination of) physical treatments, using *E. coli* as indicator.	Feb – June 2018

(1) This project was performed by researchers of Wageningen Research[8].

3.3.1 *Campylobacter* reduction in the chain (7625LIV4)

This study looked at the possibilities to reduce *Campylobacter* in primary poultry farms and during the slaughter process.

Literature studies have shown that it is not possible to fully reduce *Campylobacter*, when present in the flock. Therefore, it is examined whether it is possible to keep the flock free of *Campylobacter*. According to literature studies there are various ways for *Campylobacter* to enter the broiler house, important transmission routes are flies and staff. Figure 24 is a map made by students showing various routes of introduction of *Campylobacter* in a broiler house.

[8] H. Van Bokhorst-van de Veen, L. Berendsen, W. Roland, Inactivation of *Campylobacter* on poultry meat by nitrogen cold atmospheric plasma – A feasibility study, Wageningen Food & Biobased Research, Report 1839, Wageningen, 2018

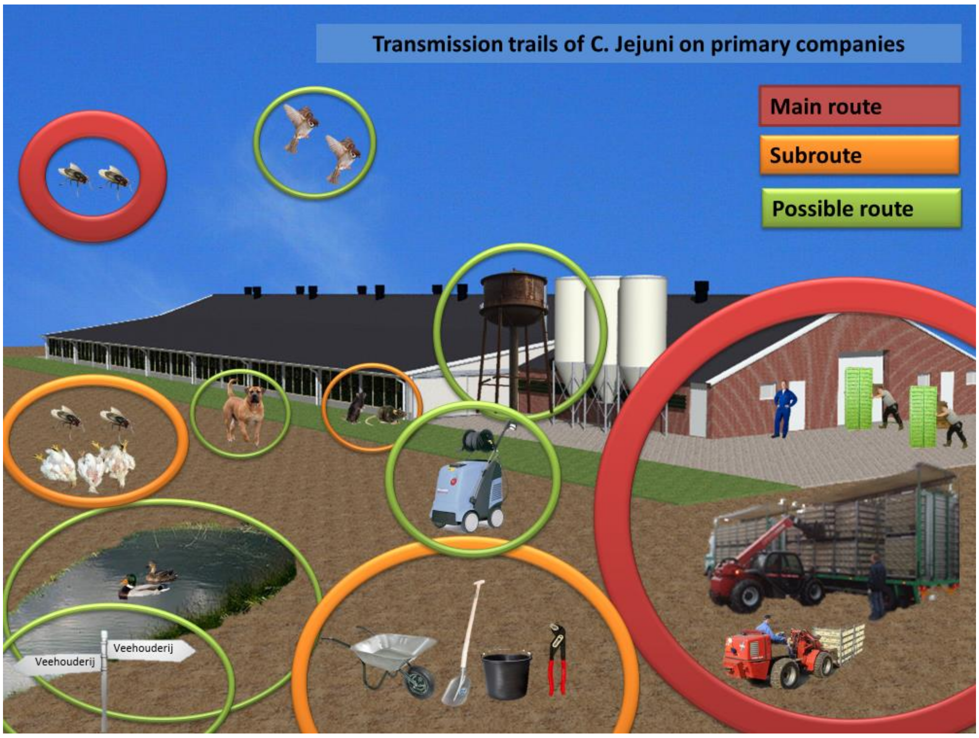

Figure 24 *Major routes of Campylobacter introduction in a broiler house.*

Campylobacter jejuni, C. coli and C. lari are thermotolerant species, with an optimal growth temperature of around 42°C. The body temperature of birds together with a lack of an effective local immune response, makes chickens an ideal amplification vessel for *Campylobacter* and leads to extremely high numbers of *Campylobacter* in their intestines. The amount of *Campylobacter* in the caecum manure of a broiler can reach up to 8 log colony forming units per gram (CFU/g). After the first chicken is colonised with *Campylobacter*, the entire flock becomes colonised within a few days, without showing any signs of disease.

Research has shown that feed additives may have a positive effect on the reduction of *Campylobacter* in broiler chickens. To examine the effect under field conditions, three most promising feed additives were selected; Original XPC, Adimix Precision and PoultryStar®. These feed additives were tested in 10 broiler houses on two broiler farms, a farm with 6 broiler houses (Farm 1) and one with 4 houses (Farm 2). Flocks received either a single feed additive Adimix Precision (n=2), Original XPC (n=4), a combination of PoultryStar® with Adimix (n=1) or Original XPC (n=1), or no feed additive (n=2). The presence and levels of *Campylobacter* in the chickens were tested using samples of caecum manure. Every week these samples were taken and *Campylobacter* counts were performed.

A possible effect of the additives on intestinal health was measured based on a footpad lesion score, visual bedding material score and technical results. Earlier research had shown that the feed additives had a positive effect on intestinal health.

Field research showed that six of the ten flocks became colonised with *Campylobacter*, 4 of which around the time of thinning of the broilers. It is plausible that at thinning *Campylobacter* is introduced in the remaining part of the flock through staff and equipment. Chicken in two broiler houses on two farms became colonised with *Campylobacter* on day 28. After the first detection, *Campylobacter* counts increased to high levels (up to 9 log) in subsequent days, regardless of the use of feed additives.

The results of the field research show that Adimix Precision was associated with wetter bedding material in the house. This is based on the footpad lesion score and the visual score bedding material. This research has not shown that feed additives have a positive effect on feed conversion and growth per day of the broilers.

In addition to the experiments on feed additives, also possible interventions to reduce *Campylobacter* during the slaughter process were investigated. A literature research was performed to identify where contamination occurs with potential for reduction within the slaughterhouse . Literature studies have shown that during the slaughter process scalding and cooling usually lead to a significant reduction of *Campylobacter*, while after plucking and evisceration often a significant increase in contamination is observed.

An overview of *Campylobacter* levels in various phases of the poultry production line was created by the students and is presented in Figure 25.

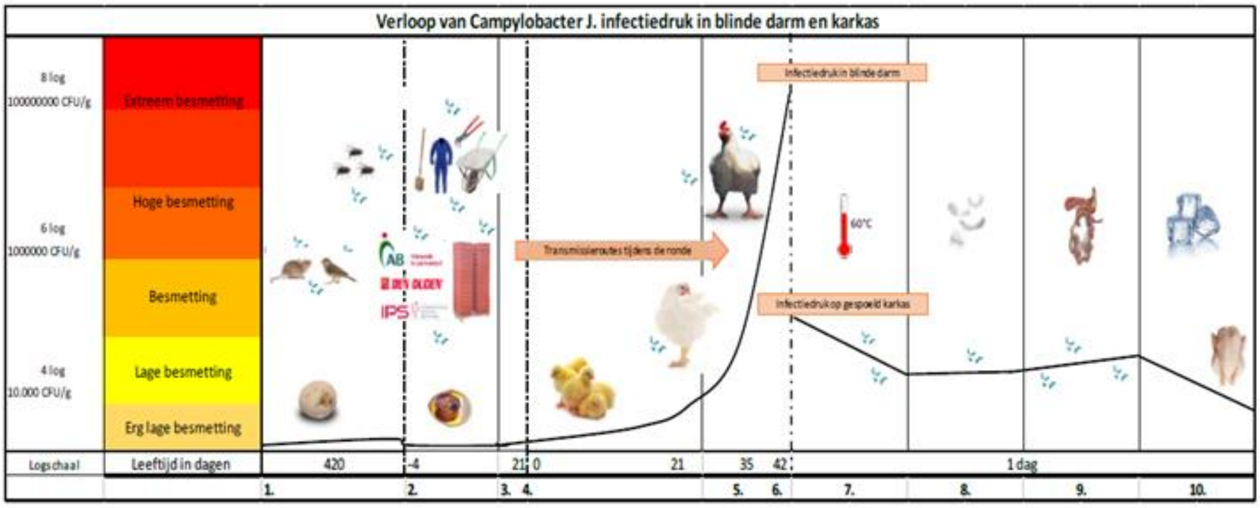

Figure 25 *Campylobacter levels in various phases of the poultry production line.*

3.3.2 *Campylobacter* reduction in the chain II (7678LRW4)

This research project focuses on obtaining more insight into the effects of feed additives on reducing *Campylobacter* and *Salmonella* in broiler chickens. Additionally, the influence of the dry matter content of the litter on *Campylobacter* concentrations was studied. Furthermore it was explored whether thinning can be confirmed as transmission route of *Campylobacter* in poultry flocks using genotyping of *Campylobacter* strains isolated around the time of thinning.

Three broiler farms were selected, two of which were sampled on two succeeding cycles. At each Farm 1 and 2, 4 broiler houses were included (in total 16 flocks) from October 2016 until December 2016. The third farm, with 6 houses, was sampled during one cycle from November until December 2016. Overall, 22 flocks were included in the study; 15 flocks were treated with Original XPC (6 from farm 1, 4 from farm 2 and 5 from farm 3), 2 flocks from farm 1 were treated with PoultryStar®, and 5 flocks (from farm 1 and 2) formed the control group. Samples were taken according to a protocol, which was adapted in November. as no farms could be visited for sampling because of regulatory restrictions due to Avian Influenza. As a result the sampling in the first cycle of house 1 and 2 differ from the second cycle for house 1, 2, and 3, in that samples were collected by the farmer instead of students. The protocol states that per house per week, 5 caecum manure samples have to be taken at a fixed location in the house. At the same location, litter samples are taken to be evaluated visually and to determine the dry matter content. Next to these samples, per house one pooled caecum sample is taken.

The analysis showed that 19 out of 22 flocks (86.4%) were colonised with *Campylobacter.* At all the *Campylobacter* positive farms, on average about 7 to 8.5 log CFU/g *Campylobacter* was detected in the caecum samples, independent of the use of feed additives or farm. For house 1, the infection was not detected until slaughtering. In house 2, antibiotics were used in the first cycle, these results could therefore not be compared with the other results. There were no differences in colonisation levels of *Campylobacter* between individual chicken in the flocks treated with or without (control) a feed additive. The results showed that levels of Campylobacter in the caeca of individual birds within a flock vary only at the start of the colonisation.

The visual evaluation of litter did not result in a reliable indication for the dry matter content of the litter. Additionally, there was no relation between the visual score or the dry matter concentration and the presence of *Campylobacter* in the caecum samples. Perhaps a sample of the surface of the litter would have resulted in a different finding, this requires more research.
For the study on *Salmonella*, the results of samples (on disposable shoe covers) taken by the veterinarians of the farms were used. Results showed the presence of *S. infantis*, both in Original XPC treated flocks from farm 2 and 3 as in the non-treated flock from Farm 3. However, based on these limited results it is not possible to draw a conclusion on the effect of the feed additive Original XPC on *Salmonella* reduction..

During the first cycle in house 1 and 2, samples for *Campylobacter* were also collected at thinning. Samples were taken from boots, the catching machine, shovel, and other equipment in the work area. Upon detection of *Campylobacter,* genotyping was performed. The genotyping did not result in a match between the strains detected on equipment at thinning and strains from chicken after thinning, which would be indicative of a transmission route for *Campylobacter* introduction in the remaining flock. Remarkably, results showed that 70% of the sampled crates carried *Campylobacter.* To obtain better insight into the transmission route of *Campylobacter,* more extensive sampling would be required.

3.3.3 The role of partial depopulation (thinning) on the introduction of *Campylobacter* in broiler houses (7696LRW4)

Partial depopulation, also called thinning, is a potential risk for contamination of broiler flocks. Therefore, the current study focused on the risk of depopulation and washing of transport containers as possible introduction routes for *Campylobacter* in broiler flocks.
Faeces samples were taken in broiler houses on broiler farms just before depopulation and in addition, the courtyard, depopulation crew, and depopulation material were sampled (Figure 26). When no *Campylobacter* was present in the broiler house before depopulation, a second faeces sample was taken a few days later. At 3 farms *Campylobacter* was only found on the courtyard and/or the depopulation equipment but not in the chicken house, 1 farm was *Campylobacter* positive after depopulation, but was not detected on the courtyard, depopulation crew or material, and at the remaining 3 farms no *Campylobacter* was found.

Figure 26 *Overview of the sampling locations at the broiler farm and on the truck.*

In addition to the study at the broiler farms the contamination risk of transport containers and the effectiveness of the container washer at the slaughterhouse was determined. Here the presence of

Campylobacter, *Enterobacteriaceae*, aerobic bacteria and protein contamination were investigated. Of the containers, 3 of the 9 drawers and all slots still contained *Campylobacter* after washing. Enterobacteriaceae were found in 4 of the 10 slots. In addition, 8 of the 10 drawers and 9 of the 10 slots contained aerobic bacteria. All drawers and slots were also contaminated with proteins.
A total of 9 environmental samples were taken from the location of the washing of the containers and crates; All locations except a puddle below the disinfection section of the washer were positive for *Campylobacter*.

From the results at the broiler farms it can be concluded that at the farms included in this survey partial depopulation did not lead to the introduction of *Campylobacter* in the broiler houses. However, because of the limited number of houses tested, this does not allow any final conclusions on the risk of thinning to introduce *Campylobacter*.
From the results regarding the container washer it can be concluded that the current washing process is not sufficient to clean transport containers because the containers are still (microbiologically) soiled after washing. Therefore the containers pose as a contamination risk. Based on this study it is recommended to make the following changes to the container washer: modify the design of the containers or extend the container washer with cleaning brushes and finally add an enzymatic protein degrading step to the washing process.

3.3.4 *Campylobacter* reduction in the slaughter line I (7695LRW4)

The main question of this research was: how to reduce *Campylobacter* by an adjustment in the slaughter process. Based on the literature, two methods which are in accordance with regulations and requirements in the Dutch law were selected for further research; cold atmospheric plasma (CAP) and crust freezing.

Plasma is created by adding energy to a gas, the electrons are moving fast and moving away from the atom. This is where the plasma phase forms. Cold plasma takes place at a temperature between 30°C and 60°C. This method can be used to disinfect surfaces and foods, however it is mainly still in an experimental phase and no large scale application for surface treatment of meat exists. The advantages of this method are; no use of high temperatures, no water consumption and no chemicals are applied.

For the cold plasma treatments investigated in this project, nitrogen was used, which is food grade and does not form ozone, which happens when air is used, and which can have negative health effects for the operators. Experiments were performed with skin from necks and legs, and total aerobic coounts were performed (colony forming units, CFU). Figure 27 shows that a treatment of 10 secondes effective plasma led to a CFU reduction of 3.4 log on average on leg skins and to a reduction of 1.7 log on neck skins. A treatment of 20 seconds effective plasma led to a CFU reduction of 3 log on average for both skins. The control treatments with normal nitrogen gas (not in plasma state) led to minor reduction, probably caused by dehydration.

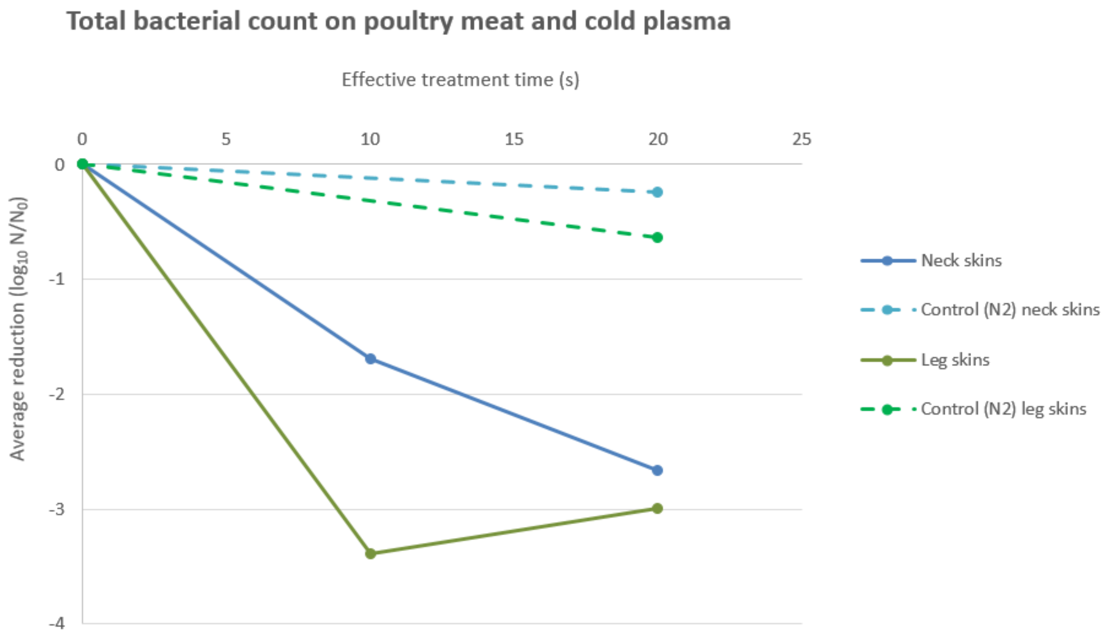

Figure 27 *Average log reduction between the different plasma treatments.*

It is plausible that CFU reduction was caused by cold plasma treatment. However, it is also possible that the reduction was caused by heat. The meat surface showed discoloration and dehydration. The cold plasma apparatus used was very "strong", leading to temperatures of possibly up to 70°C. It is therefore suggested to use this apparatus with a setting of 5 seconds effective plasma only. This setting might result in sufficient microorganism reduction and might lead to less meat damage and less thermal effects. Alternatively, another "weaker" cold plasma apparatus could be used, which is located in an ML-II laboratory and would therefore be suitable for usage with the pathogenic bacterium *Campylobacter*. Another recommendation would be to also study the reduction of not only CFU (total), but of specifically *E. Coli* and *Campylobacter*.

About the crust freezing more literature was found. During crust freezing only the crust is frozen and not the core of the broiler carcass. Literature states that the reduction of *Campylobacter* after crust freezing was about 0.5 to 1 log. The advice is to study the crust freezing line in Denmark to determine whether or not this method will be effective.

In conclusion, cold plasma and crust freezing seem the most suitable methods for reduction of *Campylobacter* in the slaughter line. For cold plasma the increase in temperature during the treatment and its effect on meat quality has to be further studied, and also the possibilities to use this technology on industrial scale.

3.3.5 Inactivation of *Campylobacter* on poultry meat by nitrogen cold atmospheric plasma (WUR)

This report describes the main results and conclusions on the research conducted with the human pathogen *Campylobacter* and its inactivation on raw poultry meat after nitrogen cold atmospheric plasma (CAP) treatment. *Campylobacter* is the causative agent of campylobacteriosis, the largest human zoonoses disease reported in the EU. Its natural habitat includes the intestinal tract of (wild) birds and other animals. During slaughtering of infected broilers, the bacteria can get in contact with the meat. Since *Campylobacter* is transmittable to humans via the meat, it is of great concern to control *Campylobacter* spp. numbers on chicken products. Moreover, EU law states that *Campylobacter* spp. numbers on broiler carcasses after chilling may not exceed 1,000 colony forming units (CFU) per gram.

After a broad literature study, which was carried out in collaboration with HAS Den Bosch, CAP and crust freezing seemed to be realistic techniques to inactivate *Campylobacter* on the surface of poultry meat (described in section 3.3.4). This report focusses on CAP. A cold plasma is an ionised gas containing active particles that have antimicrobial decontamination properties. CAP is already applied in the medical and dental field to sterilize devices and to disinfect wounds.

First, a scientific literature search was performed to check whether the resistance of *Campylobacter* spp. against CAP is already reported. It appeared that an argon or air CAP could inactivate *Campylobacter jejuni* on raw poultry meat; reductions between 1.5 and 3 log within 3 to 10 min were reported. However, argon is industrial not feasible due to the high cost price of the gas and in an air plasma, ozone might be created that is harmful for employees. Therefore, we decided to use a nitrogen CAP to investigate its feasibility to inactivate *C. jejuni* on raw poultry meat. A protocol was developed to be able to inoculate raw broiler breast meat with *C. jejuni* bacteria and to treat it with nitrogen CAP. In this case, a lab-scale CAP apparatus was used, in contrast to a "stronger" CAP apparatus described in section 3.3.4. Poultry meat samples appeared to be sticky enough to stay attached to a Petri dish during treatment. In addition, 10 µL containing 10^6 CFU *Campylobacter* bacteria per meat sample (~2x2 cm) was workable and the recovery was approximately 10^5 CFU per sample using the rubbing method; enough to perform a useful experiment. After inoculating the surface of poultry breasts with *Campylobacter*, it appeared that a lab-scale nitrogen CAP device was able to inactivate 1.5 log *Campylobacter* (illustrated in Figure 28). Although the sensorial meat quality after treatment was not acceptable due to dehydration, this study shows the potential of nitrogen CAP in inactivation of *Campylobacter* on raw broiler meat.

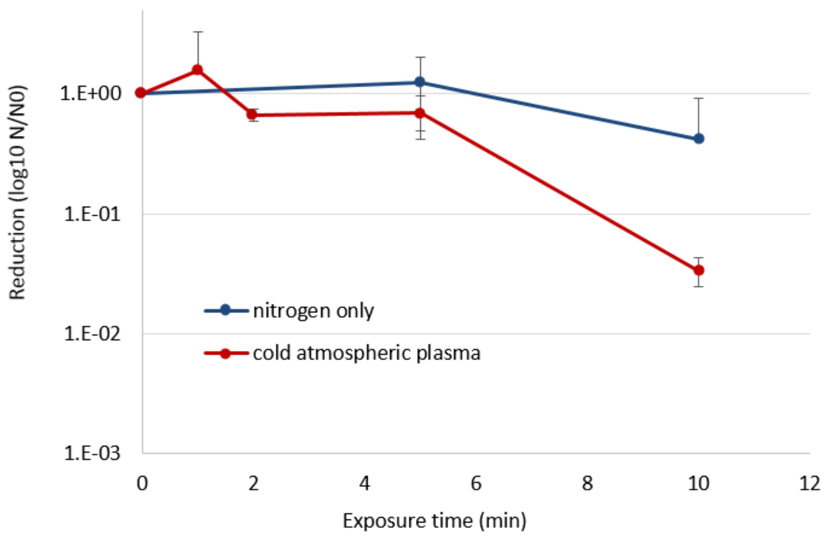

Figure 28 *Reduction of C. jejuni ATCC 33560 on broiler meat samples during CAP exposure, expressed as relative reduction in log with t=0 min set at 0 log.*

Recommendations include investigating whether the dehydration of the exposed meat is reversible. In addition, if employee's safety can be guaranteed, an air CAP might be more effective compared to a nitrogen CAP. Alternatively, in-pack treatment might be an option if *Campylobacter* numbers are sufficiently reduced. Currently at WFBR, development of pilot scale CAP equipment focusses on prevention of temperature increase during treatment and shortening of treatment times. This would lead to increased effectiveness and less dehydration of products.

3.3.6 Reduction of *Campylobacter* spp. in the poultry slaughter line (18200157)

Campylobacter spp. is a pathogenic bacterium for humans. This bacterium can be present in large numbers in the intestines of living poultry amongst others. The bacterium can contaminate poultry meat during the slaughter process. The main research question of this study was: How can *Campylobacter* spp. numbers be reduced by 1 log colony forming units/gram (CFU/g) measured on the breast skin, by means of one of more intervention methods in the poultry slaughter line of the Heijs Group, where *Escherichia coli* (*E. coli*) is used as an indicator organism?

The bacterial count of *Campylobacter* spp. on chicken meat during the slaughter process in the slaughterhouse of the Heijs group in Denmark was mapped out. During this research, three different physical intervention methods without the use of chemicals were implemented in the slaughter line and investigated. The effects of the three intervention methods on the numbers of *Campylobacter* spp. in the slaughter process were determined. The destructive sampling was performed at the slaughterhouse named Danish Finest Chicken A/S (DAFC A/S), which is located in Gedved in Denmark. The following microbiological measurements (*E. coli* and *Campylobacter* spp.) were carried out: a baseline and final measurement, and measurements for the different intervention methods.

Overall, the samples for *Campylobacter* spp. were below the detection level. Therefore, the conclusion of this research is based on the results of *E. coli* i.e. for the baseline measurement, and two physical intervention methods. For one of the physical intervention methods and final measurement, the conclusion is based on the results from both *E. coli* and *Campylobacter* spp. The results of the baseline measurement showed that in the beginning of the slaughter process (before scalding) there was a higher number of *E. coli* than at the end of the slaughter process (after chilling). Furthermore, the results showed that the processing steps of scalding and plucking together reduce the bacterial count of *E. coli* on the carcasses in the slaughter process. During the subsequent slaughter process, an equal bacterial count for *E. coli* was detected. The use of two individual physical intervention methods showed a significant difference (p=0,000) between treated and untreated carcasses. The effect of only chilling with an intervention method compared to the baseline measurement showed a significant difference (p = 0,000). One intervention method showed that there was less contamination on the carcass (p = 0,000) compared to the baseline measurement. If the three intervention methods were simultaneously applied in the line, it was observed that the bacterial count for *E. coli* and *Campylobacter* spp. at the end of the slaughter process was reduced, however not significantly (p = 0,088); the number of *E. coli* is reduced by 1.4 log and *Campylobacter* spp. is reduced by 1.3 log.

Before the intervention methods can be applied in the poultry slaughter line of the slaughterhouse DAFC A/S in Gedved and possibly in the slaughterhouse of HFP in Leek, it is important to examine the exact process conditions and the scatter. Further research must also be carried out to verify if the reduction of *E. coli* actually could be translated into the reduction of *Campylobacter* spp. during the use of the intervention methods. Furthermore, it is advisable that prior to the sampling in such research, it is known which flock is positive on *Campylobacter* spp. and therefore to sample these flocks so the chance of detection of *Campylobacter* spp. increases.

3.4 Valorisation and development of allergen-free and low in E-number products (WP4)

One of the market trends is towards more natural foods, or foods with as few additives as possible. Therefore, the use of artificial additives, E-numbers, is also of interest. Currently, one in three persons in Western Europe under the age of 18 suffer from food-related allergies, and one in every two babies is born with an allergy. In addition, the number of hospitalizations of children in Europe with a life threatening allergic reaction has shown a sevenfold increase (European Academy of Allergy and Clinical Immunology, n.d.)[9]. In total, approximately 1 million people in The Netherlands have an allergy and this number is expected to increase.

HFP also aims for having a wider portfolio of marinades to be used for poultry products. One of the products that HFP currently produces is marinated chicken wings. The production line for wings can also be used for other marinated products. With the above knowledge, it is preferred that new marinades are allergen-free, contain as less E-number additives as possible, and cover the ‘world kitchen’. Other interesting topics are reduction or replacement of additives, specifically salt and gluten components in breaded products, novel cooking technologies, and alternatives to deep frying. HFP has some marinade recipes available that can be used as a starting point for developing allergen-free and low in E-number recipes. The above topics should result in innovative production process for the

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[9] European Academy of Allergy and Clinical Immunology. (n.d.). *Food Allergy & Anaphylaxis Public Declaration*. Zurich. Retrieved from http://www.eaaci.org/attachments/FoodAllergy&AnaphylaxisPublicDeclaration.pdf

production of allergen-free and low in E-number marinades, innovative production process for the production of allergen-free and low in E-number (cooked or breaded) poultry product.
Table 7 provides an overview of the different projects performed within WP4. The projects are described in more detail in sections 3.4.1.1 to 3.4.3.4. The codes within the section titles refer to the thesis project numbers used by HAS.

Table 7 *Overview of projects executed within WP4 Valorisation and development of allergen-free and low in E-number products.*

Code	Project title	Main activities	Execution period
Allergen-free, reduced salt and E-numbers products			
7552COF1	Allergen-free and salt reduction in Hotwings	Literature study on functionalities of allergens of the recipe and possible alternatives. Assessment of wheat flour alternatives on availability, price, protein content, amylose content and starch gelatinization transition temperature and selection of most useful alternatives. First experiments with alternative flours, scoring on several product properties. Advise on salt reduction strategy with prioritisation of the reduction methods.	Oct 2017 – Feb 2016
7626LRW4	Gluten free Hotwings	Further development and optimisation of gluten free recipes to obtain quality characteristics similar to the current product. Sensory testing of samples.	Feb – June 2016
7627WUR4	Re-formulated Hotwings – Salt reduction	Further study of salt reduction strategies. Defining of reduction goals in relation to EU and UK regulations. Experiments on lowering the salt quantity and application of salt substitutes in the marinade and breading. Sensory testing of samples.	Feb – June 2016
7628LIV4	Marinades	Market, trends and literature research to define four world cuisine marinade types of interest for the UK market. Development of the allergen-free and low in salt marinades recipes, preparation concepts, packaging and consumption moments, with visualisation in infographics. Prototype marinades are evaluated on costs and sensory tests are performed.	Feb – June 2016
Valorisation of chicken leg meat			
7629LRW4	Valorisation leg meat	Survey of use of chicken meat and literature review on product properties, customers and trends to generate valorisation ideas. Concept development of one idea. Prototype development including costs estimates. Development of E-book with recipes to use the entire leg, intended for Heijs' website.	Feb – June 2016
(1)	Innova database search for chicken products	Search for recent product introductions in the Innova database on chicken thigh, formed meat products and transglutaminase meat product	May – June 2017
7706LRW4	Enzymatic treatment of leg meat	Literature research on potential to use transglutaminase (TG) to bind leg meat pieces, legal aspects and method to measure binding. Experiments with different preparation methods and additives containing TG. Reflection on possible allergen-free TG additives.	Feb – June 2017
18200140	Allergen-free restructured chicken leg meat	Use of allergen-free TG additives to bind meat pieces. Development of pre-processing protocol to enlarge access to amino acid residues needed for binding. Testing of allergen-free proteins and other additives to enhance binding. Process scale-up to batch size of 50 kilogram.	Feb – June 2018

(1) This project was performed by researchers of Wageningen Research. Results were shared with Heijs and the student groups working on leg meat.

3.4.1 Allergen-free, reduced salt and E-number products

Food trends like "allergen-free", "natural", "less salt" and "less E-numbers" are rising. These trends are partially caused by scientifically proven impact on health, and partially just trends follow by growing numbers of consumers. The demand from the consumer is getting larger and more and more people are diagnosed with food allergies or develop cardiovascular diseases. Food allergens can be harmful to the people affected. The choice of food producers to remove food allergens from their products and to replace them by other ingredients therefore makes daily nutrition easier for allergic people. Elevated salt (NaCl) consumption has been linked with hypertension and cardiovascular disease. The European Union has therefore set targets to regulate the salt content of food products. It is important to meet the customers' expectations and the salt reduction targets. Therefore Heijs Food Products aims at developing new products and would like to implement these trends in products and processes, which are applied in the poultry production chain.

3.4.1.1 Explorative study: Allergen-free and salt-reduced Hotwings (7552COF1)

This first study explores the production of allergen-free Hotwings and Hotwings with a decreased salt content. Hotwings are spicy marinated and breaded chicken wings.

<u>Allergen elimination</u>
The research on allergen-free Hotwings started with a literature study into the functionality of the components of wheat flour in the breading layer. This resulted in the following hypotheses:

- The cross-linked gluten protein provides texture in the breading layer;
- The free and cross-linked gluten protein promotes adhesion to the chicken wing;
- The starch (especially the amylose) is important for the film forming properties (increases the batter viscosity; film leads to reduction of moisture loss and production of a crispy surface)
- The amylose content of the total carbohydrate content and the starch gelatinization transition temperature influence the final texture of the breading layer; (temperature induces gelatinization of starch and therewith determines the expansion volume)

The so-called LeDa list (abbreviation stands for Levensmiddelen Databank, meaning Food Database) contains the 14 major food allergens. This list was used as a reference. The allergens in this list relevant for a study in this product area are: Gluten (Wheat, Rye, Barley, Oats, Spelt, and Kamut), Egg, Soy, Milk and Lupin. These ingredients were avoided in the alternative recipe.

Alternative allergen-free flours were studied; a pre-selection was made based on described influence on the taste, texture or colour compared to wheat flour. The remaining alternative flours were studied on their availability, price, protein content, amylose content and starch gelatinization transition temperature. Based on the desk research, the remaining alternatives that could have potential to substitute wheat flour in the allergen-free recipe are: corn flour, sorghum flour, rice flour, tapioca flour, chickpeas flour and buckwheat flour.

A first experimental trial was set up, to evaluate whether these flours have potential for the development of allergen-free Hotwings (tapioca flour not included yet). The Hotwings were analysed on: visual appearance (see Figure 29), taste, crunchiness and breading pick up. Chickpea flour shows a lot of potential, the breading pick up as well as the visual appearance, taste and crunchiness were comparable to the control with wheat flour. Buckwheat flour shows potential in the amount of breading pick up and taste. The other alternative flours show less potential to replace wheat flour.

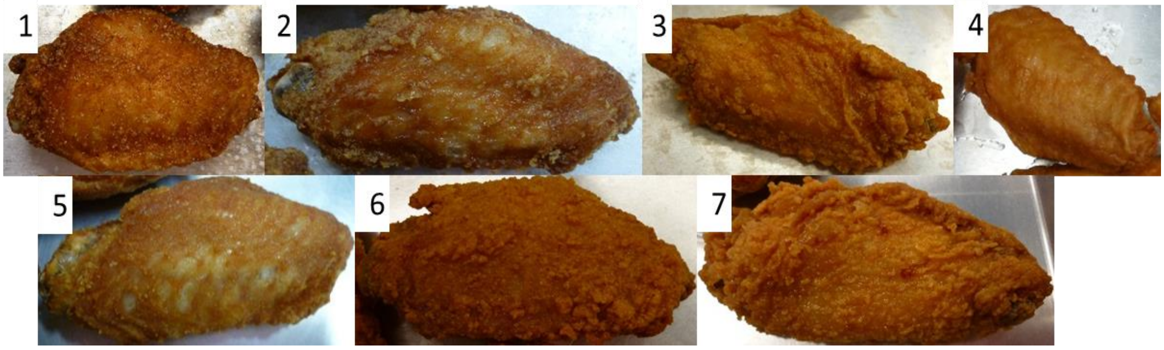

Figure 29 *Hotwings produced with the different flours: 1. Sorghum flour, 2. Oat flour, 3. Buckwheat flour, 4. Rice flour, 5. Corn flour, 6. Chickpea flour and 7. Control wheat flour.*

Based on the literature study and the experimental trial, the recommendations for further research on this topic are:

1. Extensive research into the composition of the selected alternative flours and properties of the Hotwing made with the alternative flours to verify the hypotheses;
2. Study the potential of tapioca flour;
3. Study the potential of chickpea flour into more detail;
4. Investigate the potential for changing the breading production procedure, to increase the breading pick up;
5. Study the potential of hydrocolloids (in order to improve adhesion between Hotwing and breading).

Salt reduction

The current Hotwing recipe contains about 1.8% salt, divided over the marinade and the breading; a salt reduction of 25% is desired. The reduction of the salt content should not result in a significant different product compared to the control product. A literature study was performed and showed that salt in Hotwings can contribute to the following functionalities:

- Taste
- Preservation
- Activation of meat proteins, resulting in migration of marinade into the meat, making the meat more tender (salt in the marinade)
- Improving texture by a strong network caused by interaction of positively charged Na^+ ions with negatively charged proteins (salt in the breading)

The literature study also revealed that there are several possibilities for salt reduction. Experimental trials should prove which strategy is best to use. It is recommended to start with decreasing the salt content in the marinade phase, since the impact on functionality and taste is estimated to be smaller in the marinade than in the breading. It is recommended to perform the experimental trials in the following order:

1. In the marinade: Salt reduction without substitution;
2. In the marinade: Use of smaller salt crystals;
3. In the marinade: Use of natural taste enhancers;
4. In the breading: it should be investigated salt reduction without substitution in the breading influences the texture (and taste) of the breading layer;
5. Use of salt substitutes if the other strategies do not result in the desired decrease in salt content.

When using sodium replacers or flavour enhancers in the alternative recipe, the European food law concerning the ingredient declaration should be taken into account.

3.4.1.2 Gluten-free Hotwings (7626LRW4)

During this project, research was performed to develop allergen- (gluten) and E-number free Hotwings and to develop the most potential gluten-free flour mixtures. Besides, the most cost-effective solution was researched.

Information found in the literature study showed that the term "gluten-free" may be used when the amount of gluten is ≤ 20 ppm (mg/kg). The most promising gluten-free flour types are: chickpea flour, tapioca flour, buckwheat flour and rice flour. These flour types were selected for testing in the breading layer of the Hotwings, which is applied to the Hotwing before frying. Besides, it was researched which processing steps (breading, hydration, frying) influence the structure of the crust layer of the Hotwing.

In the beginning of the product development phase, the product was evaluated on which product criteria it should meet. These criteria were: allergen- (gluten) and E-number free, having the same sensorial properties as the gluten containing Hotwing (reference), cost-effective solution (highest price-quality ratio), and it was preferred to only use the necessary ingredients in the recipe.

During the development phase, gluten-free flour types and ingredients were used to develop the product with attention to the criteria as stated above. Chickpea-, buckwheat-, tapioca- and rice flour were tested. A combination of chickpea-, rice- and tapioca flour resulted in the most potential gluten-free breading mixture with similar or better properties compared to the reference. Only the colour was different compared to the reference, so dextrin was added.

With this recipe, sensorial research with a trained panel and consumer panel was performed. The trained panel (n=4) from Heijs Food Products evaluated the attributes and found no significant difference. The results obtained from the research with the consumer panel (n=100) were: the attributes expansion, transparency, crispiness and graininess resulted in a significant difference between the developed Hotwing (gluten-free) and the reference (gluten containing) (see Figure 30 and Figure 31). It was assumed that the attributes expansion, transparency and crispiness were perceived positively by consumers. Therefore, these attributes need no optimisation. It was assumed that graininess was perceived as negative by consumers. So, only the attribute graininess needed optimisation and should be decreased.

Figure 30 *Photographs of reference Hotwings (left) and gluten-free Hotwings (right).*

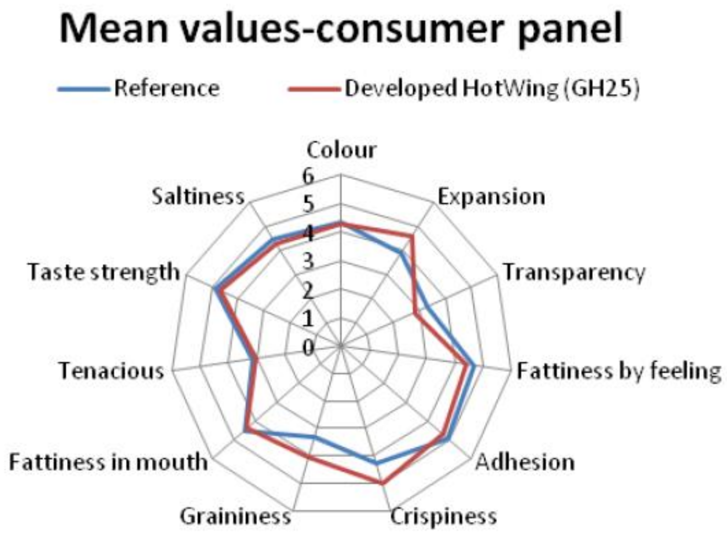

Figure 31 *Spider plot of sensory results of consumer test (n=100) of gluten-free Hotwings (red), compared to reference Hotwings (blue).*

During the optimisation phase, it was attempted to optimise the graininess and research the most cost-effective solution. The addition of β-glucan did not improve the graininess. The most cost-effective solution was studied by; the addition of more baking powder compared to dextrin and the addition of only dextrose compared to dextrin. Baking powder with dextrose is a basic ingredient present in the breading mixture and dextrin is an added ingredient, so less cost-effective. More baking powder resulted in more expansion and crispiness, but decreased the colour and graininess. Only the addition of dextrose resulted in the same properties compared to the reference, but resulted in a lower quality by the addition of β-glucan.
Also, different breading- and preparation (heating) methods were researched on the reference and the most potential developed gluten-free recipe. The breading methods which were tested, were the KFC method and a shorter breading method compared to the standard method. Only the KFC method resulted in better properties, in particular more expansion of the breading layer and a higher breading pick up. The preparation (heating) methods which were tested, were air-frying, pre-frying + oven and only oven heating. The air-frying and oven heating methods resulted in lower quality Hotwings. The pre-frying + oven method resulted in a crispier layer compared to the standard method and could possibly be used to replace the normal frying step.

The recipe costs of the most potential gluten-free recipe, the most potential gluten-free recipe without dextrin and the recipes with the addition of extra baking powder were calculated to research which recipe was most cost-effective. The price of the most potential developed gluten-free recipe was in all cases lower (€ 0.92 + X) compared to the most potential gluten-free recipe without dextrin (€ 0.96 + X) and the recipes with addition of extra baking powder (2.55%, € 0.95 +1.61* X) (3.51%, € 0.94 +2.22* X). The X stands for the price of baking powder, but this was not available.

The conclusion of this project was; an allergen- (gluten) and E-number free breading mixture was developed which contains 1.58% baking powder, 8.12% salt and 90.3% gluten-free ingredients (mostly gluten-free flours). The flour part consists of 60.40% chickpea flour, 15.10% rice flour, 7.55% tapioca flour and 7.25% dextrin (=90.30% total). An annotation to the purpose of the project is that the gluten content is not measured by an external company. The developed Hotwing has the same properties and quality as the gluten containing Hotwing with exception of the attribute graininess. The developed recipe was also the most cost-effective with an optimal price-quality ratio.

The most important recommendations for further research are:

- Execute sensorial research by consumers to find out if the graininess of the developed gluten-free recipe is unpleasant;
- Analyse the gluten content of the developed recipe by an external company, to determine if the requirement of ≤ 20 ppm gluten is achieved.

3.4.1.3 Salt-reduced Hotwings (7627WUR4)

Elevated dietary salt (sodium chloride (NaCl)) consumption has been associated with hypertension and cardiovascular disease. Therefore the EU and the UK government have passed legislations and set targets to regulate salt content of food products in the coming years. The aims of the present study were to reduce the salt content of spicy marinated and breaded chicken wings ('Hotwings') by 25% and to investigate and realize the maximal permitted salt content in Hotwings according to UK legislation without reduction of salt perception by consumers.

The following five strategies for salt reduction have been investigated in a literature study for their feasibility to be applied in Hotwings:

1. Gradual reduction of the salt content; by this strategy small quantities of salt can be omitted from the product until the consumers noticeably perceive that salt has been reduced. According to literature, it should be possible in most food products to reduce 10-15% of the salt content without perceivable sensory differences.
2. The use of salt substitutes; salt substitutes are salts (other than NaCl) that have not been associated with the development of hypertension and cardiovascular disease, for example potassium chloride (KCl), magnesium chloride ($MgCl_2$) and calcium chloride ($CaCl_2$). Their use is limited due to the fact that they can impart off-tastes at elevated concentrations.
3. The use of flavour enhancers; these compounds enhance the savoury flavour of meat which can partially counterbalance a reduced salt content.
4. The use of inhomogeneous spatial distribution of salt in the product; according to literature, the sensory contrast allows salt reduction not noticeable for consumers. It is however difficult to apply this strategy to Hotwings.
5. Increase the solubility of salt; by variation of salt crystal shape or size, the solubility of salt and therewith the perception of saltiness might be increased.

The described strategies can be combined if one strategy is not sufficiently efficient. Furthermore, the legislations which are applicable to salt, additives, and salt replacements, as well as the influences of the process variables on the Hotwings are described in this literature research. The conclusion of this literature research was that the best suitable starting point for salt reduction in Hotwings is the gradual reduction of salt content. This strategy has the least impact on the recipe and the production of the Hotwings and it should be possible to reduce salt content in Hotwings gradually without noticeable sensory differences until a certain salt concentration.

In the experimental phase, the KFC Hotwings were produced to serve as reference product. The reference Hotwings contain 2.43% salt, of which 0.21% in the chicken meat, 1.05% in the marinade and 1.17% in the breading layer. Salt reduction in the marinade was selected as starting point due to the spiciness of the marinade, which was suspected to compensate for salt perception partly. In this way, it was achievable to reduce the salt content of the Hotwings by 20.34% without causing sensory differences. The remaining 4.66% salt reduction to meet the target of 25% salt reduction has been achieved by reduction of salt in the breading. A sensory panel (n=31) evaluated the parameters saltiness, tenderness, juiciness, and colour, and perceived the 25% salt reduced Hotwings as comparable to the reference product (see Figure 32).

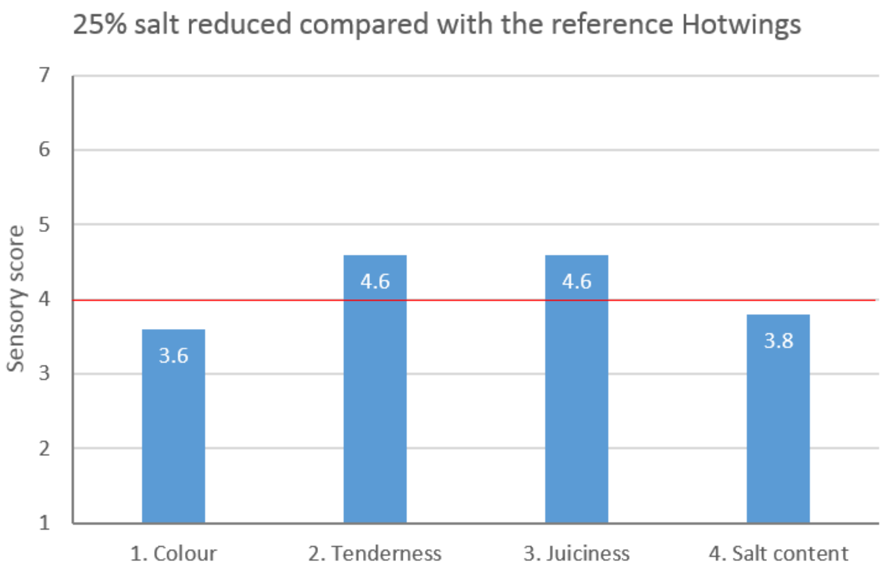

Figure 32 *Results of sensory tests with 25% salt reduced Hotwings (n = 31)**
** 4 = comparable to the reference product;*
< 4 = less than the reference product;
> 4 = more than the reference product.

The maximal allowed salt content per portion according to UK targets depends on the type of product and its nutritional value. It was investigated that Hotwings fall into category 3 'Battered or breaded chicken portions and pieces'[10]. The portion size for Hotwings was established to be six pieces of 1st part Hotwings. (See Figure 33 for a photograph of 1st part chicken wings, in comparison with 2nd part chicken wings.)

Figure 33 *1st (left) and 2nd (right) PART chicken wings.*

[10] Table 2 Eating out of home maximum per serving salt targets in Salt Reduction Targets for 2017, Public Health England, 2017, London.

Including investigated oil absorption during frying (6.15%) and moisture loss (28.1%), this portion size has a caloric value of 390 kcal and is therewith allowed to contain a maximum of 2 grams of salt per six 1st part Hotwings. One portion of 25% salt reduced Hotwings contains 2.88 gram salt and is therewith not sufficiently salt reduced to comply with the UK legislation.

It was therefore investigated whether salt replacers could be used in order to meet the target. OneGrain A50 is a homogeneous salt grain, with 50% lowered sodium content, combined with chloride, potassium and flavours. OneGrain A50 was used as full replacer of dietary salt in the marinade and as partial replacer of dietary salt in the breading. In this way, it was possible to achieve a total salt content of 2 gram NaCl per six 1st part Hotwings. A sensory panel (n=31) evaluated the parameters saltiness, tenderness, juiciness, and colour, and perceived the salt replaced Hotwings as comparable to the reference product (see Figure 34).

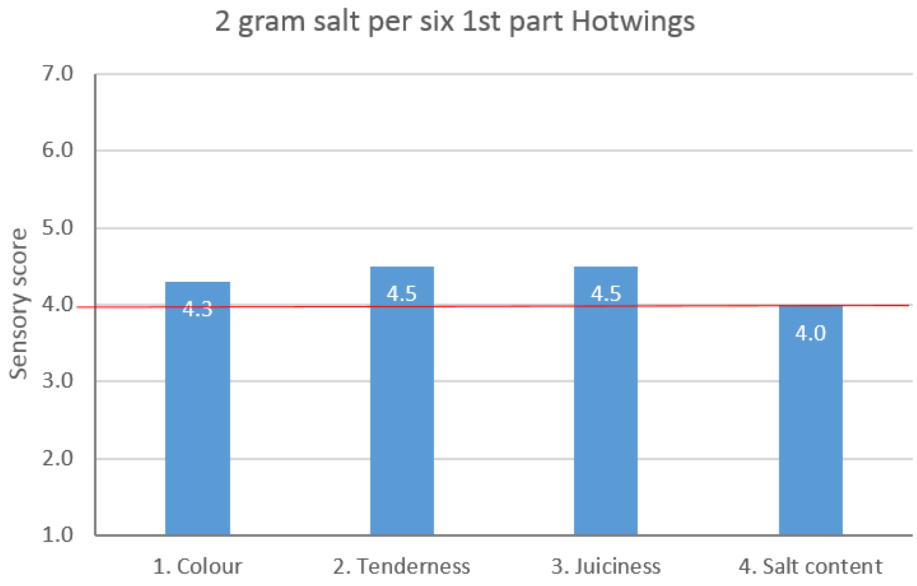

Figure 34 *Results of sensory tests with 2 grams of salt per six 1st part Hotwings (n = 31)**
** 4 = comparable to the reference product;*
< 4 = less than the reference product;
> 4 = more than the reference product.

It is recommended to evaluate both reformulated products by a larger sensory consumer panel (n=60) in order to validate the results. As the reformulated Hotwings were assessed based on sensory aspects only, it is recommended to examine the shelf-life and the food safety of the reformulated products. Furthermore, the cost aspects regarding OneGrain A50 and possible alternatives should be considered.

3.4.2 Allergen-free and E-number free marinades and seasoning mixes (7628LIV4)

HFP currently uses four different marinades. HFP wished to extend the marinade portfolio with new marinades that cover the world kitchen. Allergen-free marinades are preferred over allergen containing marinades. The functionality of alternative ingredients should be comparable to existing marinades. The marinades should be applicable to several product applications.

The aim is to develop dry seasoning mixes that meet the British salt standard and to focus on UK clients. To illustrate the added value of marinades also different ways of chicken preparations were developed combined with the marinades.

The following phases of product development have been performed. (1) Exploration Phase, with market research (desk research) for customers of HFP products, namely Sainsbury's, Tesco, Morrisons, Marks & Spencer (M & S), and Kentucky Fried Chicken (KFC); a visit to an M & S supermarket; trend research (desk research into trends in ingredients, world marinades, new restaurants and general food trends); literature research on the needs of the UK consumer; definition of target consumer segments per supermarket (visualized in Figure 35). (2) Idea Phase, with brainstorming sessions to define draft ideas for marinades and preparations; selection of preparations based on various criteria (e.g. the needs of the user, the value and feasibility for HFP). (3) Concept Phase, in which three different preparations have been developed for four marinades for the different target consumer segments. (4) Prototype Phase, with the development of the marinades (dry seasoning mixes in combination with a basic recipe consisting of functional ingredients), and testing of different ways of chicken preparations.

To determine the marinade pickup, different ratios of the starch components (functional ingredients) were tested. In this phase, a sensory consumer test of the marinades (n = 100) was performed with marinated tumbled chicken breast pieces: juiciness, flavour intensity, spiciness and salt intensity were rated on a 7-point scale, and ratings were compared with the desired assessment of these attributes. Furthermore, cost estimates were done and packaging designs were made.

In the Exploration Phase four different target segments of UK consumers have been defined: for Sainsbury's, especially young, educated consumers without children; for Tesco, families with young children, with different incomes; for Morrisons, hardworking consumers who do not have much to spend; for M & S, successful consumer with high needs. At KFC mostly consumers want to quickly eat cheap fried chicken. Most often sold in British supermarkets are pre-cut raw chicken parts, of which 55% is breast fillet. Meat and marinade products, and emerging world marinades were mapped: classic flavours such as BBQ, Piri and Chile exist, and also lesser known flavours such as lemon herb and Jamaican Jerk. Supermarkets largely overlap in their range of flavours that they offer. Allergen-free marinades are only offered to a limited extent. Increasingly popular world cuisines are e.g. Mexican, Caribbean, Indian, Peruvian, Greek, Jewish and British. For every world cuisine a mood board was created. Observed trends are e.g. craft beer, home chef, BBQ and grilling, back to the roots, fusion and sport & health.

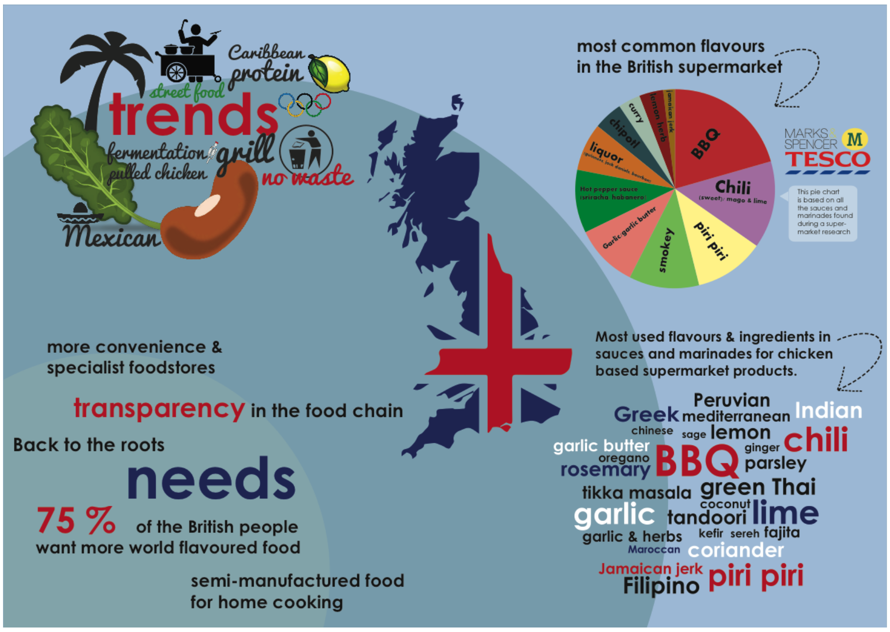

Figure 35 *Infographic exploration phase: needs of British consumers, British market, trends.*

Based on the findings in the Exploration Phase on supermarkets and trends, in the Idea Phase four world cuisines were selected. Marinades that belong to these world cuisines were developed, namely Jamaican Jerk, Mexican Mole, Pure Peru and Indian Indira. Based on the market research, three ways of chicken preparation concepts were selected, namely beer can chicken, chicken pie and pulled chicken, all to be cooked at home by the consumer. These concepts were completed by designing a suitable packing, which fits with the targeted supermarket.

In the Concept Phase, for the three preparation concepts, draft statements were written and also infographics (Figure 36) were created: the beer can chicken fits into the British Sunday roast tradition and reflects the BQQ trend, it is suitable for all target segments except for Morrisons, it is easy to prepare at home and all marinades are suitable, but most appropriate is Jamaican Jerk (in the form of a dry rub); chicken pie is a variation of the popular and classic pie and fits with the fusion trend, it is suitable for all four target groups, it is easy to prepare at home, HFP can offer marinated chicken cubes for pies in each four of the marinade flavours; pulled chicken is a new dish, for many consumers it will be the first occasion to consume chicken thigh meat, it is in line with the home chef and fusion trend, it is suitable for all target segments except Morrisons, it is easy to prepare at home and all marinades are suitable.

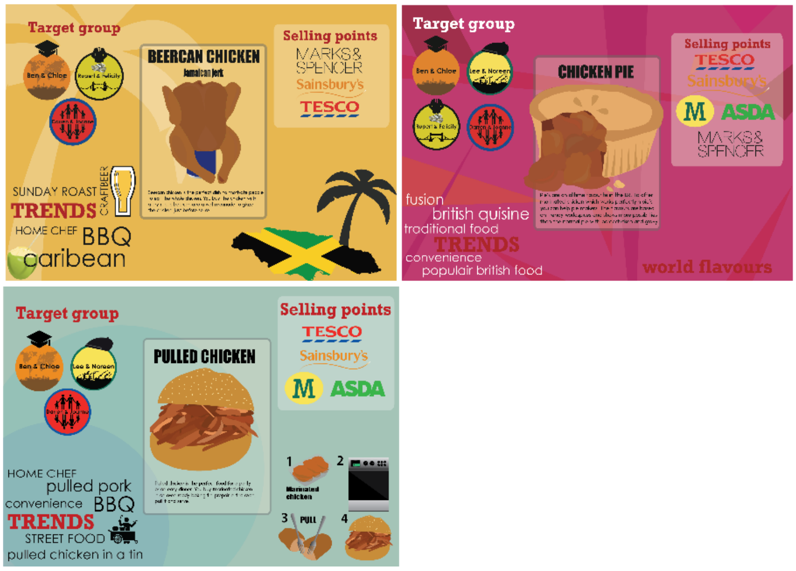

Figure 36 *Infographics of beer can chicken, chicken pie and pulled chicken.*

In the Prototype Phase, the four marinades were made composed of dry ingredients. To obtain the complete marinade, a basic recipe consisting of functional ingredients, namely, water, sodium bicarbonate (E500), salt, Nhance 57 (modified potato starch), and rice starch was added to the dry mix. This basic recipe ensures that the marinade is well absorbed by the chicken meat and results in a more tender meat. Marinade pick-up at a ratio Nhance 59:rice starch of 80:20 was 10 to 11% after tumbling during 45 minutes (desired pick-up is between 10 and 15%). The marinades are all allergen-free, contain only one ingredient with an E-number and do meet the British salt standard (0.75% salt in marinated meat). For each marinade, cost price and nutritional values were calculated. Shelf life of the marinades is extended, because only dry ingredients were used. For the three preparation concepts, also the steps were described in order to complete the concepts into a consumer product. These steps cannot be done by HFP, but need to be performed by the customer itself, namely steps such as wrapping the whole chicken with a can of beer, adding a package of sauce and dough to the marinated chicken cubes for a pie, and adding a bag of sauce for the pulled chicken preparation.

The sensory test showed that flavour intensity and spicy taste of the Jamaican Jerk marinade is close to the desired ratings, and that salt intensity and the juiciness could be higher. Mexican Mole and Pure Peru marinades scored rather low on all attributes, especially the Pure Peru marinade on juiciness and flavour intensity. All attributes of the Indian Indira marinade were scored close to desired values, only the taste could be somewhat more intense. The low salt content caused a fairly low salt intensity of marinades Pure Peru, Mexican Mole and Jamaican Jerk. On the basis of the results of the sensory test suggestions have been given for the improvement of the formulations. The cost price of Jamaica Jerk Marinade is € 0.84/100g, of Mexican Mole € 2.41/100g, of Pure Peru € 2.92/100g and of Indian Indira € 1.68/100g.

The packaging for the beer can chicken contains all the ingredients (including the can of beer). The chicken is visible through a see-through window. The preparation method is illustrated with pictures. The design of the beer can chicken is focused on the luxury segment and thus fits best at M & S and Sainsbury's. The chicken pie is packaged in an amount suitable for one person, with a luxurious appearance aimed at M & S. The pulled chicken comes in a box with a see-through window in the form of a chicken. It includes marinated thigh fillets and an added sauce sachet. It is in line with the target segments of Tesco and Morrisons. For all packaging examples, see Figure 37 and Figure 38.

Figure 37 *Packaging of Beercan chicken, front (left) and back (right).*

Figure 38 *Packaging of Chicken Pie (left) and Pulled Chicken (right).*

The four developed concepts for world marinades, combined with the three preparation concepts for the various target groups, seem promising for HFP for its market development targeting UK customers. Optimizing the flavour (possibly accompanied by a cost reduction) and increasing the salt perception of the marinades still needs some attention, preferably in consultation with the target supermarket, where after advisable is to perform a sensory test with the target consumer segment.

3.4.3 Valorisation of chicken leg meat

Heijs Food Products (HFP) is continuously investing in quality and expansion of their product portfolio. The next step is valorisation of chicken leg meat. Due to a large demand of chicken breast meat, there is an excess of chicken leg meat. Leg meat represents 50% of the chicken carcass and is sold in Europe for a low price, as nowadays it is not - yet - very popular among Western consumers. In Europe, chicken leg meat is consumed during the higher temperature months, while the consumption of chicken leg meat is relatively lower in the colder seasons. During these seasons, the chicken leg products are exported to foreign countries for low prices. Heijs Food Products wants to add extra value to their chicken leg products and therefore wants to be informed about the possibilities to differently marinate or restructure chicken leg meat with the enzyme transglutaminase (TG).

3.4.3.1 Valorisation leg meat (7629LRW4)

The aim of the reported research is to develop a leg meat marinated concept, that is allergen-free and E-number free, taking into account new trends.

The different phases of concept development have been completed. (1) Exploration phase, consisting of a literature review on leg meat (sales, product features, chicken breeds, preparation techniques, potential customers, trends), and a survey on the use of chicken meat among Dutch consumers (n = 253). (2) Idea phase, which includes three concept ideas written in draft statements. (3) Concept phase, in which one concept idea ("No Sticky Fingers") has been developed for three target segments, including packaging and preparation. Also, the concept was assessed in a survey (n = 20), and the idea for an inspiration website was developed. (4) Prototype Phase, with a cost estimation for the "No Sticky Fingers" concept, creation of packages, trying out various methods of preparation, and the development of an E-book. The development of E-number free and allergen-free marinades was done in another research project (7628LIV4, *Marinades from around the world*). Herein, four marinades in combination with the three preparation concepts for different target groups of consumers of British supermarkets were developed.

The literature review in the Exploration phase showed, that mostly chicken breast filet is chosen by Western consumers, so there is a large surplus of chicken pieces with bones. Cutting out leg meat from the bone is costly. The leg meat is somewhat darker in colour, because it contains more myoglobin as compared to breast meat. The colour is also influenced by breed and growth rate of the chicken. Dark chicken meat evokes associations with a lower health. The survey among Dutch consumers also showed that chicken breast meat is purchased in particular. Reasons are because it is lean and easy to prepare in a desired flavour. Not everyone likes to gnaw, because of the dirty fingers. Consumers like to try out new recipes with chicken. HFP indicated three potential customers to focus on during the concept development, namely Marks & Spencer (M & S), Kentucky Fried Chicken (KFC) and hospital kitchens, that all seek for marinated and easy to prepare products. Major trends that were observed were, that gnawing is allowed again, also going back to the old way of cooking (authenticity), and health.

In the Concept phase, about 25 ideas were generated, of which three were chosen in consultation with HFP: "No Sticky Fingers" (gnawing without dirty fingers; a drumstick without skin with completely clean bone), 'Kluiv &' (for the gnaw lover, an incised marinated chicken leg) and "Skinny Nuggets" (thigh pieces of which the bread-crumb layer contains crispy chicken skin). The concept "No Sticky Fingers" is elaborated on further. Also, it was decided to develop a bilingual inspiration website for HFP customers in English and Dutch. This website focuses on the "No Sticky Fingers" concept, including some concepts for thigh meat, the part of the leg that remains in the production of "No Sticky Fingers". This e-book can be accessed using the following link: https://issuu.com/femkevdklundert/docs/e-book_heijs_food_products.

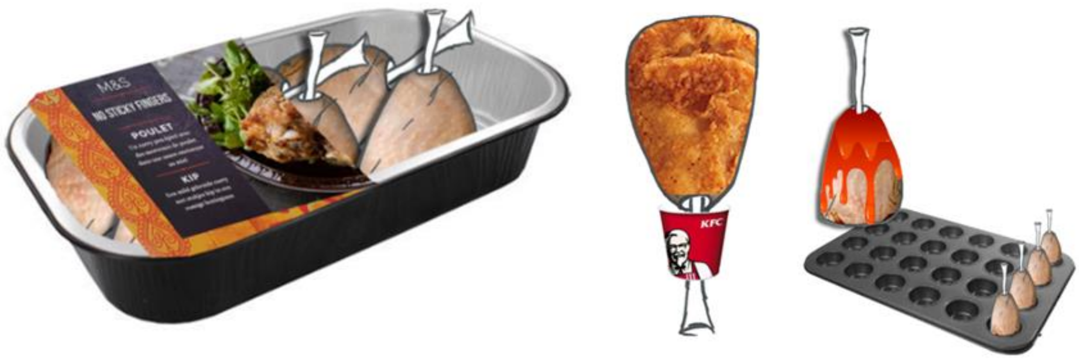

Figure 39 *"No Sticky Fingers" product suggestion for Marks & Spencer, KFC, and hospital kitchens (from left to right).*

In the Concept phase, the "No Sticky Fingers" concept is written in draft statements focussing on the three target segment consumers, which were representative for the customers. Various packaging designs emerged in this phase: for M & S a package with a sleeve; for KFC one with a cover for the bone of the drumstick that enhances keeping the fingers clean during consumption; for kitchens, drumsticks marinated in a vacuum bag (suggestions depicted in Figure 39). The marinades from the project *Marinades from around the world* (Mexican Mole, Peru, Jamaican Jerk, and India) may be used. The survey among a group of 20 consumers showed that "No Sticky Fingers" seems to have great potential: it is seen as professional, convenient, and manly, and easy to use (a party and for barbecuing were mentioned). Respondents would be willing to pay more than for normal drumsticks. Also, the conditions for the inspiration website were investigated in this phase (e.g. the design, namely an E-book; content, namely a concept book; suggestions for quality assurance were given).

The Prototype phase showed, that in addition to the costs for the marinade, in particular the cleaning of the bone will be of influence on the cost price, because this takes time and manpower. Obviously, there are also costs because of specific needs of the customer for packaging. After trying out various ways to prepare the chicken fingers (pan, oven, sous vide, frying, etc.), it was concluded that preparation in the oven at a low temperature (150°C, 40 minutes) was best (tender and brown meat). Furthermore, prototypes of packages were made and also the E-book with six concepts for thigh chicken meat was made. The "No Sticky Fingers" concept seems a promising concept for HFP, which reflects the current trend that gnawing of bones is allowed again. The E-book will inspire customers to use (the residual) thigh chicken meat.

Follow-up actions to this research may include: tests with a minimum of 60 consumers, preferably from the target segments, including assessment of sensory aspects. This consumer testing should be preceded by an optimization of the marinades, which should be done in consultation with the customers (e.g. inclusion of less expensive ingredients, no baking soda, lower costs). Furthermore, a production trial of the "No Sticky Finger" concept at HFP seems advisable, optionally making use of a machine for trimming the drumsticks. Also, potential customers of the "No Sticky Fingers" concept could be approached, preferable also using the E-book.

3.4.3.2 Innova database search for chicken products (WUR)

In order to complete the picture of existing chicken thigh product for other countries and to get an idea about reformed chicken products, an Innova database search has been performed for i) chicken thigh, ii) formed meat products, and iii) transglutaminase meat products. Innova database is a database that shows all food and beverage product launches of the last years in more than 70 countries. However, it does not give information on whether these products are still on the market or not. Also established products, which are on the market for a long time already, are not listed in this database. The search has been performed in June 2017.

i) Regarding chicken thigh products, the Innova database search revealed that within Europe and America, 457 chicken thigh products have been found, which have been launched between 2002 and 2017. These products consist of a wide variety of traditional chicken thigh products, highly processed chicken thigh products and products in marinade and sauce and in convenience meals. However, compared to chicken breast product, the number of products is relatively limited. Examples of the chicken thigh products are shown in Figure 40.

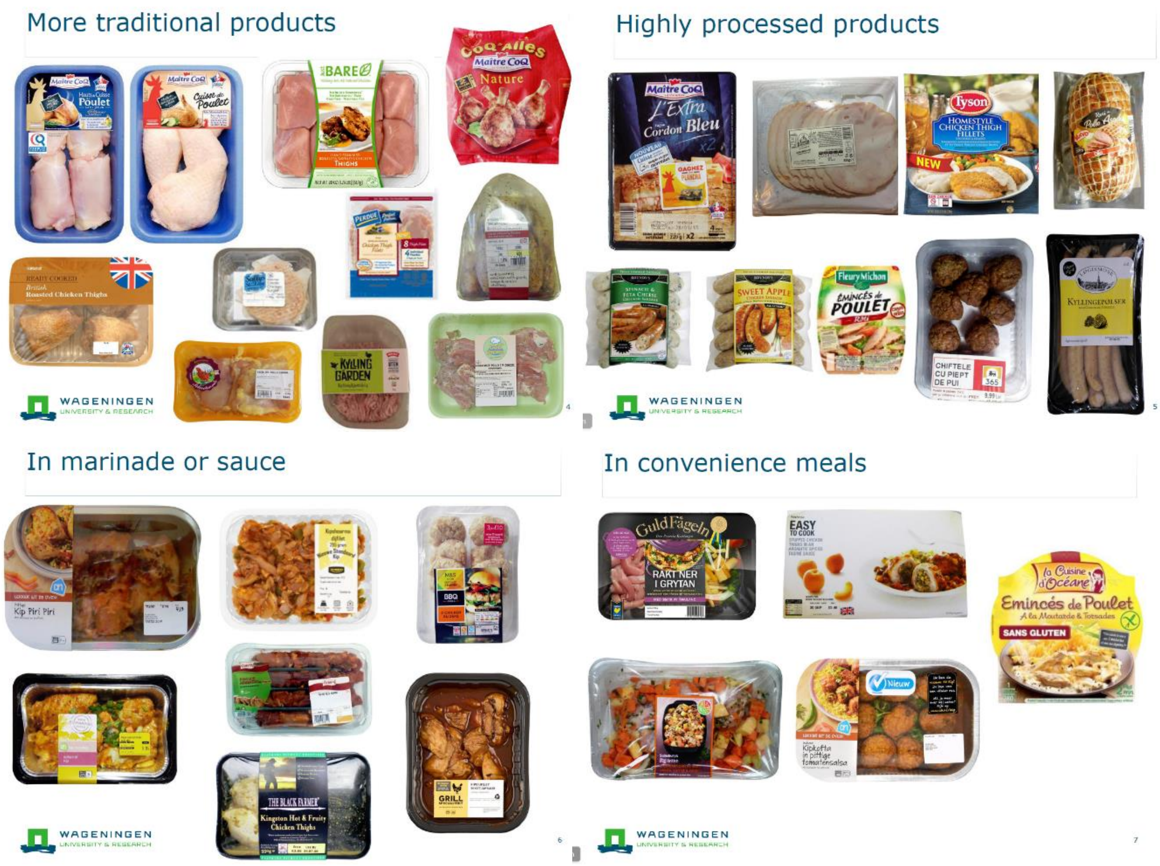

Figure 40 *Four categories of chicken thigh products identified.*

ii) For formed meat products (Figure 41), a very small number of products has been identified. Four formed chicken meat products have been identified, from which three consisted of chicken breast meat and one was not specified further regarding the part of the chicken used. These four chicken products are all breaded. Furthermore, nine formed pork meat products were found (all in the UK), which all concern ham-like products. The results suggest that the small number could also be caused by the fact that "formed meat" is a quite recent way of labelling. None of the "formed meat" products contained "transglutaminase" on its label. This could be due to forming by other methods (seven of the nine products contained phosphates), or due to no compulsory labelling when transglutaminase is used as processing aid.

Figure 41 *Formed-meat products with chicken and pork.*

iii) Regarding transglutaminase meat products, the results were also very limited. There are basically five sliced meat products, which contain transglutaminase on their labels. These are:

- Chicken meat slices (Company Bachoco, Mexico 2006, retailer price 3.71 €/kg, ingredients of interest: chicken meat, salt, sodium caseinate, transglutaminase)
- Turkey breast (Company Cencosud, Chile 2015, retailer price 19.54 €/kg, ingredients of interest: turkey breast, polyphosphates, salt, transglutaminase)
- Pheasant medallions (Company O Cool, Belgium 2013, retailer price 14.95 €/kg, ingredients of interest: pheasant, transglutaminase, sodium caseinate)
- Round chicken breast medallions (Company Bachoco, Mexico 2015, retailer price 5.63 €/kg, ingredients of interest: chicken breast fillet, salt, polyphosphates, transglutaminase, egg)
- Pork sizzle steaks (Company Woolworths, Australia 2010, retailer price 10.89 €/kg, ingredients of interest: formed pork meat, milk proteins, transglutaminase)

The first four products are depicted in Figure 42.

Figure 42 *Transglutaminase and chicken or other poultry / bird meat.*

In conclusion, Innova Database did not identify a product launch of a chicken thigh product labelled as formed meat or labelled containing transglutaminase. There are however two chicken breast products, one turkey breast product and one pheasant meat product that are similar to what is envisioned for project restructured chicken leg meat (see section 3.4.3.3).

3.4.3.3 Enzymatic treatment of leg meat (7706LRW4)

First a literature research was performed on the application and the binding properties of transglutaminase (TG), the legal aspects and to set up an objective measuring method for the quality of the binding. Afterwards, different experiments were executed regarding the use of TG to bind chicken leg meat. Research was conducted on the primary factors which could influence the binding, such as type of tissue (muscle, connective tissue, fat), the size of the meat parts, the influence of pre-processing of chicken leg meat, the dosage and type of TG. To be able to measure the binding of TG in an objective way, different measuring protocols were developed.

Microbially produced TG is used in many different applications and is provided by companies such as Ajinomoto (brand name Activa). The most used variant is Activa EB (alias RM). The ingredients of this product are sodium caseinate, maltodextrin and TG. TG is capable of crosslinking proteins via the amino acid residues of lysine and glutamine. Caseinate works synergistically with TG but has to be declared as an allergen. During this project, first Activa EB (with allergen) was researched but the final goal is an allergen-free, reconstructed product. According to the law, the caseinate in the Activa has to be labelled. Reformed meat made of pieces of leg meat has to be declared as “formed meat”. This is not the case if the product has been produced from one large piece of leg meat.

During this research project, different samples were produced (see process in Figure 43). The pre-treatment of chicken leg meat was performed with different methods, such as tenderizing, tumbling and rolling. In order to process one kg of restructured chicken leg meat, 10 grams of Activa and 40 gram water were used and mixed together. The chicken leg meat was mixed with the slurry and afterwards vacuum sealed in a casing. Lastly the casing with meat was stored for 20 hours at 4°C. Then the restructured chicken leg meat samples were frozen slightly and sliced in order to objectively measure the binding properties of TG. The binding strength was evaluated by rheological measurements on a tensile meter and by usage of a slicing machine.

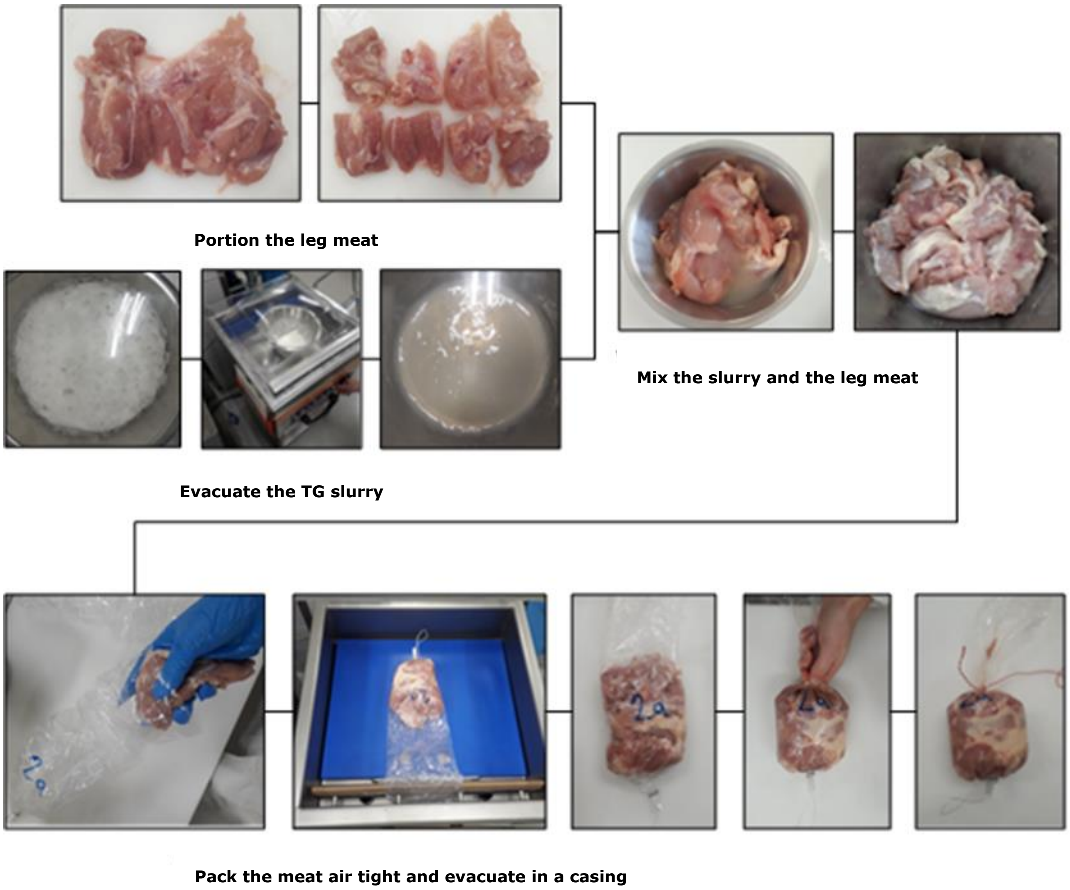

Figure 43 *Production process of enzymatically bound chicken leg meat.*

The practical research showed that the use of different application methods (slurry or powder) of TG and different chicken leg meat structures such as connective tissue, meat, and fat have a huge impact on the binding. Bare meat in combination with TG in slurry form provided the best binding properties to restructure chicken leg meat. However, the removal of connective tissue from the meat is a very labour intense processing step. Therefore, it was chosen to perform the research with meat from which connective tissue was not removed. No differences between whole meat parts and small meat parts regarding binding were found. Kitchen tests showed that the minimum dose of Activa EB should be 1% to provide an acceptable binding. Tensile tests with meat pieces of different Activa dosages revealed that a stable piece of meat (of 1 cm thickness) is achieved when the force to tear it apart is ≥1.53 N. Cutting tests revealed that the reformed meat is stable when it can be cut in pieces of 0.5 cm thickness. The pre-treatments of chicken leg meat improved the binding properties of TG, of which the rolling method shows the best improvement. Tumbling meat pieces with dry marinade or with wet marinade led to adequate products - visually as well as objectively measured The use of the allergen-free Activa TI (alias WM), with or without the addition of chicken protein (chicken collagen), resulted in no or inadequate binding of pieces of chicken leg meat. Based on the prices for chicken leg meat and the Activa EB an estimation on the raw material cost has been made. The cost price addition of the Activa EB is € 0.90 per kg. On top of this, additional costs of extra processing steps during the production of restructured chicken leg meat will have to be added.

There are multiple recommendations set for further research of restructuring chicken leg meat. Research should be done to optimize the processing time of TG. Furthermore, research needs to be conducted regarding the influence of salt concentration, pH and water activity on the binding properties of TG. The current TG product contains the allergen sodium caseinate. Therefore, further research needs to be conducted regarding the use of allergen-free Activa TI and possible alternative proteins (preferably originating from chicken) to restructure chicken leg meat. It is recommended to investigate the dosage of Activa TI and the water content of the slurry to optimize binding. Finally, the lab scale process has to be scaled up to industrial size.

3.4.3.4 Restructured chicken leg meat (18200140)

In the previous project, leg meat was bonded to each other with the enzyme transglutaminase (TG). To this end, Activa EB was used. Activa EB is a commercial form of TG that is mixed with maltodextrin (carrier of TG) and sodium caseinate. Sodium caseinate supports the function of transglutaminase. However, sodium caseinate is an allergen which means HFP has to declare this on the product label. In order to produce products which are also edible for people with food allergies, it would be preferred to use an allergen-free binding system. During the current research, it was first examined whether good binding can be obtained if only transglutaminase is used. Subsequently, alternatives were sought for sodium caseinate, using both meat and meat foreign proteins and vegetable proteins. Other binding systems, Fibrimex (based on bovine fibrinogen and thrombin) and a calcium alginate system, have been looked at.
In addition, HFP wants to scale up the product from laboratory scale to industrial scale. The binding of the allergen-free products must meet the pre-established standards to ensure that the binding between the meat parts is so stable that industrial operations and consumer actions can be carried out. In the previous study (see section 3.4.3.3) it was determined that the product must be able to withstand a minimum tensile force (maximum load of 1.53 N) during a tensile test with the Instron.

To replace sodium caseinate, use was made of Activa WM (contains TG and maltodextrin). After several tests, it appears that adding only Activa WM to leg meat does not result in a binding which meets the pre-established standards. In order to find non-allergenic alternatives for sodium caseinate, a literature research was first conducted into meat and alternative proteins. This has shown that the structure of vegetable proteins differs from the structure of sodium caseinate (globular versus open structure), which has consequences for their binding abilities. The structure of chicken collagen is fibrillar, which means that the structure is fairly open, but the solubility is very limited compared to sodium caseinate.

A possibility that has been examined to release protein from meat, is the mechanical processing of meat surfaces (tenderizing or long-term mixing) and the addition of comminuted meat (reduced by a bowl cutter). The use of the tenderizer, which releases proteins, results in better binding. Comminuted meat, with the released proteins, is added together with Activa WM in different amounts of leg meat. The use of comminuted meat resulted in poor binding.

Thereafter, various pea proteins, brown rice protein and potato protein have been analyzed. These proteins have been added, in different quantities, to water and Activa WM in a screening procedure to investigate the gelling ability. The gel-forming protein concentrations have been added to meat. The best result was obtained with the use of pea protein LAB 4462 (2.5% w/w). However, the binding properties of the product was only resistant to soft contact and could not be moved without falling apart.

Because collagen proteins have a similar, reasonable open (fibrillar), structure as sodium caseinate, these proteins were expected to support the binding of Activa WM sufficiently. However, the meat products made with chicken collagens, that showed gelling during the screening test, did not have sufficient binding ability.
In addition to adding Activa WM with alternative proteins, alternative binding systems without Activa were also tested, namely Fibrimex and Alginate. Both binding systems were tested in different ratios with leg meat. Looking at the results, it can be concluded that only Fibrimex provides a good binding (Maximum Load >1.53 N). The liquid variant results in a significantly higher binding (9.3 N) than the powder version (1.71 N). Furthermore, the liquid variant is easier to use during a production process.

When using Fibrimex, “Beef protein” must be declared, which is not preferred by HFP. In addition, a product with Fibrimex must be filled immediately, as the cross-linking reaction of Fibrimex works immediately and if the binding is destroyed at a later time no bonds will form again. The addition of alginate and calcium sulfate, in the used form, did not result in a good binding (1.01 N), but it is plausible that optimization of the alginate binding system has potential to achieve a sufficiently good binding (>1.53 N). However, alginate and calcium sulfate are E-numbers (E401 and E516, respectively), which is avoided as much as possible within this project.

During the scaling up, Activa EB was used. Activa EB is easy to process in an industrial way, because a meat mix with Activa EB can still be filled an hour after mixing, which is convenient in case of processing delays. After the breakdown of the binding, bonds are formed again. The addition of Activa EB has the disadvantage that is contains an allergen, but its use results in an excellent product with good binding (2.8 N). The cost price is € 2.33 per kilo finished product. During the scaling up, the meat is only mixed with the Activa EB slurry and filled in casings. The vacuum filler ensures that there is no air in the product and the mixer ensures that the binding aid is distributed over the binding surface. A scale-up test with 50 kilogram of meat with the addition of 1% Activa EB and 4% water has been successfully accomplished (see products in Figure 44). The measured maximum load of the formed product was 4.4 N. This value was higher than for lab scale products, which can be caused by the use of industrial machines.

Figure 44 *Products made on industrial scale.*

From this research it can be concluded that only with Fibrimex an allergen-free product can be produced which meets the pre-established standards. There are several recommendations for the progress of the research. In order to make an allergen-free product with Activa WM and pea protein LAB 4462 or alginate, it can be investigated whether the scaling up positively influences the binding by using industrial machines. Furthermore, the influences of the process steps after filling, such as: cutting, marinating, breading and packaging, must be examined.

Product suggestion for the sliced formed leg meat are: plain burger, marinated burger, breaded burger, and cordon bleu, as shown in Figure 45.

Figure 45 *Product suggestions: plain burger, marinated burger, breaded burger, cordon bleu.*

4 General conclusions and recommendations

4.1 Product transparency and recognisability (WP1)

A sustainability model was developed that shows the sustainability score of the broiler farmer, the processing industry (Heijs), and the total score of the defined poultry production chain per year for the customer. The sustainability score is composed of 36 indicators that are related to the 4 categories; People, Planet, Profit, and Poultry; the 4 Ps. All of the indicators are measureable and have a five-point ranking, based on (scientific) literature and expert interviews. Of the indicators, 34 out of 36 were validated, except for *Campylobacter* as there was not enough data available and CO_2 emission as there was a lack of information for calculating the total CO_2 emission. Colours from green (excellent) to red (very poor) were used to visualise the sustainability score. By creating confrontation matrices, we identified what effect a certain measure (the turning of a 'knob') had on all the indicators (either positive or negative). Insight into knobs that can be turned with little effort by the broiler farmer and the processing industry to improve on sustainability indicators was provided. Data security was taken into account in the information system, several recommendations for anonymity and security were done in consultation with experts in the field. This information system and sustainability model can be implemented and will improve the transparency towards customers.

To improve sustainability of the chain even further, it is recommended to encourage more communication between the chain partners and to exchange more information up- and downstream. Promoting the exchange of information would be easier with a digital tool to record all the data being collected now, like on the flock card. The design of such a tool or software was out of the scope of this project, but is food for thought for the Heijs chain.

The developed model is as good as the data (available), thereto Heijs could take a leading role in filling out the information system, and create supporting power at the broiler farmers by showing them the benefits of using the information system and the generated output, for example for benchmarking themselves against the average. Additionally, it is worthwhile to find an independent party that visits the broiler farmers and the processing industries with the model to obtain reliable data.

Currently, the model shows results per house/company per year, if it is desired that the model can show results per house per unit of time, each house should have its own energy, gas, and water meter. The model should also be maintained and updated now and then as indicators may become more or less relevant in time, rankings and their averages are subject to change, e.g. due to changing regulations, and measurability of indicators can improve (e.g. emissions).

4.2 Animal welfare (WP2)

4.2.1 Effective environmental enrichment

From the series of experiments performed for the development of effective environmental enrichment for the Heijs production chain we conclude that broilers show a preference for resting on an elevated structure and below a structure. When elevated resting structures are provided, broilers prefer to rest on a platform instead of on a wooden A-framed perch. Providing pecking objects such as plastic-wrapped wood shavings bales, pecking stones, lucerne bales and rapeseed straw bales stimulates natural behaviour such as explorative pecking, dustbathing and foraging behaviour. Reducing the stocking density from 35 kg/m^2 to 25 kg m^2 resulted in a significantly better use of enrichment objects (platforms and wood shavings bales) by broiler chickens. Combining different types of enrichments showed that wood shavings bales only stimulated activity more than providing wood shavings bales and platforms. Providing pecking stones in addition to wood shavings bales did not stimulate activity more than providing wood shavings bales only. None of the experiments indicated that environmental enrichment had a negative effect on the technical performance of the broilers.

A recommendation for the implementation of platforms in practice is to study the development of platforms that require less labour (e.g. cleaning) for the farmer. During this project indications were found on how many enrichments should be provided, but more research is needed to determine the optimal number of enrichments in a broiler house. A final recommendation is to further study the combination of natural light and environmental enrichment to further stimulate the activity of the broilers (and possibly the gait score). A pilot study on the effect of natural light on broiler behaviour was performed, but to draw firm conclusions more production cycles should be included.

4.2.2 Improving welfare in end-of-life phase

From the studies on improving welfare in the end-of-life phase we conclude that there is a positive association between average live body weight and the percentage of dead-on-arrival and between average live body weight and the percentage of wing fractures at the slaughter plant. It can also be concluded that there is a negative association between the percentage of wing fractures and the number of broilers per tray. The percentage of wing fractures increases between lairage, post-shackling, post-stunning, and post-plucking, with an average of 4.0% between lairage and post-plucking. The stunner settings did not correlate with the percentage of wing fractures post-plucking. There is a decrease in small sized bruises and an increase in large sized bruises between lairage and post-plucking. Finally, most injuries and damage occurs during the slaughter process and only a small percentage during the pre-slaughter process (catching and transport).

During transport in moderate environmental temperatures, no differences were found in environmental temperature and relative humidity in transport containers between short (2.5 hours) and long (7 hours) transport. However, in the truck, there is a large variation in temperatures in the containers dependent on the location of the containers in the truck (and the ventilation pattern when driving). Further, the temperature in the containers rose as soon as the truck stopped (during pause and at arrival at the slaughter house). Panting only occurred in two transports. Duration of transport did not affect weight loss of the broilers in the experiment.

To develop preventive measures for injuries and damage, it is recommended to determine where exactly in the process injuries and damage occur. In addition, a scoring system to accurately determine the age of bruises needs to be developed. With respect to transport, it is advised to monitor temperatures and relative humidity, as well as heat stress or cold stress in the broilers, under more extreme climatic conditions, e.g. during hot summer days and in winter. Moreover, it is advised to develop methods to increase the ventilation in certain locations in the truck, to prevent heat stress in the broiler chickens.

4.2.3 The welfare model

A welfare model was developed that includes 13 variables indicative for on-farm broiler welfare and welfare during the end-of-life process, a scoring system based on risk assessment principles, and limit values for each welfare indicator. The model is sensitive to variation in welfare performance between broiler flocks.

Further development requires the testing of the indicators for broiler behaviour. Whenever new indicators are developed, the model should be updated. It is also recommended to communicate the individual variable scores to the broiler farmers to allow them to improve their performance.

4.3 Healthy and robust broiler chickens (WP3)

4.3.1 Reducing *Campylobacter* in the chain

From the studies on the measures to reduce *Campylobacter* colonisation in broiler chickens we conclude that there are multiple introduction routes of *Campylobacter* on farm. We also conclude that feed additives do not significantly reduce *Campylobacter* in broiler chickens. Thinning is regarded as a risk-factor for *Campylobacter* colonisation of the (remaining) flock. Although comparing *Campylobacter* strains from transport crates and equipment used for thinning with strains isolated from chickens did not reveal a match, it was shown that the current process of washing the transport containers does not remove all contamination, implicating that this might involve a risk for the introduction of *Campylobacter* in the broiler house at thinning.

Recommendations for future research are to further study the thinning process to get more insight in the risk of thinning for introduction of *Campylobacter* on farms. Such study should include increased sample sizes. The washing process of containers could be improved to remove all contamination, and it is also advised to explore how an improved container design can contribute to a decreased risk of *Campylobacter* contamination.

4.3.2 Reducing *Campylobacter* in the slaughter line

The results of the baseline measurement [*Campylobacter* spp. and *E. coli* (as indicator for *Campylobacter* in the case of *Campylobacter* negative flocks) showed that in the beginning of the slaughter process (before scalding] there was a higher number of *E. coli* than at the end of the slaughter process (after chilling). *Campylobacter* numbers sometimes fell below the detection level, therefore some conclusions were based on *E. coli* determinations. Furthermore, the results showed that the processing steps of scalding and plucking together reduce the bacterial count of *E. coli* on the carcasses in the slaughter process. During the subsequent slaughter process, an equal bacterial count for *E. coli* was detected.

The intervention methods studied in these projects resulted in a reduction of *Campylobacter* and/or *E.coli* on carcasses in the slaughter line. The intervention methods studied were; cold plasma which was evaluated in a laboratory setting, and three other physical treatments of the carcasses without chemicals implemented in-line of a slaughter plant. Although the sensorial meat quality after nitrogen cold atmospheric plasma (CAP) treatment was not acceptable due to dehydration, this study showed the potential of nitrogen CAP in inactivation of *Campylobacter* on raw broiler meat. Two physical intervention methods resulted in a significant reduction of *E. coli.* The third one resulted in less contamination on the carcass compared with the baseline.

Before the intervention methods can be applied in the poultry slaughter line, it is important to examine the exact process conditions and the scatter. Further research should determine whether the reduction of *E. coli* can be translated into a reduction in *Campylobacter.* For the sampling in such research, it is advisable to screen for flocks that are positive on *Campylobacter* spp. to increase the chance of detection. Further recommendations include investigating whether the dehydration of the exposed meat treated with CAP is preventable and reversible. In addition, if employee safety can be guaranteed, an air CAP might be more effective compared to a nitrogen CAP. Alternatively, in-pack treatment might be an option if *Campylobacter* numbers are sufficiently reduced.

4.4 Valorisation and development of allergen-free and low in E-number products (WP 4)

4.4.1 Reduction of allergens and salt in chicken wing products

From the literature studies on allergen-free spicy chicken wings ("Hotwings") we conclude that alternative flours that show the potential to substitute wheat flour's functionality in the breading of Hotwings are; corn flour, sorghum flour, rice flour, tapioca flour, chickpea flour, and buckwheat flour. The experiments on allergen-free Hotwings revealed that a combination of chickpea, rice, and tapioca flour resulted in the best gluten-free breading mixture with similar or better properties compared with the reference. Colour differences could be compensated by the addition of dextrin. A trained sensory panel did not discover significant differences between the reference recipe and the gluten-free recipe. A consumer panel detected differences in expansion, transparency and crispiness (positive), and in graininess (negative). Attempts to improve graininess by changing of the recipe were not successful.

From the literature studies on salt reduction in Hotwings we conclude that the following strategies are feasible in Hotwings: a gradual reduction of the salt content, the use of salt substitutes, the use of flavour enhancers, and possibly application of different salt crystal shape or size.
Not feasible in Hotwings is the application of inhomogeneous spatial distribution of salt; in theory, the sensory contrast allows salt reduction not noticeable for consumers.

According to UK guidelines, one portion of Hotwings consists of six pieces, containing 390 kcal and has a maximally allowed salt content of 2 grams. Our experiments show that a combination of salt replacer (KCl, NaCl, flavour) and NaCl at 2 grams in such a portion, resulted in a product that was perceived comparable to the reference by a sensory panel (n=31). From the experimental studies on salt-reduced Hotwings we also conclude that a salt reduction of 25% in Hotwings (20% in the marinade and 5% in the breading) is possible, which was confirmed by a sensory panel (n=31) that perceived no difference in saltiness, tenderness, juiciness, and colour between the salt-reduced version and the reference.

In conclusion, the resulting Hotwings recipes were either free from allergens or low in salt content, or both, and except from baking powder, they were free from E-numbers.

Recommendations for further research are to analyse the gluten content of the developed gluten-free product to confirm the gluten content to be ≤ 20 ppm, which is the requirement for gluten-free products. It is also recommended to perform shelf-life tests on the reduced salt products as reduced salt content can lead to shorter shelf life. Additionally, it is recommended to execute sensorial research by a large consumer panel (n= 60 – 100) to validate appreciation of the newly developed recipes.

4.4.2 Reducing allergens and salt in marinades

From the studies on the development of marinades for the target supermarkets and consumers, four concepts for world marinades to be used on chicken thighs were developed. All marinades are free from allergens, low in salt, and low in E-numbers.

A recommendation for further research is to execute sensorial research by a large consumer panel (n= 60 – 100) with consumers from the target segments to validate appreciation of the world marinades and the intended preparations.

4.4.3 Valorisation of chicken leg meat

Chicken leg meat in the supermarkets is either the leg meat itself (with or without marinade or sauce, with or without bones, in one piece or chopped into smaller pieces), or it is processed into e.g. sausages or part of a convenience meal. There is a very small number of (enzymatically) formed poultry meat applications on the market. Chicken leg meat can be valorised with several concepts developed in the project. One concept is the "no sticky fingers" concept, which consists of drumsticks with a clean bone resulting in clean hands after consumption. Another concept developed an E-book which introduces three concepts and provides six recipes for preparation of chicken thigh meat. The next concepts focus on the application of enzymatic re-structuring of leg meat. Formed leg meat was developed by applying the enzyme transglutaminase and the (allergenic) milk protein sodium caseinate, resulting in excelling binding strength. Without the protein, the formed leg meat falls apart. Non-allergenic plant proteins or chicken proteins are not suitable as replacers for sodium caseinate, as the stability of those products was insufficient. Another binding mechanism using Fibrimex (fibrinogen and thrombin from bovine plasma) resulted in a very good and stable allergen-free product, implying labelling of beef protein as minor disadvantage. The milk protein containing product was scaled up to industrial scale production, yielding excellent sliced formed leg meat with several product suggestions.

We recommend to perform consumer tests (n≥60) for the developed concepts within the target segments, and if necessary optimize the marinades developed for the chicken thighs. It is also recommended to perform a production trial with the trimming machine for the "no sticky finger" drumsticks. For the formed leg meat, we recommend to investigate industrial scale further, regarding the possibilities to reduce costs by reduced transglutaminase dosage and to carry out the process steps after filling, such as: cutting, marinating, breading and packaging, which influences the binding.

4.5 Concluding remarks

With these four work packages, we covered a large part of the poultry production chain; starting on the farm with welfare, health and sustainability, to the processing industry where sustainability, product safety and health were addressed, to the production of products with lower salt and E-number contents, novel concepts for valorisation of the broilers, up to the customer. The projects performed contribute to an innovative poultry production chain, through the developed welfare and sustainability model for more transparency and recognisability, through the novel, high-quality product concepts developed, and the improved understanding of effective environmental enrichment that can be applied on-farm.

Lastly, to make these findings a success in the poultry production chain, the communication within the poultry production chain is very important, by exchanging more information between the different chain links, the chain as a whole can improve on e.g. sustainability, welfare, and product safety. For the sustainability and welfare model to be implemented, we recommend to invest in a software tool that allows the broiler farmers and the processing industry to digitally record their data and allow them to create outputs with which they can benchmark themselves against the average and get insight into aspects they can improve on (and how). For the developed product concepts, we recommend to strengthen the collaboration with Heijs' customers and perform sensorial tests with large(r) consumer panels.

Annex 1 Acknowledgments

Heijs Food Products

Henk Benedictus
Janet Bos Wierenga
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Richard de Vegt

Agro Food Lab

Anne van den Bosch

AC Adviseurs

Marjolijn van Aarst
Anco Bunning
Annelies de Vries

Marks & Spencer

Peter Kennedy
Tom Willings

British Research and Development Institute Food Animal Initiative

Laura Higham
Carly Scott
Kelly Watson

Wageningen University & Research

Food & Biobased Research

Jacqueline Berghout
Hermien van Bokhorst-van de Veen
Erik Esveld
Hasmik Hayrapetyan
Anke Janssen
Aard de Jong
Marcel Meinders
Wibke Roland
Theo Verkleij
Martijntje Vollebregt

Livestock Research

Marien Gerritzen
Henk Gunnink
Theo van Hattum
Ingrid de Jong
Henny Reimert

Bioveterinary Research

Miriam Koene

HAS

Project coordinators

Patricia de Cocq, Gerben Hofstra, Mariëlle Verkaik

WP1 supervisors	*WP2 supervisors*	*WP3 supervisors*	*WP4 supervisors*
Claudy Claus	Liesbeth Dingboom	Gerben Hofstra	Linda Bonte
Marga Hendriks	Annemarije van Hoek	Peter Jacobs	Annelies van Gunst
Ge-annele Hof	Gerben Hofstra	Hein van Lieverloo	Joke Overgaag
Bart Rietjens	Peter Jacobs	Martin Perescis	Jet Vossebeld
Henk van de Ven	Nina Leenders	Arnold de Vries	Gérard de Wildt
	Tamara Lohman	Gérard de Wildt	
	Bart Rietjens		
	Judith Roelofs		
	Femke Schaafstra		
	Jehan Verstappen		

WP1 students	
7525HFP4	Roel van Heugten
(WP1&2)	Stefan Krol
	Frank Vermeulen
	Jimmy Wolff
7525HFP4	Frank Vermeulen
	Jimmy Wolff
7623LRW4	Dyoni van der Leest
	Stef van de Ven
	Bart Vlemmix
	Willem van Welie
7664LRW4	Roel van Asseldonk
	Ferry de Weert
7704LRW4 &	Jop Hoeben
7705LRW4	Mark van der Spank
	Dirk Timmers
	Dominique Troisfontaine
18200144	Niels van der Cruijsen

WP2 students	
7526WUR4	Roel van Heugten
	Stefan Krol
7555LRW4	Laura van den Eertwegh
	Moniek Göertz
	Sylvia Luijten
7557LIV4	Bart Schoenmakers
	Ellen Waas
7624WUR4	Jens Vullings
	Ralph Pasman
	Ton van Welie
7676LRW4	Néomi Geurts
	Valerie Kalle
	Marinda Vale
7677LIV4	Toinina Banning
	Daan Duijmelinck
	Jeroen Snijders
18200141	Rosalie Coolen
	Ruth van Heerbeek
18200143	Pam Allers
	Susan Janssen
	Sabine Kreisel
18200144	Maartje Frankena
	Roseanne Minderhoud
	Chess Stolk
18200142	Iris Keurntjes
	Vince Verpoort

WP3 students	
7625LIV4	Corné van Boxem
	Carolien van Hooijdonk
	Tom Jansen
7678LRW4	Marieke van Doornik
	Jacobine Visser
7696LRW4	Joyce van Drunen
	Femke Gruter
	Hanneke Versteijnen
18200157	Linnea Bentlage
	Gera van den Boer
	Didi Jonkers

WP4 students	
7626LRW6	Mirte Doomen
	Marlou Wouters
7627WUR4	Doremi van Bommel
	Stephan van Wetten
7628LIV4	Olaf Boom
	Kelly van Lomwel
7629LRW4	Femke van de Klundert
	Micky Taylor
7706LRW4	Douglas Looijen
	Jolien van de Vendel
18200140	Marrit de Kok
	Wouter Lommerse

Junior researchers

WP2	7662LRW1&7734LRW1	Moniek Göertz
WP3	7695LRW4	Sophie van der Linden
WP4	7552COF1	Anne Wesseling

Annex 2 List of dissemination activities

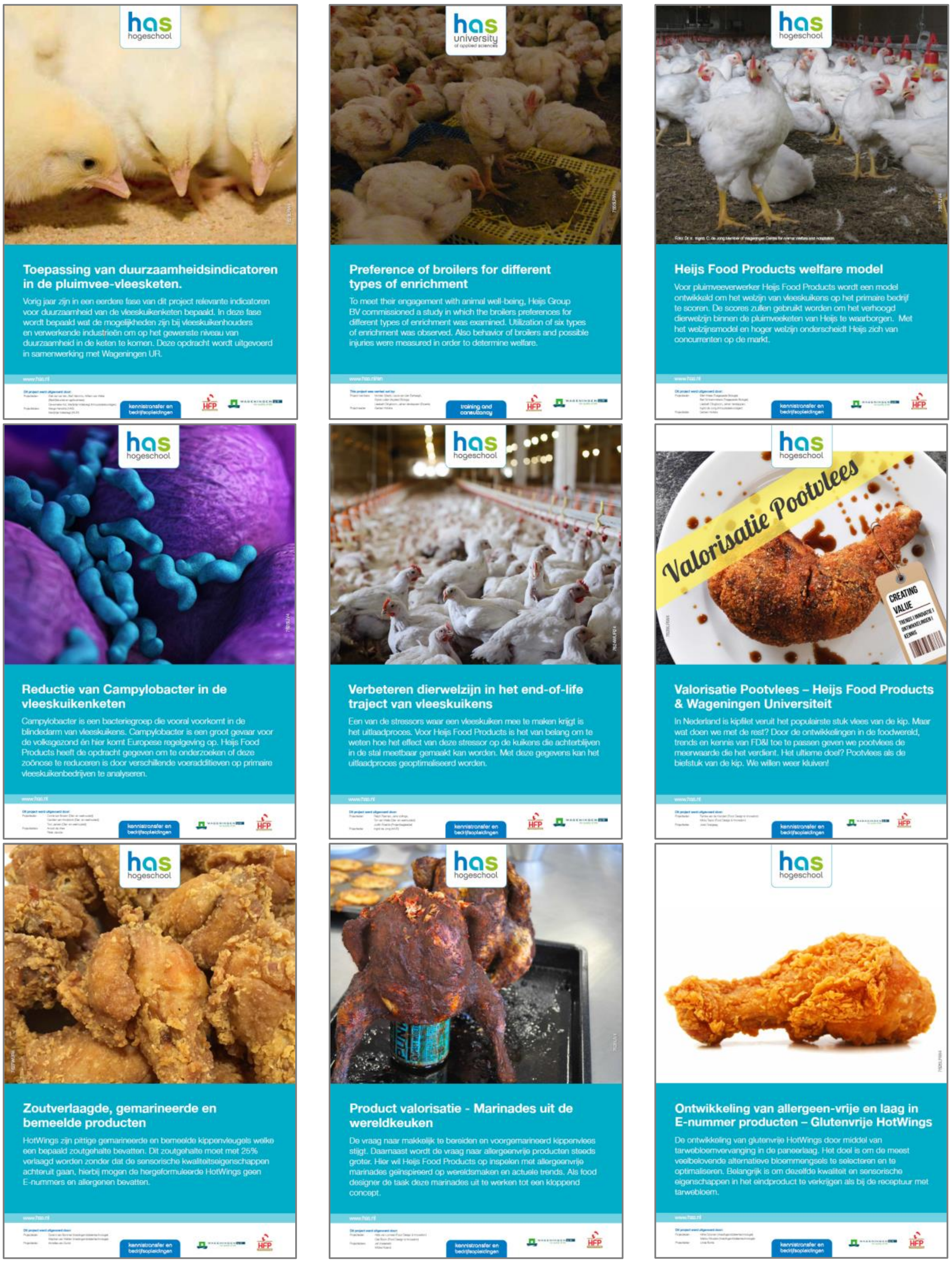
WP	Dissemination activity	Reference	(Place and) date
Websites			
WP1-4	Project description on the WUR website	http://www.wur.nl/nl/project/Farming-for-the-future-innovatie-in-vleeskuikenproductieketen-en-producten.htm	
WP1-4	Project description on the HAS website	https://has.nl/nl/showcase/has-hogeschool-werkt-aan-ontwikkeling-innovatieve-vleeskuikenketen	
WP4	E-book for leg meat concepts	https://issuu.com/femkevdklundert/docs/e-book_heijs_food_products	2016
Posters			
WP1-4	HAS year event 2016		Den Bosch, 2016
WP1-4	HAS year event 2017	https://has.nl/nl/topproject/has-hogeschool-werkt-aan-ontwikkeling-innovatieve-vleeskuikenketen	Den Bosch, 2017
WP2	Poster at the 7th International Conference on the Assessment of Animal Welfare at Farm and Group level	Jong, I.C. de; Hofstra, Gerben ; Hamming, Jan Jaap (2017) In: Proceedings of the 7th International Conference on the Assessment of Animal Welfare at Farm and Group level. - Wageningen : Wageningen Academic Publishers, WAFL 2017, Ede, 2017-09-05/2017-09-08 - p. 176 - 176.	Wageningen, 2017
WP1-4	HAS year event 2018		Den Bosch, 2018
WP4	Poster at the XVth European Poultry Conference[11]	Roland, W., van Bommel, D., van Wettern, S., van Gunst, A., de Vegt, R., Salt reduced chicken wings	Dubrovnik, 2018
Presentations			
WP1-4	Project presentation for Marks and Spencer and FAI		Oxford, 29 June 2016
WP1-4	Project presentation for Province of Groningen and Drenthe		Hoogeveen , 2 November 2016
WP2	Presentation at Xth European Symposium on Poultry Welfare	Jong, I.C. de; Goertz, M. (2017), Broiler chicken stocking density affects use of environmental enrichment objects In: Xth European Symposium on Poultry Welfare - 2 p.	Ploufragan, 19-22 June , 2017
WP2	Presentation at the XVth European Poultry Conference[12]	Jong, I.C. de, Reimert, H., Lohman, T., Gerritzen, M., Identification of risk factors and prevalence of injuries at different stages of the broiler slaughter process	Dubrovnik, 2018
WP1-4	Symposium: Concluding project presentation for Province of Groningen and Drenthe, Heijs, HAS, AC advisors, broiler farmers		Westerbork, 2018
Articles			
WP1+4	Article in VMT	Janssen, A. (2017): Heijs Food Products wil innovatieve kip producten ontwikkelen. In: VMT online, http://www.vmt.nl/Nieuws/Innovatieve_kipproducten_ontwikkelen-170317152023	2017
WP1+2	Expected Article: Journal to be determined	Welfare and sustainability model	2018

[11] Picture in Annex 3

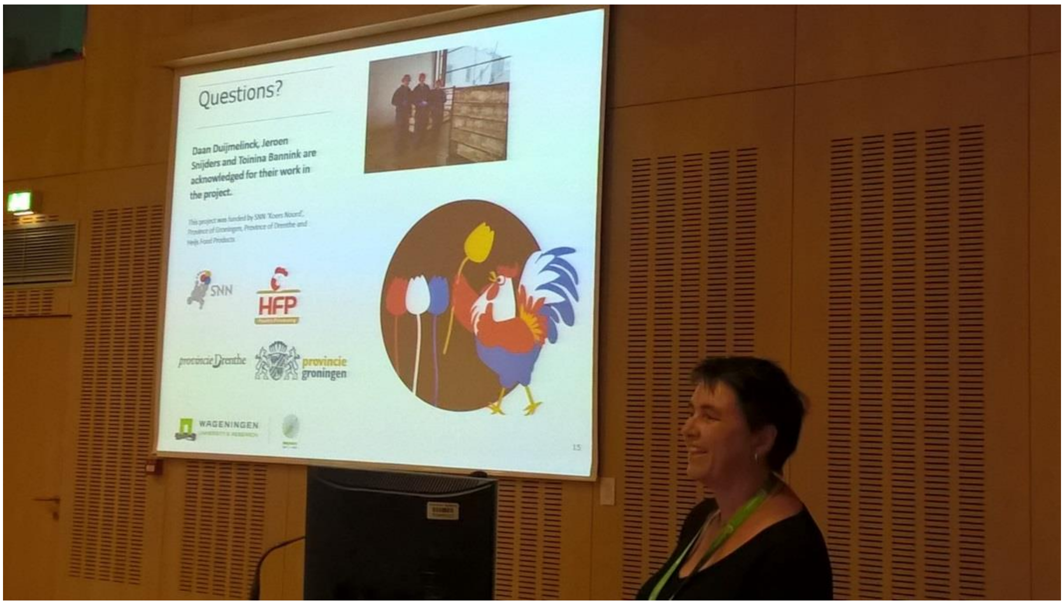
WP	Dissemination activity	Reference	(Place and) date
WP2	Expected: Article in Pluimveehouderij	Enrichment	2018
WP2	Expected: Article in Pluimveehouderij	Transportation	2018
WP2	Expected: Article in Pluimveehouderij	Damage	2018
WP4	Expected: Article in VMT	Enzymatically restructured chicken leg meat	2018

Annex 3 Examples of dissemination activities

The Innovative Poultry Production project was represented by 9 posters during the HAS Year event 2016 in Den Bosch



The Innovative Poultry Production project was represented by 2 presentations and 1 poster during the XVth European Poultry Conference (2018) in Dubrovnik



To explore the potential of nature to improve the quality of life

Wageningen Livestock Research
P.O. Box 338
6700 AH Wageningen
The Netherlands
T +31 (0)317 48 39 53
E info.livestockresearch@wur.nl
www.wur.nl/livestock-research

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