

Report of the Scientific Committee
of the Food Safety Authority of Ireland

2011

Recommendations for a Practical Control Programme for Campylobacter in the Poultry Production and Slaughter Chain



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Recommendations for a Practical Control Programme for Campylobacter in the Poultry Production and Slaughter Chain

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EXECUTIVE SUMMARY

I. Background

Campylobacteriosis is the most frequently reported gastrointestinal bacterial illness in humans in Ireland and across the EU. A number of risk factors have been associated with human campylobacteriosis. These include the consumption and/or handling of raw or undercooked poultry or other meats, raw milk, surface waters, cross-contamination of ready-to-eat foods during food preparation as well as direct contact with animals.

Poultry is regarded as one of the most important reservoirs for *Campylobacter* species and constitutes a very significant vehicle for the transmission to humans. The result of an EU wide baseline study revealed an Irish prevalence in broiler batches of 83.1% and a prevalence of 98.3% on carcasses at the end of slaughtering process. As the results of this baseline study emerged, the Food Safety Authority of Ireland (FSAI) requested its Scientific Committee to provide advice on a practical control programme for *Campylobacter* spp. in the Irish broiler production and slaughter chain. The Scientific Committee delegated the task to the Microbiology Sub-committee and a working group was formed to address five specific risk management questions.

This report contains recommendations on control measures in the Irish broiler food chain and proposes microbiological criteria in broilers (i.e. pre-harvest) and on carcasses (i.e. post-harvest) which require validation. The recommendations (in particular, the proposed pre- and post-harvest criteria) will be subject to periodic review by the FSAI. The review process will take account of the results generated by pre- and post-harvest analyses and of emerging and newly published research. If a voluntary control programme proves ineffective, a legal approach may need to be considered.

II. Recommendations Relating to Thinning/Partial Depopulation

Thinning or partial depopulation is a process whereby a portion of a flock is removed for slaughter approximately one week before the remainder of the flock. This practice increases the risk of infection with *Campylobacter* spp. The answers to the risk management questions addressed in this report are made on the basis that thinning of flocks is current industry practice and that there is no immediate likelihood that it will be eliminated. In addition, the following recommendations are made on how the risk of infection posed by thinning could be minimised:

- There should be no more than one partial depopulation prior to the final depopulation
- The period between the partial depopulation and the final depopulation should not exceed five days
- Emphasis by processors and producers should be placed on the following:
 - Effective cleaning and disinfection of forklift wheels prior to entry into broiler house
 - Improved crate, module and truck washing and disinfection in the slaughter plant
 - Thinning teams must put on clean protective clothing and clean and disinfected boots prior to entry into the broiler house. There should be dedicated footwear supplied for use by the thinning teams for each house on-farm
 - Thinning teams must comply with biosecurity measures
 - Education of thinning personnel to enforce the importance of biosecurity

III. Recommendations Relating to Risk Management Questions

The following is a summary of the answers to the five risk management questions addressed in this report and related recommendations.

RISK MANAGEMENT QUESTION 1

Q1. What is the strength of evidence that poultry is the most important source of campylobacter causing campylobacteriosis in humans?

There is compelling evidence from a number of studies in Ireland and internationally that poultry is the most important source of campylobacter causing campylobacteriosis in humans. It is therefore recommended that the poultry industry develops and implements its own voluntary code of practice taking the guidance in this report into consideration.

ADDITIONAL RECOMMENDATION

- It is recommended that the Health Service Executive (HSE) and the Department of Agriculture, Fisheries and Food's (DAFF) National Reference Laboratory for *Campylobacter* spp. (in food, feed and animals) coordinate activities to ensure that identification to species level and genotyping of a subset of human, food and animal isolates is undertaken using appropriate methods to assist with source attribution.

RISK MANAGEMENT QUESTIONS 2 AND 3

Q2. What microbiological criteria for campylobacter could be applied to results of analyses of poultry for *Campylobacter* spp. that would reduce the risk of campylobacteriosis in humans and could be used as part of a statutory/voluntary control programme as a trigger for actions aimed at reducing the contamination of broilers in a particular production/processing facility.

Q3. How should such a microbiological criterion be applied in practice to monitor campylobacter contamination rates on broiler carcasses?

Pre-harvest and post-harvest microbiological criteria are recommended in response to these questions to reduce the risk of campylobacteriosis in humans. Interventions on-farm and in the slaughterhouse should be taken when these criteria are exceeded.

A pre-harvest microbiological criterion of $\leq 7 \log_{10}$ cfu/g of *Campylobacter* spp. in 10 pooled caecal contents is recommended. Sampling should be carried out on-farm seven days (or less if feasible) before slaughter.

A post-harvest target level of $\leq 4 \log_{10}$ cfu/g of *Campylobacter* spp. on neck skin samples taken post-chill is recommended, with a sampling plan and limits of $n=5$, $c=1$, $m=4 \log_{10}$ cfu/g and $M=5 \log_{10}$ cfu/g.

These criteria should be validated by a pilot study and should be subject to periodic review based on progress made by the industry when the control programme is in place. It is recommended that data generated are collated in a central database to facilitate trend analysis.

ADDITIONAL RECOMMENDATIONS

- Sampling to test against the pre-harvest criterion should be carried out on-farm, seven days (or less if feasible) before slaughter. Testing of each house should be performed by randomly selecting 10 birds from various locations within the house which will be harvested and dispatched to the laboratory on the same day. The caecal contents of the 10 birds will be pooled for analysis in the laboratory.
- Post-harvest sampling should be carried out once a week in the slaughterhouse. One positive flock (with pre-harvest levels $\leq 7 \log_{10}$ cfu/g) should be selected for sampling. Sampling should take place after chilling and prior to further processing. It is recommended that sampling should be carried out when approximately half of the flock have been slaughtered. Fifteen carcasses should be sampled. A piece of approximately 10g of neck skin shall be obtained from each carcass. The neck skin samples from three carcasses shall be pooled before examination in order to form 5×25 g final sampling units.
- The laboratory used to analyse pre- and post-harvest samples should have accreditation specifically for the international standard method for the enumeration of *Campylobacter* species (ISO/TS 10272-2:2006 or equivalent). The laboratory should be capable of demonstrating reproducibility of their testing methods. Participation and good performance in ring trials would be required.

- Results should be collated in a central database to facilitate trend analysis.
- Exceeding the pre-harvest criterion:
 - Will invoke a number of interventions at farm level including a review of biosecurity
 - A scheduling approach towards slaughtering chickens from these flocks should be adopted in the slaughterhouse, whereby carcasses from these flocks (with higher counts of *Campylobacter* spp. in the caeca) are subjected to treatments or interventions which can reduce campylobacter counts
- It is recommended that an incentive scheme is adopted by the industry to reward high standards of biosecurity and compliance with the pre-harvest microbiological criterion. The incentive scheme could take the form of a bonus and/or penalty scheme based on producer performance as demonstrated by the pre-harvest sampling.
- Repeated breaches of the pre-harvest criterion should result in removal from the Bord Bia Quality Assurance scheme.
- Exceeding the post-harvest criterion will require a review of hygiene practices in the slaughterhouse.
- Where persistent, repeated breaches of the post-harvest criterion occur (more than four breaches in eight batches sampled), a comprehensive review of slaughterhouse procedures and sourcing of broilers is necessary. In addition, further interventions may be necessary and it is recommended that freezing is considered in these cases.
- It is recommended that isolates are sent to the National Reference Laboratory to ensure that a subset of caecal and carcass isolates are identified to species level and genotyped by the same methods applied to human isolates. This will provide data for comparison with human data in source attribution studies.

RISK MANAGEMENT QUESTION 4

Q4. What are the practical and legally permissible measures that processors and producers could implement to reduce campylobacter contamination on broilers if the microbiological criteria were exceeded?

In order to achieve the recommended pre- and post-harvest criteria, the poultry industry should develop and implement its own voluntary code of practice, taking the guidance in this report into consideration.

ADDITIONAL RECOMMENDATIONS

- It is recommended that a **campaign to improve producers' and catchers' understanding of biosecurity measures**, and their importance, is developed and delivered to coincide with the introduction of the voluntary campylobacter control programme. Processors are responsible for the sourcing of their raw materials and for the food they produce and they should therefore be involved in this campaign with support from the FSAI and DAFF.
- It is recommended that an **incentive scheme** is implemented by the processors to encourage compliance with biosecurity measures on-farm. The incentive scheme may take the form of a bonus or penalty system.

Measures on-farm in the event of the pre-harvest criterion being exceeded (i.e. $>7 \log_{10}$ cfu/g)

- A full review of biosecurity measures should take place. In addition to ensuring that the Bord Bia quality assurance standard requirements are met, implementation of one or a combination of the following measures will lead to greater reductions in levels of campylobacter in flocks:
 - **Personnel access and hygiene:** A system of boot changes and hygiene measures in the anterior room as outlined in Appendix 4 is recommended in preference to the boot dipping system currently practiced on many Irish broiler farms. It is recommended that broiler farmers who also farm other animal species observe the guidelines on personnel access and hygiene, particularly when carrying out routine husbandry procedures on their farm
 - **Personnel access and hygiene during non-routine events:** Poultry producers should consider keeping dedicated maintenance tools (which would be used regularly in the house) in the *ante-room* to each house

Recommendations for a Practical Control Programme for *Campylobacter* in the Poultry Production and Slaughter Chain

- **Focus on effective implementation of biosecurity:** Effective biosecurity requires (i) installation of physical hygiene barriers; (ii) training to ensure that existing measures are consistently and effectively applied; (iii) monitoring by processors and third party auditors that every measure is being consistently applied. Training of producers and provision of booklets and signage, e.g. to be displayed in the *ante-room*, could be used to reinforce the need for 100% compliance with biosecurity measures
- **Surrounds of broiler house:** A concrete apron (maintained in good condition) is recommended in front of the doors to the broiler house and it is recommended that pebbles/gravel are placed along the sides of the houses to permit drainage, prevent dust circulation and deter rodents
- **Fly control:** Wherever possible, fitting of fly screens is recommended, particularly in high prevalence flocks. Alternative methods of fly and insect control may be appropriate where fly screening is not possible
- **Biosecurity during preparation and stocking of house:** It is recommended that the truck ramp is thoroughly cleaned and then disinfected and that the ramp is placed very close to the entrance of the house to ensure that the modules carrying the chicks are not a source of contamination
- **Reduction of slaughter age:** Taking the association between increasing age and risk of colonisation into account, a policy of slaughtering flocks at a younger age particularly during the summer months, may lead to a reduction in the prevalence of *Campylobacter* in broilers and is recommended for flocks with persistently high concentrations of *Campylobacter* spp. in the caecal contents
- **Disposal and storage of waste:** Adequate storage and disposal of broiler farm waste is important to eliminate the potential of waste being a source of contamination for subsequent flocks. On mixed farms, careful management of waste from all species of animals is recommended
- **Sexing of birds prior to placement:** Consideration could be given to sexing of flocks and slaughter of flocks based on sex which will permit greater size consistency of birds at time of slaughter

The following measures may provide additional options for reducing *Campylobacter* levels in broilers prior to slaughter. Extensive studies on the effectiveness of these controls have not been completed to date:

- **Addition of 0.44% lactic acid** to drinking water during the period of feed withdrawal
- **Addition of organic acids to the feed**

Measures in the slaughterhouse for flocks which have exceeded the pre-harvest criterion (i.e. $>7 \log_{10}$ cfu/g)

- A scheduling approach to the slaughter of broilers from flocks which have exceeded the pre-harvest microbiological criterion is recommended. Scheduled slaughter, which is practiced in some countries, means identifying flocks that are positive for *Campylobacter* spp. before they are slaughtered and subjecting carcasses from these flocks to treatments or interventions which can reduce *Campylobacter* counts. Given the high prevalence in Irish broiler flocks currently, a scheduling approach is recommended, whereby carcasses from flocks which exceed the pre-harvest criterion are subjected to treatments or interventions (outlined below) which can reduce *Campylobacter* counts.
 - It may take a period of time to introduce and establish the scheduling approach in the slaughterhouse. In the interim, it is recommended that whole birds from flocks which have exceeded the pre-harvest microbiological criterion ($>7 \log_{10}$ cfu/g) are clearly labelled:
“containing bacteria at potentially harmful levels”

One or a combination of the following interventions can be used to reduce the concentration of *Campylobacter* spp. on the final product:

- **Freezing of chicken:** In Ireland, the market for frozen product is relatively small when compared to the fresh poultry market. Where possible, the processor should ensure that flocks with $>7 \log_{10}$ cfu/g are used to produce frozen product. It is also recommended that where other interventions have failed, or in the case of repeated breaches by the producer of the pre-harvest microbiological criterion, freezing of resulting carcasses should be considered
- **Adjustment and monitoring of slaughtering equipment:** It is recommended that processors ensure defeathering and evisceration equipment is appropriately adjusted to the size of the broilers before processing. Monitoring of the number of evisceration failures (i.e. intestinal rupture as a percentage of birds slaughtered within each batch) will highlight problems with the alignment of equipment and permit corrective action to be taken
- **Crust-freezing of chicken:** Crust-freezing is a technique for rapid chilling of meat. Chicken meat which has undergone the process of crust-freezing can be sold as fresh meat, provided the temperature of the meat remains greater than or equal to -2°C (Regulation (EC) No. 543/2008). The CO_2 crust-freezing technique has been shown to produce consistent reductions (approx. $0.5 \log_{10}$) in campylobacter concentrations. Crust-freezing is recommended as an intervention which should be considered by processors
- **Removal of skin from chicken:** Most campylobacter contamination is located on the skin; therefore, skinless products have counts several logs lower than the respective meat products with skin
- **Logistical approach to slaughter:** Logistic slaughter is an intervention measure intended to reduce cross-contamination during slaughter by slaughtering positive flocks after negative flocks. Given the high prevalence in Irish broiler flocks currently, logistic slaughter is not feasible. However, a logistical approach to slaughter is recommended whereby broilers from these farms (with higher counts of *Campylobacter* spp. in the caeca) will be slaughtered later in the day, after flocks with caecal counts below the criterion limit (although not necessarily negative) are slaughtered
- **Hot water treatment of carcasses:** Studies have shown that hot water treatment can reduce campylobacter numbers while not adversely affecting the appearance and quality of the meat
- **Steam treatment of carcasses:** The main advantage of using steam is the large amount of heat transferred to the food when steam condenses. In addition, steam can penetrate cavities, crevices and feather follicles
- **Combined steam and ultrasound:** Greater reductions can be achieved when different interventions are combined
- **Diversion of carcasses to value added products – marinating and cooking:** The composition of marinade, e.g. a low enough pH, is a key factor in achieving a decrease in campylobacter concentration on the surface of the chicken. Normal cooking temperatures (where a core temperature of 75°C is achieved) will kill campylobacter and therefore cooking is an option for chickens from highly contaminated flocks
- **Modified atmosphere packaging (MAP):** Given the right combination of gases, reduction of campylobacter levels has been achieved after approximately a week of storage in MAP

Measures if the flock is positive for campylobacter but the result is $\leq 7 \log_{10}$ cfu/g prior to the first depopulation:

- These flocks may be slaughtered in the usual manner; however, it is recommended that broilers from the subsequent depopulation of this flock should be processed in the same manner as broilers from flocks that have exceeded the pre-harvest criterion, unless the producer decides to re-test the remaining broilers. If the result of the re-testing is $\leq 7 \log_{10}$ cfu/g, then the broilers can proceed to slaughter without a scheduling approach

Flocks negative for campylobacter

- Broilers from these flocks will be processed as normal and re-testing will be unnecessary during the same flock cycle

Measures if the post-harvest criterion is not met:

- It is recommended that a review of GHP (good hygiene practices) in the slaughterhouse be undertaken
- It is recommended that de-feathering and evisceration equipment is appropriately adjusted to the size of the broilers before processing. Consideration should be given to replacing equipment with models shown to have better performance
- A review of line speed is recommended
- Where persistent, repeated breaches of the post-harvest microbiological criterion occur (more than four breaches in eight batches sampled), a comprehensive review of slaughterhouse procedures and sourcing of broilers is necessary. In addition, further interventions may be necessary and it is recommended that freezing is considered in these cases. Freezing is a safe, effective and readily available method to reduce the number of viable campylobacter on poultry.

RISK MANAGEMENT QUESTION 5

Q5. What, if any, additional packaging and labelling measures could the poultry processing industry adopt to reduce the risk of campylobacteriosis for consumers?

It is recommended that raw chicken is packaged in leak-proof packaging. It is recommended that safe handling and cooking instructions should be clearly visible at time of purchase. Instructions should not be on the reverse of a label or otherwise obscured.

ADDITIONAL RECOMMENDATIONS

- Packaging solutions which reduce the amount of handling of the raw poultry, e.g. oven-ready trays, cooking bags, should be explored.
- Labels on whole birds should advise consumers that carcasses are ready-to-cook and in the interests of safe handling, that washing of the carcass should be avoided. In addition, as many Irish people have a tendency to wash chicken meat, the advice not to wash chicken should be carried on portions as well.

IV. Recommendations Regarding GHP which should be Implemented by All Processors

Recommendations to reduce contamination by crates, modules and vehicles:

- Food business operators should ensure that crates, modules and vehicles are effectively cleaned and disinfected. Appropriate temperatures for cleaning (hot water) and disinfection and adequate concentrations of disinfectants should be used. This cleaning should be validated through periodic microbiological testing and incorporated into the food safety management system
- If crates and modules cannot be cleaned effectively because of the material used in their manufacture then they should be replaced with crates and modules manufactured from materials that are easily cleaned
- Placing modules onto dirty flatbed trucks can serve as another source of contamination for the modules. Flatbed trucks should be adequately cleaned and disinfected before transportation of modules. Where a tarpaulin is used during transport – this must also be cleaned and disinfected adequately between uses

Recommendations to reduce contamination during the scalding step include:

- Counter-current scald tanks
- High water flow rates in the tanks
- Multi-stage scald tanks

Recommendations to minimise cross-contamination during de-feathering include:

- Correct alignment of machinery based on bird size
- Adequate flow rates of water
- Regular equipment sanitation and maintenance

Recommendations to minimise cross-contamination during evisceration include:

- Control alignment of equipment
- Visual inspection of carcasses to identify problems with evisceration
- Replace old/dysfunctional equipment
- Regular maintenance of equipment

Recommendations to minimise cross-contamination during chilling:

- Effective air chilling requires appropriate design and maintenance of equipment
- Appropriate line speeds should be used, as inappropriate line speeds may result in inadequately chilled carcasses exiting the chill
- Thorough washing of carcasses prior to chilling is recommended to remove surface debris and contamination

Recommendations for sanitation and process hygiene:

- Verification of effective cleaning and disinfection can be carried out through the microbiological sampling plan in the plant

CHAPTER 1. INTRODUCTION

1.1 Background

Campylobacteriosis is the most frequently reported gastrointestinal bacterial illness in humans in Ireland and across the EU (EFSA, 2010a; HPSC, 2009). The European Food Safety Authority (EFSA) is of the view that there is considerable under-ascertainment and under-reporting of campylobacteriosis within the EU, and that the true incidence is likely to be 10-100 times higher than the reported number (EFSA, 2010a). In 2004, campylobacteriosis became a notifiable disease in humans in Ireland under the Infectious Diseases Regulations (S.I. No. 707 of 2003). The annual incidence rate reported in Ireland in 2008 was 41.4 per 100,000 of the population (HPSC, 2009); the average EU incidence that year was 40.7 per 100,000 (EFSA 2010a). In Ireland, the highest incidence has been observed in children aged 1-4 years, followed by adults aged 20-24 years. Reports generally peak during the early summer months, with fewer cases recorded between December and April.

A number of risk factors have been associated with human campylobacteriosis. These include the consumption and/or handling of raw or undercooked poultry or other meats, raw milk, surface waters, cross-contamination of ready-to-eat foods during food preparation as well as direct contact with animals (EFSA 2010b; Whyte *et al.*, 2004). Poultry is regarded as one of the most important reservoirs for campylobacter and constitutes a very significant vehicle for the transmission of campylobacter to humans (Humphrey *et al.*, 2007). An EFSA opinion estimated that handling, preparation and consumption of chicken meat may account for 20% to 30% of cases, while 50% to 80% may be attributed to the chicken reservoir as a whole (EFSA, 2010b).

An EFSA baseline survey on the prevalence of *Campylobacter* species in broiler batches and of *Campylobacter* spp. and *Salmonella* spp. on broiler carcasses in the EU during 2008 found the community prevalence of campylobacter-colonised broiler batches was 71.2% and the prevalence of campylobacter-contaminated broiler carcasses was 75.8% (EFSA, 2010c). In Ireland, the prevalence in broiler batches was 83.1% and the prevalence of contaminated carcasses was 98.3%. The results of the campylobacter enumeration on broiler carcasses revealed that 65.2% of Irish broiler carcasses had counts between 100 cfu/g and 10,000 cfu/g and 8.9% greater than 10,000 cfu/g. This report suggests significant exposure of the Irish consumer to *Campylobacter* spp. from broilers. In Ireland, the consumption of poultry meat is quite significant. The gross human apparent consumption¹ of poultry meat per capita in 2005, 2006 and 2007 was 32 kg, 30 kg and 27 kg, respectively (Eurostat).

In 2002, the FSAI's Scientific Committee drafted a report on the control of *Campylobacter* spp., which carried 38 recommendations throughout the food chain (FSAI, 2002). In 2009, the FSAI requested the Scientific Committee to advise specifically on a practical control programme for campylobacter in the Irish broiler production and slaughter chain. The Scientific Committee delegated the task to the Microbiology Subcommittee and a working group was formed to draft a report. This document outlines the findings of the Scientific Committee.

1.2 Scope and Objectives

This report addresses control of *Campylobacter* spp. in broilers produced and slaughtered in Ireland, including whole carcass but excluding spent hens. The document focuses on commercially produced broilers and excludes free range and organic broiler production. Free-range and organic productions are likely to be addressed in the future. The report addresses the following risk management questions:

1. What is the strength of evidence that poultry is the most important source of campylobacter causing campylobacteriosis in humans?
2. What microbiological criteria for campylobacter could be applied to results of analyses of poultry for *Campylobacter* spp. that would reduce the risk of campylobacteriosis in humans and could be used as part of a statutory/voluntary control programme as a trigger for actions aimed at reducing the contamination of broilers in a particular production/processing facility?
3. How should such microbiological criteria be applied in practice to monitor campylobacter contamination rates on broiler carcasses?
4. What are the practical and legally permissible measures that processors and producers could implement out to reduce campylobacter contamination on broilers if the microbiological criteria were exceeded?
5. What, if any, additional packaging and labelling measures could the poultry processing industry adopt to reduce the risk of campylobacteriosis for consumers?

¹Gross human apparent consumption: Apparent consumption = (commercial production + estimated own account production for self-consumption + imports + opening stocks) – (exports + usage input for processed food + feed + non-food usage + wastage + closing stocks)

The recommendations made in this report (in particular, the recommended pre- and post-harvest criteria) will be subject to periodic review by the FSAI. The review process will take account of the results generated by pre- and post-harvest analyses and of emerging and newly published research. If a voluntary control programme proves ineffective, a legal approach may need to be considered.

1.2.1 Thinning/partial depopulation

Thinning or partial depopulation is a process whereby a portion of a flock is removed for slaughter approximately one week before the remainder of the flock. In the 2002 report on campylobacter, it was recommended that the practice of partial depopulation (called thinning) should be discontinued as it was a breach of biosecurity. While the Scientific Committee continues to make this recommendation, it recognises that for commercial reasons, industry has not yet implemented this recommendation. The answers to the risk management questions in Chapter 2 are made on the basis that thinning is current industry practice and that there is no immediate likelihood that it will be eliminated. Therefore, recommendations on how the risk posed by thinning could be minimised are given in Appendix 1.

CHAPTER 2. RISK MANAGEMENT QUESTIONS

2.1 RISK MANAGEMENT QUESTION 1

Q1. What is the strength of evidence that poultry is the most important source of campylobacter causing campylobacteriosis in humans?

There is compelling evidence from a number of studies in Ireland and internationally that poultry is the most important source of campylobacter causing campylobacteriosis in humans. It is therefore recommended that the poultry industry develops its own voluntary code of practice taking the guidance in this report into consideration.

A number of risk factors have been associated with human campylobacteriosis. These include the consumption and/or handling of raw or undercooked poultry or other meats, raw milk, surface waters, cross-contamination of ready-to-eat foods during food preparation as well as direct contact with animals (Whyte *et al.*, 2004). Poultry is regarded as one of the most important reservoirs for campylobacter and constitutes a very significant vehicle for the transmission of campylobacter to humans (Humphrey *et al.*, 2007). Various studies have demonstrated high levels of campylobacter in broilers (Stern *et al.*, 1995), on broiler carcasses (Cantor, 1997) and retail chickens (Zhao *et al.* 2001). Surveys of Irish retail poultry have reported contamination rates of approximately 50% (FSAI, 2002; Whyte *et al.*, 2004). Similar results have been reported in the UK (FSA, 2001) with mean prevalence ranging from 46%, 42%, 75% and 77% for England, Wales, Scotland and Northern Ireland, respectively. The reported level of contamination at 50% in Irish retail poultry is lower than the level of contamination (98.3%) reported in the EFSA baseline survey of chickens in slaughterhouses.

The European Food Safety Authority (EFSA) published a scientific opinion on quantification of the contribution of broiler meat to the burden of human campylobacteriosis in the EU in January 2010. EFSA concluded, following a meta-analysis of case-control studies, that handling, preparation and consumption of broiler meat may account for 24-29% of human cases of campylobacteriosis, while 50% to 80% may be attributed to the chicken reservoir as a whole (EFSA, 2010b).

In Ireland, research has shown evidence that poultry is a significant source of infection with *Campylobacter* spp. for humans. In one study, genotyping by pulse field gel electrophoresis (PFGE), followed by cluster analysis revealed that 28.3% of human clinical campylobacter isolates and 29.9% of food isolates shared common genotypic profiles. This study reported the analysis of a total of 2,391 retail food samples and the isolation of *Campylobacter* spp. from raw chicken (49.9%); turkey (37.5 %) and duck (45.8%). Lower frequencies of isolation of 3.2%, 5.1% and 11.8%, were observed for raw beef, pork and lamb, respectively. This study demonstrates that poultry products play an important role in the epidemiology of human campylobacteriosis (Whyte *et al.*, 2006).

In an all-Ireland case-control study, the most important risk factors identified for contracting sporadic campylobacter infection were consumption of chicken ([adjusted matched² (am) odds ratio (OR) of 6.8; 95% confidence interval (CI) 2.1-21.9]), consumption of lettuce (amOR 3.3; 95% CI 1.5-7.1) and eating in takeaways (amOR 3.1; 95% CI 1.4-6.6). Chicken consumption showed a dose-response relationship, whereby more frequent consumption of chicken increased the risk of infection by 20% per time of consumption (Danis *et al.*, 2009).

The Food Standards Agency (FSA) in Scotland funded a project with the goal of using multi-locus sequence typing (MLST) to provide quantitative attributions of clinical campylobacter infections to infection sources. A total of 31% of human clinical isolates were attributed to retail chicken. This study clearly identified retail chicken as the single largest source of clinical campylobacter infection in Scotland (FSA Scotland, 2009).

In Canada, analysis of outbreak datasets over a period of 30 years was carried out. The study revealed that 56% of outbreak cases of campylobacteriosis were associated with poultry (Ravel *et al.*, 2009).

In June 1999, the dioxin crisis in Belgium caused the withdrawal of Belgian chicken and eggs from the market. The sentinel surveillance system showed a decrease in campylobacter infections during June 1999. A model was generated based on the number of infections in previous years (1994-1998) and a prediction of the number of infections expected in 1999 was calculated. The model showed a 40% decline in the numbers of infections in 1999 by comparison with the number of predicted infections. Vellinga and Van Loock (2002) attribute this to the withdrawal from the market of Belgian poultry. Foreign poultry, which accounted for 41% of poultry available for human consumption in 1999, remained available on the market during this time.

²In this study, age was chosen as a matching variable because (i) potential high-risk exposures, e.g. food habits, leisure activities, vary considerably among different age groups and (ii) the age profile of campylobacteriosis in Ireland, both ROI and NI, peaks in some age groups, namely 0-4 and 20-34 years

In Iceland, prior to 1996, only frozen poultry products had been sold. After 1996, markets increasingly demanded fresh poultry which contained comparatively higher levels of campylobacter per carcass and provided greater public exposure to the contaminated products. The rate of reported human cases in 1997 was 13.8 cases/100,000. This rate increased in 1998 to 52 cases/100,000 and peaked in 1999 at 116 cases/100,000 (Stern *et al.*, 2003). In 2008, the number of reported cases was 31.1/100,000 (EFSA, 2010a).

In Lancashire, England, a method of source attribution was applied to 1,231 human isolates that had been subjected to molecular typing (MLST) and the proportion of cases attributable to different sources was estimated (Wilson *et al.*, 2008). The method involved modelling the DNA sequence evolution and zoonotic transmission of *C. jejuni* between host species and the environment and assigning human cases probabilistically, to source populations. The study estimated that chicken was a source of infection in the majority (56.6%) of cases.

In New Zealand, source attribution modelling was used to estimate the contribution of poultry to the burden of campylobacteriosis. Three different models were used: Dutch model, modified Hald model and Island model. All models suggest that the majority of cases (from March, 2005 – February, 2008) could be attributed to poultry (French, 2008). Given the conclusions of international and Irish studies on the link between campylobacteriosis and poultry, it can be concluded that there is compelling evidence that poultry is an important source and most likely the principal source of campylobacteriosis in Ireland. It is therefore recommended that the poultry industry develop its own voluntary code of practice taking the guidance in this report into consideration.

2.1.1 Note on identification of *Campylobacter* spp. to species level in relation to source attribution

Many of the Irish clinical laboratories do not identify campylobacter isolated from humans to species level, or do not identify them by robust methods, as this is not immediately relevant to management of the patients' illness. The Scientific Committee acknowledged that the traditional biochemical test is unreliable, but noted that there is an effective PCR (polymerase chain reaction) method. The issue of accurate identification to species level is important for source attribution. Where clinical laboratories do not have the capacity to perform identification to species level by a reliable method, isolates could be sent to a reference laboratory for speciation and typing. It has been demonstrated that differences in risk factors exist for different *Campylobacter* species, suggesting that the aggregation of these bacterial species, as done in most case-control studies, may mask important species-specific risk factors (EFSA, 2010d; Doorduyn *et al.*, 2009; Gillespie *et al.*, 2002). Consequently, in its opinion, EFSA recommended that future studies on risk factors for campylobacteriosis take into account differences in the ecology of *Campylobacter* species, and subtypes within species (EFSA, 2010d).

It is recommended the HSE and DAFF's National Reference Laboratory for *Campylobacter* spp. (for food, feed and animal health) coordinate activities to ensure that identification to species level and genotyping of a subset of human, food and animal isolates is undertaken using appropriate methods to assist with source attribution.

2.2 RISK MANAGEMENT QUESTIONS 2 AND 3

Q2. What microbiological criteria for campylobacter could be applied to results of analyses of poultry for *Campylobacter* spp. that would reduce the risk of campylobacteriosis in humans and could be used as part of a statutory/voluntary control programme as a trigger for actions aimed at reducing the contamination of broilers in a particular production/processing facility.

Q3. How should such a microbiological criterion be applied in practice to monitor campylobacter contamination rates on broiler carcasses?

Pre-harvest and post-harvest microbiological criteria are recommended in response to these questions to reduce the risk of campylobacteriosis in humans. Interventions on-farm and in the slaughterhouse should be taken when these criteria are exceeded.

A pre-harvest microbiological criterion of $\leq 7 \log_{10}$ cfu/g of *Campylobacter* spp. in 10 pooled caecal contents is recommended. Sampling should be carried out on-farm seven days (or less if feasible) before slaughter.

A post-harvest target of level of $\leq 4 \log_{10}$ cfu/g of *Campylobacter* spp. on neck skin samples taken post-chill is recommended, with a sampling plan and limits of $n=5$, $c=1$, $m=4 \log_{10}$ cfu/g and $M=5 \log_{10}$ cfu/g.

These criteria should be validated by a pilot study and should be subject to periodic review based on progress made by the industry when the control programme is in place. It is recommended that data generated are collated in a central database to facilitate trend analysis.

Reducing the contamination of broilers and the subsequent risk to human health can be achieved by reducing the concentration of campylobacter in the intestines of broilers on-farm and reducing the concentration of campylobacter on the surface of processed chickens in the slaughterhouse.

A number of studies have identified a relationship between the concentration of campylobacter in the intestines of colonised birds and the levels found on the surface of carcasses. It has been reported that campylobacter levels in caeca (paired blind pouches forming the beginning of the large intestine) are approximately 1 log₁₀ higher than faecal levels (Nauta *et al.*, 2007). Reich *et al.* (2008) investigated the effect of concentrations of *Campylobacter* spp. in the caeca of broilers on contamination levels on carcasses during slaughter and processing. A positive correlation was found between high levels of colonisation in the caeca and concentrations on corresponding carcasses and portioned products. Rosenquist *et al.* (2006) documented a correlation between campylobacter concentration in intestinal content and on chicken carcasses after defeathering. This finding implies that the mean concentrations in neck skin samples after defeathering are closely related to the mean concentration in the intestinal samples. The mean number of campylobacter per gram in positive intestinal samples ranged from 4.74 log₁₀ cfu/g to 8.2 log₁₀ cfu/g, compared to the mean number of campylobacter per gram in neck skin after defeathering which ranged from (1.90 log₁₀ cfu/g – 3.93 log₁₀ cfu/g). Similar observations were made for both slaughterhouses included in this study irrespective of differences in scalding and defeathering practices. Based on concentration data, it was extrapolated that the concentration on neck skin (after defeathering) was 4.2 log₁₀ cfu/g less than levels recovered in the intestines (Rosenquist *et al.*, 2006).

A UK study found that following processing, all carcasses from fully colonised flocks were contaminated with campylobacter and fully colonised flocks had significantly higher numbers per carcass (average of 5.3 log₁₀ cfu; range 1.3 to >8.0 log₁₀ cfu) than carcasses originating from low prevalence flocks (average of 2.3 log₁₀ cfu; range <1.1 to 4.1 log₁₀ cfu). The study also showed that during processing, cross-contamination from previously processed flocks was significant, especially on carcasses of low prevalence flocks (Allen *et al.*, 2007a).

A review of risk assessments models used in Denmark, the Netherlands, UK and Germany indicates that a change in flock prevalence is expected to have a linear effect on human health risk. The review also concluded that a reduction in the concentration of campylobacter on carcasses after evisceration is found to be a more effective intervention measure than prevalence reduction (Nauta *et al.*, 2009). The Danish risk assessment carried out simulations designed to predict the effect of different mitigation strategies. The simulation showed that the incidence of campylobacteriosis associated with consumption of chicken meals could be reduced 30 times by introducing a 2 log₁₀ reduction of the number of *Campylobacter* spp. on chicken carcasses. To obtain a similar reduction of the incidence, the flock prevalence should be reduced approximately by a factor of 30 or the kitchen hygiene improved by a factor of 30 (Rosenquist *et al.*, 2003).

These studies demonstrate that a reduction in the concentration of campylobacter on the surface of the chicken is, in part, dependent on (i) the initial concentration of campylobacter in the chicken's intestines; and also on (ii) cross-contamination during processing. Therefore, in order to reduce the human health risk, an approach involving both the reduction of levels in the intestines of broilers on-farm and the reduction of campylobacter concentration on the surface of broilers in the slaughterhouse is recommended.

Consistent with the recommendation on identification and typing of human isolates for the purpose of source attribution it is recommended that a subset of carcass isolates are identified to species level and genotyped by the same methods applied to human isolates. This will provide data for comparison with human data in source attribution studies.

2.2.1 Rationale for pre- and post-harvest criteria chosen

Applying a microbiological criterion close to the end point of the food chain is recommended because this point in the processing chain is relatively close to the stage of human exposure (Nauta *et al.*, 2008). A post-harvest target approach was successfully used in New Zealand leading to significant reductions in the numbers of human campylobacter infections. A moving window target of 3.78 log₁₀ cfu was set for carcass rinsates with a 98th percentile target of 5.88 log₁₀ cfu/carcass rinsate and a quarterly at median value of 4.16 log₁₀ cfu/carcass rinsate.

A **post-harvest** target level of ≤4 log₁₀ cfu/g on neck skin samples, taken post-chill, is recommended following consideration of the Irish results of the EFSA baseline study (EFSA, 2010c), specifically taking into account the data for concentrations on broilers, where 4 log₁₀ represents the 86th percentile on a distribution of *Campylobacter* spp. concentrations on Irish broiler neck skin samples. To assess achievement of this ≤4 log₁₀ cfu/g target, the following microbiological criterion sampling plan and limits are proposed:

n=5 samples; c=1 sample between m and M; m=4 log₁₀ cfu/g; and M=5 log₁₀ cfu/g

Where n = number of units comprising the sample and c = number of sample units giving values between the limits m and M. An assessment of the performance of this criterion was made using Montecarlo simulation software (see Appendix 2).

A **pre-harvest** criterion was considered important for two reasons: (i) to provide producers with feedback on the effectiveness of their control measures and (ii) to enable the slaughterhouse to implement a scheduling approach to slaughter. Scheduled slaughter, which is practiced in some countries, e.g. Norway, Iceland and Denmark, means identifying flocks that are positive for *Campylobacter* spp. before they are slaughtered and subjecting carcasses from these flocks to treatments or interventions which can reduce campylobacter counts. Given the high prevalence in Irish broiler flocks currently, a scheduling approach is recommended, whereby carcasses from flocks which exceed the pre-harvest criterion are subjected to treatments/interventions which can reduce campylobacter counts.

A pre-harvest criterion of $\leq 7 \log_{10}$ cfu/g in pooled caeca is recommended and is intended to relate to the target agreed for the post-harvest criterion. Caecal samples are recommended to facilitate consistency with the methodology used in the EFSA baseline study (2010c). It is accepted that carcass contamination arises as a result of faecal contamination of the bird during processing and from faecal contamination of the surface of the bird during rearing, transportation and lairage. It is important to note that caecal levels of campylobacter are reported to be approximately $1 \log_{10}$ higher than faecal levels (Nauta *et al.*, 2007). Based on the Dutch risk assessment (Nauta *et al.*, 2005), processing is expected to yield approximately a $2 \log_{10}$ reduction from the initial campylobacter concentration in faeces, prior to slaughter, to the final concentration on the dressed carcass. As a post-harvest target of $4 \log_{10}$ cfu/g is being recommended on the dressed carcass, therefore a target level of $6 \log_{10}$ cfu/g would be set for faecal samples. But as the pre-harvest criterion is being set for caecal samples (to be consistent with the EFSA baseline study) and caecal levels are reported to be approximately $1 \log_{10}$ higher than faecal levels, the recommended pre-harvest criterion of $7 \log_{10}$ cfu/g in pooled caecal samples is recommended.

It is accepted that there are many variables which can influence the average \log_{10} reduction of contamination on broiler carcasses and a reduction of $3 \log_{10}$ (between the concentration in the caeca and the final concentration on the dressed carcasses) may not be consistently achieved. The level on the carcass after chilling depends on the prevalence and distribution of concentrations within and between flocks, in both the faeces and on the exterior of the birds prior to slaughter. A three-class attribute plan was chosen for the post-harvest criterion to allow for this.

The recommended pre- and post-harvest microbiological criteria should be validated by a pilot study and should be subject to periodic review based on progress made by the industry when the control programme is in place. It is recommended that data generated are collated in a central database to facilitate trend analysis (see Appendix 2).

2.2.2 Pre-harvest criterion

Microbiological criteria could be applied to reduce the risk to humans at the pre-harvest level by setting a quantitative limit for *Campylobacter* spp. in the caeca of a random sample within a flock prior to the first depopulation or thinning. A limit of $\leq 7 \log_{10}$ cfu/g in pooled caecal samples is recommended. Sampling should be carried out on-farm seven days (or less if feasible) before slaughter. Testing of each house should be performed by randomly selecting 10 birds from various locations within the house which will be harvested and dispatched to the laboratory on the same day. The caecal contents of the 10 birds will be pooled for analysis in the laboratory. Enumeration of campylobacter in the pooled samples will be carried out by a laboratory with accreditation specifically for the international standard method for the enumeration of *Campylobacter* spp. (ISO/TS 10272-2:2006 or equivalent). The laboratory should be capable of demonstrating reproducibility of their testing methods and carrying out testing to a high standard. Participation and good performance in ring trials would be required. The testing laboratory should submit the results to the central database.

Such an approach will generate ongoing farm-specific data for each crop of birds and enable stakeholders (farmers, processors and regulators) to assess the effectiveness of the controls from a sample taken prior to the biosecurity breach of thinning. This approach would also permit the processor to arrange the logistics of a scheduling approach to the slaughter of flocks exceeding the criterion.

The efficacy of a scheduling approach is dependent on the testing of flocks in advance, and its effectiveness can be affected by limitations associated with the testing programmes implemented. The ideal scenario would be the availability of a rapid test, where the infection status of a flock could be ascertained on the day of sampling, which would provide a low limit of detection (ability to detect low numbers of campylobacter) and high specificity. The availability of such tests is limited or may not provide the required level of performance. Havelaar *et al.*, 2007 estimated that 50-75% of infected flocks would be correctly identified by culture if tested one week in advance of processing, and this monitoring would be effective if combined with scheduling. Therefore, an approach involving testing of samples seven days (or less if feasible) before slaughter is recommended. This approach will be reviewed based on the availability of a rapid test for use on-farm with satisfactory limits of detection and specificity.

Exceeding the criterion will invoke a number of interventions at farm level including a review of biosecurity (see Section 2.3.1). A scheduling approach towards slaughtering chickens from these flocks would be adopted in the slaughterhouse (see Section 2.3.2). A diagram of the actions to be taken depending on the pre-harvest result obtained is outlined in Figure 1.

It is recommended that an incentive scheme be adopted by the industry to reward high standards of biosecurity and compliance with the pre-harvest microbiological criterion. The incentive scheme could take the form of a bonus and/or penalty scheme based on producer performance as demonstrated by the pre-harvest sampling. Repeated breaches of the pre-harvest criterion should result in removal from the Bord Bia Quality Assurance scheme.

2.2.3 Post-harvest criterion

A post-harvest target of level of $\leq 4 \log_{10}$ cfu/g of *Campylobacter* spp. on neck skin samples taken post-chill is recommended, with a sampling plan and limits of $n=5$, $c=1$, $m=4 \log_{10}$ cfu/g and $M=5 \log_{10}$ cfu/g (see Appendix 2 for estimated criterion performance).

This criterion is considered to have been met (i.e. a satisfactory result) when:

- All 5 samples $\leq 4 \log_{10}$ cfu/g or
- One of the 5 samples $> 4 \log_{10}$ cfu/g and $\leq 5 \log_{10}$ cfu/g and the other 4 samples $\leq 4 \log_{10}$ cfu/g

This criterion is considered not to have been met (i.e. unsatisfactory result) when:

- Any of the 5 samples $> 5 \log_{10}$ cfu/g or
- More than one of the 5 samples $> 4 \log_{10}$ cfu/g and $\leq 5 \log_{10}$ cfu/g

Sampling in the processing plant should be confined to broilers from those farms whose pre-depopulation sampling results are positive and have levels that are $\leq 7 \log_{10}$ cfu/g. This sampling approach is recommended to verify that flocks with caecal levels of campylobacter $\leq 7 \log_{10}$ cfu/g meet the post-harvest criterion. This sampling provides a measure of process hygiene in the slaughterhouse. It is considered unnecessary to sample broilers in the slaughterhouse from flocks whose pre-depopulation sampling levels are $> 7 \log_{10}$ cfu/g because it is assumed that these carcasses would not meet the post-harvest criterion and as such, they have been flagged for additional control measures (see Section 2.3.2). Sampling of these carcasses would therefore represent an unnecessary burden of sampling and costs.

Post-harvest sampling should be carried out once a week in the slaughterhouse. One positive flock (with pre-harvest levels $\leq 7 \log_{10}$ cfu/g) should be selected for sampling. Sampling should take place after chilling and prior to further processing. It is recommended that sampling should be carried out when approximately half of the flock have been slaughtered. The time of day and frequency of sampling will depend on when these particular flocks are slaughtered. Fifteen carcasses should be sampled. A piece of approximately 10g of neck skin shall be obtained from each carcass. The neck skin samples from three carcasses shall be pooled before examination in order to form 5×25 g final sampling units. The samples should be dispatched to a suitable laboratory (as outlined in Section 2.2.2) for isolation and enumeration of *Campylobacter* spp. It is recommended that a pilot study should be conducted to validate this criterion.

Breaches of the post-harvest criterion would require a review of hygiene practices in the slaughterhouse and on-farm, including biosecurity. A diagram of the actions to be taken depending on the post-harvest result obtained is outlined in Figure 2. The processor should carry out trend analysis of the post-harvest results (see Appendix 2).

A comprehensive review of slaughterhouse procedures and sourcing of broilers is necessary where repeated breaches of the criterion occur (more than four breaches in eight batches sampled). An overview of good hygiene practices during processing is provided in Appendix 5. Where persistent, repeated breaches of the post-harvest microbiological criterion occur, further interventions may be necessary and it is recommended that freezing is considered in these cases (see Section 2.3.2).

Figure 1. Proposed Pre-harvest Microbiological Criterion set for 10 Pooled Caecal Samples from each Flock (subject to validation by pilot study)

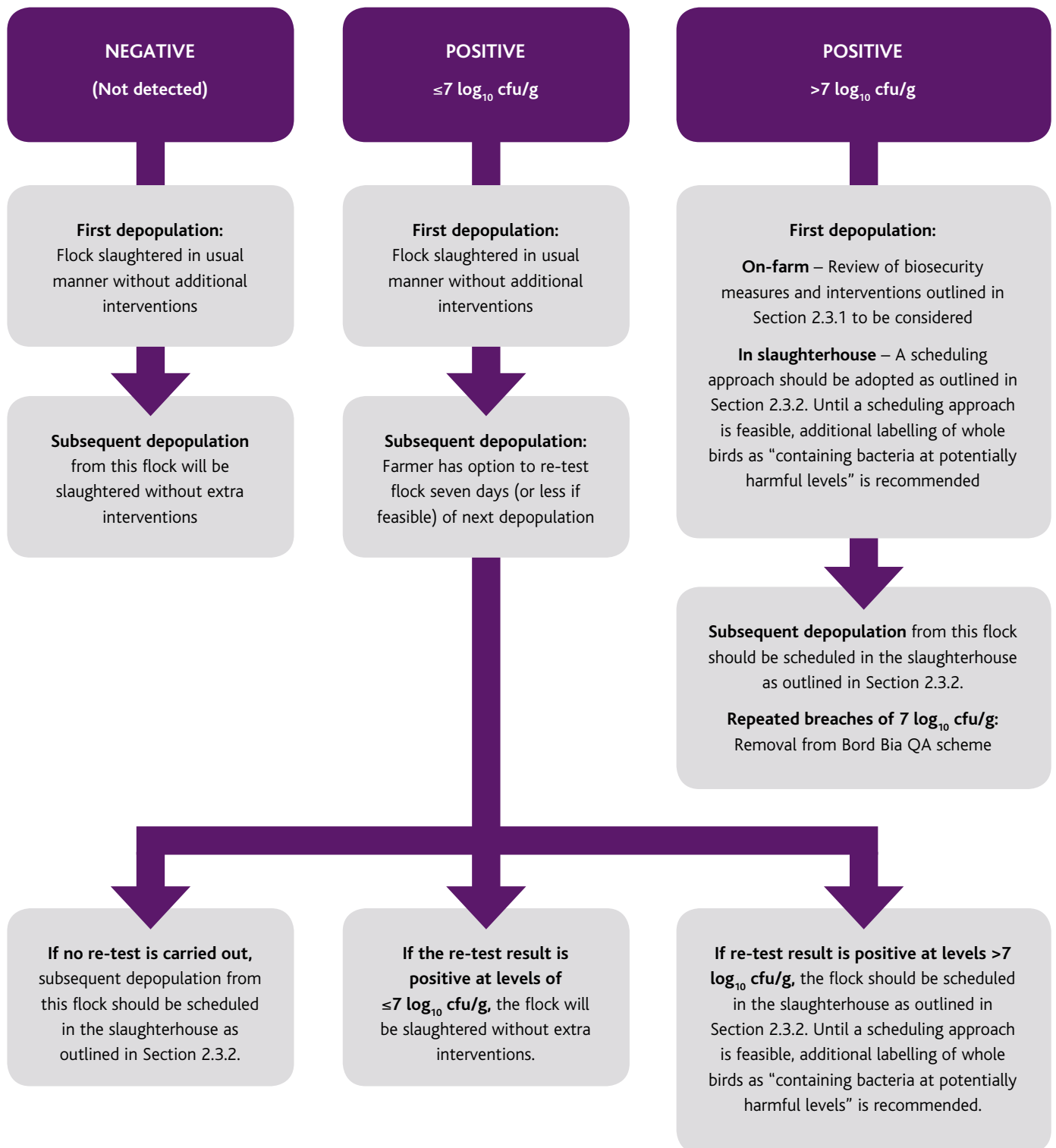
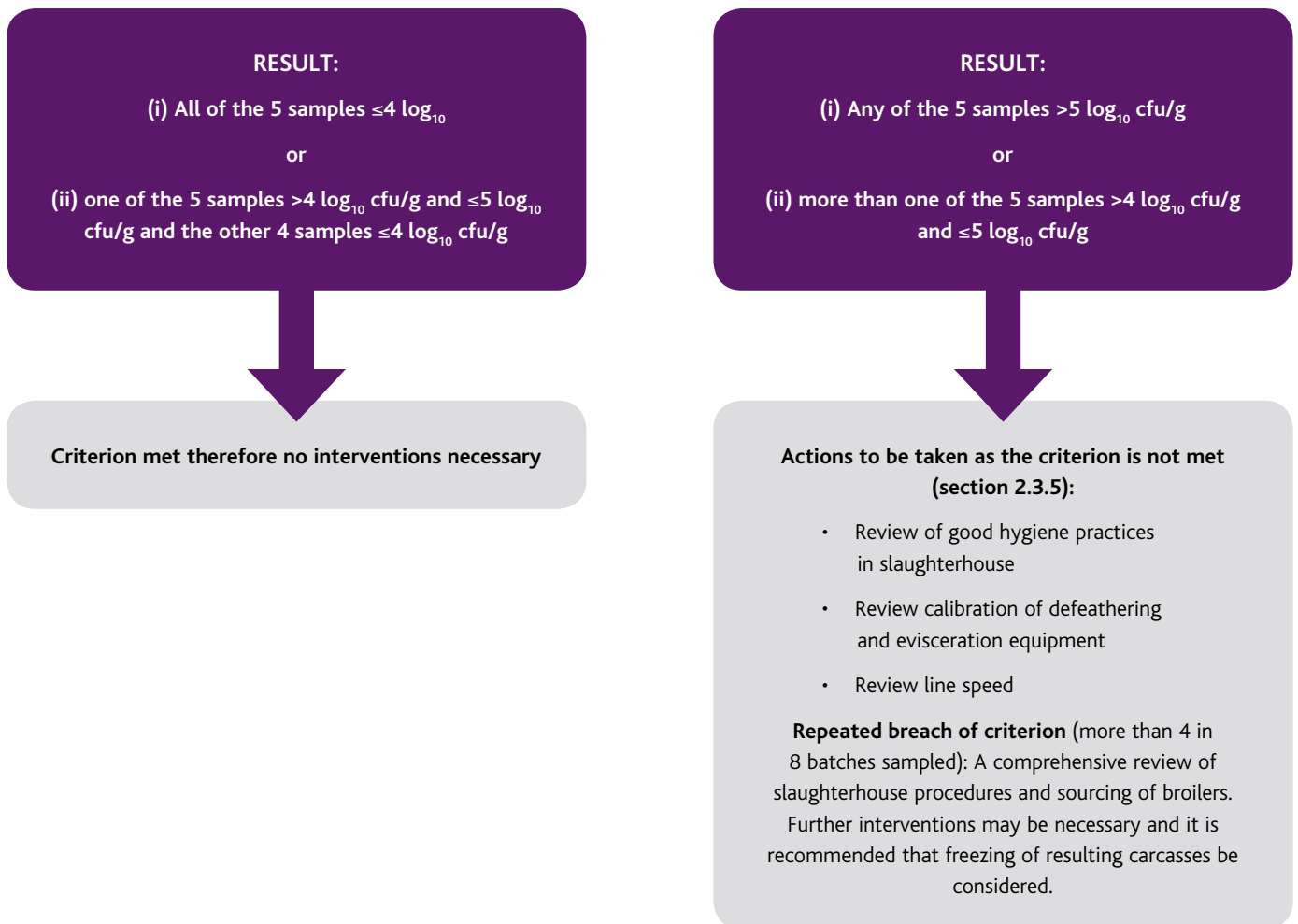


Figure 2. Proposed Post-harvest Microbiological Criterion Set for Neck Skin Samples (subject to validation by pilot study)

Post-harvest sampling should be conducted once a week on a carcass from a flock whose pre-harvest result is positive and $\leq 7 \log_{10}$ cfu/g.

Sampling plan and limits: n=5 samples; c=1 sample between m and M; m= $4 \log_{10}$ cfu/g; and M= $5 \log_{10}$ cfu/g (for details see section 2.2.3 and Appendix 2)



2.3 RISK MANAGEMENT QUESTION 4

Q4. What are the practical and legally permissible measures that processors and producers could implement to reduce campylobacter contamination on broilers if the microbiological criteria were exceeded?

In order to achieve the recommended pre- and post-harvest criteria, the poultry industry should develop and implement its own voluntary code of practice, taking the guidance in this report into consideration.

It is recommended that a **campaign to improve producers' and catchers' understanding of biosecurity measures**, and their importance, is developed and delivered to coincide with the introduction of the voluntary campylobacter control programme. Processors are responsible for the sourcing of their raw materials and for the food they produce and they should therefore be involved in this campaign with support from the FSAI and DAFF.

It is recommended that an **incentive scheme** is implemented by the processors to encourage compliance with biosecurity measures on-farm. The incentive scheme may take the form of a bonus or penalty system. In Denmark, the implementation of an incentive scheme has resulted in increased compliance with biosecurity by producers. The Danish system involves an annual audit of biosecurity measures on-farm. Findings of non-compliance during the audit result in a financial penalty applied per kilo of live bird until the non-compliances are closed out satisfactorily. Sampling of the broiler house prior to depopulation and cloacal sampling at the slaughterhouse is also undertaken and if either sample is positive a penalty is also enforced. It is interesting to note that under the Danish control programme, biosecurity breaches incur a higher penalty per bird than a positive sample result.

2.3.1 Measures on-farm in the event of the pre-harvest criterion being exceeded

If the campylobacter count in the pooled caecal sample exceeds 7 log cfu/gram, on-farm controls should be reviewed and implementation of additional measures (outlined in this section) should be considered. Biosecurity is a very important step in preventing campylobacter colonisation of poultry flocks. Preventing the introduction of campylobacter into the broiler house or delaying its introduction for as long as possible will impact significantly on the caecal counts of campylobacter. It is important that **processors continually emphasise the importance of biosecurity to producers and catching teams**. In the event of the caecal samples exceeding 7 log₁₀ cfu/g, a **full review of biosecurity measures should take place**. Strict adherence to a high standard of biosecurity is required as a minimum. There are a number of measures relevant to campylobacter in the Bord Bia Quality Assurance Standard (Bord Bia, 2008). These are highlighted in Appendix 3. **In addition to ensuring that the Bord Bia quality assurance standard requirements are met, implementation of one or a combination of the following measures will lead to greater reductions in levels of campylobacter in flocks:**

- **Personnel access and hygiene:** Boot changes before entering the broiler house have been shown to be an effective measure when combined with other biosecurity measures (Pattison, 2001). Van de Giessen *et al.* (1998) demonstrated a reduction in flock prevalence from 66% to 22% when boot changes and other biosecurity measures were implemented for personnel entering the broiler house. A system of boot changes and hygiene measures in the anterior room as outlined in Appendix 4 is recommended in preference to the boot dipping system currently practiced on many Irish broiler farms. These measures include dividing the anteroom into three zones using 40 cm barriers – 1. An unclean zone where outside clothes are removed; 2. A changing zone where hand-washing (lasting at least 10 seconds) is undertaken and dedicated inside clothes are put on and 3. A clean zone where dedicated inside boots are put on. Once a hygiene barrier system is in place, it requires less effort to use routinely than maintaining and using an effective boot dipping system. Boots with deep cleats tend to hold litter, and unless this compacted litter is removed from the cleats (best done by using a brush) before dipping, it is unlikely that disinfectants will penetrate through the litter to kill all of the organisms present. Contact time between the disinfectant and the boot is critical to ensuring adequate disinfection. Adequate contact time may be difficult to achieve in practice. **Mixed farms:** Farms with mixed animal species also run the risk of increased flock infection because strains of campylobacter found in cattle and pigs can also be isolated from chickens (Jacobs-Reitsma *et al.*, 1995, Nesbit *et al.*, 2001). It is recommended that broiler farmers who also farm other animal species observe the above guidelines on personnel access and hygiene, particularly when carrying out routine husbandry procedures on their farm
- **Personnel access and hygiene during non-routine events:** It is likely to be the non-routine events, such as fan or power failure, or use of relief staff that result in biosecurity procedures being ignored. Producers themselves should never let their guard down, even when potentially distracted by a problem which needs urgent attention. They should ensure that all visitors (relief staff, maintenance workers) are not sources of contamination. Poultry producers should consider keeping dedicated maintenance tools (which would be used regularly in the house) in the *ante-room* to each house

Recommendations for a Practical Control Programme for *Campylobacter* in the Poultry Production and Slaughter Chain

- **Focus on effective implementation of biosecurity:** In countries that practice strict biosecurity, low prevalence in flocks has been achieved. Biosecurity measures are currently applied on Irish farms, however, it appears that their application and effectiveness varies between farms. The challenge is twofold – to improve physical biosecurity and to ensure that operational biosecurity is employed 100% of the time. Effective biosecurity requires: (i) installation of physical hygiene barriers, e.g. *ante-rooms* with two boot changes; (ii) training to ensure that existing measures are consistently and effectively applied, e.g. that farmers always wash their hands every time they enter the house; and (iii) monitoring by processors and third party auditors that every measure (including those for chick placement and thinning) is being consistently applied. Training of producers and provision of booklets and signage, e.g. to be displayed in the *ante-room*, could be used to reinforce the need for 100% compliance with biosecurity measures. See booklet and poster from FSA, UK – <http://www.food.gov.uk/safereating/microbiology/flocks/>
- **Surrounds of broiler house:** Genotyping studies have shown that isolates of *Campylobacter jejuni* located outside the broiler house of negative flocks had the same genotypes as isolates subsequently recovered from the flock (Hielt *et al.*, 2002; Newell *et al.*, 2001). Clean and intact concrete aprons can reduce the risk of carrying contaminated material into the broiler house (Newell and Fearnley, 2003). The Bord Bia Standard (Bord Bia, 2008) requires the area around the broiler house to be kept tidy and free of vegetation. In Denmark, there are similar requirements in terms of vegetation and cleanliness but also additional recommendations: A concrete apron (maintained in good condition) is recommended in front of the doors to the house and it is recommended that pebbles/gravel are placed along the sides of the houses to permit drainage, prevent dust circulation and deter rodents, as they do not like the surface (Report of an international expert consultation, Denmark, 2007). The gravelled area is expected to be a minimum of one metre in width, with a vegetation-free area extending three metres from the gravelled area. Good drainage is important because puddles can harbour campylobacter. It is therefore recommended that a concrete apron is put in place outside the entrance to broiler houses and robust measures are taken to eliminate vegetation from the area immediately surrounding the broiler house (see Appendix 4)
- **Fly control:** In Norway, campylobacter have been detected in 50.7% of flies sampled in the autumn in the environs of poultry rearing units (Rosef and Kapperud, 1983). A feature of the epidemiology of campylobacter in broiler flocks is its pronounced and consistent seasonal pattern. EFSA (2010d) identified that there is increased risk of campylobacter-contaminated carcasses and campylobacter-colonised broiler batches during certain months of the year, with the period of July-September being the quarter most at risk. The seasonality observed may be associated with temperature related factors, increased ventilation of broiler houses during these months and increased numbers of insects during the summer and autumn months (WHO 2009). Studies in Iceland and Denmark, where fly screens were applied to broiler houses, showed significant decreases in the number of positive flocks. In the Danish study, 51.4% of control flocks (without fly screens) were positive compared to 15.4% of flocks housed in units fitted with fly screens (Hald *et al.*, 2007). In Iceland, a 62% reduction of infection in flocks was reported following the use of fly screens (Lowman *et al.*, 2009). Wherever possible, fitting of fly screens is recommended, particularly in high prevalence flocks. Alternative methods of fly and insect control may be appropriate where fly screening is not possible
- **Biosecurity during preparation and stocking of house:** Machinery used to deliver bedding or chicks may be a source of contamination of the broiler house. It is recommended that the truck ramp is thoroughly cleaned and then disinfected and that the ramp is placed very close to the entrance of the house to ensure that the modules carrying the chicks are not a source of contamination. As per recommendations for all personnel entering a broiler house, it is important that those delivering the chicks wear clean protective clothing and clean, disinfected boots prior to entry into the broiler house
- **Reduction of slaughter age:** Studies show a higher risk of campylobacter-colonisation associated with increasing age at slaughter (Barrios *et al.*, 2006; Berndtson *et al.*, 1996b; Bouwknecht *et al.*, 2004; Evans and Sayers, 2000; Hansson *et al.*, 2010a; McDowell *et al.*, 2008). In Iceland, during peak periods of campylobacter infection, birds are slaughtered at <34 days (presented at FSA UK international meeting). Taking the association between increasing age and risk of colonisation into account, a policy of slaughtering flocks at a younger age particularly during the summer months, may lead to a reduction in the prevalence of campylobacter on broilers and is recommended for flocks with persistently high concentrations of *Campylobacter* spp. in the caecal contents
- **Disposal and storage of waste:** Campylobacter survival in broiler litter has been determined to have a D value (the time required at a certain temperature to kill 90% of a particular organism) of 2.53 days (based on spread on grass pasture in summertime; Hutchinson *et al.*, 2005). Therefore, adequate storage and disposal of broiler farm waste is important to eliminate the potential of waste being a source of contamination for subsequent flocks. Waste storage is also an important consideration for mixed farms (poultry farms where other species of animals are farmed in close proximity to the broiler houses). Studies show that the prevalence of campylobacter in cattle ranges from 0 to 80% (Munroe *et al.*, 1983; Rosef and Kapperud *et al.*, 1983; Giacoboni *et al.*, 1993). An Irish study of feed lot cattle demonstrated a campylobacter prevalence of 54% (Minihan *et al.*, 2004). Domestic animals and pets can also be a significant

reservoir of infection for broilers. This is a problem particularly in the summer months when flies can act as vehicles for the spread of campylobacter. Careful management of waste from all species of animals is recommended

- **Sexing of birds prior to placement:** Variability in the size of broilers creates problems in the slaughterhouse, particularly in relation to plucking and evisceration (see Appendix 5). Consideration could be given to sexing of flocks and slaughter of flocks based on sex which will permit greater size consistency of birds at time of slaughter

The following measures may provide additional options for reducing campylobacter levels in broilers prior to slaughter. Extensive studies on the effectiveness of these controls have not been completed to date:

- Addition of 0.44% lactic acid to drinking water during the period of feed withdrawal: Crop contamination with campylobacter was significantly reduced by lactic acid treatment (campylobacter was detected in 62.3% (109/175) of crops of treated broilers compared to detection in 85.1% (149/175) of controls $P \leq 0.001$; Byrd *et al.*, 2001)
- Addition of organic acids to the feed: has been shown to reduce campylobacter colonisation. Skanseng *et al.* (2010) found little or no effect on *Campylobacter jejuni* colonisation levels in chickens that were given feed supplemented with formic acid alone. A combination of 1.5% formic acid and 0.1% sorbate reduced the colonisation of *Campylobacter jejuni* significantly while a concentration of 2% formic acid in combination with 0.1% sorbate prevented *Campylobacter jejuni* colonisation in chickens. The data suggest that growth rate may be impaired by supplementation of the feed with 2% formic acid

2.3.2 Measures in the slaughterhouse for flocks which have exceeded the pre-harvest criterion

A scheduling approach to the slaughter of broilers from flocks which have exceeded the pre-harvest microbiological criterion (i.e. $>7 \log_{10}$ cfu/g) is recommended, whereby carcasses from these flocks are subjected to treatments or interventions (outlined below) which can reduce campylobacter counts. It may take a period of time to introduce and establish the scheduling approach in the slaughterhouse. In the interim, it is recommended that whole birds from flocks which have exceeded the pre-harvest microbiological criterion ($>7 \log_{10}$ cfu/g) are clearly labelled:

"containing bacteria at potentially harmful levels"

One, or a combination of the following interventions, can be used to reduce the concentration of *Campylobacter* spp. on the final product:

- **Freezing of chicken:** This control measure has been shown to effectively reduce campylobacters on carcasses between 1.3 to 2.2 logs in several studies (Georgsson *et al.*, 2006; Rosenquist *et al.*, 2006; Rosenquist *et al.*, 2008; Boysen and Rosenquist, 2009; El-Shibiny *et al.*, 2009; Loretz *et al.*, 2010; Sampers *et al.*, 2010). This measure is used as a control in Norway (Hofshagen and Kruse, 2003); Iceland (Reiersen *et al.*, 2003) and Denmark. In Ireland, the market for frozen product is relatively small when compared to the fresh poultry market. Where possible, the processor should ensure that flocks with $>7 \log_{10}$ cfu/g (rather than negative or $\leq 7 \log_{10}$ cfu/g flocks) are used to produce frozen product. It is also recommended that where other interventions have failed, or in the case of repeated breaches by the producer of the pre-harvest microbiological criterion, freezing of resulting carcasses should be considered
- **Slaughtering equipment:** It is recommended that processors ensure defeathering and evisceration equipment is appropriately adjusted to the size of the broilers before processing. Monitoring of the number of evisceration failures (i.e. intestinal rupture as a percentage of birds slaughtered within each batch) will highlight problems with the alignment of equipment and permit corrective action to be taken (see Appendix 5 for further details)
- **Crust-freezing of chicken:** Crust-freezing is a technique for rapid chilling of meat. The technique is based on rapid ice crystallisation on the meat surface, resulting in a thin frozen crust followed by temperature equalisation. The CO₂ crust-freezing technique has been shown to produce consistent reductions (approx. $0.5 \log_{10}$) in campylobacter concentrations (Corry *et al.*, 2003). Boysen and Rosenquist, (2009) showed a reduction in numbers on the carcass of $0.42 \log_{10}$. Published evidence shows that crust-freezing has little adverse effect on meat quality and the process known as 'deep or 'super' chilling has been commonly used in the USA where chicken meat is stored close to its freezing point at -1 to -2°C (James *et al.*, 2007; Jul, 1986). James *et al.* (2007) investigated the effects of physical decontamination of poultry carcasses using steam or hot water in combination with rapid cooling, chilling and freezing. The optimum combination of treatments reported was treatment with water at 80°C for 20 seconds followed by crust-freezing. This combination resulted in a reduction in the numbers of *Campylobacter jejuni* of $2.9 \log_{10}$ cfu/cm² without extensive degradation of the carcass appearance. Chicken meat which has undergone the process of crust-freezing can be sold as fresh meat, provided the temperature of the meat remains greater than or equal to -2°C (Regulation (EC) No. 543/2008). Crust-freezing is recommended as an intervention which should be considered by processors

Recommendations for a Practical Control Programme for *Campylobacter* in the Poultry Production and Slaughter Chain

- **Removal of skin from chicken:** Most campylobacter contamination is located on the skin; therefore skinless products have counts several logs lower than the respective meat products with skin (Uyttendaele *et al.*, 1999). *Campylobacter jejuni* is retained in a liquid film on the skin and becomes entrapped in skin ridges and crevices. In this way, chicken skin provides a micro-environment suitable for the survival of *Campylobacter jejuni* (Chantarapanont *et al.*, 2003). A study by Sampers *et al.* (2008) found that the presence or addition of skin during production of chicken meat preparations resulted in almost a 2.2 fold increase in the probability of a sample being positive for campylobacter. Davis and Conner, (2007) found that populations of campylobacter remained consistently higher (0.4 to 0.9 log₁₀ cfu/g) on skin versus meat. Consideration could be given to the removal of skin as an intervention to reduce the concentration of campylobacter on the surface of chicken
- **Logistical approach to slaughter:** Logistic slaughter is an intervention measure intended to reduce cross-contamination during slaughter by slaughtering positive flocks after negative flocks (Evers, 2004). Given the high prevalence in Irish broiler flocks currently, logistic slaughter is not feasible. However, it is recommended that a logistical approach to slaughter is adopted whereby broilers from these farms (with higher counts of *Campylobacter* spp. in the caeca) will be slaughtered later in the day, after flocks with caecal counts below the criterion limit (although not necessarily negative) are slaughtered. While a number of studies have shown that logistic slaughter (i.e. where positive flocks are slaughtered after negative flocks) does not significantly impact on human health risk (Nauta and Havelaar, 2008; Tandrup *et al.*, 2009), reducing the level of cross-contamination of campylobacter-free flocks or flocks with low levels of campylobacter carriage by heavily contaminated flocks has been shown to reduce the final carcass burden of campylobacter (Allen *et al.*, 2007a; Reich *et al.*, 2008). Following the baseline survey on the prevalence of campylobacter in broiler batches and of campylobacter and *salmonella* on broiler carcasses in the EU in 2008, EFSA analysed the risk factors associated with campylobacter colonisation of broiler batches and campylobacter contamination of broiler carcasses. This analysis found that there is an association between campylobacter contamination on broiler carcasses and the colonisation status of the broiler batches (EFSA, 2010d). This highlights the need to limit the extent of cross-contamination in the slaughterhouse between broiler batches with a heavy burden of campylobacter and batches which are free or have low levels of campylobacter
- **Hot water treatment of carcasses:** Corry *et al.* (2007) showed a decrease of 1.66 log₁₀ cfu/cm² following hot water immersion treatment at 75°C for 30 seconds. The physical changes on the chicken were not considered generally unacceptable. A study by Purnell *et al.* (2004) reported reductions in campylobacter contamination of carcasses following a hot wash at 70°C for 40 seconds, without detrimentally affecting the chicken skin. Hot water treatment of carcasses is therefore recommended as a potential intervention to reduce concentration of campylobacter on carcasses
- **Steam treatment of carcasses:** The main advantage of using steam is the large amount of heat transferred to the food when steam condenses (James *et al.*, 2007). In addition, steam can penetrate cavities, crevices and feather follicles (Morgan *et al.*, 1996). Whyte *et al.* (2003) reported a 1.3 log₁₀ cfu/g decrease in campylobacter numbers following exposure of carcasses to 90°C atmospheric steam for 12 seconds. Significant campylobacter reduction and maintenance of product quality has been difficult to achieve with steam treatments because of the denatured appearance of the skin or meat surface (James *et al.*, 2007; Whyte *et al.*, 2003)
- **Combined steam and ultrasound:** This treatment was investigated by Boysen and Rosenquist, (2009). Reductions of ≥2.52 log₁₀ cfu/carcass were reported. An adverse effect of this technique was a slightly boiled appearance of the carcass. The steam and ultrasound system of carcass treatment has been developed and used in Denmark. The issues surrounding the boiled appearance of the chicken after treatment have been resolved since its introduction. Achieving significant reductions in contamination levels on naturally infected carcasses has been a challenge in practice and further investigation of this treatment may be necessary
- **Diversion of carcasses to value added products:**

The following processes could be considered as interventions to reduce campylobacter on chicken. It is important to note that chicken which has undergone marinating or heat treatment is no longer considered fresh meat and must be marketed as a meat preparation or meat product respectively

 - **Marinating** is the process by which a brine solution composed of water, food ingredients, spices, salt and acids are used as marinade for broiler fillets. Birk *et al.* (2010) reported a 1.2 log₁₀ unit reduction in campylobacter on chicken fillets after three days of storage in low pH marinades (pH < 3). Perko-Makela *et al.* (2000) reported the survival of *Campylobacter jejuni* was similar in marinated (vegetable oil, water, spices, NaCl (5.9%), lactic and acetic acid at pH4.5) and non-marinated chicken drumsticks and strips. The composition of marinade is a key factor in achieving a decrease in campylobacter concentration on the surface of the chicken.

- **Heat treatment of chicken:** The D-value (decimal reduction time – the time required at a certain temperature to kill 90% of a particular organism) for *Campylobacter* spp. in cooked chicken at 55°C is 2.12-2.25 minutes and at 57°C 0.79-0.98 minutes (ICMSF, 1996). Normal cooking temperatures (where a core temperature of 75°C is achieved) will kill campylobacter and therefore, cooking is an option for chickens from highly contaminated flocks
- **Modified atmosphere packaging (MAP):** MAP has been examined as a method to reduce the numbers of viable campylobacters. Boysen *et al.*, 2007 reported reductions in campylobacter numbers of 2.0-2.6 log₁₀ cfu/g after eight days in 70%/30% O₂/CO₂ mixture (p<0.0001) compared to no reduction observed in a gas mixture of 70%N₂/30%CO₂. Modified atmosphere of 80% O₂/20% N₂ resulted in a reduction of approximately 1.2 log₁₀ cfu/g (Rajkovic *et al.*, 2010) however, different strains of campylobacter may exhibit different oxygen tolerances (Kaakoush *et al.*, 2007)

2.3.3 Measures if the flock is positive for campylobacter but has not exceeded the pre-harvest criterion prior to the first depopulation

If a flock is campylobacter positive, based on the results of the sampling carried out prior to the first depopulation, due to the dynamics of campylobacter colonisation of flocks it is expected that the level of colonisation will increase significantly by the second or subsequent depopulation. It is therefore recommended that broilers from positive flocks with caecal levels ≤ 7 log₁₀ cfu/g prior to first depopulation, are scheduled for slaughter during the subsequent depopulation and processed as outlined in Section 2.3.2.

It is also recommended that producers are given an option to re-test the remaining broilers prior to the subsequent depopulation. If the result of the re-testing of pooled caeca is ≤ 7 log₁₀ cfu/g, then the broilers can proceed to slaughter without employing a scheduling approach. Carcasses from such flocks are subject to sampling and testing against the post-harvest criterion (section 2.2.3).

2.3.4 Flocks negative for campylobacter

Flocks which show a negative result following analysis of pooled caecal samples will be treated as low-risk flocks. The broilers from these flocks will be processed in a normal fashion and a scheduling approach or carcass sampling will not be necessary. The subsequent depopulation from these flocks will also be slaughtered in a normal fashion. Re-testing of these flocks during the same growing period will not be necessary.

2.3.5 Measures if the post-harvest criterion is not met:

- It is recommended that a review of GHP in the slaughterhouse be undertaken. The GHP measures which should be implemented in all plants are outlined in Appendix 5
- It is recommended that defeathering and evisceration equipment is appropriately adjusted to the size of the broilers before processing. Consideration should be given to replacing equipment with models shown to have better performance
- A review of line speed is recommended
- Where persistent, repeated breaches of the post-harvest microbiological criterion occur (more than four breaches in eight batches sampled), a comprehensive review of slaughterhouse procedures and sourcing of broilers is necessary. In addition, further interventions may be necessary and it is recommended that freezing is considered in these cases. Freezing is a safe, effective and readily available method to reduce the number of viable campylobacter on poultry

2.3.6 Measures not currently permitted under EU law

A number of control measures used in countries outside the EU, but not currently permitted under EU law are outlined in Appendix 6. In the future, chemical decontamination may be considered as a supplement to biosecurity and good hygiene practice (GHP), for substances shown to be safe and effective at significantly reducing microbial contamination (European Commission, 2004; EFSA, 2008a).

2.4 RISK MANAGEMENT QUESTION 5

Q5. What, if any, additional packaging and labelling measures could the poultry processing industry adopt to reduce the risk of campylobacteriosis for consumers?

It is recommended that raw chicken is packaged in leak-proof packaging. It is recommended that safe handling and cooking instructions should be clearly visible at the time of purchase. Instructions should not be on the reverse of a label or otherwise obscured.

2.4.1 Packaging

The EFSA baseline study has revealed a very high level of contamination (98.3%) in Irish broiler carcasses at the end of the slaughter process (EFSA, 2010c). Given this level of contamination on carcasses, it is not surprising that a study published by the FSAI, (2010) found 13.2% of the external surfaces of chicken packaging sampled at retail level were contaminated with campylobacter. In addition to the overall level of contamination on packaging, the study compared contamination on conventional and leak-proof packaging. The type of leak-proof packaging described in the study was where the plastic wrapping sealed onto the tray. Of course it is possible that other types of leak-proof packaging have been/will be developed.

The 2010 FSAI study showed that the contamination on conventional packaging (18.9%) was higher than that on leak-proof packaging (2.1%), providing convincing evidence for the need for processors to change to using the latter. The study also found that campylobacter was detected on 19.5% of packages containing whole birds compared to 3.2% of packages containing chicken portions. This may be attributed in part to the fact that 84.1% of whole birds were packaged in the conventional manner compared to 23.1% of chicken portions.

In New Zealand, a survey enumerating campylobacter and *salmonella* on retail packs of chicken found that whole birds (68%) and offal trays (66%) were more likely to have leakage than portion trays (27%; Wong *et al.*, 2004). The poultry industry and supermarkets in New Zealand have since introduced leak-proof packaging for retail sale of whole birds and a proportion of packaged portions (Wong, 2008). Fluid absorbing packaging or edible films may help in preventing cross-contamination in the kitchen.

It is recommended that raw chicken is packaged in leak-proof packaging. Packaging solutions which reduce the amount of handling of the raw poultry, e.g. oven-ready trays, cooking bags, should be explored.

2.4.2 Labelling

The FSAI packaging study (2010) also examined labelling to establish whether handling and cooking instructions deviated from accepted best practice. Approximately one-third of chicken packages provided handling, preparation and/or cooking instructions on the front of the label. However, 63% of labels carried these instructions on the reverse of the label (i.e. not visible at time of purchase). To view this information, the consumer must either peel off the label (which can be difficult to do), or once they have opened the packaging, look at the label through the plastic film. This latter practice would result in increased handling and risk of contamination. It is recommended that handling and cooking instructions on the reverse of a label should be discontinued. All instructions should be clearly visible at time of purchase.

Of the 365 samples which were identified as whole birds, in the FSAI study (2010), 6.8% carried instructions advising customers to wash the whole bird or the cavity of the bird prior to cooking. This instruction is contrary to current best practice advice and can lead to the spread of campylobacter within the kitchen environment. **Labels on whole birds should advise consumers that carcasses are ready to cook and in the interests of safe handling, that washing of the carcass should be avoided. In addition, as many Irish people have a tendency to wash chicken meat, the advice not to wash chicken should be carried on portions as well.**

APPENDIX 1. MINIMISING THE RISK OF *CAMPYLOBACTER* SPP. INFECTION DURING THINNING/PARTIAL DEPOPULATION

'Thinning' is the term given to the process whereby a proportion of the broiler flock is removed early for slaughter at about 5 weeks of age, whereas normal slaughter age is around 6 weeks. This reduces stocking density within the poultry house and allows higher initial stocking densities to be used. Feed is withdrawn from the birds for up to eight hours before thinning begins (usually 4-6 hours) and water is withheld for a maximum of one hour (usually 15 minutes or less); Allen *et al.* (2008). The procedure is practiced by most companies in Ireland and increases profitability by permitting increased stocking densities in the poultry house. It is however associated with an increased risk of campylobacter infection in the flock through the passive transfer of organisms from previously visited farms, or the processing plant on clothes, boots, gloves, crates and vehicles on to the farm and subsequently into the broiler house during catching.

Once campylobacter has gained access to a flock, it spreads rapidly among the birds (Newell and Fearnley, 2003). Evans and Sayers (2000) showed that when a flock was infected, virtually all cloacal swabs were positive within a week indicating that campylobacter infection spreads very rapidly amongst housed broiler chickens. Shanker *et al.* (1990) found that 67% of the batch was contaminated within three days. Allen *et al.* (2008) studied flocks which had undetectable levels of campylobacter at the time of depopulation and found that in the case of some flocks the caecal samples were positive within 3-4 days of thinning. Twenty-seven out of 30 flocks which were not colonised at the first depopulation were positive by 6 days post-thinning (there were two flocks in this study which displayed a different pattern of infection and were campylobacter-negative at slaughter despite being tested positive previously – this phenomenon was highlighted as needing further investigation). It has been hypothesised that if final depopulation occurs 5 days after the first thin, the within flock prevalence will still be low at slaughter minimising the importance of this potential route (FSA, 2008). In Ireland, the period between initial and final depopulation often exceeds five days and provides sufficient time for flock colonisation.

A number of surveys have found statistically significant risks associated with thinning. Studies in Denmark found that thinning significantly increases the risk of campylobacter infection in the birds remaining in the flock (Hald *et al.*, 2001) as it compromises biosecurity due to breach of hygiene barriers by catching personnel and equipment during collection of birds. The results strongly suggest that the introduction occurred during catching of the first batch.

In the UK a systematic review was carried out by Adkin *et al.* (2006) in the Veterinary Laboratories Agency (VLA). This review identified the depopulation schedule (thinning) and multiple houses on-farm as contributing factors associated with increasing the risk of campylobacter in the flock, whilst a depopulation (thinning) event, cross-house transfer and on-farm staff were found to be the highest ranked sources associated with campylobacter infection. Allen *et al.* (2008) provided more evidence that flock thinning can cause campylobacter infection. A total of 27 of 51 flocks studied became campylobacter positive within a few days of thinning, and molecular typing of isolates was able to identify their likely sources. For seven flocks colonised post-thinning, there was evidence from PFGE typing of an association between flock infection and prior contamination of specific items of equipment, vehicles and personnel. A UK national prevalence survey carried out in 2007 reported that thinning increases the risk of campylobacter colonising a flock by a factor of eight (Powell *et al.*, 2009). In Sweden, Hansson *et al.* (2010) reported that thinning increases the proportion of campylobacter positive flocks. A Danish study by Wedderkropp *et al.* (2000) found that the risk of producing campylobacter positive flocks was associated with the number of thinnings used.

A number of studies suggest that the risk associated with thinning may be further confounded with the increasing age of the birds. In some of these studies, it is unclear if thinning influenced the observed effect. Evans and Sayers (2000) reported that more than 40% of flocks were infected with campylobacter by the time the chicks were four weeks old and >90% by seven weeks. The power of the study to detect an association between infection and visits by abattoir personnel was relatively poor, because nearly three-quarters of the flocks were infected prior to any birds being slaughtered. Bouwknegt *et al.* (2004) found a marked effect of increasing age on the presence of campylobacter however, the effect of thinning was not estimated and this may have interfered with the observed effect of age. An Icelandic study by Barrios *et al.* (2006) showed a higher risk of campylobacter colonisation associated with increasing age at slaughter. It also concluded that due to special hygienic measures taken by catching crews during thinning, crews did not play an important role in introducing campylobacter to most of the positive flocks.

Two Dutch studies found that thinning was not significantly associated with campylobacter status at slaughter, whereas age and season were (Bouma *et al.*, 2003; Russa *et al.*, 2005). Russa *et al.* (2005) carried out a comprehensive study on partial depopulation (thinning) of flocks. They questioned the importance of thinning on flock prevalence and reported an odds ratio (O.R.) of 0.8 when adjusted for the confounders of age and season, and concluded that partial depopulation was not a significant risk factor for campylobacter prevalence at slaughter. The Dutch risk assessment model (Katsma *et al.*, 2005) found that the effect of thinning is not as large as previously thought, particularly if the thinning is carried out shortly before the final depopulation. The flocks may become infected post-thinning but at lower levels. This is based on the assumption that the bacterium is introduced at low levels during thinning. When there is an indication that many animals become infected during the thinning process, the risk of high level prevalence in the flock increases.

It is reasonable to conclude that campylobacter introduction into the broiler house occurs during thinning – there are many studies documenting the presence of campylobacter on crates, modules, crew members clothing and footwear and machinery entering the broiler house. Van de Giessen *et al.* (1998) reported the isolation of campylobacter from a lorry and slaughterhouse crates used during thinning which suggests that, where such materials are not properly cleaned and disinfected, they may transmit campylobacter. Hansson *et al.* (2005) reported that campylobacter was isolated from 57% of transport crates which had been washed and disinfected, and concluded that crates could contaminate subsequent chicken batches being transported for slaughter. Slader *et al.* (2002) found that the process of catching and putting birds in crates significantly increased the chance of contamination with campylobacter ($P < 0.001$). Newell *et al.* (2001) and Hiatt *et al.* (2002) demonstrated the presence of campylobacter on crates prior to their use on-farm. Ramabu *et al.* (2004) identified campylobacter on 75% of pallets; more than 50% of catchers' boots, drivers' boots, crates and truck wheels were positive, while 47% and 31% of truck beds and forklift wheels, respectively were also contaminated.

The potential ingress of campylobacter is compounded by the fact the birds often become stressed as a result of the catching process and feed withdrawal. This may render those remaining in the house more susceptible to colonisation with campylobacter (FSA UK, 2005).

Results from Iceland suggest that thinning may be carried out without infecting the birds remaining in the house, if a number of hygienic precautions are taken. The area of the house where thinning has taken place should be left to dry properly before the remaining birds are given access. This may be accomplished by setting up temporary separating barriers or walls (Interventions to control campylobacter in broiler production – report of international expert consultation, 2007).

Evidence from UK studies indicates that the elimination of thinning practices should significantly reduce later slaughter batches from flocks becoming positive (FSA, 2008). EFSA analysed the risk factors associated with campylobacter colonisation and contamination of carcasses and found, that batches of broilers from previously thinned flocks were at higher risk of being colonised with campylobacter compared to non-thinned flocks (EFSA, 2010d). However, following analysis, no association was found between thinning and higher campylobacter counts on carcasses. It was suggested that this failure to find a statistically significant link may be due to the power of the analyses being too low due to too few samples.

Recommendations

It is recommended that the practice of thinning is avoided because thinning is recognised as an important risk factor for the introduction of campylobacter into poultry flocks. In the event that thinning is practiced, the following recommendations may help to minimise the potential negative impact of thinning:

- It is recommended that there should be no more than one partial depopulation prior to the final depopulation
- The period between the partial depopulation and the final depopulation should not exceed five days
- Emphasis by processors and producers should be placed on the following:
- Effective cleaning and disinfection of forklift wheels and forks prior to entry into broiler house
- Improved crate, module and truck washing and disinfection in the slaughter plant
- Thinning teams must put on clean protective clothing and clean, disinfected boots prior to entry into the broiler house. There should be dedicated footwear supplied for use by the thinning teams for each house on-farm
- Thinning teams must comply with biosecurity measures as outlined in Appendix 3
- Education of thinning personnel to enforce the importance of biosecurity

APPENDIX 2. POST-HARVEST CRITERION – ESTIMATED PERFORMANCE AND TREND ANALYSIS

A level of less than or equal to $4 \log_{10}$ cfu/g on carcasses at the end of slaughter is considered achievable when the mean concentration of *Campylobacter* spp. in a positive flock is less than or equal to $7 \log_{10}$ cfu/g caecal content. Therefore a target level of $\leq 4 \log_{10}$ cfu/g has been proposed in this report, pending validation by a pilot study.

There are several approaches to monitoring the achievement of a target level:

- A microbiological criterion that is valid for a single sampling window
- A microbiological criterion that is valid for a set sampling time frame, e.g. several sampling sessions over several weeks
- A moving average approach that monitors trends in performance

These approaches were evaluated using Montecarlo simulation software (@Risk™) and the following proposal was considered the most practical while providing reasonable performance.

Post-harvest Microbiological Criterion using a Single Sampling Window

Sample	Carcass neck skin
Point of sampling	After chill and before further processing
Sampling plan	n=5 and c=1 Where n = number of units comprising the sample and c = number of sample units giving values between the limits m and M (see row below) Once a week, sample 15 carcasses from a flock that meets the $\leq 7 \log_{10}$ cfu/g standard. A piece of approximately 10g from neck skin shall be obtained from each carcass. On each occasion, the neck skin samples from 3 carcasses shall be pooled before examination in order to form 5×25 g final sampling units.
Limits	m= $4 \log_{10}$ cfu/g M= $5 \log_{10}$ cfu/g
Method of analysis	ISO/TS 10272-2:2006 a horizontal method for the enumeration of <i>Campylobacter</i> spp.
Action to be taken	Action, as outlined in section 2.3.5, should be taken if any of the sampling units is $> 5 \log_{10}$ cfu/g or more than one of the 5 sampling units is $> 4 \log_{10}$ and $\leq 5 \log_{10}$

The expected performance of this microbiological criterion, assuming that the microbiological logarithmic counts on carcasses are normally distributed, is shown in Table 1.

Table 1. Probability of Failure/Pass of Microbiological Criterion given Performance of the Process

Mean log ₁₀ count /g achieved		Standard deviation log ₁₀ counts/g achieved by the slaughter process		
		0.3	0.6	0.9
2.5	P* failure	0.00	0.00	0.01
	P pass	1.00	1.00	0.99
3.0	P failure	0.00	0.01	0.13
	P pass	1.00	0.99	0.87
3.5	P failure	0.01	0.32	0.60
	P pass	0.99	0.68	0.40
4.0	P failure	0.68	0.69	0.70
	P pass	0.32	0.31	0.30

*P= probability, e.g. p=1 means 100% chance, p=0.5 means 50% chance

For example, a slaughter process that is capable of producing poultry carcasses with a microbial load that is distributed normally with a mean of log₁₀ 3.5cfu/g and a variability of standard deviation log 0.6 cfu/g will on average fail the microbiological criterion 32% of the time and pass it 68% of the time. Improvements in the process to lower means or lower variability will decrease the chance of failure.

Process monitoring/trend analysis

Whilst the number of times a process fails a criterion is a measure of the performance of the process, it cannot be used to predict if the process is moving out of control. Most manufacturers prefer to make such predictions and take corrective action before the process fails a criterion. It is suggested that data from the post-harvest microbiological sampling can be used to do this using a weighted moving average approach, which means that the latest data have more of a bearing on the moving average. Such charts of historic data are more responsive to changes in the process performance than simple moving averages, where no weighting is applied.

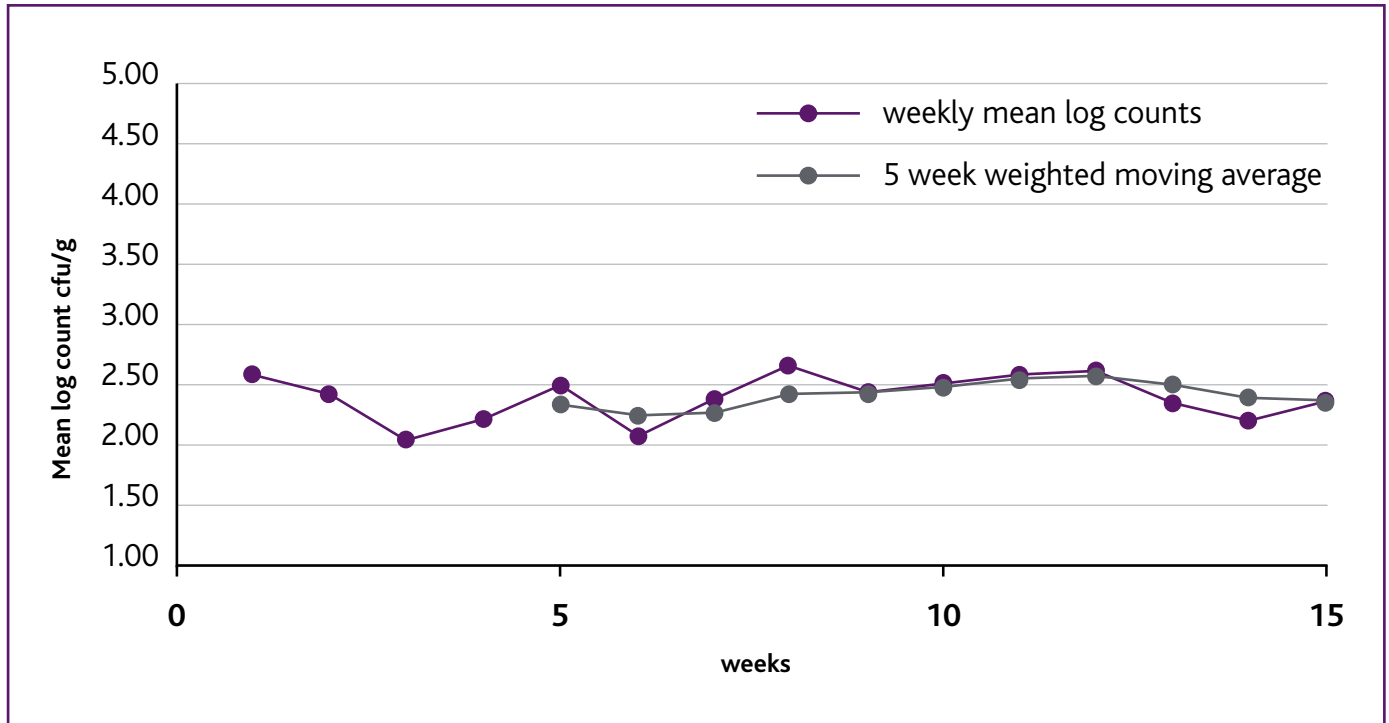
Each plant can plot its weighted moving average over a five week period. The moving average is computed as follows:

$$\text{Weighted moving average} = (\text{mean log count week 1} * 0.067) + (\text{mean log count week 2} * 0.133) + (\text{mean log count week 3} * 0.2) + (\text{mean log count week 4} * 0.267) + (\text{mean log count week 5} * 0.333)$$

Note: Week 5 represents the most recent and week 1 the most remote sampling point.

As each week's count is added, the oldest count is dropped from the moving average. Control charts or process behaviour charts can be used to spot trends as shown in Figure 1. If the trend shows successive increases, this could indicate that the slaughter process is moving out of control, especially if it crosses the 4 log₁₀ line. Operators should aim to maintain their moving average below 3.5 log₁₀ cfu/g.

Figure 1. Example of a Control Chart/Process Behaviour Chart



Web-based database to facilitate trend analysis

The report recommends trend analysis. It is suggested that the FSAI should set up a secure web database where processors can enter their weekly data. The site could calculate and display the moving average and also show a comparison with the national performance of all plants. The latter would be calculated by taking a mean of all plants' weekly means and plotting as a five week weighted moving average. Plants would be able to access only their data but would be able to compare their data to the national results. The data would not be used by the authorities in any form from which plants could be identified. Over time, the data would provide an indication of the performance of the industry as a whole and be a basis for calculating improvements in the microbiological criterion set by agreement.

APPENDIX 3. SECTIONS OF THE BORD BIA QUALITY ASSURANCE STANDARD OF PARTICULAR RELEVANCE TO CONTROL OF CAMPYLOBACTER

The following sections are of particular relevance to the control of campylobacteriosis in flocks:

3.1 General

(c)

3.2 Chicken Production Site

(c) – (n)

3.6 Flock Health

(a)

3.7 Feed and Water

(p)

3.9 Site Hygiene and Biosecurity

All of this section: (a)-(t)

3.10 Catching and Transport

All of this section: (a)-(d)

Appendix 3. House Preparation Checklist

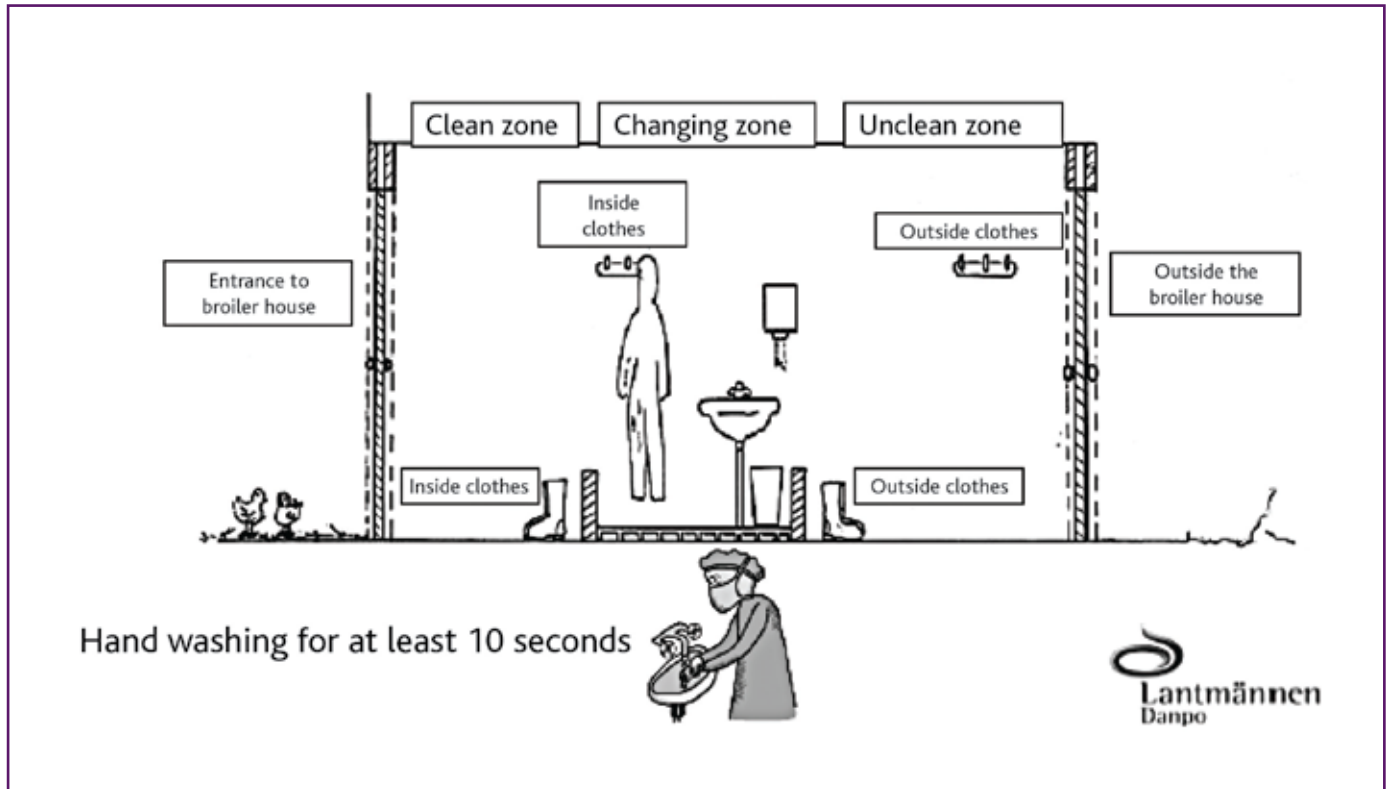
Appendix 4. Hygiene and Welfare for Catching Teams

Appendix 5. HACCP Plan

Appendix 8. Terminal Hygiene Programme

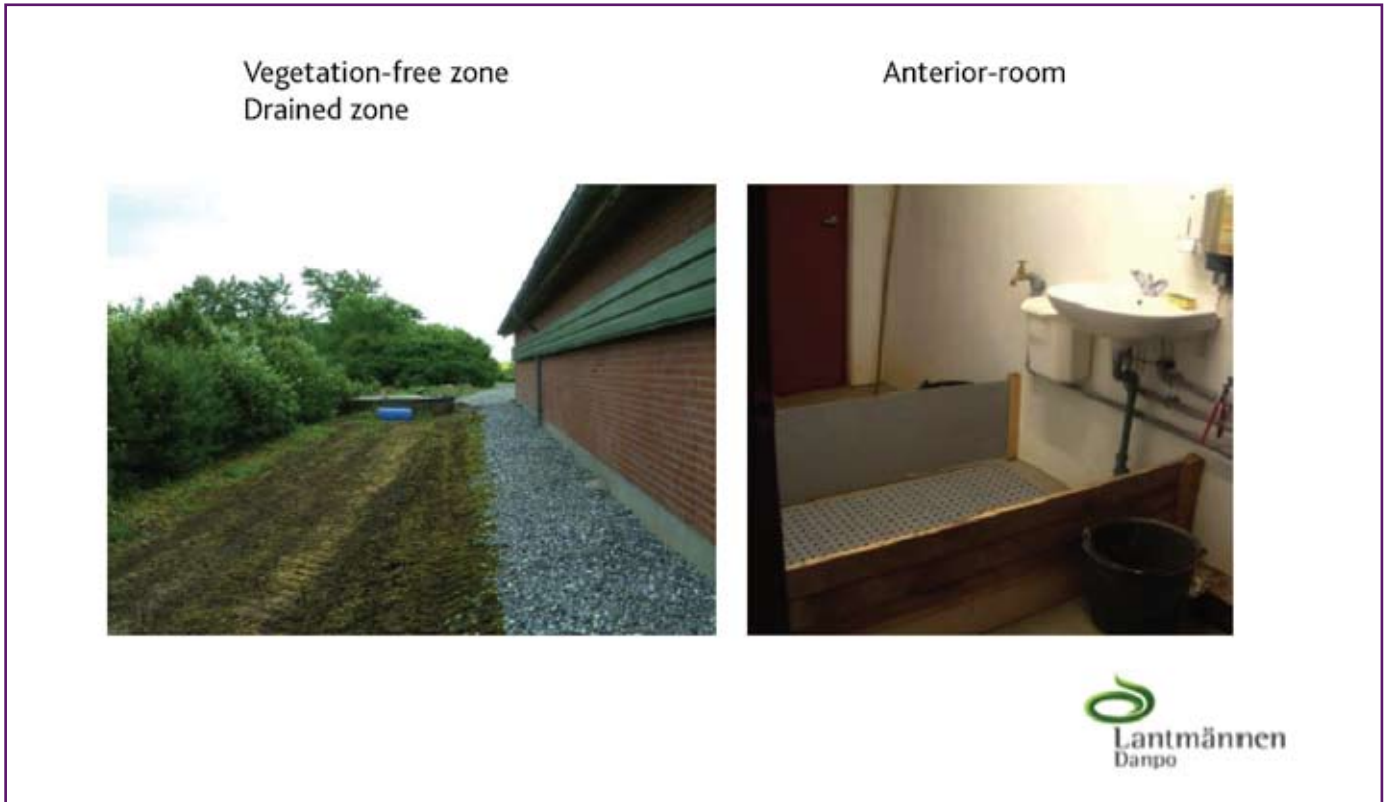
APPENDIX 4. BIOSECURITY – PHYSICAL BARRIERS

Poultry House Anterior Room with 3 Zones³



³ Source: Jacob Roland Pedersen, Lantmännen Danpo A/S

Examples of Biosecurity around the Poultry House and Prior to Entry⁴



Note on Vegetation-Free Zone

The area of pebbles/gravel is expected to be a minimum of one metre and the area that is kept free of vegetation is expected to be a minimum of three metres.

Note on Anteroom

The barrier in the anteroom is expected to be a minimum of 40cm in height.

⁴Source: Jacob Roland Pedersen, Lantmännen Danpo A/S

APPENDIX 5. GHP MEASURES DURING PROCESSING

The following GHP measures should be implemented by all processors

Module, Crate and Vehicle Washing

Current methods of crate washing to remove campylobacter are recognised as being largely ineffective (Berndtson *et al.*, 1996a; Slader *et al.*, 2002; Ramabu *et al.*, 2004) and represent a substantial risk of introducing the pathogen to subsequent broiler houses. The time between the cleaning and disinfection process and the reuse of the transport containers may range from hours to days. High campylobacter numbers have been recovered of up to 10⁸ cfu on the inside crate base and module base. However, there was little correlation between visual assessment of cleanliness and numbers recovered (Allen *et al.*, 2007b). Insufficiently cleaned and disinfected crates may also be a source of poultry flock contamination during transport to the slaughterhouse (Hansson *et al.*, 2005; Newell *et al.*, 2001; Slader *et al.*, 2002).

Recommendations to reduce contamination by crates, modules and vehicles:

- Food business operators should ensure that crates, modules and vehicles are effectively cleaned and disinfected. Appropriate temperatures for cleaning (hot water) and disinfection and adequate concentrations of disinfectants should be used. This cleaning should be validated through periodic microbiological testing and incorporated into the food safety management system. For further details, see the FSA UK funded report on improved crate washing - http://www.foodbase.org.uk/admintools/reportdocuments/540-1-943_M01023_Report_and_appendices_complete_revised.pdf
- If crates and modules cannot be cleaned effectively because of the material used in their manufacture, they should be replaced with crates and modules manufactured from materials that are easily cleaned
- Placing modules onto dirty flatbed trucks can serve as another source of contamination for the modules. Flatbed trucks should be adequately cleaned and disinfected before transportation of modules. Where a tarpaulin is used during transport, this must also be cleaned and disinfected adequately between uses

Scalding

Scalding is the immersion of carcasses in scalding water tanks. The scalding temperatures used vary from 50-52°C (soft scald) to 56-68°C (hard scald). In Ireland, a soft scald is used as it is more suitable for fresh poultry meat production (Gracey *et al.*, 1999). Substantial contamination with enteropathogens including campylobacter may occur during the scalding step. Humphrey *et al.* (2007) reported that the death rate in scald tank water maintained at 52°C was 9 minutes. Chickens remain in the scald tanks for much shorter periods during normal processing. This emphasises the potential of this processing stage for the dissemination of campylobacter.

Recommendations to reduce contamination during the scalding step include:

- **Counter-current scald tanks:** This is a system whereby water in the tank should move through the system flowing against incoming carcasses. This flow creates a 'dirty to clean' gradient. Carcasses moving through the tank are washed by even cleaner water. In the US, the National Chicken Council (NCC) recommends that best management practices include using counter-current systems with adequate water replacement (NCC, 1992)
- **High water flow rates in the tanks:** High flow rates of water and adequate agitation dilute dry matter and bacterial load in the tank (Cason *et al.*, 2001)
- **Multi-stage scald tanks:** Multiple stage tanks are better than single stage tanks because they create more opportunities to clean the carcasses (Cason *et al.*, 2000). When combined with a counter-current water flow system, the multi-stage scald tank provides a dilution effect (Veerkamp, 1991). Consequently, there is a marked reduction in carcass contamination (3.0-3.9 log₁₀ reduction per carcass) even at 'soft scald' temperatures. With such a system, Berrang and Dickens (2000) obtained almost a thousand-fold reduction in campylobacter contamination of carcasses, though much of the advantage was reported to be lost during subsequent stages of processing. Hinton *et al.* (2004) showed that significantly less campylobacter was recovered from the final tank of a multiple-tank counter-flow scald system than from the first tank

Defeathering

Defeathering is a mechanical process that is carried out immediately after scalding, while the carcasses remain warm. It involves a series of in-line machines containing banks of counter-rotating metal domes or discs bearing multiple rubber 'fingers' that rotate at high speed and scour the surface of each carcass. These machines incorporate continuous water sprays that help to flush away feathers as they are removed. Any remaining feathers are removed by hand. Cross-contamination of carcasses by the machinery may occur at this step. The feather follicles in the skin at this stage are open and may lead to movement of campylobacter cells inside the follicles, which may decrease the efficacy of subsequent carcass rinses (Cason *et al.*, 2004). Guerin *et al.* (2010) and Son *et al.* (2007) found that in contrast to scalding, the defeathering step consistently increased the prevalence and level of campylobacter contamination. It is generally well accepted that contamination increases largely due to the escape of faecal material through the cloaca by the action of the picker fingers pressing on the abdomen (Oosterom *et al.*, 1983; Berrang *et al.*, 2001; Allen *et al.*, 2003).

Recommendations to minimise cross-contamination at this step include:

- Correct alignment of machinery based on bird size: This will prevent excessive pressure being applied to the birds during defeathering causing expulsion of increased amounts of faecal material from the cloaca. Segregation and slaughter of the flock based on sex will permit more consistency in the size of bird at slaughter
- Adequate flow rates of water: adequate flow rates of water are necessary to wash away the feathers and contamination from the surface of the carcass
- Regular equipment sanitation and maintenance are recommended to minimise cross-contamination during defeathering

Evisceration

The evisceration process involves removal of the feet, head and viscera of the birds and the harvesting of edible offal. The important challenge during evisceration is to minimise the rupture of the exposed intestines and prevent the spread of faecal bacteria, such as campylobacter, which can occur at high concentrations in the intestines of positive birds. Several studies show that campylobacter contamination levels increase during evisceration (Oosterom *et al.*, 1983; Izat *et al.*, 1988; Rosenquist *et al.*, 2006).

Generally, carcasses visibly contaminated with faecal material have significantly higher campylobacter counts than carcasses without contamination (Berrang *et al.*, 2004; Boysen and Rosenquist, 2009). Damage to the intestines can occur because the machines are sometimes unable to adjust to the natural variation in carcass size that is associated with poultry flocks. This can result in excessive pressure by the equipment on the abdominal cavity (Berrang *et al.*, 2004) or an accidental cut may occur. As a result, significant amounts of campylobacters can be introduced during this process step (Takahashi *et al.*, 2006) potentially resulting in extensive cross-contamination.

Recommendations to minimise cross-contamination during this step include:

- Control alignment of equipment: Due to the size variability within and between flocks, evisceration machines must be properly aligned to avoid damage to the intestines of birds and subsequent cross-contamination. If machines are set for the median weight of the flock, carcasses that are heavier or lighter may not be properly eviscerated (FSIS USDA, 2010). Segregation and slaughter of the flock based on sex will permit more consistency in the size of bird at slaughter
- Visual inspection of carcasses to identify problems with evisceration: perforation of intestines and leakage of gut contents is a significant vehicle for spread of contamination between carcasses. For the evisceration process to work properly, carcasses must be placed on the shackles correctly and monitored as they move through the system. Visual monitoring of the evisceration process will highlight problems with equipment and enable swift intervention to remedy problems. Monitoring of the number of evisceration failures (perforated or leaking intestines) as a percentage of birds slaughtered within each batch will highlight problems with the alignment of equipment and permit corrective action to be taken
- Replace old/dysfunctional equipment
- Regular maintenance of equipment: Machines should be kept in good working order. Blades should be kept sharpened and attention should be given to thorough cleaning. Maintaining the equipment in a clean condition, free from intestinal contents is important for maintaining good process control

Chilling

Freshly eviscerated carcasses are still warm and must be chilled as soon as possible to inhibit microbial growth. In Ireland, air chilling systems are used. This system has shackles or tiered chains that move the carcasses through a chilled room until they are correctly chilled (<4°C). A number of studies have found that air chilling results in a decrease in campylobacter levels on the carcass (Sanchez *et al.*, 2002; Hinton *et al.*, 2004; Allen *et al.*, 2007a; Berrang *et al.*, 2007; Huezio *et al.*, 2007; Boysen and Rosenquist 2009). An approximate decrease in the counts of campylobacter of 1 log₁₀ unit has been reported following chilling, although in some studies, chilling was preceded by carcass washing (Rosenquist *et al.*, 2006; Allen *et al.*, 2007a; Berrang *et al.*, 2007).

Recommendations to minimise cross-contamination during this step:

- Effective air chilling requires appropriate design and maintenance of equipment
- Appropriate line speeds should be used as inappropriate line speeds may result in inadequately chilled carcasses exiting the chill
- Thorough washing of carcasses prior to chilling is recommended to remove surface debris and contamination

Packaging

The results of the recent FSAI survey on campylobacter contamination of poultry packaging, highlights the need for a change to leak-proof packaging. See Section 2.4.1.

Labelling

Labelling of chicken with safe handling and cooking instructions is required under the General Labelling legislation (Directive 2000/13/EC). Clear and legible instructions are required. See Section 2.4.2.

While this report makes recommendations about labelling of meat from flocks which have exceeded the pre-harvest criterion (Section 2.3.2), processors should consider that all products carry an instruction on the label that the product “may contain harmful bacteria”.

Sanitation and Process Hygiene

Peyrat *et al.* (2008) reported that *Campylobacter jejuni* is able to survive overnight on food processing equipment surfaces after cleaning and disinfection procedures and that these strains may contaminate carcasses during the slaughter process. This study highlights the importance of effective cleaning and correct use of detergents and disinfectants in the slaughterhouse.

Recommendations for sanitation and process hygiene:

- Verification of effective cleaning and disinfection can be carried out through the microbiological sampling plan in the plant. Monitoring of the effectiveness of cleaning can be carried out by swabbing food contact surfaces and equipment. Evidence of inadequate disinfection should be followed by a thorough review of cleaning procedures and a review of the use of detergents and disinfectants (to ensure that the correct dose and contact time were employed)

APPENDIX 6. INTERVENTIONS NOT CURRENTLY PERMITTED UNDER EU LAW

The following interventions are **not currently permitted under EU law** but may be used in the future if legislation changes.

Currently, the use of chemical decontaminants during poultry slaughter and dressing within the EU is not permitted. In the future, chemical decontamination may be permitted as a **supplement** to biosecurity and good hygiene practice (GHP), provided chemicals are shown to be safe and effective at significantly reducing microbial contamination (European Commission, 2004; EFSA, 2008a). EFSA has published Scientific Opinions on the evaluation of the efficacy of several compounds for the decontamination of poultry carcasses, including lactic acid, peroxyacids, chlorine dioxide, acidified sodium chlorite and trisodium phosphate (EFSA, 2005a&b, 2006a&b). EFSA has also published opinions on the effects of applying a number of these chemicals as carcass decontaminants and the emergence of antimicrobial resistance, and have concluded that no data currently exist showing tolerance or resistance among bacterial populations to these specific compounds (EFSA, 2008a). However, EFSA does state that there is some evidence of tolerance to other antimicrobial substances not covered in the study and as a consequence has proposed that this be examined alongside microbial efficacy and safety criteria (EFSA, 2008a).

The following presents results of efficacy studies for a range of treatments not currently permitted under EU law.

• **Antimicrobial treatments:**

- **Lactic acid:** Ellerbroek *et al.* (2007) reported *Campylobacter jejuni* reductions ranging from 0.2 to 1.5 log₁₀ cfu/g when broiler carcasses were immersed in lactic acid solutions (10 and 15% v/v); while Riedel *et al.* (2009) observed reductions of 1.7 log₁₀ cfu/ml on retail carcasses immersed in a 2.5% solution of lactic acid for one minute. Lactic acid also reduced the incidence of finding campylobacter on pre-chill carcass rinses by 14.7% compared with the controls (Byrd *et al.*, 2001)
- **Acetic acid:** Immersion of broiler carcasses in 2% acetic acid reduced *Campylobacter jejuni* populations by 1.2-1.4 log₁₀ cfu/g (Zhao *et al.*, 2006)
- **Trisodium phosphate:** Immersion of broiler carcasses in 10% trisodium phosphate solutions reduced campylobacter by up to 1.7 log₁₀ cfu/g (Slavik *et al.*, 1994; Whyte *et al.*, 2001; Riedel *et al.*, 2009)
- **Chlorine/Acidified chlorine:** The application of chlorine or acidified sodium chlorite at varying concentrations and exposure times can reduce campylobacter populations on broiler carcasses at rates ranging from 0.5 to 3.0 log₁₀ cfu as summarised by Loretz *et al.*, 2010
- **Electrolysed water:** Electrolysed water (EO) is produced by passing a current of electricity through a dilute sodium chloride solution. One product of the reaction is sodium hydroxide and the other is hypochlorous acid, which has a low pH, contains active chlorine, and has a strong oxidation-reduction potential similar to that of ozone. EO has been shown to give good reductions in *campylobacter jejuni* on poultry carcasses: 3 log₁₀ reduction (Park *et al.*, 2002) 2.33 log₁₀ cfu/g reduction (Kim *et al.*, 2005)
- **Ozone:** Ozone (O₃) is a strong antimicrobial agent with numerous potential applications in the food industry. High reactivity, penetrability, and spontaneous decomposition to a non-toxic product (i.e. O₂) make ozone a viable disinfectant for ensuring the microbiological safety of food products. Ozone, either in the gaseous phase or in an aqueous solution, is used in the food industry for decontamination of product surface and water treatment. Ozone has been used with mixed success to inactivate contaminating microflora on meat, poultry, eggs, fish, fruits, vegetables, and dry foods. Excessive use of ozone, however, may cause oxidation of some ingredients on food surface. This usually results in discoloration and deterioration of food flavour. Additional research is needed to elucidate the kinetics and mechanisms of microbial inactivation by ozone and to optimise its use in food applications (Kim *et al.*, 1999)
- **Irradiation:** Gamma irradiation can also improve the microbiological safety of raw poultry meat. *Campylobacter* and most other non-spore forming bacteria are relatively susceptible to irradiation. A dose of 0.08-0.2 and 0.18-0.32kGy has been reported to be sufficient to produce a D-10 reduction (decimal reduction radiation dose (i.e. dose required to reduce the number of microorganisms to one-tenth of the initial value) in *Campylobacter jejuni* in fresh and frozen foods (Farkas, 2005). For fresh and frozen poultry meat, doses of 2 or 3-5kGy would be sufficient to reduce populations of sensitive bacteria, including *Campylobacter jejuni* by more than 6 logs (Sudarmandji *et al.*, 1972; Klinger *et al.*, 1993; Farkas and Mohacsi-Farkas, 2011).

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