



The risk of handling poultry meat with *Campylobacter jejuni* from the consumer's perspective

Risken att hantera kycklingfilé med Campylobacter jejuni ur ett konsumentperspektiv

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Abstract

Campylobacteriosis is the most reported gastrointestinal infection in humans, within the EU, since 2005. Campylobacteriosis is a zoonosis and caused by *Campylobacter* spp. Poultry is believed to be a vehicle for human exposure to *Campylobacter*. Foodborne outbreaks of campylobacteriosis have been connected to private households. This thesis aimed to study simulated risk factors for transmission of *C. jejuni* from poultry meat to humans in the kitchen.

Broiler chicken filets were artificially contaminated with *C. jejuni* sequence type 257 and 918. Sampling was conducted on a glove (to simulate hands), a washed glove (to simulate washed hands), a first sampling of a used cutting board, a second sampling of a used cutting board, and utensils (a scissor and tweezer). Concentrations of *Campylobacter* on the chicken meat used in this study varied between 2.7 log₁₀ CFU/g and 5.3 log₁₀ CFU/g when the transfer of both sequence types onto these objects was analysed.

Campylobacter were isolated in all samples but in various concentrations. The highest transfer of both sequence types was in unwashed glove and in the first sampling of the cutting board. The transfer was lower when the gloves were washed and in the second sampling of the cutting board. The lowest transfer found from meat was found in utensils that were used take chicken meat.

This thesis further emphasises the significant risk of cross-contamination when handling chicken contaminated with *Campylobacter*. It is important to prevent cross-contamination during the handling of *Campylobacter* contaminated chicken to prevent or reduce campylobacteriosis in humans.

Keywords: *Campylobacter jejuni*, ST-257, ST-918, cross-contamination, chicken meat

Sammanfattning

Campylobacterios är den mest rapporterade gastrointestinala sjukdomen i EU sedan 2005. Campylobacterios är en zoonos och orsakas av *Campylobacter* spp. *Campylobacter* anses huvudsakligen spridas till människor via fågelkött. Många livsmedelsburna utbrott av campylobacterios har tidigare kopplats till privata hushåll. Syftet med den här studien var att simulera riskfaktorer för överföringen av *C. jejuni* från fågelkött till människa i köksmiljö.

Bröstfiléer av slaktkyckling kontaminerades artificiellt med *C. jejuni* sekvenstyp 257 och 918. Provtagning genomfördes på vanligt förekommande objekt vid matlagning, en handske (för att simulera händer), en sköljd handske (för att simulera tvättade händer), en första och andra provtagning av en skärbräda, och köksredskap (sax och pincett). När överföringen av båda sekvenstyperna till miljöproverna analyserades varierade koncentrationen av *Campylobacter* i kycklingen som hanterades i studien mellan 2.7 log₁₀ CFU/g och 5.3 log₁₀ CFU/g.

Campylobacter påvisades i alla proverna men i olika koncentrationer. Den högsta överföringen av båda sekvenstyperna var till de otvättade handskarna och från första provtagningen av skärbrädan. Överföringen av *Campylobacter* var lägre i de sköljda handskarna och i andra provtagningen av skärbrädan. Den lägsta överföringen av *Campylobacter* påvisades i redskapen som användes för uttagning av kycklingkött.

Den här studien understryker den signifikanta risken med kors-kontaminering vid hantering av kycklingkött som är kontaminerat med *Campylobacter*. Det är viktigt att förhindra kors-kontaminering vid hantering av kycklingkött, för att förhindra och minska antalet smittade med campylobacterios.

Nyckelord: Campylobacter jejuni, ST-257, ST-918, kors-kontaminering, kyckling

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Abbreviations

BHI	Brain heart infusion
BPW	Buffered peptone water
CDT	Cytolethal distending toxin
CFU	Colony forming units
ECDC	European Centre for Disease and Prevention and Control
EFSA	European Food Safety Authority
GBS	Guillain-Barré Syndrome
HACCP	Hazard Analysis and Critical Control Points
IBS	Irritable bowel syndrome
LOS	Lipooligosaccharides
MALDI-TOF	Matrix Assisted Laser Desorption Ionization – Time of Flight
mCCDA	Modified charcoal-cefoperazone-deoxycholate agar
PHC	Process hygiene criterion
RT-PCR	Real time polymerase chain reaction
ST	Sequence type

1. Introduction

1.1. *Campylobacter*

1.1.1. History

The *Campylobacter* species have been a known veterinary problem since the beginning of the 20th century. Infectious abortion in sheep and cattle was associated with the bacterium *Vibrio* (now *Campylobacter*) *fetus* (Adams et al. 2016). Later, *Vibrio* (now *Campylobacter*) *jejuni* was isolated from the intestine of calves and cattle with enteritis (Jones et al. 1931). However, in 1938 a similar organism to *Vibrio jejuni* was isolated from humans, connected to a milk-borne outbreak (Levy 1946). Two decades later, King (1957) isolated *Campylobacter* spp. from blood samples of children with diarrhoea. She observed that the isolated bacteria had an optimal growth temperature at 42°C. The isolated strains from the human blood samples were similar to other strains isolated from chickens (King 1957). The genus *Campylobacter* was established in 1963, to reclassify *V. fetus* due to phenotypic reasons. Thus, the *C. fetus* became the type species for the genus (Sebald & Veron 1963).

1.1.2. Taxonomy

The genus *Campylobacter*, originating from the Greek *kampulos* (curved) and *bacter* (rod), belongs to the family *Campylobacteraceae*, the order *Campylobacterales*, the class *Epsilonproteobacteria*, and the phylum *Proteobacteria*. Since the establishment of *C. fetus*, the genus has grown to include 34 species (<https://lpsn.dsmz.de/genus/campylobacter> 2021-04-19). The most important in terms of foodborne disease are *Campylobacter jejuni* subsp. *jejuni* (hereafter called *Campylobacter jejuni*), *Campylobacter lari*, and *Campylobacter coli* (EFSA & ECDC 2021).

1.1.3. Morphology

General traits for the members of the *Campylobacter* genus are non-sporeforming, gram-negative, and oxidase-positive rods. Cells varies in length (0.5-0.8 µm) and width (0.2-0.5 µm). They are pleomorphic, and when in log-phase, they exhibit a slender, curved, or spiral shape together with one or more flagella which is either polar or amphitrichous, hence rapid and darting motility (Adams et al. 2016).

1.1.4. Culture

Campylobacter cannot ferment or oxidize carbohydrates, instead utilises amino acids or tricarboxylic acid cycle intermediates to obtain their energy (Kaakoush et al. 2015). They are microaerophilic and have an optimal growth rate in an atmosphere containing 5-10% carbon dioxide and 1-10% oxygen (Bolton & Coates 1983). *Campylobacter* are sensitive to other extrinsic factors; temperature above 72 °C, extremes of pH, freezing, UV, disinfectants, and drying (Adams et al. 2016; Hansson et al. 2018). All species within the genus grow at 37 °C; however, as King (1957) observed, some can grow at 42 °C, these are *C. coli*, *C. lari*, *C. upsaliensis*, and *C. jejuni*. These thermotolerant species cannot multiply at temperatures below 30 °C and are preferably incubated at 42 °C to inhibit the growth of other bacteria (Adams et al. 2016).

Direct plating or enrichment followed by plating is the standard isolation method (Hansson et al. 2018). Chromogenic agar, blood-containing, and charcoal-containing media are the three main groups of specific media (Corry et al. 1995). There are different standards for detection of *Campylobacter*, the most commonly used are the International Standard Organisation 10272 (2017), Nordic Committee on Food Analysis 119 (2007), World Organization for Animal Health (2004), and United States Department of Agriculture (2007). Thermotolerant *Campylobacter* can be difficult to culture in a laboratory due to their sensitivity to many extrinsic factors. When analysing foods, the enrichment procedure can be necessary due to low concentrations of *Campylobacter*. This step is time-consuming, and the needed time may exceed five days, hence introducing difficulties during outbreak investigations. A solution to this is the real-time polymerase chain reaction (RT-PCR), which is used more frequently and enables more rapid detection of *Campylobacter* (Josefsen et al. 2015). However, there is a shortage of standardised RT-PCR methods and the method is connected to low sensitivity and specificity (Hansson et al. 2018).

1.1.5. Sources

Humans are mostly infected by *Campylobacter* after consuming contaminated or undercooked meats, unpasteurised milk, or unwashed vegetables (Hansson et al. 2018). The principal reservoirs of *Campylobacter* are the alimentary tracts of wild and domesticated birds and mammals (EFSA Panel on Biological Hazards 2010). Birds are the principal reservoir, presumably due to their high body temperature (Hansson 2007). The preparation and consumption of broiler meat may account for 20% to 30% of human cases of campylobacteriosis. The chicken reservoir may contribute 50% to 80% of human campylobacteriosis cases (EFSA Panel on Biological Hazards 2010). Hence, chicken meat is a major vehicle for human exposure to *Campylobacter*. Besides poultry meat, *Campylobacter* has been isolated in various meats at the retail level, pork, lamb, shellfish, and beef (Whyte et al. 2004; Korsak et al. 2015; Walker et al. 2019; EFSA & ECDC 2021). Furthermore, consumption of unpasteurised milk, contaminated water, and direct or indirect contact with colonised animal faeces may lead to an infection (Hansson et al. 2020).

1.1.6. Subtypes

There is evidence that suggests a variation in survival rates between strains of *Campylobacter*. *Campylobacter* is regarded as sensitive to desiccation compared to other foodborne pathogens (Fernández et al. 1985). However, one report shows that hyper-aerotolerant strains of *C. jejuni* are highly prevalent in raw chicken meats (Oh et al. 2015). Strains of aerotolerant *C. coli* are found in various retail meats (Karki et al. 2018). While under aerobic conditions, some strains of *C. jejuni* have exhibited an improved acid tolerance (Murphy et al. 2003). Therefore, these abilities among strains could increase the impact on human health by enhancing their capability to endure during the food process.

1.1.7. Virulence

In a scientific opinion from the European Food Safety Authority (EFSA) (2010), it is stated that the mechanisms of disease caused by *Campylobacter* are poorly understood. It is predominantly in the human small intestine that infection of *Campylobacter* occurs. Virulence factors of *Campylobacter* are still poorly understood, despite being one of the most prevalent foodborne diseases and the conducted research on the species. Attributes connected to the *Campylobacter* species host cell invasion and disease pathogenesis are; flagellar apparatus, various genes and proteins linked to adhesion and invasion, lipooligosaccharide (LOS) capsule, and cytolethal distending toxin (CDT) (Guerry 2007; Dasti et al. 2010; Bolton 2015; Hansson et al. 2018).

1.2. Campylobacteriosis

Campylobacter causes the human infection campylobacteriosis, a zoonosis, which is the most reported gastrointestinal infection in humans, within the EU, since 2005. The EU notification rate of campylobacteriosis in 2019 was 59.7 cases per 100.000 population, and the number of confirmed cases was 220.682. The number of confirmed cases has remained stable during the past five years (EFSA & ECDC 2021). In Sweden, the number of confirmed cases was 6.693 individuals in 2019, corresponding to a notification rate of 65 cases per 100.000 citizens (Public Health Agency of Sweden 2020).

Few studies have been conducted regarding the infectious dose of *Campylobacter* in humans. In a study by Robinson (1981), an oral dose of 500 CFU caused an infection; however, the study was only performed on one participant. Black et al. (1988) reported an infective dose of 800 CFU, the lowest dose given in that study. The usual incubation period for campylobacteriosis is 2-5 days, and common symptoms are diarrhoea, abdominal pain, nausea, headache, fever, and vomiting. Symptoms may endure for up to three weeks, though they usually last for one week. Relapses and hospitalisation may occur. Secondary diseases, e.g., Guillain-Barré Syndrome (GBS), irritable bowel syndrome (IBS), and septicemia, may occur (Allos & Blaser 1995). A previous study based on data from the Swedish Institute for Infectious Disease Control showed that the GBS incidence following campylobacteriosis was 30.4 per 100.000 cases (McCarthy & Giesecke 2001). The incidence rate of IBS following campylobacteriosis was 33.1 per 1.000 cases in an American study (Scallan Walter et al. 2019). EFSA has (2021) reported that the EU case fatality for campylobacteriosis is 0.03%. Individuals at risk are young children, the elderly, and immunosuppressed (WHO 2020).

1.3. Outbreaks

EFSA defines a foodborne outbreak as "two or more people developing the same foodborne illness after eating or drinking the same food" (EFSA 2021b). In the latest report on zoonoses in the EU, 319 outbreaks of *Campylobacter* were communicated to EFSA (EFSA & ECDC 2021). In context, a total of 3.101 outbreaks were reported, of which unknown agents caused 40% of outbreaks. *Salmonella* was the most frequently reported causative agent regarding foodborne outbreaks, followed by norovirus and other caliciviruses. With 319 outbreaks, *Campylobacter* was the third most reported cause of foodborne outbreaks. The outbreaks of *Campylobacter* led to 1.254 cases of illness, 125 hospitalisations, and no deaths. Most of the reported outbreaks due to *Campylobacter* were without speciation information. When there was substantial evidence, *C. jejuni* was the most

isolated causative agent, followed by *C. coli*. Cases with solid evidence were related to broiler meat and milk. During the last decade, broiler meat and milk have been the most common groups connected with solid evidence foodborne outbreaks of campylobacteriosis.

1.3.1. The outbreak in Sweden, 2016–2017

In 2016, nearly 7.000 cases of confirmed campylobacteriosis were reported in Sweden, which is the highest annually reported number in Sweden since the zoonosis became notifiable by law. A summary report on trends and sources of foodborne outbreaks conducted by EFSA and the European Centre for Disease Prevention and Control (ECDC) (2017) stated that the outbreak in Sweden was the most prominent foodborne outbreak within the EU. Genetic comparisons between *Campylobacter* in humans and broilers indicated a connection between the humans with campylobacteriosis and Swedish produced conventional chicken. The largest abattoir in Sweden sent out a press release which stated that a faulty installation in the cleaning system of transport crates resulted in that the crates were rinsed with contaminated water. Analyses of *Campylobacter* strains showed clear evidence that *C. jejuni* ST-918 was believed to be one of the primary causative agents for the widespread outbreak (Swedish Food Agency & Public Health Agency of Sweden 2018).

1.4. Seasonal variation

A seasonal variation in cases of campylobacteriosis is a prominent characteristic of *Campylobacter* (Humphrey et al. 2007). Clear seasonality of reported cases with campylobacteriosis has been observed within the EU between 2015-2019, with peaks in the summer months. During the past eight years, distinct peaks of reported cases within the EU have been observed during the winter (EFSA & ECDC 2021). It has proven to be challenging to explain the reason for the seasonality (Strachan et al. 2013). The seasonal peak in human cases often precedes the seasonal peaks in the prevalence of positive broiler flocks (Hansson et al. 2018). Compared to northern European countries, the seasonal effect seems to be of lesser importance in a temperate and warmer climate. A theory is, summertime changes human behaviour, and they get altered eating habits, such as conducting more barbeques, thus increasing the risk of consuming contaminated food (Strachan et al. 2013). A study by Djennad et al. (2019) reports a strong association between *Campylobacter* and temperature, though further research is needed to draw further conclusions. The main reasons for the seasonality of *Campylobacter* remain uncertain (Hansson et al. 2018).

1.5. Underreporting

Since campylobacteriosis is a zoonosis, it goes under the Zoonoses Directive 2003/99/EC (European Commission 2003). The directive obliges the EU Member States to collect data on the occurrence of zoonoses, zoonotic agents, antimicrobial resistance, animal populations, and foodborne outbreaks. Surveillance, monitoring, and reporting of this data are not harmonised within the EU, hence the risk of underestimation of the true incidence. The collected data are based on national passive surveillance, meaning there is no active search for cases. Differences in healthcare usage and laboratory practices contribute to the variation of underreporting between countries (Haagsma et al. 2013). EFSA and ECDC (2021) reported that 0.6% of human cases of campylobacteriosis came from investigations concerning foodborne outbreaks. It is difficult and time-consuming to conduct cohort studies that estimate the true prevalence of campylobacteriosis cases. There are few reports which have investigated the extent of underreporting and under-diagnosis. Studies have developed multipliers to estimate the actual number of cases. A study in the United Kingdom reported a multiplier of 9.3 for campylobacteriosis (Tam et al. 2012). There are uncertainties in these datasets, though the reports visualise that the reported incidence is underestimated (Boqvist et al. 2018).

1.6. *Campylobacter* in broilers

As previously mentioned, the preparation and consumption of broiler meat may account for 20% to 30% of human cases of campylobacteriosis. The chicken reservoir may contribute 50% to 80% of human campylobacteriosis cases (EFSA Panel on Biological Hazards 2010). There are several risk factors connected to the routes of transmission by *Campylobacter* to broiler flocks before harvest. The risk factors are connected to inadequate disinfection between chick placements, age disposition, use of multi-unit sites, proximity to other livestock, season, and failing biosecurity measures in the stable (Bouwknegt et al. 2004). One reason for the high prevalence of *Campylobacter* in broilers is the required dose of *C. jejuni* to colonise chicks and chickens, which in one study has been reported as 40 CFU (Cawthraw et al. 1996). The gastrointestinal tract is where the broilers are colonised, and they are often asymptomatic carriers when colonised. *Campylobacter* can be found in significant amounts in the faeces of colonised broilers, up to 8 log₁₀ CFU/g faeces (Stern & Robach 2003). Hence, the risk of bird-to-bird colonisation through faeces.

Later in the food chain, the colonised broilers may contaminate the meat during further processing (EFSA 2011). Faecal contamination and gut tissue damage have proven to be common risks which could lead to contamination of the carcass.

Processes at the abattoir, such as scolding, defeathering, and evisceration, are connected to contamination of the carcasses. The risk of contamination during slaughter is more significant if the chickens are colonised before slaughter, though colonised birds may contaminate the equipment in the abattoir. Hence, the risk of transferring *Campylobacter* onto uncolonised birds (Newell et al. 2017). During the slaughter of beef, pork, and lamb, water is regarded as a potential risk and is therefore avoided, hence reducing the concentrations of *Campylobacter*. However, water is regularly used during the slaughter of chickens, allowing more *Campylobacter* to endure and end up in private households (Bolton 2015).

1.7. Swedish *Campylobacter* Program

In Sweden, findings of thermotolerant *Campylobacter* are notifiable by law according to SJVFS 2012:24 (National Veterinary Institute et al. 2019). Furthermore, in 1991, the Swedish Poultry Meat Association initiated a voluntary surveillance program for broilers. The program was introduced to minimise the number of *Campylobacter*-positive broiler flocks, from primary production to consumer. A flock meaning, all broilers kept within the same enclosure and constituting a single epidemiological unit. The Swedish Poultry Meat Association is the program's operator, and it is mainly financed by the Swedish Board of Agriculture. The program is carried out by collecting ten individual ceca samples from all flocks at the slaughterhouse and analysing them as a pooled sample. The trend for *Campylobacter*-positive broiler flocks has been reduced after implementation until 2014-2016, when the prevalence increased. During 2019, the prevalence of *Campylobacter*-positive in 4 423 conventional broiler flocks was 4.6%, which is the lowest measured prevalence since the introduction of the program. The prevalence of *Campylobacter* positive organic and other broiler flocks with outdoor access was 36%, though only 72 ceca samples were analysed (Swedish Poultry Meat Association 2021).

1.8. Prevalence

In 2008, a European Union-wide baseline survey was conducted at abattoirs to determine the prevalence of *Campylobacter* and *Salmonella* within the Union. It was the first-ever baseline survey in the EU which directly investigated foodstuffs. Sampling was carried out at 561 abattoirs in 26 member states, together with Norway and Switzerland. Samples were taken from 10.132 broiler batches, and one batch included caecal contents of 10 broilers plus neck skin and breast skin from one chilled broiler carcass. Of the chilled carcasses, 75.8% were contaminated with *Campylobacter*. The prevalence of contaminated carcasses varied from 4.9% to

100% between member states. Half of the contaminated carcasses contained less than 1 log₁₀ CFU/g, and 16% contained between 3 log₁₀ CFU/g and 4 log₁₀ CFU/g. The highest count was 4 log₁₀ CFU/g, which six percent exceeded, and their actual count is not stated. Overall, *C. jejuni* was the most isolated species from the caeca and skins samples, followed by *C. coli* (EFSA 2010).

1.9. Legislation

The Baseline survey (2010) clarified that *Campylobacter* were present in all reporting member states, though in various levels. The data from the study were to be used as information for potential intervention methods and setting reduction and performance objectives. Furthermore, the panel on Biological Hazards et al. (2012) delivered a Scientific Opinion on public health hazards in poultry meat. They constructed a decision tree for risk ranking hazards in poultry meat based on the impact on human health, the number of potential human cases, and the likeliness of occurrence in poultry flocks' carcasses. Results showed that *Salmonella* spp. and *Campylobacter* spp. were considered high public health relevance for poultry meat inspection. The panel proposed introducing risk-based interventions, which would include clear and measurable targets at the carcass level concerning a particular hazard. The abattoirs were to prevent or reduce faecal contamination by using installed equipment together with their hazard analysis and critical control points (HAACP) programmes. It was also concluded that a Process Hygiene Criterion (PHC) could be introduced.

A PHC is a microbiological criterion implemented somewhere along the food chain for a specific food group to reduce the number of bacteria and improve the food safety. Codex Alimentarius (2013) defines a microbiological criterion as "a risk management metric which indicates the acceptability of a food, or the performance of either a process or a food safety control system following the outcome of sampling and testing for microorganisms, their toxins/metabolites or markers associated with pathogenicity or other traits at a specified point of the food chain." A criterion verifies the performance of a food safety control system, among other things HACCP systems, or prerequisite programs. The criterion provides information on levels of microbiological contents to food business operators, which enables the determination of the acceptance or rejection of the food (Codex Alimentarius Commission 2013).

In 2005, Regulation (EC) No 2073/2005 was introduced (European Commission 2005). With it came microbiological criteria for pathogens, such as *Listeria monocytogenes* and *Salmonella*, and established a PHC for various food categories. According to the regulation, a PHC gives an indicative value of contamination and

indicates the proper functioning of a production process. If the value is above the criterion, corrective actions are needed to maintain compliance with the food law. Regulation (EC) No 2073/2005 introduced a PHC for *Salmonella* in poultry carcasses of broilers and turkeys. However, when first introduced, the regulation did not state a PHC for *Campylobacter*.

Regulation (EC) No 2073/2005 was later amended with Regulation (EC) No 2017/1495, and a PHC for *Campylobacter* in carcasses of broilers was introduced and went into force on 1 January 2018. The regulation obligates food business operators, the abattoirs, once every week to take samples from the slaughtered broilers. The limit for *Campylobacter* was set to 3 log₁₀ CFU/g after the carcasses have been chilled. Samples are pooled from five neck skins and taken from 10 consecutive sampling sessions. The international standard EN ISO 10272-2 is the reference method for verifying compliance with the criterion (European Commission 2018).

If the criterion limit is exceeded, the food business operator must improve slaughter hygiene, review process controls of the animals, and the biosecurity measures in the farms of origin. Sampling results of the PHC became mandatory to report during the end of 2019 (EFSA & ECDC 2021). EFSA (2011) concluded, if all batches of broiler meat sold as fresh meat would comply with the 3 log₁₀ CFU/g limits, > 50% public health risk reduction could be achieved. A criterion limit of 2.7 log₁₀ CFU/g allowed a possible health risk reduction of > 90%. Data from the baseline study (2010) were used, and it was noted that 15% and 45% of all tested batches would not comply with the microbiological limit, respectively. Placing the Regulation (EC) No 2073/2005 in context, the latest zoonosis report from EFSA and ECDC (2021) presents data from the reporting member states on the prevalence of *Campylobacter* in the year 2019. Seven member-states reported results from their monitoring programs, and out of the 15.323 neck skin samples, 2.038 samples were tested positive, of which 1.033 samples exceeded the limit of 3 log₁₀ CFU/g. A report from the National Veterinary Institute et al. (2019) showed that no abattoirs in Sweden had problems meeting the criterion.

The Dutch government considered establishing a microbiological criterion for *Campylobacter* before the European Commission made the regulation. In a report by Swart et al. (2013), different criterion limits were evaluated based on their impact on public health and cost for the industry. It was concluded that a stricter criterion would improve public health and be a cost for the industry due to the higher ratio of noncomplying batches. How the cost would be distributed through price adjustments or taxes remained to be answered.

1.10. Consumer

There are many outbreaks of foodborne gastroenteritis with private households as the known source of origin (EFSA & ECDC 2021). However, the number of cases originating from private households are highly underreported (de Jong et al. 2008). The European food law only obliges food business operators to comply with regulations, and there are no surveillance systems for private households. Introducing a risk, such as *C. jejuni*, into private households puts the consumer at risk and calls for knowledge in safe hygiene practices. In Sweden, few studies have been conducted regarding consumer's knowledge of food safety. However, these studies have shown crucial consumer knowledge gaps regarding storage temperatures, pathogens, reheating, cleaning, and handling of risk foods (Marklinder et al. 2004, Marklinder et al. 2013; Lange et al. 2016).

Campylobacter is frequently introduced into private households, and broiler meat is assumed to be the most significant vehicle for distribution (EFSA & ECDC 2021). In 2020, Sweden's average total meat consumption was 78.6 kg per capita, and of which poultry was 21.5 kg. Pork is the most consumed meat, followed by beef, poultry, lamb, and other meats (Swedish Board of Agriculture 2021).

The lack of studies conducted regarding the Swedish knowledge within food safety makes it difficult to know how well informed the consumer is on the prevalence of *Campylobacter* in retail. A study performed in New Zealand reported that 15% of the interviewed individuals knew that 60-90% of retail chicken meat in New Zealand is contaminated with *Campylobacter* (Allan et al. 2018). To underestimate the level of contamination, together with lousy hygiene practices, could lead to more campylobacteriosis cases. Domestic kitchen practices are of utmost importance when *Campylobacter* is introduced into the kitchen environment (Langsrud et al. 2020). It is generally acknowledged, that good hygiene practices are essential when handling a raw product. The World Health Organization (2006) has outlined five keys to prevent foodborne diseases:

- Keep clean.
- Separate raw and cooked.
- Cook thoroughly.
- Keep food at safe temperatures.
- Use safe water and raw materials.

When preparing meat, it must be adequately cooked to avoid a direct human infection of *Campylobacter* (EFSA 2010). EFSA (2011) concluded that proper heat treatment could reduce the number of *Campylobacter* by more than 6 log₁₀ units, though a fixed temperature for an amount of time is not stated. In a study by

Sampers et al. (2010), the concentration went below 10 CFU/g when the internal temperature of a minced poultry meat patty reached 57.5 °C. According to the Swedish Food Agency an internal temperature of 70 °C for one minute will suffice if the carcass contains 6 log₁₀ units (Swedish Food Agency 2017). A study conducted on European consumers reported that consumers might assess the doneness of the chicken based on its inner colour or texture, a method that does not ensure the inactivation of pathogens. In some cases, was the juiciness of the cooked chicken more prioritised than any safety concerns (Langsrud et al. 2020).

Besides the undercooking of meat, cross-contamination is essential in transmitting *Campylobacter* from poultry meat during food preparation (EFSA 2011). EFSA's (2021a) definition of cross-contamination is "The process by which microbes are unintentionally transferred from one substance or object to another, with harmful effect." According to EFSA (2010), the contaminated meat could act as a vehicle for the distribution of *Campylobacter* and quickly spread to the kitchen equipment, among other things cutting boards, plates, and knives. Thus, *Campylobacter* could contaminate other foods in the kitchen, causing human infection if consumed without further bacteriocidal treatment.

During the years, multiple risk assessments and theoretical models of cross-contamination and the transfer rate of *Campylobacter* have been conducted (Kusumaningru et al. 2004; Uyttendaele et al. 2006; Lindqvist & Lindblad 2008; Habib et al. 2020). Lindqvist and Lindblad (2008) reported that many consumers' kitchen practices might lead to cross-contamination. Kusmaningru et al. (2004) concluded a higher probability of *Campylobacter* cross-contaminating salads than *Salmonella* and a reduction in the rate of campylobacteriosis by improving the private households' kitchen hygiene. Practical studies have been made on how *Campylobacter* are transferred from chicken meat to kitchen equipment (DE Boer & Hahné 1990; Lubber et al. 2006; Bai et al. 2020; Cardoso et al. 2021). These studies have concluded that there is a significant cross-contamination risk when handling *Campylobacter* contaminated chicken. Despite numerous studies, there are knowledge gaps within cross-contamination and the transfer of *Campylobacter*.

1.11. Aim

The purpose of this study was to simulate risk factors for transmission of *C. jejuni* from poultry meat to humans in the kitchen. To obtain more knowledge regarding the risks for the consumer to handle poultry meat contaminated with *Campylobacter*.

2. Materials and Methods

2.1. Bacterial culture preparation

In this study, two sequence types (ST) of *C. jejuni* were used, ST-257 and ST-918. ST-257 has been isolated from the water pipes in a broiler house where broiler chickens were colonised by ST-257, whereas ST-918 was isolated from transport crates after cleaning and disinfection (Frosth et al. 2020). The sequence types were chosen due to their connection with human campylobacteriosis (Public Health Agency of Sweden 2017).

The isolates were taken out of the freezer, -80 °C, and cultured on blood agar plates (SVA, Uppsala, Sweden), incubated at 37 °C h in a microaerophilic atmosphere by CampyGenTM (Oxoid, Basingstoke, UK) for 44 ± 4 h. The isolates were identified by MALDI-TOF to confirm that the isolates were *C. jejuni*. Cell suspension for the respective sequence type was made with Brain heart infusion (BHI).

2.2. Sample preparation and quality control

Swedish frozen chicken breast filet was purchased from a grocery store. Chicken breasts were thawed in a refrigerator 24 hours before cut into pieces of approximately 50 g. The cell suspension of *C. jejuni* ST-257 or ST-918 was mixed with buffered peptone water (BPW) and the pieces of chicken meat. Samples were kept at room temperature for one hour to allow an even distribution of cell suspension. Samples of chicken meat and 5 ml aliquot of cell and BPW suspension were put in individual stomacher bags. The bags were frozen and kept at -22 °C throughout the study.

From each bag of chicken breast filet, 10 g of meat were removed and used for a qualitative analysis with enrichment, to determine if the breast filets were naturally contaminated with *Campylobacter* or not. The piece of 10 g was placed in a stomacher bag together with 90 ml Bolton broth and placed in a stomacher

(easyMIX® Lab Blender AES-Chemunex, Weber Scientific, Hamilton, New Jersey, USA) and homogenized at 240 rpm for 60 seconds. The sample was incubated at 41,5 °C in a microaerophilic atmosphere for 44 ± 4 h. The enriched culture was surface spread on modified Charcoal Cephoperazone Desoxycholat Agar (mCCDA) (Oxoid, Basingstoke, UK) and incubated at 41,5 °C in a microaerophilic atmosphere for 44 ± 4 h. The analysed samples of the chicken meat chicken were tested negative for *Campylobacter*.

2.3. Environment sampling

Different sampling risk objects found in a kitchen environment were used as sampling locations in the laboratory; nitrile gloves (to simulate hands), a previously used plastic cutting board, and utensils (scissor and tweezer). The gloves were sampled with and without rinsing under tap water. The cutting board was sampled twice using two different pieces of a Wettex dishcloth. In total, five environment samples were analysed: glove before rinsing in water, glove after rinsing in water, first sampling of the cutting board, second sampling of cutting board, and utensils. Henceforth, the samples will be called: glove before, glove after, cutting board first, cutting board second, and utensils. Sampling was performed in singles, on five objects, and on 20 occasions, giving 20 samples for each environment sample and 100 samples per ST.

The day before each sampling occasion, inoculated chicken broiler filet was thawed in a refrigerator overnight. On the day of analysis, five pieces of broiler filet were placed on a plastic cutting board and sampled within another Master thesis performed by Ella Råhlén. The last sampled filet was used for the environmental sampling.

The sampling of glove before (simulated unwashed hand) was performed by the piece of meat was held (Figure 1), put down, and held once again. The nitrile glove was removed from the hand and placed into a stomacher bag, Figure 1. The sampling of glove after (simulated washed hand) was repeated as before, except the glove was rinsed under running tap water for one second before removal. For the remaining samples, a Wettex dishcloth, 20 cm*17.5 cm, was cut into four pieces, wholly soaked in cold tap water, squeezed hard once, and then used for sampling. A piece of Wettex dishcloth was used to swab the contact area of the utensils (a scissor and a tweezer), which had recently been used on the chicken sample (Figure 1). The dishcloth was placed in a stomacher bag and used for quantitative and qualitative analysis. In the first sampling of the cutting board, a Wettex dishcloth was used to swab the contaminated area of the plastic cutting board, Figure 1, and the dishcloth was placed in a stomacher bag. The second sampling of the cutting

board was swabbed with another Wettex dishcloth, and the dishcloth was placed in another stomacher bag.

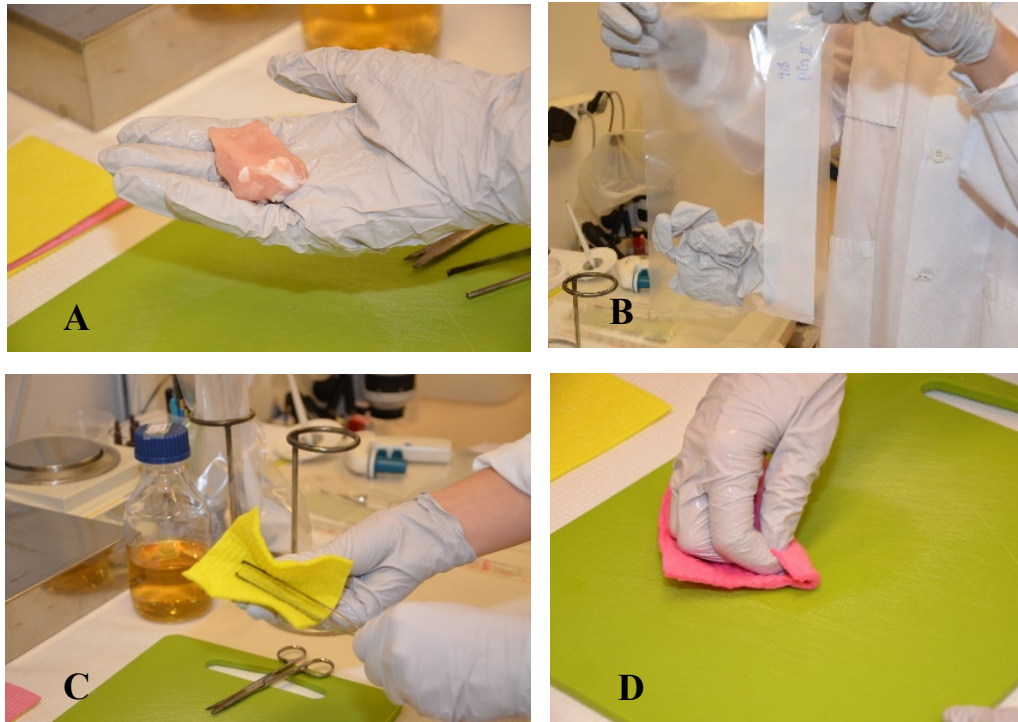


Figure 1. A: Simulation of how the hands (nitrile gloves) get contaminated with the inoculated meat. B: After the glove “before” had handled the chicken meat. C: Depicts how the sampling was conducted on the tweezers. D: Sampling conducted on the cutting board, using a Wettex dishcloth.

2.4. Quantitative analysis

For the quantitative analysis of each environment sample, 10 ml of BPW was added into each stomacher bag with a nitrile glove or Wettex dishcloth and placed in a stomacher and blended at 240 rpm for 60 seconds. A 10-fold serial dilution in 0.1% (v/v) peptone water (Dilucups, LabRobot Products AB, Stenungsund, Sweden) was prepared, and 0.1 ml from dilution in the range 10^{-1} - 10^{-3} , was surface spread on mCCDA, and incubated at 41.5 °C in microaerophilic atmosphere for 44 ± 4 h. The number of *C. jejuni* was expressed as \log_{10} CFU/ml, the detection limit was 1 \log_{10} CFU/ml, the countable range was determined to ≤ 150 CFU in line with ISO 10272 (2017).

The CFU was calculated using the standard formula from ISO 7218 (2014):

$$N = \sum C / (V \times 1.1 \times d)$$

- N : number of microorganisms.
- $\sum C$: sum of the colonies on two plates from successive dilutions.
- V : volume of the inoculum/plate, in ml.
- d : the first countable dilution retained.
- 1.1: a factor used when the weighed mean is calculated from two plates, if only one plate is used, the factor is 1.0 and 1.11 if three plate is used.

2.5. Qualitative analysis

In addition to the quantitative analysis, qualitative analysis with enrichment of each environment sample was carried out, 90 ml Bolton broth was transferred to each stomacher bag, where the dishcloth remained, and incubated at 41,5 °C in a microaerophilic atmosphere for 44 ± 4 h. If the quantitative analysis was below the detection limit, $1 \log_{10}$ CFU/ml, the corresponding enriched culture was plated on mCCDA and incubated at 41,5 °C in a microaerophilic atmosphere for 44 ± 4 h. Samples below detection limit but detected after enrichment, were reported as $(0 + \text{detection limit}) / 2$ meaning $0.7 \log_{10}$ CFU/ml.

2.6. Statistical analysis

The number of *C. jejuni* in the environmental samples are compared with the corresponding concentration of *C. jejuni* in the meat. The concentration of *C. jejuni* in the meat is the mean value of the five sampled pieces of chicken broiler filet, taken from Ella Råhlen's Master Thesis at each sampling occasion.

The mean transfer rate of *C. jejuni* from the chicken to respective environmental sample was calculated by:

$$\frac{\sum ((CFU/ml \text{ environmental sample}) / (CFU/g \text{ meat sample}))}{\text{number of samples}}$$

The standard deviation was calculated for each transfer rate. The transfer rate represents the transfer of *C. jejuni* from the inoculated chicken to each environmental sample expressed in percent. Statistical analysis was performed using Excel (version 2103, Microsoft, Redmond, Washington, USA).

3. Results

ST-257 was isolated from all the 20 samples although from four of the samples, the amount of *Campylobacter* was below the detection limit for quantification. Those samples were from the utensils and twice in the second sampling of the cutting board (Figure 2). The highest number of *Campylobacter* quantified were samples from gloves before washing and the first sampling of the cutting board. Glove after and the second sampling of the cutting board had a lower concentration of ST-257 than sampling before. In some cases, a higher number of *Campylobacter* could be quantified from the first sampling of the cutting board and glove before washing than the corresponding mean value of concentration of ST-257 in the meat. Utensils corresponded with the lowest transfer of ST-257.

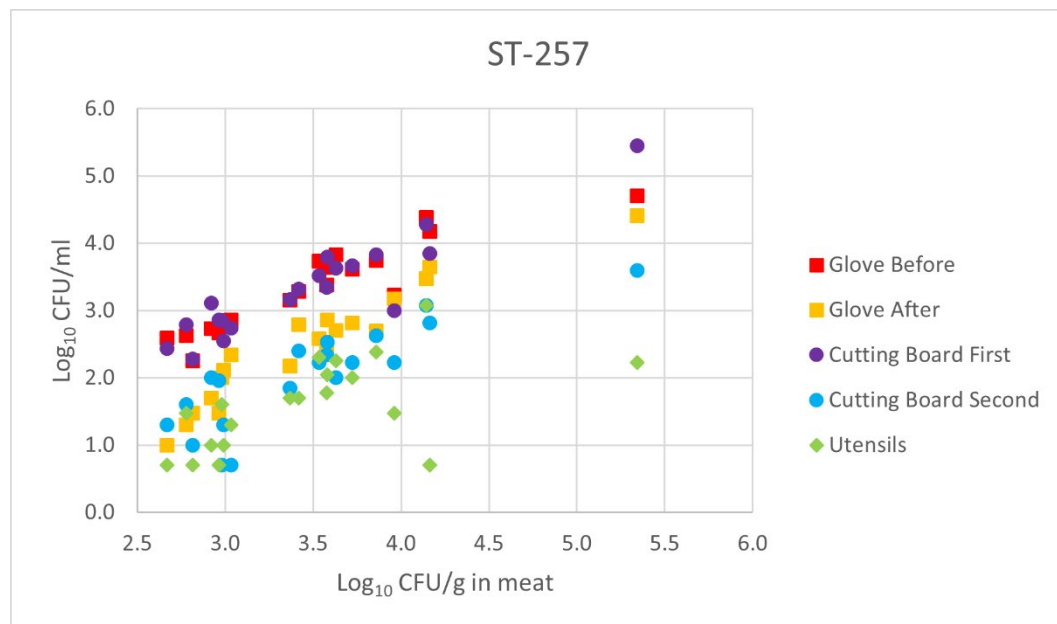


Figure 2. Distribution of *C. jejuni* ST-257 (\log_{10} CFU/ml) in environment sample compared with the concentration in the meat. Concentrations below the detection limit were set at 0.7 \log_{10} CFU/ml, according to the material and method.

ST-918 could be quantified in 18 of the 20 samples, since, two utensils samples were below the detection limit (Figure 3). However, ST-918 was isolated after enrichment in those below the detection limit. Utensils and cutting board after corresponded with the lowest transfer of ST-918 from the meat to the environmental samples. Glove before washing and the first sampling of the cutting board had the highest transfer of ST-918, and in some cases, a higher concentration observed than in the meat. Glove after and cutting board after had lower concentrations of ST-918 compared with the corresponding before in all samples (Figure 3).

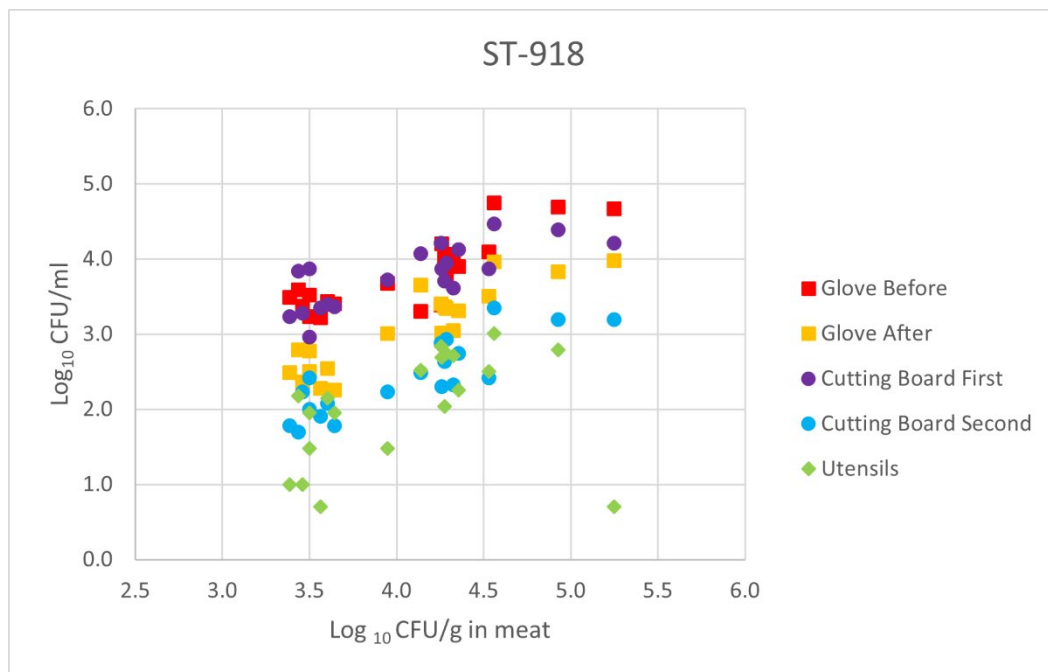


Figure 3. Distribution of *C. jejuni* ST-918 (\log_{10} CFU/ml) in environment sample compared with the concentration in the meat. Concentrations below the detection limit were set at $0.7 \log_{10}$ CFU/ml, according to the material and method.

A mean transfer rate for each environmental sample and sequence type was calculated from the artificial contaminated chicken meat and the concentration on each environmental sample (Table 1). The transfer rate varies between 82% in the first sampling of the cutting board ST-257 and 2% in utensils ST-918. There is a significant reduction of the transfer rate in the glove after when it has been rinsed in tap water for one second. The second sampling of the cutting board presents a significant reduction in transfer rate. The reduction of transfer rate observed in glove after and the second sampling of the cutting board is presented in both sequence types. Overall, the ST-257 has a larger or equal transfer rate in the samples compared with the ST-918.

Table 1. Mean transfer rates of C. jejuni ST-257 and ST-918 from the meat to the kitchen equipment. SD = Standard deviation.

Sample	ST-257 (%±SD)	ST-918 (%±SD)
Glove Before	80±0.43	65±0.40
Glove After	12±0.08	12±0.07
Cutting Board First	82±0.41	70±0.63
Cutting Board Second	5±0.03	3±0.02
Utensils	2±0.02	2±0.01

4. Discussion

This thesis aimed to study simulated risk factors for transmission of *C. jejuni* from poultry meat to humans in the kitchen. The measured transfer of *C. jejuni* emphasizes the significant risk of cross-contamination when handling poultry contaminated with *C. jejuni*.

The transfer of ST-257 and ST-918 differ depending on sampling location and sequence type. The glove before always had a high transfer, in ST-257, the transfer rate was 80%, whereas in ST-918 it was 65% (Table 1). In some cases, the concentration of *Campylobacter* was higher in the environment sample than in the chicken meat (Figure 1 and Figure 2). When analysing chicken meat, studies have found external contamination to be higher than internal contamination (Hansson et al. 2015). In the analyses of chicken meat, the meat samples were collected from both internal and external part of the chicken meat. The used samples of internal muscle could explain why the environmental sample had higher concentrations than the chicken meat. The transfer rate of ST-257 and ST-918 was significantly lower when the gloves had been washed (Figure 1 and Figure 2). The gloves were rinsed in tap water for one second, which apparently was enough to reduce the concentration in the gloves (hands) of both sequence types. Nitrile gloves were used to simulate hands, these gloves have a smoother surface than hands which may lead to a more extensive removal of *Campylobacter* when rinsing in running tap water. Studies indicate that consumers do not always wash their hands after handling raw poultry meat (Worsfold & J. Griffith 1996; Li-Cohen & Bruhn 2002; Cardoso et al. 2021). The transfer of ST-257 and ST-918 onto gloves in this study show the risk of cross-contamination handling chicken meat and the importance of washing the hands.

The transfer rate from the cutting board was higher, 82% for ST-257 compared with ST-918, 70% (Table 1). A few of the first samplings of the cutting board had a higher concentration of *Campylobacter* than the corresponding mean value of the five pieces of chicken broiler filet (Figure 1 and Figure 2). The high concentration of *Campylobacter* might be due to the higher external rate of contamination on the meat than the internal contamination. The second sampling of the cutting board had a significantly reduced transfer of *Campylobacter*, irrespective of the sequence type

(Figure 1 and Figure 2). The sampling from the cutting board was below the detection limit twice in the ST-257, even though it was quantified in the same concentration and lower mean concentrations of *Campylobacter* in the chicken. The reason may be that the piece of meat used for sampling had a higher concentration of ST-257 than the mean value of ST-257 from the five pieces of meat. Alternatively, the amount of meat juice varies between sampling occasions, depending on the meat's capability to retain water. If the cutting board remains unwashed, there is a likelihood of cross-contaminating other foods (Habib et al. 2020). It is essential to wash or replace the cutting board to reduce the risk of cross-contaminating, even small failures in doing so may lead to human infection, since the infectious dose may be as low as 500 CFU (Robinson 1981; Verhoeff-Bakkenes et al. 2008). The washed glove and the second sampling of the cutting board show a lower transfer of *Campylobacter* (Figure 1 and Figure 2). In future studies, different cleaning scenarios of the hands and equipment should be analysed. By prolonging the glove's washing time or using a detergent on the cutting board, there might be an even lower transfer of *Campylobacter* to the equipment.

A previously used plastic cutting board was used to mimic the private kitchen. The National Institute of Public Health and the Environment concluded that a cutting board presents a significant risk of cross-contamination (National Institute for Public Health and the Environment 2020). Bai et al. (2020) found that the plastic cutting board had a statistically significantly lower transfer rate than a wooden cutting board. Hence, there might be a difference in cross-contamination depending on the material of the cutting board. There is also a need for further studies with lower concentrations of *Campylobacter* in the meat. In a French study, using less than 1 log₁₀ CFU/g *Campylobacter* in the meat, *Campylobacter* were transferred via a cutting board to cooked chicken meat (Guyard-Nicodème et al. 2013).

The first and second sampling of the cutting board and the utensils were sampled with a Wettex dishcloth. In previous studies, the domestic dishcloth has been shown to harbour a significant concentration of bacteria (Hilton & Austin 2000; Gillies 2020). Cardoso et al. (2021) were able to isolate *Campylobacter* from a kitchen cloth and connected it to unsafe handling practices by the consumer. A sterile cotton swab could have been used to sample the cutting board and utensils, though the Wettex dishcloth was used to mimic the kitchen environment.

The utensils corresponded with the lowest transfer in both sequence types. It fell below the detection limit twice in ST-918 and four times in ST-257. The low transfer could be explained by the small surface area of stainless steel of the scissor and tweezer. According to Kusumaningrum et al. (2003), *C. jejuni* can endure on stainless steel surfaces. However, their susceptibility to drying hinders them from

enduring for more extended periods and allows for a shorter survival time than *Salmonella*. It is important to remember the variation between strains and aerotolerant strains of *C. jejuni* (Oh et al. 2015). It may not be common practice in private households to use a scissor and tweezer, suitable for laboratory work when handling raw chicken. Thus, to have kept the endeavour to mimic the kitchen environment consistent, one might have used a kitchen knife instead. The kitchen knife has a larger surface area that may have retained more meat juices than the scissor and tweezer, allowing a more extensive transfer of *Campylobacter*. It was deemed more practical to use scissors and tweezers since they were used to sample the chicken broiler filets in the Master thesis performed by Ella Råhlén. Furthermore, in a study on naturally *Campylobacter* contaminated chicken breast filets, the transfer of *Campylobacter* were sometimes higher to the knife than to the cutting board (Luber et al. 2006). The importance of adequate cleaning and replace used cutlery has previously been established (de Jong et al. 2008; Verhoeff-Bakkenes et al. 2008). The highest observed transfer was 3.1 and 3 log₁₀ CFU/g for ST-257 and ST-918, respectively (Figure 1 and Figure 2). The transfer visualises the significant risk of cross-contamination when using small unwashed utensils.

No studies have previously analysed the transfer rate of ST-918 and ST-257 to the kitchen equipment, though, multiple studies have used other sequence types (de Jong et al. 2008; Verhoeff-Bakkenes et al. 2008; Bai et al. 2020). Bai et al. (2020) measured a transfer rate of 12% from artificially contaminated chicken onto gloves. However, in that study they used another ST of *C. jejuni*, and a 6 log₁₀ CFU/g suspension to contaminate the meat. In this thesis, the highest measured concentration of *C. jejuni* in the meat was 5.3 log₁₀ CFU/g and 5.2 log₁₀ CFU/g for ST-257 and ST-918, respectively. When the aim is to determine the transfer rate, it is crucial to use broiler meat with a contamination level that are closer to natural contaminated broiler meat. When the transfer rate is expressed in percent, the transfer rate will become lower if a high concentration of *C. jejuni* is used to contaminate the meat. Vice versa, the transfer rate will be higher if a lower concentration of *C. jejuni* is used to contaminate the meat. The concentration of *Campylobacter* in the contaminated meat should therefore be closely related to reality to accurately determine the transfer rates (Montville & Schaffner 2003). According to the Baseline survey (2010), half of the contaminated carcasses contained less than 1 log₁₀ CFU/g, and 16% contained between 3 log₁₀ CFU/g and 4 log₁₀ CFU/g. At the same time, 6% exceeded 4 log₁₀ CFU/g. The latest report on zoonoses by EFSA and ECDC (2021) confirms that the counts of *Campylobacter* remain on a high level since half of the contaminated chicken carcasses exceeded the PHC of 3 log₁₀ CFU/g. In this thesis, the concentration of *C. jejuni* in the chicken meat used for the environmental sampling was more than 4 log₁₀ CFU/g on 15% and 55% of the 20 sampling occasions for ST-257 and ST-918, respectively (Figure

1 and Figure 2). The used concentrations of *C. jejuni* in the chicken meat are high, though it is essential to understand the significant risk of cross-contamination when the concentration of *Campylobacter* is high. In future studies, lower concentrations of *Campylobacter* could be used to investigate the risk of cross-contamination further.

There was a difference in transfer rate between ST-257 and ST-918 (Table 1). The difference might be due to higher concentrations of ST-918 in the broiler chicken filets compared to the concentrations of ST-257, thus lowering the transfer rate of ST-918. Another reason, there might be a difference in their ability to adhere to different surfaces. In a study on naturally contaminated chicken, *C. jejuni* isolates were significantly more adherent to inert surfaces than isolates of *C. coli*. Though isolates of *C. jejuni* and *C. coli* had a solid or moderate adhesion ability, further research was needed to explain if adhesion was linked to intrinsic properties of the strains (Guyard-Nicodème et al. 2013). In the outbreak that occurred 2016-2017 in Sweden, ST-918, was believed to be one of the primary causative agents. ST-918 connection to the outbreak may indicate that the sequence type exhibits abilities to survive stress better than other sequence types of *Campylobacter*. The ability to survive stress does not explain the difference in transfer rate between ST-918 and ST-257, only that there exist different intrinsic abilities of the sequence types, which needs to be investigated further.

A limitation of the method used in this study is that it only measures the transfer of *Campylobacter* from the chicken meat to hands and equipment and not to the product which is consumed. The transfer of *Campylobacter* from chicken meat to ready-to-eat products, cucumber or cooked chicken, via kitchen equipment has previously been established (de Wit et al. 1979; Luber et al. 2006; de Jong et al. 2008; Verhoeff-Bakkenes et al. 2008). The result of this thesis does not state the concentration of *Campylobacter* in any consumable product, though it further specifies the significant risk of cross-contamination if the chicken is contaminated with *Campylobacter*. Since the infectious dose of *Campylobacter* is low, 500 CFU, it is highly relevant to extend the knowledge within cross-contamination (Robinson 1981). Future studies would be of interest to analyse the complete route of cross-contamination and analyse the transfer of different sequence types on ready-to-eat products.

The two sequence types, ST-257 and ST-918, have previously been isolated from infected humans and related to foodborne outbreaks in Sweden (Public Health Agency of Sweden 2017). Other sequence types, which have not been related to human infection or foodborne outbreaks, could be used in future studies. Since there

is a variation between sequence types, there might be a difference in *Campylobacter* abilities to cross-contaminate kitchen equipment and therefore cause illness.

5. Conclusion

This thesis presents the transfer of two different sequence types of *Campylobacter* onto hands and kitchen equipment during the handling of artificially contaminated chicken meat. *Campylobacter* were isolated from all samples, and the glove (simulated hands) and cutting board were presented as the highest risk factors for transmission of *C. jejuni* from poultry meat to humans by kitchen equipment. It is of utmost importance as a consumer to prevent cross-contamination during the handling of *Campylobacter* contaminated chicken, to prevent or reduce the risk of campylobacteriosis.

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Appendix 1



The risk of handling poultry meat with *Campylobacter jejuni* - from the consumer's perspective

CONCLUSION

The results emphasizes on the significant risk of cross-contamination when handling poultry contaminated with *Campylobacter*.



Sampling conducted on the cutting board, using a Wettex dishcloth.

Aim

The purpose of this study was to simulate risk factors for transmission of *C. jejuni* from poultry meat to kitchen equipment.

Introduction

Campylobacteriosis, caused by *Campylobacter* spp., is the most reported gastrointestinal infection in humans, within the EU, since 2005. Poultry is believed to be a significant vehicle for human exposure to *Campylobacter*. Many foodborne outbreaks of campylobacteriosis have been connected to private households.

Method

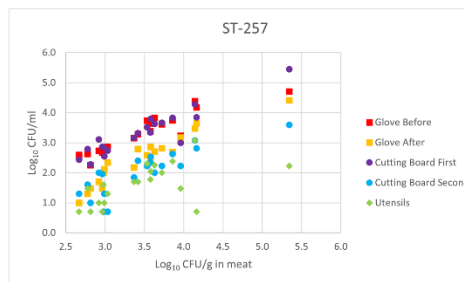
Chicken filets were artificially contaminated with two *C. jejuni* sequence types, ST-257 and ST-918. Five objects were sampled; gloves and washed gloves (to simulate hands), first and second sampling of cutting board, and utensils (scissor and tweezer). The analysis was performed according to ISO 10272 part 1 and 2.



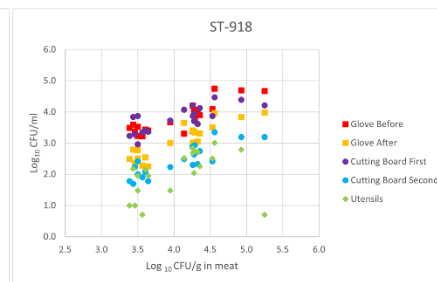
Simulation of how the hands (nitrile gloves) get contaminated with the chicken meat and washed.

Results

Campylobacter were isolated in all samples. The unwashed glove and the first sampling of the cutting board presented the highest transfer of *Campylobacter*. There was no significant difference in transfer rate between ST-918 and ST-257.



Distribution of *C. jejuni* ST-257 in environment sample compared with the concentration in the meat.



Distribution of *C. jejuni* ST-918 in environment sample compared with the concentration in the meat.



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