OCURRENCE OF *Campylobacter* SPP. IN REFRIGERATED POULTRY PRODUCTS IN CENTRAL REGION OF RIO GRANDE DO SUL

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**ABSTRACT**

Foodborne infections in humans caused by *Campylobacter* spp. result in large economic losses and are related to the production and slaughter of poultry, important steps in the spread of bacteria. The objective was to assess the occurrence of *Campylobacter* spp. into refrigerated chicken products in the central region of Rio Grande do Sul and analyze the effect of freezing on the degree of contamination. One hundred thirty five samples of giblet and poultry meat were purchased at supermarkets in the central region of Rio Grande do Sul and forwarded to LCDPA, UFSM. The samples were then enriched and plated on appropriate medium for the isolation of bacteria. When the bacteria culture were positive we performed the process of Gram stain and the colonies were then analyzed qualitatively by optical microscopy (1000x). All refrigerated samples were analyzed and the remaining material was frozen at -20 °C for analysis after a certain period. Thus sixty samples were frozen for a week and seventy five samples were frozen for two months. After that they were analyzed in the same way as cold samples. Of the total samples cooled 57% were positive for *Campylobacter* spp. After freezing the same material showed positivity of 35.5%. There was a 21.5% reduction in contamination by this bacterium after the freezing process. The samples frozen for one week had 23.4% reduced contamination, while the samples that were frozen during two months had the decrease contamination in 20%.

**KEYWORDS:** foodborne disease, bacterial infection, *Campylobacter*iosis, chicken.

**INTRODUCTION**

Foodborne infections in humans caused by *Campylobacter* spp. represent an important public health issue (HERMANS, 2011), result in large economic losses (CHAVES, 2007) and are related to the production and slaughter of poultry, important steps in the spread of bacteria (RAUT et al., 2012).

*Campylobacter* spp. is a Gram negative bacterium and belongs to the family *Campylobacteriacea* that requires microaerophilic ambient for growth. They are curved rods and usually found in the gastrointestinal tract of mammalian and birds. When two or more bacterial cells are grouped together they form an “S” shape or the format of gull-wing (FONSECA, 2006). Infection in humans is extensive and is related to the consumption of raw poultry and the cross contamination of uncooked food. The most important species related to human infection are *Campylobacter jejuni* and *Campylobacter coli* (RAHIMI, 2011).

*Campylobacter* spp. grows between 30 and 45°C, with an optimum range between 41 and 45°C and pH between 5.6 and 8. Thus, the body temperature (41-42 °C) and the intestinal environment make the chicken an ideal host to the agent. The role of *Campylobacter* spp. as foodborne pathogen is associated to the ability of survival in food during storage and manipulation. Therefore, the temperature plays
a key role for survival (GONZÁLEZ, 2009).

Studies have shown high levels of contamination by Campylobacter spp. properties in poultry, a variation between 40% and 100% in the contamination of commercialized chicken meat. The contamination of poultry carcasses occurs directly through the intestinal contents or indirectly via equipment and water at the time of processing (RAHIMI, 2011).

Studies performed to evaluate contamination by Campylobacter spp. in raw and later frozen chicken found no significant reduction after freezing the samples (SAMPERS et al., 2010). There was no difference in the recovery of bacteria from this gender of samples of fresh, refrigerated or frozen poultry meat after selective enrichment, showing that these samples can survive under storage conditions (MAZIERO et al., 2010).

The objective of this study is to assess the occurrence of Campylobacter spp. in refrigerated chicken products in the central region of Rio Grande do Sul and analyze the effect of freezing on the degree of contamination.

MATERIALS AND METHODS

Refrigerated chicken cuts (thigh and drumstick, wing, heart, liver, and gizzard) were purchased in supermarkets located in the central region of Rio Grande do Sul, totaling 135 samples. Of this total, 60 refrigerated samples were analyzed immediately after collection and also after freezing for a week, and 75 cooled samples were analyzed immediately after collection and after freezing for two months.

Ten grams of each sample was placed in 100mL of enrichment medium selective for Campylobacter (TECRA®), and incubated at 42°C for 24h. The remaining material of each sample was frozen at -21°C for one week (168h), or two months (1440h). After this period the samples were defrosted and passed also through the same process carried out with the refrigerated material.

After the enrichment, samples were plated in selective culture media composed of Columbia Agar plus 0.4g% of activated charcoal, antibiotic supplement (rifampicin cefsulodina and polymyxin B sulfate) FBP supplement (ferrous sulfate, sodium bisulfite and sodium pyruvate). The plates were incubated in microaerophilic conditions (85% N₂, 10% CO₂ and 5% O₂) for 72h. at 42°C. The atmosphere necessary for the proper development was obtained using microaerobac gas generator (PROBAC®). Then, the mixture was immediately placed into a special atmospheres jar sealed to the gas levels, reaching the correct proportions.

The colonies were evaluated for morphology and staining characteristics on Gram staining, using as contrast basic fuchsin instead of safranin due to inability of Campylobacter spp. in flushing by the latter. The stained slides were analyzed by optical microscopy (1000x) searching for curved rods or spiral cells, shaped “s” or gull-wing characteristics of the genre.

RESULTS AND DISCUSSION

Of the 135 samples cooled, 77 (57%) were positive for Campylobacter spp. After freezing for one week or for two months the same material showed average positivity of 48 samples (35.5%). There was a reduction in contamination of 29 samples (21.5%) after the freezing process.

All the 60 samples that were frozen for one week showed a medium reduction contamination of 23.4%, 45 refrigerated samples (75%) were positive and after the freezing period 31 samples (51.6%) were positive. In turn, the reduction on contamination of the 75 samples that were frozen for two months was 20%, where 32 refrigerated samples (42.6%) were positive before freezing, and 17 samples (22.6%) were positive after freezing.

After freezing there was reduction of positive samples with no significant difference between the period used for freezing during one week and two months. The average positive samples after freezing for a week and for two months were 44.44% in thigh of the wing;
33.33% in thigh and drumstick; 37.03% in liver; 40.74% in heart and 22.22% in gizzard. This way, there was a medium reduction of contamination of 37.04% in thigh of the wing; 37.04% in thigh and drumstick; 11.11% in liver; 7.4% in heart, and 14.81% in gizzard.

The temperature plays a key role in the survival of Campylobacter spp. The reduction of viability at -20°C is associated to the effect of heat treatment in the process of this microorganism cell susceptible to oxidative stress (GONZÁLEZ, 2009). Campylobacter jejuni was able to survive up to 56 days, frozen at -20 °C, which is the temperature of most domestic freezers (MAZIEIRO, 2010).

CONCLUSION
Among the refrigerated samples, 57% were positive for Campylobacter spp. There was a reduction of 21.5% in average contamination after the freezing process. Freezing the samples for 7 and 60 did not significantly reduce the positivity to this agent, indicating that this thermal method does not effectively control the contamination within this period of time.

REFERENCES