

Original Research Article

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Detecting the Occurrence of *Campylobacter jejuni* in Chicken Meat by PCR in Retail Outlets of Chennai, India

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ABSTRACT

Keywords

C.jejuni, *Hyp* gene, PCR, Chicken meat

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A study was conducted to identify the presence of *Campylobacter jejuni* in chicken meat by polymerase chain reaction. Campylobacteriosis is a food-borne bacterial illness due to the consumption of poultry meats and its by-products. Contaminated raw or undercooked poultry meats and/or by-products are particularly important to cause food-borne. Due to improper slaughtering method, carcass may get contaminated with intestinal contents. Around 36 samples were collected from different parts of Chennai and were tested for the presence of *Campylobacter jejuni* by targeting *Hyp* gene with 500bp by PCR. No samples were shown to be positive for *Campylobacter jejuni*. The test concludes that the processing of chicken was done in perfect manner without any cross contamination.

Introduction

Campylobacter jejuni is present in the gastro intestinal tract of all animals. Due to improper slaughtering method, the carcass may get contaminated with intestinal contents.

Contaminated raw or undercooked poultry meats and/or by-products are particularly important to cause food-borne Campylobacteriosis in humans (CDC, 2005).

The prevalence of contamination with *Campylobacter* species were approximately 60% in any portions of raw poultry meats and by-products except for fillets and hearts. In most of the countries (both developed and developing countries), in spite of their sanitary conditions, a majority of retail poultry meats

and by-products were contaminated with *Campylobacter spp.* *C.jejuni* was usually the dominant one, but the ratio of *C.coli* to *C.jejuni* varied among developed countries (Suzuki and Shigeki, 2008). The raw chicken meat has very high *Campylobacter* contamination levels.

Campylobacter spp. was found in higher levels in carcass-rinse or carcass-rinse plus whole skin samples (Jorgensen *et al.*, 2002). Polymerase chain reaction (PCR) is a rapid method with both high sensitivity and specificity for rapid detection and identification of pathogenic bacteria from different food matrix. Hence, the present study is undertaken for detection of *Campylobacter jejuni* from chicken by using polymerase chain reaction in different zones of Chennai.

Materials and Methods

Around 36 chicken meat samples were collected from different retail outlets of Chennai city. The samples placed in sterile polythene bags and transported hygienically to the Department of Meat Science and Technology, Madras Veterinary College, Chennai – 7 in clean insulated box with ice packs. Before screening, 25 gram of meat sample was homogenized in 225 ml of BPW and incubated at 37°C for 18 hours. The meat homogenate obtained was then subjected to DNA extraction using Bacterial DNA extraction kit and PCR analysis for the presence of *C.jejuni* by targeting *Hyp* gene with 500bp. A 20 µl of reaction mixture was set up in 0.2 ml PCR tube with following components such as master mix - 10µl, forward primer-1 µl, reverse primer-1 µl, template DNA-1 µl and nuclease free water-7 µl.

The PCR amplification was carried out in Master Cycler Gradient Thermo cycler (M/s. Eppendorf, Germany) with the following cycling conditions of initial denaturation at 94°C for 5 minutes, followed by 30 cycles of denaturation (94°C for 30 seconds), annealing (52°C for 30 seconds) and extension (72°C for 30 seconds) and subsequently a final extension at 72°C for 7 minutes. The PCR product obtained was subjected to electrophoresis in 2% Agarose gel. Ethidium bromide with concentration of 10mg/ml was added at the rate of 5µl / 100 ml of Agarose. Electrophoresis is carried out using 1X TAE buffer at 100 volts for 30 minutes. The gel was viewed under UV illuminator and documented using gel documentation system.

Results and Discussion

Chicken meat samples of around 36 numbers were collected from different retail outlets of Chennai. The meat homogenate obtained was

subjected to DNA extraction using Bacterial DNA extraction kit and the developed PCR was used to detect *Campylobacter jejuni*. None of the sample showed positive for the presence of *Campylobacter jejuni* in the retail chicken meat by PCR (Figure 1). Screening of chicken meat from different zone wise details were given below (Table 1).

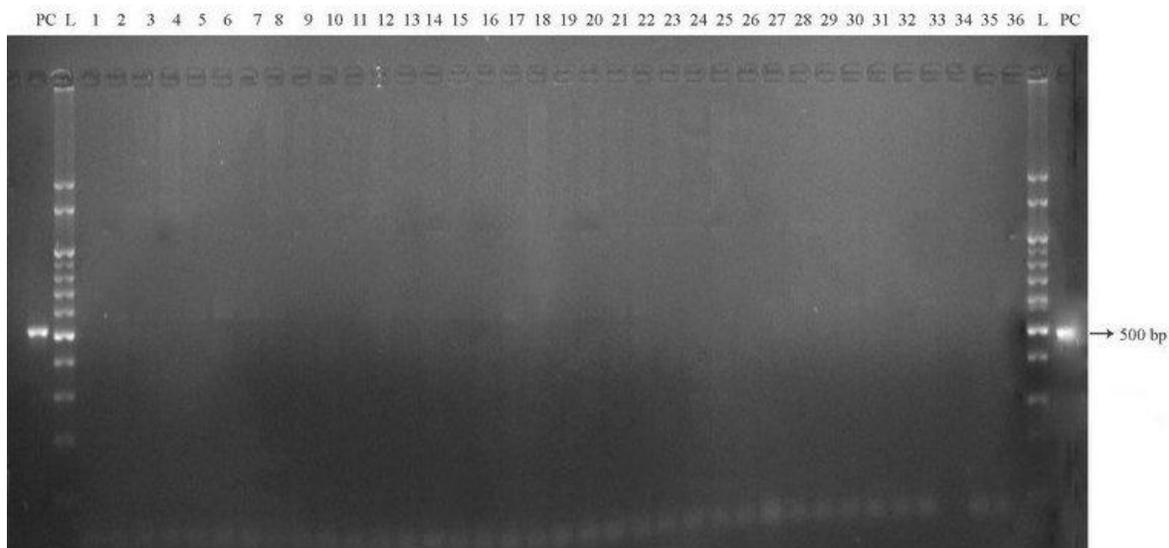
This screening study reveals that chicken meat samples collected from retail outlets for the presence of *C.jejuni* is to assess the effectiveness and efficiency of the PCR technique and the level of processing of retail chicken meat. The 36 chicken meat samples collected from different areas of Chennai city were not positive for *Campylobacter jejuni*. *Campylobacter jejuni* are found in domestic animals and also in raw poultry meat (Humphrey *et al.*, 2007). In this study, the *Hyp* (Hippurate) gene was targeted for PCR amplification of *Campylobacter jejuni* since the gene was specific and is responsible for hippurate hydrolysis which is a specific biochemical and confirmatory test. Results of this study revealed species specific bands of 500 bp in 2% agarose gel (Nakari *et al.*, 2008).

Similar results were reported by targeting *hipO* gene for detection of *C. jejuni subsp. jejuni* (Rahimi *et al.*, 2010). He found higher prevalence of *Campylobacter* (61.7%) in chicken meat from retail markets of Iran. A study was conducted and found that the level of contamination of poultry meat with *Campylobacter jejuni* was 50.9% (Atanassova and Ring, 1997). Around 1 in 1000 infections leads to Guillain–Barre syndrome (GBS), the risk increased to around 1 in 200 for patients infected with a particular *C. jejuni*, Penner type HS:19 (Nachamkin, 2002). An overlap of 34% between sero-/genotype combinations in sporadic *C. jejuni* infections in chicken flocks at slaughter during a seasonal peak (Karenlampi *et al.*, 2003).

Table.1 Screening of chicken meat samples collected from different retail outlets of Chennai

Zone	Name	No. of samples	No. of positive samples by m-PCR
			<i>Campylobacter jejuni</i>
1	Thiruvottiyur	5	-
2	Madhavaram	5	-
3	Royapuram	5	-
4	Ambattur	5	-
5	Anna nagar	5	-
6	Teynapet	5	-
7	Kodambakkam	6	-
Total		36	0

Fig.1



Note: L: 100 bp DNA Ladder, 1-36: Sample result showing absence of *Campylobacter jejuni* in chicken meat, PC: Positive control of *Campylobacter jejuni* with 500bp, NC: Negative control

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