

Screening of Chicken Meat in Chennai for Emerging Food Pathogens by Multiplex Pcr



Veterinary Science

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ABSTRACT

Meat can be contaminated at any stage of processing. *Campylobacter jejuni* and *Listeria monocytogenes* are the emerging food pathogens in chicken meat. These microorganisms are located in the intestine of chicken and can contaminate the meat due to improper handling. A study was conducted to collect the samples from retail outlet of all zones of Chennai. 90 samples were collected from 15 zones and are tested for the presence of *C.jejuni* and *L.monocytogenes* by targeting *Hyp* and *prfA* gene with 500bp and 290 bp respectively by multiplex PCR. None of the samples were shown to be positive for both microorganisms. The test reveals that the processing can be performed hygienically.

INTRODUCTION

Food Safety and Inspection Service (FSIS) and other regulatory agencies are framing new rules and regulations to produce hygienic meat for consumers. Moreover, microbial quality of meat is an important aspect for industries engaged in production, processing and distribution. Meat can be contaminated at various stages of production by most of the microorganisms. Recently many pathogens such as *Listeria monocytogenes*, *Campylobacter spp.*, *Vibrio spp.*, etc. have been identified as emerging meat borne diseases. In most of the countries (both developed and developing countries), inspite 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).

Listeria monocytogenes is widely distributed and found in many food commodities. It is very persistent microorganism that survives on surfaces and equipment of food processing units in conditions of insufficient cleaning. Post processing contamination is the major source and cross contamination may also occur at the retail shop and also in products due to improper hygienic practices. Ingestion of uncooked meat contaminated during processing can produce infection. Consumption of poultry meat had been identified as a risk factor for *Campylobacteriosis* in almost all types of chicken meat either raw or undercooked (Friedman *et al.*, 2004). Rahimi *et al.*, (2010) found that higher prevalence of *C. jejuni* in their study was due to cross contamination during manual skinning, evisceration and processing in the slaughterhouse or in the butcher shops. *L. monocytogenes* was found prevalently high in poultry meat (24.5%), intermediate in beef meat (24.4%) and less prevalent in pork meat (21.4%). Osaili *et al.*, (2011) conducted a study to detect the prevalence of *Listeria spp.* in raw chicken and ready-to-eat (RTE) chicken products in Amman, Jordan and they found that *L. monocytogenes* was present in 9.4% of fresh dressed broiler chickens.

Hence a study is planned to conduct a screening of *C.jejuni* and *L.monocytogenes* on different zones of Chennai with multiplex PCR. Multiplex PCR is a type of PCR which enables simultaneous amplification of many target sites (template DNA) in one reaction by using more than one pair of primers for different organisms. It helps in minimizing the use of chemicals and also time needed for detecting single organism by normal PCR.

MATERIAL METHODS

A total of 90 chicken meat samples were collected from different retail outlets located in fifteen corporation zones of Chennai city. The samples placed in sterile polythene bags and transported hygienically to the Department of Meat Science and Technol-

ogy, 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 Phenol-Chloroform-Isoamylalcohol mixture (25:24:1) and PCR analysis for the presence of *C. jejuni* and *L.monocytogenes* by targeting *Hyp* and *prfA* gene with 500bp and 290bp respectively. The PCR amplification was carried out in Master Cycler Gradient Thermo cycler (M/s. Eppendorf, Germany). 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.

RESULT AND CONCLUSION

None of the sample showed positive for the presence of *Campylobacter jejuni* and *Listeria monocytogenes* in the retail chicken meat by multiplex PCR (Plate 1, 2, 3). Screening of chicken meat from different zone wise details were presented in Table 1. It indicates that the process is carried out in a hygienic manner or there may be some other reason for the absence of these microorganism in chicken meat.

Table 1

Screening of chicken meat samples collected from different retail outlets located in 15 corporation zones of Chennai city

Zone	Name	No. of samples	No. of positive samples by m-PCR	
			Campylobacter jejuni	Listeria monocytogenes
1	Thiruvottiyur	6	-	-
2	Manali	6	-	-
3	Madhavaram	6	-	-
4	Tondiarpet	6	-	-
5	Royapuram	6	-	-
6	Thiru-vi-ka nagar	6	-	-
7	Ambattur	6	-	-
8	Anna nagar	6	-	-
9	Teynapet	6	-	-
10	Kodambakkam	6	-	-
11	Valasaravakkam	6	-	-
12	Alandur	6	-	-
13	Adyar	6	-	-
14	Perungudi	6	-	-
15	Shozhinganallur	6	-	-
Total		90	0	0

Plate 1

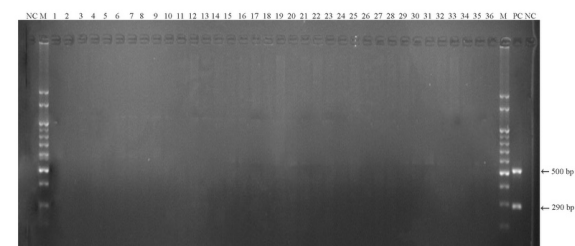


Plate 2

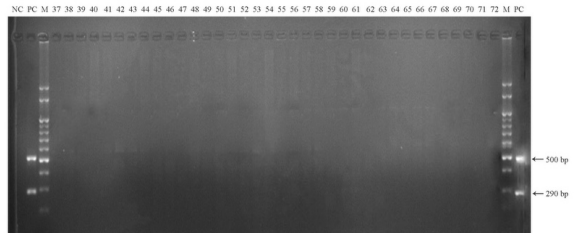


Plate 3

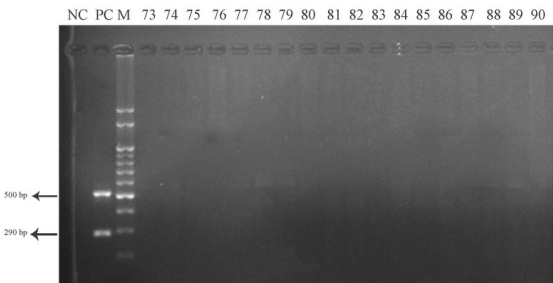


Plate 1,2,3 : Multiplex PCR (m-PCR) amplification for chicken meat samples collected from retail outlets

M : 100 bp DNA Ladder

1-36 : No amplification for *Campylobacter jejuni* and *Listeria monocytogenes*

37-72 : No amplification for *Campylobacter jejuni* and *Listeria monocytogenes*

73-90 : No amplification for *Campylobacter jejuni* and *Listeria monocytogenes*

PC : Positive control

NC : Negative control

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