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Public Health Significance of *Listeria* spp. Isolated from Vegetables Sold in Retail Markets of Thrissur, Kerala

B. Sunil[1], C. Latha[2], R. Remya[3], K. Vrinda Menon[4] and V.J. Ajaykumar[5]

Department of Veterinary Public Health, College of Veterinary and Animal Sciences, Mannuthy, Thrissur - 680 651, India.

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A total of 120 vegetable samples were collected from retail markets in Thrissur districts for over a period of two months. These samples were examined for the presence of *Listeria* spp. to provide information on the occurrence of organisms in such vegetables. Following a two step enrichment procedure and plating on selective agar, confirmation of the isolates was based on the biochemical tests. *Listeria* spp. could be detected in 8.3 % samples. Of the 10 isolates obtained 8 were *L.innocua* and the rest were *L.welshimeri.* These organisms were also isolated from human infections earlier. It was also reported that *L.innocua* and *L.monocytogenes* share the same ecological niche and on selective enrichment, *L.innocua* inhibit the growth of *L.monocytogenes* producing false negative results. So screening of more samples is necessary because vegetables are considered as part of healthy diet worldwide.

Key words: *Listeria* spp., Vegetables, UVM, PALCAM, GLISA.

Listeriosis is a fatal foodborne zoonotic infection caused by the intracellular bacterial pathogen, *Listeria monocytogenes*. The fatality rate of the infection (30 %) is very high compared with other bacterial foodborne infections. The genus Listeria includes other five closely related species *viz*. *L. ivanovii, L. seeligeri, L. innocua, L. welshimeri and L. grayi.* Among this *L.ivanovii, L. welshimeri* and *L.innocua* have been isolated from human infections. The disease is of worldwide distribution and there are also reports from India. The organism has been isolated from various products such as meat, milk, vegetables and fish [1]. However, there are no reports of isolation of the organism from Kerala. Occurrence of these organisms in fresh vegetables is of particular interest since raw vegetables are consumed as ready to eat foods like salads. The purpose of the present study was to generate information on the incidence of *Listeria* spp. in the vegetables, especially those consumed with minimal or no cooking, from the retail markets in Thrissur, Kerala.

* To whom all correspondence should be addressed.

MATERIALS AND METHODS

Fresh samples of cabbage, coriander leaves, cucumber, carrot, mint, cauliflower, amaranthus and celery were collected for screening. Fifteen numbers of each sample were collected over a period of two months from retail markets in and around Thrissur. The samples were collected in sterile polythene sachets and were processed soon after collection. Screening of the samples for the presence of *Listeria* spp. was done following the procedure of USDA [2] with necessary modifications. The vegetables were washed in peptone water and 10 ml of the washing was

transferred to 90 ml of University of Vermount Medium I (UVM I, Himedia, India) and incubated at 30 ° C for 24 hr. 0.1 ml of UVM I was transferred to UVM II and incubated at 30°C for 24 hr. Then it was streaked to Polymixin acriflavin lithium chloride ceftazidime asculin mannitol (PALCAM, Himedia, India) agar plates and incubated at 37 ° C for 48 hrs.

Identification of *Listeria* spp.

The colonies suspected for *Listeria* spp. were grayish green glistening colonies surrounded by a black zone of aesculin hydrolysis in medium. After subculturing the suspected colonies on Brain heart infusion (BHI) agar, colonies were subjected to further confirmation tests like gram's staining, catalase reaction, tumbling motility at 20-25 ° C, Methyl red – Voges Proskauer (MR-VP) reactions, nitrate reduction, sugar fermentation (dextrose, xylose, mannitol, rhamnose) and hemolysis on horse blood agar.

All the samples were also screened for the presence of *Listeria monocytogenes* using GLISA (Gold labeled immunosorbent assay) rapid test kit (Merck, Germany).

RESULTS AND DISCUSSION

Of all the 120 vegetable samples screened, 10 (8.3 per cent) samples were positive for the presence of *Listeria spp*. No isolates were obtained from amaranthus, cauliflower and celery. The details of isolates obtained from different categories of vegetables are given in the Table 1.

Table 1. Incidence of *Listeria* contamination on vegetables

Vegetable Samples (15 each)	Different Species found (with the number of each isolates)	Total Number
Cabbage	*L. innocua* (3)	
	L. welshimeri (1)	4
Coriander leaves	*L. innocua* (1)	1
Mint	*L. innocua* (1)	1
Carrot	*L. innocua* (1)	1
Amaranthus	Nil	0
Cucumber	*L. innocua* (2)	
	L. welshimeri (1)	3
Cauliflower	Nil	0
Celery	Nil	0

Among all the vegetables screened, the cucumber had maximum (33.3 per cent) number of isolates followed by cabbage (26.6 percent). The screening of coriander leaves, mint and carrot yielded same number of isolates (6.66 per cent). The source of *Listeria* spp. in these vegetables may be soil as the organism is found in soil and surface water. The vegetables which recorded the highest occurrence have close proximity to the soil or it may be due to post harvest cross contamination. The presence of these organisms indicates the chances of *Listeria monocytogenes* being present in the area as the organisms share the same ecological niche [3]. *Listeria innocua* is physiologically close to *Listeria monocytogenes* and could mask the later following enrichment procedure leading to many false negative results[4]. The results probably indicate the contamination of the soil at the area of cultivation. Among the 10 isolates, 8 (80 per cent) was *L. innocua* and remaining (20 per cent) was *L.welshimeri*. Although not regarded as highly pathogenic these two organisms have been isolated from human infections [5,6]. The results of the study clearly points to the need of pre harvest food safety with respect to soil quality. As the vegetables screened in this study included the ones which are consumed with minimal or no cooking, the results are significant. Moreover previous studies have shown that invasion into the roots by human pathogens could lead to systemic spreading and contamination of seeds and fruits of plants [7]. Further studies are also needed to establish a protocol for production, marketing and cleaning of such vegetables before consumption. Screening of more number of vegetable samples which are meant for direct consumption is very important as the vegetables are promoted as healthy foods worldwide.

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