A STUDY ON PRESENCE OF *LISTERIA* SPP. IN PORK.

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Abstract- The record of listeriosis in pig is very limited in India particularly in different pork consuming areas. Therefore, the study is directed to develop a maiden attempt towards developing a record of such disease prevalence. It is a zoonotic one and needs further work also. The percentage of occurrence though seems to be in and around 12% but this cannot be overlooked. The pathogenicity and identification of the organism from such cosmopolitan thickly populated area of the world got immense importance for investigation and further emphasise.

Key words: Listeriosis, zoonotic disease, pork

I- INTRODUCTION

Listeria monocytogenes is one of the most important species in the genus *Listeria* creating human health hazards and having a worldwide distribution with a wide range of hostswhich includes mammals, poultry, fish, crustacean and ticks. It affects primarily pregnant patients, neonates, elderly and immunocompromised individuals. Their significant roles as food borne human pathogen became evident only in 1980s, when documented reports of listeriosis out-break were traced to contaminated food (Schlech*et al.* 1983). The member of the genus *Listeria* is mesophilic, facultatively anaerobic, non-sporeforming, non acid-fast, gram-positive, non-branching, regular, short (0.4 to 0.5μ m in diameter by 0.5 to 2μm in length) rods with rounded ends. In pig, the primary manifestation is septicaemia, with encephalitis reported less frequently and abortion rarely (Blend, 1986). Numerous animal species are susceptible to listerialinfection, with a large population of healthy asymptomatic animals shedding *L. monocytogenes* in their faeces and may cause human health hazard (Low and Donachie, 1997). In India Giridhar and Garg (2002) reported presence of *Listeria* spp. in different samples like faces, sewage, fodder, feed, soil, water and milk of sheep and cattle farms. Presence of *Listeria spp.* as well as *L. monocytogenes*in beef in and around Kolkata city was reported by Biswas (2010).In India little work has been done as reflected in the scanty literature. Therefore, it is felt that a study on presence of listeria species in pork could be explored as the study on this has not been documented and that is ample chance of occurrence of listeriosis in India as pork get entered through different international borders and to the cosmopolitan cities like Kolkata.

The present study was undertaken with the following objectives:

- To study the prevalence of *Listeria* spp. Particularly *L. monocytogenes* in pork.
- To study the pathogenesis of *L. monocytogenes* isolates in animal models.

II - MATERIALS AND METHODS:

A total of 80 pork and 80 pork swab samples were collected from dressed carcasses of pig from different meat retailers of Kolkata city. Forty pork and 40 pork swab samples were also collected from organized pig slaughterhouses of Kolkata city. About 25 gm of pork was collected for each sample and four sq. cm. areas were used for taking the swabs.

PALCAM *Listeria* Selective Enrichment Broth and Agar [*Listeria* Identification Broth (PALCAM)] were from Hi Media. Methyl Red test, Voges-Proskauer Test, Catalase test, Oxidase test,

Sugar Fermentation test, Haemolysis test and CAMP test were performed as per OIE 2000. Different species of *Listeria* were identified by biochemical tests (Recourt and Remount, 1983).

Adult healthy Swiss albino mice were used for the study of pathogenicity. The mice were inoculated intraperitonealy with 0.5 ml of tryptose broth with10^9cfu/ml of *L. monocytogenes*. A batch of ten mice was kept as control and was observed for ten days. During the experimentation, the dead mice were placed for Histopathological examination where study of heart, liver, spleen and brain was done using Haematoxylin and Eosin staining method (Moshtaghi*et al.* 2002).

Scanning Electron Microscopy was performed following the method described by Dewar (1982) with some modifications.

III- RESULTS AND DISCUSSION:

Among 80 pork and 80 pork swab samples from retail market, were examined and one pork and one pork swab sample had the presence of *L. monocytogenes*. Among other pork samples, one each was positive for *L. innocua* and *L. seeligeri*. Among other pork swab samples, one each was found positive for *L. innocua* and*L.welshimeri*. Besides, another two samples showed the growth of *L. seeligeri* **(Table –I).**

Among40 pork and 40 pork-swab samplesfrom organized slaughter house, were examined and one pork and three pork swab samples were positive for *L. monocytogenes*. Among other pork samples, three were positive for *L. Ivanovii* and one each was positive for*L. innocua,L. seeligeri* and *L. grayi*. Among other pork swab samples, three showed presence of *L. ivanovii*, two each were positive for *L. innocua*, for *L. seeligeri* and four samples were positive for *L. grayii* also (**Table –I).**

Table –I: Distribution of *Listeria* spp. in pork meat and meat swab.

Source	Type	Nos	*L. monocytogenes*	*L. ivanovii*	*L. innocua*	*L. welshimeri*	*L. seeligeri*	L. grayii	*Total*
Organized Slaughter House	Meat	40	1	3	1	0	1	1	7
	Swab	40	3	3	2	0	2	4	14
Retail Market	Meat	80	1	0	1	0	1	0	3
	Swab	80	1	0	1	1	2	0	5
Total		240	6	6	5	1	6	5	29

Among 240 pork and pork swab samples tested, 12% samples, were *Listeria* spp. positive. Among these positive samples, 20% was found positive for *L. Monocytogenes* only.

Pathogenicity test in Swiss albino mice model:

The post mortem materials i.e. heart, liver, spleen and brain were examined for histopathological changes and observation in view of listeriosis.

Heart: Diffuse haemorrhages in between muscle fibers along with congestion and cellular infiltration were found. The visa vesorum was severely congested and edematous fluids separated some portion of muscles. Infiltrating cells were strictly mononuclear and neutrophils. (Fig:1)

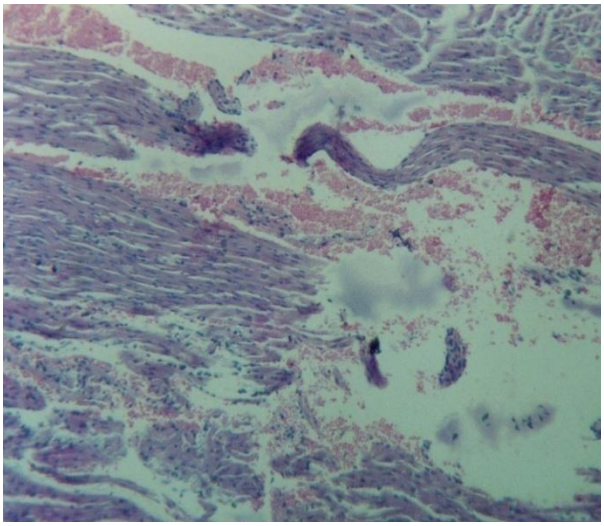

Fig.-1: Muscle showing extensive haemorrhage, disorganization and presence of oedematous fluid of mice heart.

Liver: Massive haemorrhage in the liver parenchyma and degenerative changes were prominent. The hepatic blood vessels showed congestions. The bile canaliculi were dilated and infiltrated with blood cells and inflammatory cells. (Fig:2)

Spleen: There were proliferations of spleenic pulp, especially with lymphocytes. Lymphocytic proliferation, and spleenic capsules showed focal haemorrhage. (Fig:3)

Brain: Micro abscesses; degenerative changes along with accumulation of mononuclear cells were found in the brain tissue. Perivascular cuffing was also noted. Meningeal blood vessels showed severe congestion and infiltration with mononuclear cells and maningolethiolal cells were also found. (Fig:4)

All these findings were corroborated to the observations of Miller and Burn (1970),Nigam *et al.* (1998),Moshtaghi (2002)and Biswas (2010).

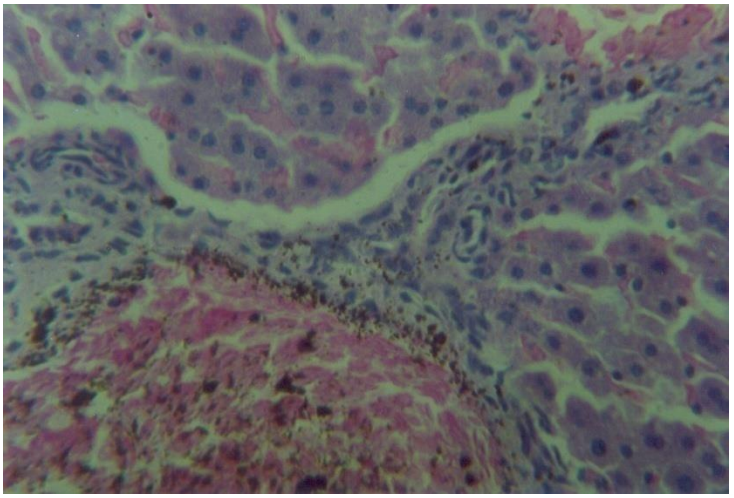

Fig.-2: Haemorrhage, congestion, necrosis and dilation of bile canaliculi are present in mice liver.

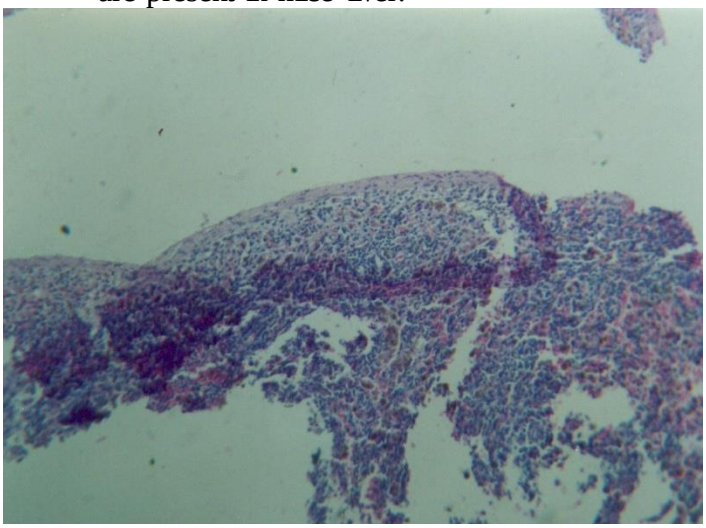

Fig.-3: Haemorrhage, lymphatic proliferation and thickening of capsule are present in mice spleen.

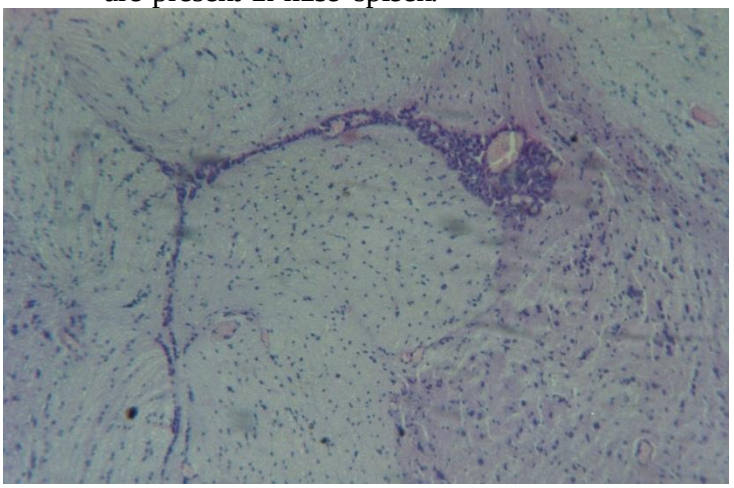

Fig.-4: Perivascular cuffing, congestion with moderate degenerative changes of mice brain.

Study of *L. monocytogenes* through scanning Electron Micros copy:

The morphology of *L.monocytogenes* was studied through electron microscopy. The average length of the bacteria was measured 4 μm and diameter was 0.4 μm. The bacterial cells were in 'V' and 'Y' arrangement. Cells were found in the process of division. And the cell surface was smooth. The findings were identical to the findings of earlier worker Ritz *et al.* (2001). (Fig:5).

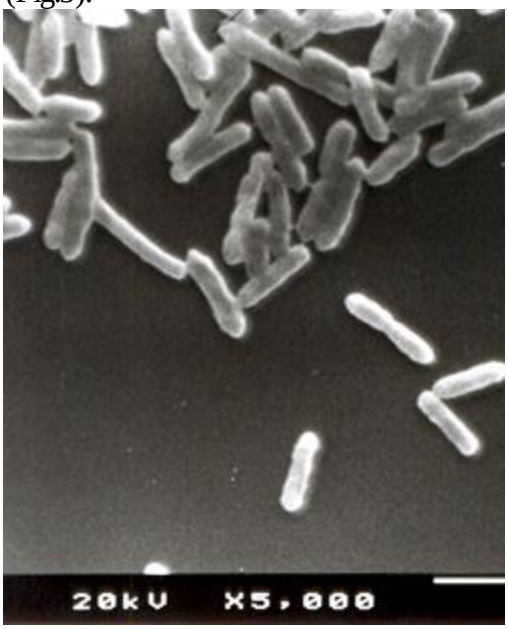

Fig.-5: *L. monocytogenes* in 'V' arrangement and cell is in process of division.

III. CONCLUSION:

It can be concluded that the presence of *Listeria* Spp. particularly *L. monocytogenes* in pork is alarming in the city of Kolkata. Occurrences of Listeriosis in animal and human are to be studied through screening the samples from animal and human patient showing specific symptoms.

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