RESEARCH ARTICLE

Prevalence of Listeria Species in Dairy Cows and Pregnant Women with Reference to Virulotyping of Listeria monocytogenes in Egypt

EL-Sayed Y. M. EL-Naenaeey,1 Ashraf M.O. Abdel-Wahab*, Abdallah M. A. Merwad and Hadeer M. A. Abdou

1Department of Microbiology, Faculty of Veterinary Medicine, Zagazig University, 44511, Egypt
2Department of Zoonoses, Faculty of Veterinary Medicine, Zagazig University, Egypt

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Abstract

This study was conducted to determine the prevalence of Listeria spp. in milk and feces of dairy cows as well as stool of pregnant women in Sharkia and Dakahlia Governorates, Egypt. Three hundred and fifty samples including normal raw milk (n=200), mastitis milk (n=50), feces of dairy cattle (n=50) and stool of pregnant women (n=50) were randomly collected. Listeria spp. were isolated on Oxford agar and then subjected to biochemical and molecular identification. The overall isolation rate of Listeria spp. was 16%. The prevalence rates of L. monocytogenes were 8%, 4%, 2% and 4% in feces of dairy cows, normal milk, mastitis milk and stool of pregnant women, respectively. The prevalence of L. ivanovii and L. welshimeri in milk of dairy cows were 6% and 4%, respectively. L. monocytogenes isolates were molecularly confirmed to the species level by amplification of 16S rRNA gene. The distribution of internalin A (inlA) and internalin B (inlB) was determined in 15 identified L. monocytogenes isolates. The overall prevalence of inlA gene and inlB gene in L. monocytogenes isolates from different sources were 80% and 40%, respectively. Two L. monocytogenes isolates from normal raw milk and feces of dairy cows didn’t harbor both inlA and inlB genes. Listeria spp. isolated from normal milk, feces, mastitis milk and stool of pregnant women, potentially can cause human illness and abortion in pregnant women. Internalin (A&B) genes are considered the best indicator for virulence determination of L. monocytogenes isolated from different sources.

Keywords: Listeria monocytogenes, 16S rRNA gene, Internalin A&B, Milk, Pregnant women.

Introduction

Listeria species are ubiquitous bacteria, widely spread in the nature such as soil, mud, plant, decaying vegetation, raw and treated sewage, fecal material, water, food processing facilities [1]. The genus Listeria comprises of 6 species including: Listeria monocytogenes, L. ivanovii, L. innocua, L. welshimeri, L. seeligeri, and L. grayi. These organisms are short Gram-positive bacilli, facultative anaerobe, non-capsulated and non-spore former. The main two pathogenic Listeria spp. for humans and animals are L. monocytogenes and L. ivanovii in spite of occasional human cases reported by L. ivanovii infection [2]. The ubiquitous nature of L. monocytogenes enables its entrance and proliferation into human food chain. Listeria may be a common contaminant in the dairy farms [3].

Raw milk and animals’ manure have been considered as vital sources for Listeria in the dairy environment. The oral-fecal cycle resulted in persistence of such pathogen in the farm [4].

Diseases attributed to food borne pathogens represent a worldwide hazard to public health. Listeriosis is one of most important zoonotic disease with fatality rates up to 20-30% [5]. Listeriosis is a foodborne disease transmitted through the consumption of raw or insufficiently cooked food. L. monocytogenes is the main etiological agent frequently incriminated in such illness. Food recognized as common vehicle for Listeria infection

*Corresponding author email: (ashrafomar_2004@yahoo.com), Department of Microbiology, Faculty of Veterinary Medicine, Zagazig University, 44511, Egypt
include milk, dairy products, raw vegetables, poultry, seafood, meat and meat products [1]. The individuals at high risk for listeriosis are pregnant women, neonates, older age and individuals with poor immunity. Wide ranges of infections were reported in human including: gastroenteritis, fever, diarrhea, muscle aches, nausea, vomiting and fatigue; however, severe forms may result in encephalitis, meningitis and abortion in pregnant women [6].

Sporadic cases and epidemic outbreaks of listeriosis in humans and large number of animal species were recorded. Latent listeriosis in pregnant women resulted in spontaneous abortions, placental necrosis, intrauterine deaths and fetal malformations. The prevalence rate of listeriosis in general population is 0.7 in 100000 while the incidence rate is 12 in 100000 in pregnant women (that is a 17-fold increase) [6-9].

Mastitis is a multi-etiological disease that induces major economic losses in the dairy industry all over the world. *Listeria monocytogenes* could infect the udder from localization after systemic infection or from outside penetration of the teat [10]. Moreover, *L. monocytogenes* might infect one or all of quarters and the bacteria are excreted in milk causing a potential risk to the public health. Listeric mastitis is difficult to treat and many cases of Listeric mastitis go unobserved or unnoticed [11].

PCR assays are one of the preferable ways for detection and confirmation of such pathogen. Lastly, researchers have been attracted toward molecular techniques relying on 16S rRNA sequences as a reliable method for identification and differentiation of wide ranges of bacteria at the genus level across almost all phyla and also at species and sub species levels. It is large enough (1,500 bp) for genetic information [12,13].

Internalins have an essential role in pathogenicity of *L. monocytogenes* and its success as facultative intracellular pathogen. Internalin genes (*inLA & inLB*) are a panel of *Listeria* virulence-specific genes that encoded by proteins in inner bacterial surface which involved in *L. monocytogenes* passage through epithelial cells of the host as well as through the intestinal barriers for the successive stages of infection [14]. Internalin A binds to the host cell receptor E-cadherin which is required for the entry into human epithelial cells such as enterocytic Caco-2 cells and crossing the intestinal barrier to host blood stream [15]. Besides, internalin B is an invasion surface protein on *L. monocytogenes* which could play a great role in the entrance into multiple host cells as hepatocytes, endothelial and epithelial cells [16]. Differential expression levels of *inLA* and *inLB* may have an important role in the virulence capacities of *L. monocytogenes* strains [17].

This study was aimed to investigate the prevalence of *Listeria* species in milk, feces of dairy cows as well as stool of pregnant women in two Governorates (Sharkia and Dakahlia) in Egypt, and also to determine the prevalence of some virulence genes (*inLA* and *inLB*) in molecularly identified isolates of *L. monocytogenes*.

**Materials and Methods**

**Sample collection**

A total of 350 samples including; normal milk (n=200), mastitis milk (n=50), feces of dairy cows (n=50) and stool of pregnant women (n=50) were randomly collected from different dairy farms and hospitals in Sharkia and Dakahlia Governorates, Egypt during the period from January 2018 to January 2019. The collected samples were transferred in an ice box immediately to the Microbiology and Zoonoses Departments laboratories, Faculty of Veterinary Medicine, Zagazig University, Egypt until isolation and identification.

Milk samples were randomly screened for mastitis by using California Mastitis Test (CMT). Fifty milk samples were mastitis positive and involved in this study [18].

**Isolation and identification of Listeria spp.**

Twenty-five milliliter of milk samples either normal raw milk or mastitis milk was pre-enriched in 225 mL of Tryptone soya broth (TSB) (Oxoid, CMO129, UK). Twenty-five grams either from feces of dairy cows or stool of pregnant women were also pre-enriched in 225 mL of Tryptone soya broth (TSB) (Oxoid, CMO129, UK). All previous samples were incubated at 37°C for 18-24h. Ten milliliter of an initial suspension was placed into a sterile tube contained 90 ml of Listeria enrichment broth base (LEB) (Oxoid, CM0862,UK) with Listeria enrichment selective supplement (Oxoid, SR0141,UK) and incubated for 2-7days at 30°C for enrichment...
of Listeria spp. Finally, a loopful of incubated enrichment broth was streaked onto petri dish containing Listeria selective agar (Oxford formulation) (Oxoid, CM0856, UK) with Listeria selective supplement (Oxoid, SR0140, UK) and placed in an incubator at 37°C for 24-48 h. Presumptive gray colonies surrounded by black zone were purified by subculture on Tryptic soy agar yeast extract (TSAye) and incubated at 37°C for 18-24 h [19].

**Identification of Listeria spp.**

For identification of isolates to species level, typical colonies were subjected to the following tests (Gram staining, catalase, hemolysis on blood agar, oxidase, motility, fermentation of Rhamnose, Xylose and Mannitol sugars) [20, 21].

**Molecular characterization of isolated L. monocytogenes**

Genomic DNA from biochemically identified *L. monocytogenes* strains was extracted by using QIAamp DNA Mini Kit Catalogue no.51304 (QIAGEN GmbH, Hilden, Germany) according to the manufacturer's guidelines. Extracted DNA was subjected to PCR using primers targeting 1200 bp of the 16S rRNA specific for *L. monocytogenes*. The oligonucleotide primers 16S rRNA was carried out as previously described [22] in Table 1.

**Table 1: Oligonucleotide primers used for amplification of 16S rRNA gene, inlA and inlB genes in isolates of *L. monocytogenes* from dairy cows and pregnant women.**

<table>
<thead>
<tr>
<th>Gene</th>
<th>Primer sequence (5'→3')</th>
<th>Expected product size (bp)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>16S rRNA</td>
<td>16S rRNA-Forward GGA CCG GGG CTA ATA CCG AAT GAT AA</td>
<td>1200</td>
<td>22</td>
</tr>
<tr>
<td></td>
<td>16S rRNA-Reverse TTC ATG TAG GCG AGT TGC AGC CTA</td>
<td>22</td>
<td></td>
</tr>
<tr>
<td>inlA</td>
<td>inlA-Forward ACG AGT AAC GGG ACA AAT GC</td>
<td>800</td>
<td>23</td>
</tr>
<tr>
<td></td>
<td>inlA-Reverse CCC GAC AGT GGT GCT AGA TT</td>
<td>343</td>
<td></td>
</tr>
<tr>
<td>inlB</td>
<td>inlB-Forward CTGGAAAGTTTGTATTTGGGAAA</td>
<td>1200</td>
<td>24</td>
</tr>
<tr>
<td></td>
<td>inlB-Reverse TTTCATAATGCCTCATCACT</td>
<td>22</td>
<td></td>
</tr>
</tbody>
</table>

1The 16S rRNA primers were designed for species-specific recognition and the inlA and inlB primers for virulence determination of *L. monocytogenes*.

All PCR reactions were carried out in a final total volume of 25μl (12.5 μl of Emerald Amp GT PCR master mix (2x premix) (Takara, Code No. RR310Akit, Japan), 1 μl of 20 pmol of forward primer, 1 μl of 20 pmol of reverse primer, 5.5 μl PCR grade water and 5μl Template DNA). The cycling conditions for 16S rRNA PCR involved a primary denaturation of DNA at 94°C for 5 min followed by 35 cycles of secondary denaturation at 94°C for 30 sec, annealing at 60°C for 1 min, extension at 72°C for 1 min, then final extension at 72°C for 12 min. PCR products stained with ethidium bromide (Sigma, US) were electrophoresed on 1.5 % agarose gel at 1.5volts/cm for 30 min in Tris borate EDTA (TBE) buffer. The amplified DNA products were seen using UV transilluminator. A positive control isolate of *L. monocytogenes* was kindly provided from Biotechnology unit, Reference laboratory for Veterinary Quality Control on Poultry Production, Animal Research Institute, Dokki, Giza, Egypt.

**Molecular detection of internalin genes in *L. monocytogenes* isolates**

Isolates identified as *L. monocytogenes* were further subjected to screening for the presence of some virulence genes (inlA & inlB). In Table 1, the primers sequences for amplification of internalin (A & B) genes were designed as previously described [23, 24], respectively. The amplification conditions were 35 cycles of primary denaturation for 5 min at 94°C, secondary denaturation for 30 sec at 94°C, annealing for 40 sec at 55°C, extension for 50 sec at 72°C and final extension at 72°C for 10 min.

**Statistical analysis**

The data was analyzed using SPSS version 22 for Windows (IBM corp, 2013, Armonk, NY) to compute PV values for Pearson chi-square.
Results

Out of 350 examined samples, 56 (16%) were contaminated with *Listeria* spp. by using culture techniques, and biochemical tests. Among the 56 isolated *Listeria* spp.; 38 (19%), 6 (12%), 8 (16%) and 4 (8%) were recovered from normal raw milk, mastitis milk, feces of dairy cows and stool of pregnant women, respectively. The *Listeria* spp. isolates from different sources included *L. monocytogenes* (n = 15, 4.28%), *L. ivanovii* (n = 18, 5%), *L. innocua* (n = 5, 1%), *L. grayi* (n = 9, 2.5%) and *L. welshimeri* (n = 9, 2.5%).

The prevalence rates of *Listeria monocytogenes* were 8%, 4%, 2% and 4% in feces of dairy cows, normal milk, mastitis milk and stool of pregnant women, respectively (Table 2). *L. ivanovii* was isolated from normal raw milk and mastitis milk (6%, each). The prevalence rates of *L. ivanovii* were 4% and 2% in feces of dairy cow samples and stool of pregnant women samples, respectively. *L. innocua* was only isolated from raw milk and mastitis milk samples (2%, each). *L. grayi* was isolated from 4%, 3% and 2% of feces of dairy cows, normal raw milk and mastitis milk samples, respectively. The prevalence rates of *L. welshimeri* were 4% and 2% in normal raw milk and stool of pregnant women, respectively. *L. welshimeri* was not isolated from mastitis milk and feces of dairy cow (Table 2).

<table>
<thead>
<tr>
<th>Source of Samples</th>
<th>No. of examined samples</th>
<th>No. of positive samples</th>
<th><em>L. monocytogenes</em></th>
<th><em>L. ivanovii</em></th>
<th><em>L. innocua</em></th>
<th><em>L. grayi</em></th>
<th><em>L. welshimeri</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Milk of dairy cows</td>
<td>200</td>
<td>38(19%)</td>
<td>8(4%)</td>
<td>12(6%)</td>
<td>4(2%)</td>
<td>6(3%)</td>
<td>8(4%)</td>
</tr>
<tr>
<td>Mastitis milk</td>
<td>50</td>
<td>6(12%)</td>
<td>1(2%)</td>
<td>3(6%)</td>
<td>1(2%)</td>
<td>1(2%)</td>
<td>0(0%)</td>
</tr>
<tr>
<td>Feces of dairy cows</td>
<td>50</td>
<td>8(16%)</td>
<td>4(8%)</td>
<td>2(4%)</td>
<td>0(0%)</td>
<td>2(4%)</td>
<td>0(0%)</td>
</tr>
<tr>
<td>Stool of pregnant women</td>
<td>50</td>
<td>4(8%)</td>
<td>2(4%)</td>
<td>1(2%)</td>
<td>0(0%)</td>
<td>0(0%)</td>
<td>1(2%)</td>
</tr>
<tr>
<td>Total</td>
<td>350</td>
<td>56(16%)</td>
<td>15(4.28%)</td>
<td>18(5%)</td>
<td>5(1%)</td>
<td>9(2.5%)</td>
<td>9(2.5%)</td>
</tr>
</tbody>
</table>

NS: Non specific

The statistical analysis revealed that there was no significant differences between the of *L. monocytogenes* isolates recovered from different sources (P>0.05) as shown in Table 2.

All the 15 biochemically characterized *L. monocytogenes* isolates were confirmed by PCR assay, using the species-specific 16S rRNA gene (100%). *L. monocytogenes* strains revealed an amplified product of 1200 bp as illustrated in Figure 1.

PCR amplification of *inlA* and *inlB* genes revealed their presence with correct size 800 bp and 343 bp, respectively (Figure 2). *InlA* gene was molecularly detected in 12 *L. monocytogenes* isolates (80%); (7/8) of normal raw milk, (1/1) of mastitis milk, (3/4) of feces of dairy cows and (1/2) of stool of pregnant women. However, *inlB* gene was observed in only 6 *L. monocytogenes* isolates (40%); (3/8) of normal raw milk, (1/1) of mastitis milk, (1/4) of feces of dairy cows and (1/2) of stool of pregnant women (Table 3).

<table>
<thead>
<tr>
<th>Source of samples</th>
<th>No. of positive samples</th>
<th>16S rRNA</th>
<th><em>inlA</em></th>
<th><em>inlB</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Milk of dairy cows</td>
<td>8</td>
<td>8/8</td>
<td>7/8</td>
<td>3/8</td>
</tr>
<tr>
<td>Mastitis milk</td>
<td>1</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
</tr>
<tr>
<td>Feces of dairy cows</td>
<td>4</td>
<td>4/4</td>
<td>3/4</td>
<td>1/4</td>
</tr>
<tr>
<td>Stool of pregnant women</td>
<td>2</td>
<td>2/2</td>
<td>1/2</td>
<td>1/2</td>
</tr>
<tr>
<td>Total</td>
<td>15</td>
<td>15/15(100%)</td>
<td>12/15(80%)</td>
<td>6/15(40%)</td>
</tr>
</tbody>
</table>

1Biochemically suspected isolates of *L. monocytogenes*.

2The *L. monocytogenes* isolates were molecularly confirmed by PCR targeting species specific 16S rRNA gene.

Table 3: Distribution of genus species specific 16S rRNA and internalin (*inlA, inlB*) genes in isolates of *L. monocytogenes* from normal milk, mastitis milk, feces of dairy cows and stool of pregnant women at Sharkia and Dakahlia Governorates, Egypt from January 2018 to January 2019.

Table 2: Prevalence of *Listeria* spp. in normal milk, mastitis milk, feces of dairy cows and stool of pregnant women at Sharkia and Dakahlia Governorates, Egypt from January 2018 to January 2019.
Listeria monocytogenes is currently considered as the third major pathogen frequently transmitted from food. The high prevalence of Listeria spp. in milk, feces and dairy products has a great hazard on public health and dairy industry [25]. Milk and dairy products are implicated in most outbreaks of listeriosis all over the world. The first report of listeriosis case in USA was resulted from consumption of pasteurized milk in 1983 [26].

In the current study, the higher isolation rate of Listeria spp. was detected in milk of dairy cows (19%) followed by feces of dairy cows (16%), mastitis milk (12%) and then stool of pregnant women (8%). Therefore, higher prevalence of Listeria spp. in milk samples in this study might be attributed to insufficient hygienic practice in dairy farms together with the poor hygiene of surrounding environments. Dairy farms are an important reservoir of Listerias pp., so, contamination of milk by such pathogens may occurs through direct contact with contaminated sources in the dairy farm environment as the milking machines, the piping system, milking utensils, teat cups and milk handlers which are...
considered as critical points of milk contamination at the farm. So, cleanliness of dairy farms' environment seems to be an important risk factor associated with milk contamination by *L. monocytogenes* as previously supported by Hassan et al. [27], Oliver et al. [28] and Vilar et al. [29].

Regarding the overall prevalence of *Listeria* spp. in milk samples (19%), similar result (16.7%) was reported in Egypt by Abdel-Aal [30]. However, lower percentages of 13.3% and 6% were respectively recorded in milk samples of dairy cows in Giza by Reda et al. [31] and in Dakahlia Governorate by Abd El Aal and Atta [32]. These differences in the prevalence of *Listeria* spp. could be attributed to sample size and locality [33,34]. The prevalence of *L. ivanovii* and *L. welshimeri* from milk of dairy cows were 6% and 4%, respectively in this study. In Turkey, nearly similar finding for *L. ivanovii* (5.4%) and *L. welshimeri* (4.5%) isolates from milk and milk products were detected by Sanlibaba et al. [35]. On the other hand, lower prevalence of *L. ivanovii* (1.4%) and *L. welshimeri* (0.7%) in previous study were detected from cow milk samples by El-Sherbini et al. [36] in Sharkia Governorate.

The prevalence rate of *L. monocytogenes* in milk of dairy cows was 4% in this study. This finding was concurred with previous result detected in India [37]. Also, nearly similar report (5.4%) for isolates of *L. monocytogenes* from milk was recorded in Iran [38]. On the contrary, lower prevalence of *L. monocytogenes* was detected in cow milk samples (2%) by Khedr et al. [39] in Egypt, and (1.1%) by Almeida et al. [40] in Portugal. However, higher prevalence of *L. monocytogenes* (8.3%) from cow milk samples was cited by El-Marnissi et al. [41] in Morocco, and (6%) by Abd El-Tawab et al. [42] in Egypt.

Milk is a proper medium for growth of *Listeria* spp. that account for the occurrence of food poisoning. Differences in the prevalence rate of *L. monocytogenes* in previous studies might be due to several factors affecting the level of *Listeria* contamination in raw milk as health status of dairy cattle, methods of detection, environmental conditions of dairy farms and hygienic managements of dairy environments and dairy farmers [39,43].

*L. monocytogenes* was detected in 2% of mastitis milk samples of dairy cows. This result was in agreement with the finding of Yadav et al. [11] who isolated *L. monocytogenes* with a percentage of 3.5 in Gujarat state, India. While higher percentage (6%) was recorded by Sunitha et al. [44] in Kerala, India. This may suggest that *L. monocytogenes*, *L. ivanovii* and *L. welshimeri* are considered as predisposing causes of mastitis in dairy cows. Little literatures are available about the prevalence of *Listeria* spp. in mastitis milk. This was the first report for isolation of *L. monocytogenes*, *L. ivanovii* and *L. welshimeri* from mastitis milk in Egypt.

The overall prevalence of *Listeria* spp. was 16% in feces of dairy cows. Nearly similar result of 15.6% was reported in Egypt [45]. In contrary, lower percentages of 3.3% and 9.6% were reported in Egypt [46] and Finland [47], respectively. While, higher prevalence rate of *Listeria* spp. (68%) was previously reported [48]. However, Ahmed [49] didn't recover any *Listeria* spp. from feces of dairy cattle in Egypt.

The prevalence rate of *L. monocytogenes* in feces of dairy cows was 8% in the current study. Nearly similar percentage of 7.5% for *L. monocytogenes* in feces of dairy cows was previously reported in New York State [50]. However, lower isolation rates of 6% in Sweden [51] and 1.5% in Turkey [52] were documented.

This finding could be explained on the fact that milk could be contaminated by feces of infected farm animals or asymptomatic carriers. Also, feces is considered as an important source of raw milk contamination by *L. monocytogenes* or using these fecal samples as fertilizers which facilitate contamination of vegetables, plants and crops with *L. monocytogenes* and creating a hazard on dairy cows as well as milk consumers [4].

In this study, *Listeria* spp. was isolated from stool of pregnant women with prevalence of 8%. *L. monocytogenes* was isolated from stool of pregnant women with a percentage of 4% (2 out of 50).
Lower percentage (2%) was detected in stool of pregnant women in Scotland [53]. In Egypt, Abuhatab [45] recovered *Listeria* spp. in 11.1% from feces of aborted women.

Other studies revealed that isolation rate of *Listeria* spp. was 7.14 % from stool of children [54] in Assiut city. *L. monocytogenes* was recovered from stool samples of dairy handlers with a percentage of 5 in Mansoura city [55]. In India, fecal samples, vaginal swabs, placental bits, blood and urine samples of aborted women showed isolation rate (3.3%) of *L. monocytogenes* by Kaur et al. [8].

The higher prevalence of *L. monocytogenes* in this study could be attributed to decline in cell-mediated immunity in pregnant women which is the most predisposing factor for *L. monocytogenes* infection as previously advocated by Pouillot et al. [56].

In this study, 15 isolates of *L. monocytogenes* from different sources were molecularly identified to the species level using PCR amplification of 16S rRNA gene. Similarly *L. monocytogenes* from different raw milk samples based on 16S rRNA gene was recorded in Egypt [39].

In this study, *inlA* and *inlB* were reported in 80% and 40% of isolated *L. monocytogenes*, respectively. In previous studies, Khedr et al. [39] detected *inlA* and *inlB* genes in 100% and 75% of examined *L. monocytogenes* isolates, respectively. However, Coroneo et al. [57] recorded *inlA* and *inlB* genes in 60% and 47% of *L. monocytogenes* strains recovered from Ricotta salata cheese, respectively.

The differences in the distribution of virulence internalin genes could be attributed to the source of samples or *L. monocytogenes* serotype or mutations in these genes resulted in reduction in invasion or virulence in animal model studies or tissue culture, assuring the significant role of such genes in the pathogenesis and virulence of *L. monocytogenes* [57-59]. It was noticed that 12 isolates of *L. monocytogenes* carry *inlA* gene and 6 isolates carry *inlB* gene that indicates virulent strains of *L. monocytogenes* and creates hazard in dairy farms as well as milk handlers, milk consumers and pregnant women.

**Conclusion**

Prevalence of *L. monocytogenes* in raw milk could constitute a public health hazard for milk handlers, workers in dairy farms and milk consumers. The molecular detection of *inlA* and *inlB* in isolates of *L. monocytogenes* are considered as virulent determinants for such zoonotic pathogen. Therefore, to control the risk of milk and dairy products contamination with *L. monocytogenes*, it is important to emphasize on the hygiene measures in dairy farms in addition to screening of *Listeria* spp.in dairy farms, milk of dairy cows, milk handlers.

**Conflict of interest**

The authors declare no conflict of interest.

**Acknowledgments:**

Staff members of Biotechnology Unit, Reference laboratory for Veterinary Quality Control on Poultry Production, Animal Research Institute, Doki, Giza, Egypt.

**References:**


(FoodNet) sites according to age, pregnancy, and ethnicity. Clin Infect Dis, 54: S405-S410.


**The Arabic Abstract**

انتشار انواع الليستريا في الأبقار الحلوب والنساء الحوامل مع الإشارة إلى تصنيف الأنواع ذات الضايحة من الليستريا

مونوسيتوجينز في مصر

السيد نعاعي، أشرف محمود عمر عبادولهاب، عبادان محمد، هدير عهد محمد

قسم الميكروبيولوجيا - كلية الطب البيطري جامعة الزقاقيق

*melts the study to identify on the prevalence of strains of Listeria monocytogenes isolated from buffalo and pregnant cows and to classify strains that cause disease.*

A study was conducted to determine the prevalence of strains of Listeria monocytogenes isolated from buffalo and pregnant cows and to classify strains that cause disease.

The prevalence of strains of Listeria monocytogenes isolated from buffalo and pregnant cows and to classify strains that cause disease.

**Internalin (A & B)**

Based on the above results, the study found that 6% of the isolates were Listeria monocytogenes, and 16SrRNA analysis of the isolates showed that 92% of the isolates were Listeria monocytogenes. Moreover, the analysis of the isolates showed that 80% of the isolates were Listeria monocytogenes, and 20% were not. Therefore, it can be concluded that the prevalence of strains of Listeria monocytogenes isolated from buffalo and pregnant cows is high, and the study recommends further research to determine the factors that contribute to the high prevalence of strains of Listeria monocytogenes in buffalo and pregnant cows.

**References**

1. EL-Nauneey et al., (2019).

**Summary**

The study aimed to determine the prevalence of strains of Listeria monocytogenes isolated from buffalo and pregnant cows and to classify strains that cause disease. The study found that 6% of the isolates were Listeria monocytogenes, and 16SrRNA analysis of the isolates showed that 92% of the isolates were Listeria monocytogenes. Moreover, the analysis of the isolates showed that 80% of the isolates were Listeria monocytogenes, and 20% were not. Therefore, it can be concluded that the prevalence of strains of Listeria monocytogenes isolated from buffalo and pregnant cows is high, and the study recommends further research to determine the factors that contribute to the high prevalence of strains of Listeria monocytogenes in buffalo and pregnant cows.