Identification and characterization of *Yersinia enterocolitica* isolated from raw chicken meat based on molecular and biological techniques

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**SUMMARY**

A PCR-based assay was developed to detect the occurrence of *Yersinia enterocolitica* in Iranian chicken meat samples and to evaluate plasmid- and chromosome-borne virulence genes. This feasible and informative method was able to provide a rapid and reliable characterization of field isolates. A total of 720 chicken meat samples were collected randomly from abattoirs in western Iran and tested by culturing and PCR methods. Of these, 132 (18.33%) were found to be positive for *Y. enterocolitica* by both methods. Isolates included biotypes 1A (0%), 1B (0%), 2 (18.18%), 3 (52.27%), 4 (17.42%), and 5 (12.12%), and serotypes included O:3 (36.84%), O:5,27 (59.84%), O:8 (5.30%), and O:9 (0%). Of the 46 *Y. enterocolitica* serotype O:3 isolates, the prevalence of virulence genes included *yadA* (82.60%), *inv* (100%), *ail* (95.65%), *ystA* (93.47%), and *virF* (58.69%). This study highlighted the importance of chicken meat as potential sources of *Y. enterocolitica* infection in Iran.

**Key words:** biotyping, chicken meat, polymerase chain reaction, serotyping, virulence gene, *Yersinia enterocolitica*

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Foodborne diseases are a widespread and growing public health concern in developed and developing countries [1]. *Yersinia enterocolitica* is a gram-negative bacterium belonging to the genus *Yersinia*, family *Enterobacteriaceae*. Of the 12 species that comprise the genus, 3 are important in human pathogenicity, namely, *Yersinia pestis*, *Yersinia pseudotuberculosis*, and *Y. enterocolitica*. *Yersinia pestis* is the causative agent of the bubonic plague, whereas *Y. pseudotuberculosis* and *Y. enterocolitica* are intestinal pathogens. *Yersinia enterocolitica* are widely distributed throughout the environment and have been isolated from raw milk, sewage-contaminated water, soil, seafood, humans, and many warm-blooded animals, such as poultry and, most important, pigs. The serotypes O:3, O:5,27, O:8, and O:9 are the most frequent causative agents of human illness [2].

A high prevalence of gastrointestinal illness, including fatal cases attributable to yersiniosis, is also observed in many developing countries, including Bangladesh [3], Iraq [4], Iran [5], and Nigeria [6], indicating major underlying food
safety problems in low- and middle-income countries. Worldwide, *Y. enterocolitica* infection occurs most often in infants and young children, with common symptoms such as fever, abdominal pain, and diarrhea, which is often bloody. Older children and young adults are not risk free. Occasionally, *Y. enterocolitica*-associated complications, such as skin rash, joint pains, or septicemia, can also occur [7].

The identification and further subspecies typing aimed at recognition of pathogenic *Yersinia* spp. are traditionally based on phenotypic tests. *Yersinia enterocolitica* can be classified into bio-type 1A, generally regarded as nonpathogenic [8], and the pathogenic biotypes 1B, 2, 3, 4, and 5. *Yersinia pseudotuberculosis* and *Y. enterocolitica* can also be divided into serotypes with predictive values for pathogenicity. Serological and biochemical classifications, however, are time consuming and not generally available in routine laboratories. Alternative phenotypical tests, including calcium-dependent growth at 37°C [9], Congo red binding [10], pyrazinamidase testing [11], autoagglutination testing, and serum resistance testing [12-17], have limited predictive value for *Y. enterocolitica* pathogenicity. Test results are frequently ambiguous and their outcomes may be unreliable because they depend on the presence and expression of (plasmid-borne) virulence genes and the virulence plasmid pYV can easily be lost, depending on the culture conditions [10]. Therefore, pathogenic strain differentiation should not rely solely on the expression or detection of the virulence factors [10].

The detection of pathogenic *Yersinia* from these sources and laboratories of *Y. enterocolitica* infections are based mainly on the isolation of bacteria from food and clinical specimens. Nevertheless, these methods, which require time-consuming enrichments and isolation procedures to distinguish pathogenic from nonpathogenic *Yersinia*, were shown to depend on the presence of the virulence plasmid, which can be lost during bacteriological isolation and enrichment procedures [18]. Rapid methods for the detection of pathogenic *Yersinia* species by PCR techniques have been reported previously [19, 20]. In other studies, the PCR method was developed to detect both *Y. pseudotuberculosis* and pathogenic *Y. enterocolitica* and to differentiate between these 2 species [18, 21].

The importance of poultry meat as a vehicle for the transmission of various diseases has been well documented, especially in countries where standards of hygiene are not strictly enforced. Chicken meat is a specific food category with respect to yersiniosis risk assessment. Currently, information is limited regarding the prevalence of *Yersinia* spp. in foods in Iran. Therefore, the present study was undertaken to determine the prevalence rate and serotyping of *Y. enterocolitica* in chicken meat in Shahrekord and Isfahan townships, Iran, by using culturing methods and PCR analysis to describe the distribution of *Y. enterocolitica* serotype O:3 virulence factors.

**MATERIALS AND METHODS**

**Sample Collection and Identification of *Y. enterocolitica* Biotypes**

In this study, which was conducted from December 2010 to September 2011, a total of 720 fresh raw chicken meat samples were randomly collected from 543 chicken shops in the cities of Isfahan and Shahrekord. All the chickens from which meat samples were taken at the Shahrekord and Isfahan abattoirs (located in the western part of Iran) were healthy.

Separate 10-g breast muscle samples were collected using sterile scissors and tissue forceps and were put into a bag containing 90 mL of Peptone Sorbitol Bile broth (prepared in the laboratory as described in ISO method 10273:2003 [22]). The samples were transferred to the Food Microbiology Laboratory, Islamic Azad University–Shahrekord Branch, Shahrekord, Iran, in a portable insulated cooler. Samples were cultured on the day they were collected. After stomaching, 10 μL was streaked directly onto *Y. enterocolitica* chromogenic agar (YECA) [23] and cefsulodin-irgasan-novobiocin (CIN) plates [24], and 1 mL was transferred into 9 mL of irgasan-ticarcillin-potassium chlorate (ITC) broth [25]. The peptone sorbitol bile broth and ITC were incubated at 25°C for 48 h before a second streaking onto YECA and CIN. In addition, after 24 h of enrichment in ITC broth, an extra streaking on YECA and CIN was performed. All the plates were incubated at 30°C for 24 h.
The presence of typical colonies on CIN (small and smooth, with a red center and translucent red) and on YECA (small and fuchsia red) were checked. At least 2 typical colonies per plate were streaked onto *Y. enterocolitica* chromogenic medium [26], and these plates were incubated at 30°C for 24 h. This step on *Y. enterocolitica* chromogenic medium permitted rapid differentiation of the pathogenic *Y. enterocolitica* (red bull’s-eye-like colonies) from the nonpathogenic *Y. enterocolitica* (blue-purple colonies) [27].

Confirmation and biotyping of *Y. enterocolitica* were then done by biochemical assays. For this reason, the colonies were collected and put through several common tests, including oxidase and urease production, deamination of tryptophan, the behavior in Kligler agar, glucose fermentation, hydrogen sulfide production, gas formation from glucose, and lactose fermentation. The colonies that were urease and glucose positive, capable of the deamination of tryptophan and of lactose fermentation, oxidase negative, and did not produce hydrogen sulfide and gas from glucose were selected for further testing. Biochemical confirmation of the presence of lysine, ornithine, sucrose, rhamnose, and citrate was carried out, and the positive colonies underwent further testing for pathogens using salicin [26], esculin [26], and pyrazinamidase [26]. These individual biochemical tests allowed the samples to be divided into various biotypes [8]. Table 1 shows the reactions of the individual biotypes to various tests.

**Serotyping**

Selected strains with typical characters were serotyped using the commercial antisera O:3, O:5, O:8, and O:9 [28]. The individual colonies were isolated and suspended in physiological solution. A drop of antiserum specific to the individual pathogenic serotype O:3, O:5, O:8, or O:9 was then added to the suspensions.

**PCR Identification of *Y. enterocolitica***

The bacterial strains biochemically identified as *Y. enterocolitica* and cultivated on CIN medium were tested using the PCR method. Purification of DNA from bacterial colonies was achieved using a Genomic DNA Purification Kit [29] according to the manufacturer’s instructions.

The multiplex PCR method, using the following pair of specific primers, was used for the identification of specific pathogens in culture: primer A1 (5’-TTAATGTGTACGCTGGGAGTG-3’) and primer A2 (5’-GGAGTATTCAAT-GAAGCGTC-3’). On the basis of this sequence, a PCR product of 425 bp was expected. To specifically amplify the *Y. enterocolitica* 16S ribosomal RNA gene, a second set of primers, Y1 (5’-AATACCGCATAACGTCTTGC-3’) and Y2 (5’-CTTCTCTCTGCGAGTAACGTC-3’), was used, resulting in a PCR product of 330 bp [30]. The PCR using primers rfbC1 5’-CGCATCTGGGACACTAATTCG-3’ and rfbC2 5’-CCACGAATTCATCAAAACCACC-3’ [18] was used specifically for detection of the *Y. enterocolitica* serotype O:3.

Polymerase chain reaction were performed in a total volume of 25 µL, including 1.5 mM MgCl₂, 50 mM KCl, 10 mM Tris-HCl (pH 9.0), 0.1% Triton X-100, 200 µM each deoxynucleotide 5’-triphosphate [29], 25 pmol of each primer, 1.5 U of Taq DNA polymerase [29], and 3 µL (40 to 260 ng/µL) of DNA. Amplification reactions were carried out using a DNA thermocycler [31] as follows: heat denaturation at 94°C for 5 min, followed by 36 cycles of heat denaturation at 94°C for 45 s, primer annealing at 62°C for 45 s, and DNA extension at 72°C for 45 s. After the last cycle, the samples were kept at 72°C for 7 min to complete the synthesis of all strands [2].
Table 2. Primers used for detection of the various genes of *Yersinia enterocolitica*

<table>
<thead>
<tr>
<th>Gene</th>
<th>Primer name</th>
<th>Primer sequence, 5′–3′</th>
<th>Amplicon length, bp</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>yadA</td>
<td>yadA1</td>
<td>CTTCAGATACCTGGTGTCGCTGT</td>
<td>849</td>
<td>[10]</td>
</tr>
<tr>
<td></td>
<td>yadA2</td>
<td>ATGCTGACTAGAGCGATATCC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>inv</td>
<td>YC1</td>
<td>CTGTGGGGAGAGTGGGGAAGTT</td>
<td>570</td>
<td>[49]</td>
</tr>
<tr>
<td></td>
<td>YC2</td>
<td>GAACTGCTTGATCCCTGAAAA</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ail</td>
<td>Ail1</td>
<td>ACTCGATGATAACTGGGGAG</td>
<td>170</td>
<td>[21]</td>
</tr>
<tr>
<td></td>
<td>Ail2</td>
<td>CCCCAAGTAAATCCATAAAGG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ystA</td>
<td>Pr2a</td>
<td>AATGCTGTCTTCATTTGGAGCA</td>
<td>145</td>
<td>[50]</td>
</tr>
<tr>
<td></td>
<td>Pr2c</td>
<td>ATCCCAATCCTACTTGACTTC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>virF</td>
<td>VirF1</td>
<td>TCATGGCAGAACACGAGTCAG</td>
<td>590</td>
<td>[20]</td>
</tr>
<tr>
<td></td>
<td>VirF2</td>
<td>ACTCACTTACCATTAAAGAAG</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**RESULTS AND DISCUSSION**

Biochemical and PCR results showed that 132 *Y. enterocolitica* strains (18.33%) were isolated (representative of 132 different colony morphologies) out of 720 samples. The highest occurrence belonged to the *Y. enterocolitica* O:5,27 serotype, although strains O:3 and O:8 were also detected. However, serotype O:9 was not found. Additionally, biotype 3 was detected in samples the most often, occurring in up to 52.27% of samples. The second most frequent biotype of the chicken strain *Y. enterocolitica* 2 also occurred frequently (18.18%). However, biotypes 1A and 1B were not detected in the poultry strain *Y. enterocolitica* (Table 3).

The most common serotype or biotype found in the samples was O:5,27/2, which occurred in 42 out of 132 samples (31.8%). Other common strains included O:3/4 and O:5,27/3, with occurrences of 36 (27.3%) and 17 (12.9%), respectively, out of 132 samples (Table 4).

For gene detection, *ail*, *yadA*, *inv*, *ystA*, and *virF* were PCR-amplified, and the individual amplified fragments were subjected to agarose gel electrophoresis (Figure 1). Table 5 shows the occurrence of virulence genes (*ail*, *yadA*, *inv*, *ystA*, *virF*) in isolates of *Y. enterocolitica* O:3 strains.

In the current study, the *ystA* gene was found in 93.47% of isolates. The genes *yadA* and *virF* were identified in 82.60 and 58.69% of *Y. enterocolitica* O:3 isolates, respectively. The *ail* gene was found in 95.65% of isolates, and the *inv* gene was identified in all the strains.

*Yersinia enterocolitica* is a common gram-negative foodborne enteric pathogen found in water, dairy products, and meats. It is one of the most common causes of foodborne gastro-
entersitis in Western and Northern Europe. The incidence is also increasing in the United States and Canada, although this may be a result of improved surveillance and detection methods [32, 33]. Foodborne outbreaks have been associated with virtually all pathogenic serovars, but serovar O:8 has characteristically been associated with more catastrophic human infections, whereas O:3 and O:9 have been linked to milder cases [8, 32]. It seems that this present study is the first prevalence report of direct detection and identification of \textit{Y. enterocolitica} in poultry meat in the west of Iran. Our results revealed that the contaminated raw chicken meat is one of the main sources of human infection caused by \textit{Y. enterocolitica}.

### Table 4. Prevalence of \textit{Yersinia enterocolitica} serotypes/biotypes isolated from chicken meat

<table>
<thead>
<tr>
<th>Serotype/biotype</th>
<th>Positive serotype/biotype, no.</th>
</tr>
</thead>
<tbody>
<tr>
<td>O:3/2</td>
<td>3/4</td>
</tr>
<tr>
<td>O:3/3</td>
<td>3/34</td>
</tr>
<tr>
<td>O:3/4</td>
<td>36/14</td>
</tr>
<tr>
<td>O:3/5</td>
<td>4/4</td>
</tr>
<tr>
<td>O:5.27/2</td>
<td>42/16</td>
</tr>
<tr>
<td>O:5.27/3</td>
<td>17/24</td>
</tr>
<tr>
<td>O:5.27/4</td>
<td>12/4</td>
</tr>
<tr>
<td>O:5.27/5</td>
<td>8/2</td>
</tr>
<tr>
<td>O:8/2</td>
<td>3/4</td>
</tr>
<tr>
<td>O:8/3</td>
<td>2/11</td>
</tr>
<tr>
<td>O:8/4</td>
<td>2/5</td>
</tr>
<tr>
<td>O:8/5</td>
<td>0/10</td>
</tr>
</tbody>
</table>

1\(^n\) = 132 isolates.

### Table 5. Prevalence of \textit{Yersinia enterocolitica} serotype O:3 virulence genes isolated from chicken meat

<table>
<thead>
<tr>
<th>Gene</th>
<th>Positive samples, no. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>yadA</td>
<td>38 (82.60)</td>
</tr>
<tr>
<td>inv</td>
<td>46 (100)</td>
</tr>
<tr>
<td>ail</td>
<td>44 (95.65)</td>
</tr>
<tr>
<td>ystA</td>
<td>43 (93.47)</td>
</tr>
<tr>
<td>virF</td>
<td>27 (58.69)</td>
</tr>
</tbody>
</table>

1\(^n\) = 46 isolates.

Figure 1. Agarose gel electrophoresis of PCR products amplified with a multiplex PCR method for the \textit{ail} (170 bp), \textit{yadA} (849 bp), \textit{inv} (570 bp), \textit{ystA} (145 bp), and \textit{virF} (590 bp) genes from \textit{Yersinia enterocolitica} serotype O:3. M = 100-bp DNA ladder [29].
The aim of the present study was to evaluate the occurrence and antimicrobial resistance of *Y. enterocolitica* and to identify and characterize chromosomal and plasmid-encoded virulence genes.

The recent development of sensitive, specific PCR assays for the detection of *Yersinia* organisms has greatly improved the ability to study this organism in fresh and fixed samples. Because *Y. enterocolitica* is a common foodborne pathogen and is implicated in such a wide range of gastrointestinal diseases, the development of a PCR assay that could be used to assign *Y. enterocolitica*-positive specimens to a particular biogroup has significant implications for clinical diagnosis, microbiological research, and epidemiological studies [34–36].

In the present study, both molecular and cultural methods were used to characterize *Y. enterocolitica* isolates in chicken meat. Results showed that 132 of the 720 (18.3%) chicken meat samples studied were found to be infected with *Y. enterocolitica*. Out of 132 samples positive for *Y. enterocolitica*, the most prevalent biotypes were 1A (0%), 1B (0%), 2 (18.18%), 3 (52.27%), 4 (17.42%), and 5 (12.12%), and the most prevalent serotypes were O:3 (34.84%) and yadA (58.69%) out of the total.

Multiplex PCR assay results showed that the chromosomal virulence genes included virulence-associated genes, including *ystA* (82.60%) and *virF* (58.69%) out of the total of 46 *Y. enterocolitica* serotype O:3 strains.

The results of our study are the same as those of other studies [37] and showed that of the 6 biotypes and 50 serotypes of *Y. enterocolitica* isolated, the incidences of *Y. enterocolitica* se-rotype O:5,27 were the highest in chicken meat. However, the results of other studies showed that biotype 1A was the most heterogeneous and encompassed a wide range of serotypes, and the serotypes O:5, O:6,30, O:6,31, O:7,8, and O:10, as well as O-nontypable strains, were isolated the most often. Another study [38] showed that the pig is the major reservoir of pathogenic *Y. enterocolitica* of bioserotype 4/O:3, the most common type found in humans; however, our study revealed that O:5,27/2 was the most frequent serobiotype of *Y. enterocolitica* isolated from chicken meat samples. Therefore, our study showed that the O:5,27/2 serobiotype of *Y. enterocolitica* caused the most human infections through consumption of contaminated chicken meat. Our results indicated that *ystA*, with an incidence of 93.47%, was one of the most prevalent virulence genes of *Y. enterocolitica* that was isolated from chicken meat samples. However, in previous studies, pathogenic *yst*-positive *Y. enterocolitica* strains were isolated from ground beef [39] but were not detected in chicken samples [40], which was inconsistent with our results. In addition to chicken meat, chicken eggshell surfaces are a source of *Y. enterocolitica* [41].

A study conducted in Argentina [42] showed that 38.65% of meat samples were contaminated with *Y. enterocolitica*, which was higher than our results (18.33%). All *Y. enterocolitica* 2/O:9 strains gave results related to virulence by phenotypic tests and exhibited the genotype *virF*+*myfA*+*ail*+*ystA*+, whereas biotype 1A strains showed the genotype *virF*+*myfA*−*ail*+*ystA*+*ystB*+. Another study [43] showed that the positive rates of virulence genes tested in 160 *Y. enterocolitica* isolates were *inv* (100%), *ail* (94%), *ystA* (93%), *ystB* (7.5%), *ystC* (5%), *yadA* (89%), and *virF* (82%) in patients with diarrhea.

In an Indian study [44], 81 strains of *Y. enterocolitica* biovar 1A were isolated from diarrheic human stools (51 strains), wastewater (18 strains), pig throats (7 strains), and pork (5 strains). Virulence-associated genes, including *ail*, *virF*, *inv*, *myfA*, *ystA*, *ystB*, *ystC*, *tccC*, *hreP*, *fepA*, *fepD*, *fes*, *ymoA*, and *sat*, were detected by PCR amplification in 81 clinical and nonclinical strains of *Y. enterocolitica* biovar 1A. All strains lacked *ail*, *virF*, *ystA*, and *ystC* genes. The distribution of other genes with respect to clonal groups revealed that 4 genes (*ystB*, *hreP*, *myfA*, and *sat*) were associated exclusively with strains belonging to clonal group A [44]. In a German study, a total of 151 *Y. enterocolitica* strains, isolated from humans, animals, and the environment, were identified and found to belong to biovars 1A (n = 22), 1B (n = 13), 2 (n = 12), 3 (n = 31), 4 (n = 63), and 5 (n = 10). Additionally, their susceptibilities to 71 antibiotics were examined, and 99% of all *Yersinia* strains were found to be resistant to amoxicillin. The results of this study showed that unambiguous state-
ments cannot be made about the natural antibiotic susceptibility of *Y. enterocolitica* to certain β-lactams and fosfomycin. The data indicated a complex regulation of β-lactamases in *Y. enterocolitica*. Some β-lactamases were found more frequently or were expressed predominantly in specific biovars [45]. In a study on pathogenic *Y. pseudotuberculosis* and *Y. enterocolitica* found in migratory birds in Sweden [46], 468 fecal samples from 57 different species of migratory birds were collected. In total, *Yersinia* spp. were isolated from 12.8% of the collected samples. The most commonly found species was *Y. enterocolitica*, which was isolated from 5.6% of samples. In addition, 10 *Y. enterocolitica* strains, all from barnacle geese, belonged to bioserotype 3/O:3, which is associated with human disease. Two of the strains were pathogenic, carrying the virF gene within their plasmids [46]. In a Dutch study [30], a duplex PCR assay targeting the *ail* and 16S ribosomal RNA genes of *Y. enterocolitica* was developed specifically to identify pathogenic *Y. enterocolitica* from pure culture. Validation of the assay was performed with 215 clinical *Yersinia* strains and 40 strains of other bacterial species. Within an assay time of 4 h, this assay offered a very specific, reliable, and inexpensive alternative to conventional phenotypic assays used in clinical laboratories to identify pathogenic *Y. enterocolitica*. The specificity of the duplex PCR assay was examined by isolating genomic DNA from 27 different *Y. enterocolitica* serogroups [30]. Lambertz et al. [47] showed that current methods for detection of the pathogenic *Y. enterocolitica* bacterium in food are time consuming and inefficient. Thus, they developed and evaluated an in-house TaqMan probe-based real-time PCR method for the detection of this pathogen. The complete method consists of overnight enrichment, DNA extraction, and real-time PCR amplification. In addition, the method was tested on naturally contaminated food. In all, 18 out of 125 samples were positive for the *ail* gene.

A study by Thisted Lambertz and Danielsson-Tham [48] showed that pigs were the main reservoir of foodborne pathogenic *Y. enterocolitica*. Pork meat collected from refrigerators and local shops frequented by yersiniosis patients (n = 48) were examined for the presence of pathogenic *Yersinia* spp. The researchers used combined culturing and PCR methods for detection, and a multiplex PCR was developed for detection of virulence genes (*yst*, *rfbC*, *ail*, *virF*). In all, 118 pork products (91 raw and 27 ready-to-eat) were collected. Pathogenic *Yersinia* spp. were detected by PCR in 9.89% (9 of 91) of the raw pork samples but in none of the ready-to-eat products.

Thoermer et al. [10] developed a PCR-based assay for the detection of plasmid- and chromosome-borne virulence genes in *Y. enterocolitica* and *Y. pseudotuberculosis* to investigate the distribution of these genes in isolates from various sources. They compared results of PCR genotyping, based on 5 virulence-associated genes of 140 strains of *Y. enterocolitica*, with phenotypic tests, such as biotyping and serotyping, and with virulence plasmid-associated properties, such as calcium-dependent growth at 37°C and Congo red uptake. They suggested that genotypical data correlated well with biotypical data. The study found that only *ystB* was present in biotype 1A, although 40 *Y. pseudotuberculosis* isolates were PCR-tested for the presence of *inv*, *yadA*, and *lcrF*. All isolates were *inv* positive, and 88% of the isolates contained the virulence plasmid genes *yadA* and *lcrF*.

To the authors’ knowledge, contact with chicken feces and a lack of hygiene in chicken slaughterhouses are the 2 most frequent reasons chicken meat is contaminated with *Y. enterocolitica*, which can easily spread to and cause yersiniosis in humans.

**CONCLUSIONS AND APPLICATIONS**

1. The existence of *Y. enterocolitica* in poultry meat indicates a carrier state and the possibility of shedding the microorganism into the environment. It shows that chickens are an important bacterial reservoir and a potential source of human infection.

2. A limited number of studies have investigated the occurrence and characterization of *Y. enterocolitica* in Iran. These findings support the hypothesis that chicken meat is a possible source of gastroenteritis caused by *Y. enterocolitica* and may be a reservoir of infection in Iran.
3. We conclude that control of *Y. enterocolitica* in chicken meat is crucial for managing infection. Maintaining hygiene at poultry slaughterhouses; checking and monitoring the slaughterhouse staff; preventing the contact of poultry carcasses with the slaughterhouse bed, soil, and feces; using clean and sanitary water for washing the carcasses; and finally, thoroughly cooking the chicken for oral use are the main principles for preventing human disease.

**REFERENCES AND NOTES**


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