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Molecular Characterization of Foodborne Pathogens in Some Meat Products

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CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF DIAGRAMS

7. Summary

A significant number of 100 random samples of raw meat products (minced meat, sausage), ready-to-eat meat products (luncheon and cured basterma) 25 samples from each product were gathered for bacteriological analysis for the identification of *S. aureus*, *L. monocytogenes*, *E. coli* and *Salmonella* microorganisms from various markets in the provinces of Cairo and Giza.

For the detection of *S. aureus*, *L. monocytogenes*, *E. coli* and *Salmonella;* these samples were tested by bacteriological, m-PCR and sequencing techniques.

The percentages of *S. aureus* isolated from the samples of minced meat, sausage, luncheon and basterma analyzed were 24%, 28%, 16%, 12%, respectively. In addition, 20 random positive meat product samples by conventional method were re-examined by m-PCR, there were great agreement between results of conventional method and m-PCR technique in 18 random samples.

The percentage of *L. monocytogenes* isolated from the minced meat analyzed was 4% by both conventional method and m-PCR technique. But it was not isolated from samples of sausage, luncheon and basterma.

The percentages of *E. coli* isolated from the samples of minced meat, sausage, luncheon and basterma analyzed were 28%, 20%, 12%, 4%, respectively. In addition, 16 random positive meat products samples by conventional method were re-examined by m-PCR, there were great agreement between results of conventional method and m-PCR technique in 15 random samples.

The percentages of *Salmoenlla* isolated from the samples of minced meat was 16% and from sausage was 8%, but it was not isolated from treated meat products (luncheon and basterma). In addition, 5 random positive meat products samples by conventional method were re-examined by m-PCR, there were agreement between results of conventional method and m-PCR technique in 5 random samples.

One positive sample (minced meat) showed mixed infection with *E. coli* and *Salmonella* by both conventional method and m-PCR technique.

The results showed that m-PCR is an excellent method for identifying foodborne pathogens because it is less labor, more sensitive, and saves time.

For *S.aureus,* the *nuc* gene was sequenced where 2 positive samples were sequenced and submitted on GenBank one sample from sausage with accession number MT647589 and one sample from basterma with accession number MT647590. Multiple Sequence Alignment of the amplified fragment (270 bp) for *nuc* gene of *S.aureus* (GeneBank: MN7042871, CP053183, CP039162, KR080211, LC089736, CP053075, MN704287, CP047851, CP010943, CP043920) showed sequence similarity 98-100% for sample 1 and 97-100% for sample 2. Phylogenetic tree based on nucleotide sequences of 10 strains by using neighbor joinning method. Where *S. aureus* sample 1 was close related to MN704287, CP053183, and CP047851, MN704287. While *S. aureus* sample 2 was close related to CP053075 and CP039162.

For *L.monocytogenes,* the *hlyA* gene was sequenced where 1 sample from minced meat was sequenced and submitted on Genbank with accession number MT647591. Multiple Sequence Alignment of the amplified fragment (456bp) for *hlyA* gene of *L.monocytogenes* (GeneBank: CP048400, CP030813, CP025442, CP054042, CP025222, CP032669, AY512405, CP007583, CP007462,

CP013287) showed sequence similarity 97-100%. Phylogenetic tree based on nucleotide sequences of 10 strains by using neighbor joinning method. Where *L. monocytogenes* sample was close related to CP048400.

For *E.coli,* the *uspA* gene was sequenced where 2 positive samples were sequenced and submitted on GenBank one sample from minced meat with accession number MT671369 and one sample from luncheon with accession number MT671370. Multiple Sequence Alignment of the amplified fragment (884bp) for *uspA* gene of *E.coli* (GeneBank: AP023237, CP025967, CP018995, CP048647, X67639, KF765740, MH138303, AF346731, CP041002, CP043539) showed sequence similarity 97-100% for sample 1 and 96-100% for sample 2. Phylogenetic tree based on nucleotide sequences of 10 strains by using neighbor joinning method. Where *E.coli* sample 1 was close related to AP023237 while *E.coli* sample 2 was close related to CP048647.

For *Salmonella,* the *invA* gene was sequenced where 2 positive samples were sequenced and submitted on GenBank one sample from minced meat with accession number MT662113 and one sample from sausage with accession number MT662114. Multiple Sequence Alignment of the amplified fragment (284bp) for *invA* gene of *Salmonella* (GeneBank: CP053865, CP043667, KX524161, CP055130, CP051286, CP034233, CP030203, KJ718880, CP053409, CP030219) showed sequence similarity 93-100% for sample 1 and 92-99%. Phylogenetic tree based on nucleotide sequences of 10 strains by using neighbor joinning method. *Salmonella* sample 1 was close related to CP053865 and CP055130. While *Salmonella* sample 2 was close related to CP051286.

To sum up, biotechnological and bioinformatics tools considered as indispensable ways which help in the detection and genomic studies of foodborne pathogens as they are convinient, fast, and accurate tools used nowadays in modern science in accordance to traditional method especially in case outbreaks.