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Molecular Characterization of Foodborne Pathogens in Some Meat Products

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CONTENTS

Title	Page
1. INTRODUCTION.....	1
2. REVIEW OF LITERATURE.....	5
2.1. Sources of contamination of some meat products with foodborne pathogens.....	5
2.2. Incidence of foodborne pathogens in some meat products.....	8
2.3. Public health hazards of foodborne pathogens.....	15
2.4. Uses of Multiplex PCR in the diagnosis of foodborne pathogens.....	21
2.5. Molecular Sequencing in the diagnosis of foodborne pathogens.....	26
3. MATERIAL AND METHODS	31
Part I: Conventional Recovery Methods	31
Part II: Polymerase Chain Reaction (PCR).....	46
Part III: Sequencing Analysis.....	55
4. RESULTS.....	66
5. DISCUSSION.....	92
6. CONCLUSION AND RECOMMENDATIONS.....	108
7. SUMMARY.....	114
8. REFERENCES	117
9. ARABIC SUMMARY	-

LIST OF TABLES

Table No.	Title	Page
A	Detailed description of the designed oligonucleotide primers used	49
B	PCR Mastermix for Uniplex PCR	52
C	PCR Mastermix for Multiplex PCR	52
D	Temperature and time conditions of the different primers during PCR	53
E	Quality of DNA used based on product size	60
F	Sequence reaction protocol	61
1	Incidence of <i>S. aureus</i> in the examined meat product samples	66
2	Incidence of <i>L. monocytogenes</i> in the examined meat product samples	67
3	Incidence of <i>E. coli</i> in the examined meat product samples	68
4	Incidence of <i>Salmonella</i> in the examined meat product samples	69
5	Total incidence of foodborne pathogens in the examined meat product samples	70
6	Summary of bacteriological results and m- PCR results	79
7	Identity Matrix of <i>S.aureus</i> Sequence Alignment	88
8	Identity Matrix of <i>L.monocytogenes</i> Sequence Alignment	88
9	Identity Matrix of <i>E.coli</i> Sequence Alignment	89
10	Identity Matrix of <i>Salmonella</i> Sequence Alignment	89

LIST OF FIGURES

Figure No.	Title	Page
A	100 bp DNA Ladder Ready to Load (GeneDirex)	49
B	Applied Biosystems 3500 Genetic Analyzer	57
C	Steps of BLAST using National Center of Biotechnology Information NCBI Webste	63
1	Incidence of <i>S. aureus</i> in the examined meat product samples	66
2	Incidence of <i>L. monocytogenes</i> in the examined meat product samples	67
3	Incidence of <i>E. coli</i> in the examined meat product samples	68
4	Incidence of <i>Salmonella</i> in the examined meat product samples	69
5	Total incidence of foodborne pathogens in the examined meat product samples	70
6	Uniplex PCR validation	71
7	Multiplex PCR validation for Gram +ve bacteria (<i>S.aureus</i> & <i>L. monocytognes</i>)	72
8	Multiplex PCR validation for Gram -ve bacteria (<i>Salmonella</i> & <i>E. coli</i>)	73
9	Multiplex PCR for <i>S. aureus</i> positive meat product samples	74
10	Multiplex PCR for <i>L.monocytogenes</i> positive meat product sample	75
11	Multiplex PCR for <i>E.coli</i> positive meat product samples	76
12	Multiplex PCR for <i>Salmonella</i> positive meat product samples	77
13	Multiplex PCR for <i>E. coli</i> & <i>Salmonella</i> on positive meat product sample (mixed infection) from minced meat	78
14	Graphic view of <i>S.aureus</i> Multiple Sequence Alignment	83
15	Graphic view of <i>L.monocytogenes</i> Multiple Sequence Alignment	84
16	Graphic view of <i>E.coli</i> Multiple Sequence Alignment	85

17	Graphic view of <i>Salmonella</i> Multiple Sequence Alignment	87
18	Phylogenetic Analysis of <i>S.aureus</i> samples (Neighbor Joining Bootstrap consensus tree)	90
19	Phylogenetic Analysis of <i>L.monocytogenes</i> sample (Neighbor Joining Bootstrap consensus tree)	90
20	Phylogenetic Analysis of <i>E.coli</i> samples (Neighbor Joining Bootstrap consensus tree)	91
21	Phylogenetic Analysis of <i>Salmonella</i> samples (Neighbor Joining Bootstrap consensus tree)	91

LIST OF DIAGRAMS

Diagram No.	Title	Page
1	Standard procedure used for detection of <i>S. aureus</i>	34
2	Standard procedure used for detection of <i>L. monocytogenes</i>	38
3	Standard procedure used for detection of <i>E. coli</i>	42
4	Standard procedure used for detection of <i>Salmonella</i>	45
5	The main steps for sequencing	65

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 joining 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 joining 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 joining 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 joining 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 convenient, fast, and accurate tools used nowadays in modern science in accordance to traditional method especially in case outbreaks.