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PASTRY FOODS AS POTENTIAL SOURCE OF PATHOGENIC *E.COLI*

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A B S T R A C T

A total of 90 random samples of ready to-eat-pastry foods of meat pie, meat pizza and pasta pashamil. 30 samples of each were collected from different fast foods services at Kalyobia governorate to be examined bacteriologically for detection of pathogenic *Escherichia coli* microorganisms. The obtained results indicate that incidence of *E.coli* isolated from examined samples of pastry foods were 20 %, 26.76 %, 36.67% of meat pizza, meat pie and pasta pashamil respectively. Concerning Enterohaemorrhagic *E.coli* strains O26 and O111 H4 incidence were 3.33% and 6.67%in meat pizza while Enterohaemorrhagic *E.coli* O111 H4 incidence were 10 % in meat pie but the incidence of Enterohaemorrhagic strains O26 and O111 H4 were 10% and 6.67% in pasta pashamil respectively. Enteropathogenic *E.coli* strains O44 H18 incidence were 3.33% in meat pizza samples while Enteropathogenic *E.coli* strains O55 H7, O114 H21 and O119 H6 were 3.33% in meat pie samples. The incidence of Enteropathogenic *E.coli* strains O114H21 and O119 H6 were 3.33% in pasta pashamil. Incidence of toxigenic *E.coli* O127H6 were 6.67% in meat pizza while in pasta pashamil was 10.00%.Enteroinavsive *E.coli* O124 incidence in meat pie was 6.67% but in pasta pashamil its incidence was 3.38%.The public health significance of isolated organisms from the examined ready-to-eat pastry foods was discussed as well as some recommendations to ensure the safety and the quality of meat pie, meat pizza and pasta pashamil prepared in fast foods services were outlines.

Keywords: Pastry foods, *E.coli*, fast foods

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1. INTRODUCTION

Concerning the field of meat fast foods and pastry restaurants *E.coli* was found to dominate in raw meat handling subsequent to service contamination accompanied with variable hygienic conditions of manufacturing fresh meat products, Bryan, (1988). Moreover sources of contamination with *E.coli* are hands of staff, primary habitat of E.coli in gastrointestinal tract of mammals and (outcome of fecal contamination occurs), foods utensils, air, soil and unclean vegetables. Ready prepared foods are fried and held at room temperature for considerable period of time and later reheated without reaching the prescribed temperature, thus the existing microorganisms reach high levels sufficient to produce food -borne diseases (Primo et al. 1993). Therefore the objectives of the present study was directed to find the level of contamination of different pathogenic E.coli groups in ready-to-eat meat pastry products (meat pie, meat containing pizza and pasta pashamil) and, the following items were detailed.

- Isolation of *E.coli* from the examined samples of pastry foods.

- Identification of the different serotypes of pathogenic *E.coli* isolated from the examined samples of pastry foods.
- Demonstration of *E.coli* in pastry foods by application of ELISA technique.
- Interpretation of the action of antimicrobial discs on the isolated *E.coli* strains.

2. MATERIALS AND METHODS

2.1. Samples:

Ninety random samples of pastry foods represented by meat pizza, meat pie and pasta pashamil (30 of each) were collected from different restaurants in Benha city, Kalyobia governorate, Egypt. The collected samples were subjected to the bacteriological examination to detect pathogenic *E.coli* in such examined pastry foods.

2.2. Screening of E.coli

Preparation of samples was applied according to (International commission of Microbiological Specification for Foods "ICMSF" 1996) from the original dilution, 1ml was inoculated into MacConkey, broth tubes supplemented with inverted Durham, s tubes. The inoculated tubes were incubated at 37oc for 24 hours. Loop from positive MacConkey, broth tubes were separated streaked onto Eosin Methylene Blue agar plates (EMB) which were then incubated at 37oc for 24 hours. Suspected colonies were metallic green on color. Suspected colonies were purified and inoculated into slope nutrient agar tubes for further identification.

2.3. Morphological examination

Identification of *E.coli* by application gram staining (Cruickshank et al., 1975) and motility test. (Collin and Lyne1984).

2.4. Biochemical identification

It was carried out

- Indole production test (Kovacs, 1928).
- Methyl red test (Ljutov, 1961).
- Voges-praskauer test (Ljutov, 1963).
- Citrate utilization test (Simmon, 1926).
- Gelatin hydrolysis test (Collins and lyne, 1984).
- Hydrogen Sulphide production test (Macfaddin, 1976).
- Oxidation- fermentation test (Hugh and Leifsol, 1953).
- Urease test (Edwards and Ewing, 1986).
- Arginine hydrolysis (Collins and lyne, 1984).
- Eijkman test (Collins and lyne, 1984).
- Nitrate reduction test (Collins and lyne, 1984).
- Fermentation of sugars (Macfaddin,1976)

2.5. Serological identification

Isolates were serologically identified according to Koch et al. (1996) by using rapid diagnostic E.coli sets (DENKA SEIKEN CO., Japan) for diagnosis of Enteropathogenic types. Two separate drops of saline were put on a glass slide and a portion of the colony from the suspected culture was emulsified with the saline solution to give a smooth fairly dense suspension. To one suspension, control, one loop of saline was added and mixed. To the other suspension one loop of undiluted antiserum was added and titled back and forward for one minute. Agglutination was observed using indirect lighting over a dark background. When a colony gave a strongly positive agglutination with one of the pools of polyvalent serum, a further portion of it was inoculated onto a nutrient agar slant and incubated at 37°C for 24 hours to grow as a culture for testing with mono-valent sera.

A heavy suspension of bacteria from each slope culture was prepared in saline, and slide agglutination tests were performed with the diagnostic sera to identify the O-antigen..

2.6. Statistical Analysis

The obtained results were statistically evaluated by application of Analysis of Variance (ANOVA) test according to Feldman, et al. (2003).

3. RESULTS

It is evident from the results recorded in table (1) that *E. coli* was isolated from 20%, 26.67 and 36.67% in the examined samples of meat pizza, meat pie and pasta pashamil, respectively. Totally 27.78% of the examined samples of pastry foods were contaminated with E.coli

Table (1): Incidence of *E. coli* organisms isolated from the examined samples of pastry foods (n=30).

Pastry food	No	%
Meat pizza	6	20.00
Meat pie	8	26.67
Pasta pashamil	11	36.67
Total (90)	25	27.78

Table (2): Serotyping of *E.coli* organisms isolated from the examined samples of Meat pizza (n=30)

E.coli Strains	Meat pizza		Strain characteristics
	No	%	
O26	1	3.33	EHEC
O44:H18	1	3.33	EPEC
O111:H4	2	6.67	EHEC
O127:H6	2	6.67	ETEC
Total	6	20.00	

EPEC= Enteropathogenic *E.coli* ETEC = Enterotoxigenic *E.coli* EHEC = Enterohaemorrhagic *E.coli*

Results achieved in table (2) that the isolated *E.coli* was serotyped from the examined samples of meat pizza as O26 (3.33%) EHEC Enteropathogenic *E.coli* O44 H18 (3.33%) EPEC and O111 H4 (6.67%) EHEC and O127 H6 (6.67%) ETEC (Enterotoxegenic).

Table (3) has given that *E.coli* was serotyped from the examined samples of meat pie as O55H7 (3.33%) EPEC, O111 H4 (10.00%) EHEC, O114 H21 (3.33%) EPEC, O119 H6 (3.33%) EPEC and O124 (6.67%) EIEC (Entero-inavsive) E.coli.

Table (3): Serotyping of *E.coli* organisms isolated from the examined samples of Meat pie (n=30)

E.coli Strains	Meat pie		Strain characteristics
	No	%	
O55:H7	1	3.33	EPEC
O111:H4	3	10.00	EHEC
O114:H21	1	3.33	EPEC
O119:H6	1	3.33	EPEC
O124	2	6.67	EIEC
Total	8	26.67	

EPEC= Enteropathogenic *E.coli* ETEC = Enterotoxigenic *E.coli* EHEC = Enterohaemorrhagic *E.coli*

Table (4): Serotyping of *E.coli* organisms isolated from the examined samples of Pasta pashamil (n=30)

E.coli Strains	Pasta pashamil		Strain characteristics
	No	%	
O26	3	10.00	EHEC
O111:H4	2	6.67	EHEC
O114:H21	1	3.33	EPEC
O119:H6	1	3.33	EPEC
O124	1	3.33	EIEC
O127:H6	3	10.00	ETEC
Total	11	36.67	

EPEC= Enteropathogenic *E.coli* ETEC = Enterotoxigenic *E.coli* EHEC = Enterohaemorrhagic *E.coli.*

4. DISCUSSION

Pastry foods have been described as commercially prepared ready-to cook (RTC) and ready to-eat foods containing major ingredients from two or more categories, the combination of these ingredients into a single products presents not only the original hazards of each ingredient but also the possibility of additional hazards due to further handling, processing or modification of the environment, National academy of

sciences (NAS). (1985). Table one gives the incidence of *E.coli* organisms isolated from the examined samples of pastry foods.

Actually , *E.coli* was previously isolated from fast foods by Chibber et al. (1990)was examined one hundred and one (101) samples from meat, food handlers and equipments, they found that 24.8% were enterotoxegenic *E.coli.* Bensink and Bothman (1991) Concluded that about 10% of the four hundreds (400) meat samples were contaminated with *E.coli*. Mousa et al., (1993) Isolated *E.coli* from 33% of examined samples of minced meat with difference percentages.

However E.coli have probably received more attention than the most groups of bacteria in meat for their significance as indicator organisms of fecal contamination and their ability to grow well over wide range of temperature below 10oc up to 46oc Gill, McGinnis and Bryan(1998).

Tables 2,3 and 4 give serotyping of *E.coli* organisms isolated from examined samples of meat pizza, meat pie and pasta pashamil respectively. E.coli classified according to serotypes into Bryan (1982), Enterotoxigenic E. coli (ETEC) cells adhere to epithelial cells and produce toxin but do not invade epithelial cells. The predominant sero-groups are O6, O8, O11, O15, O20, O25, O27, O78, O128, O148, O149, O159, and O173. Enteropathogenic *E.coli* (EPEC) cells adhere to epithelial cells intimately, produce attachment/effacement lesion and are invasive; however, they do not produce toxin. The notable sero-groups are O55, O86, O111, O119, O125, O126, O127, O128, and O142. Enterohaemorrhagic E. coli (EHEC) also binds strongly to epithelial cells, produce attachment/effacement lesions and produce toxins. The serogroups are O4, O5, O16, O26, O55, O111ab, O113, O117, O157, and O172. Several recently identified serogroups belong to EHEC include O176, O177, O178, O180, and O181.

Enteroaggregative *E.coli* (EAEC) adheres to epithelial cells, form aggregates, produce toxin, but do not invade. Virotype O3, O15, O44, O86, O77, O111 and O127.

Enteroinavsive *E.coli* (EIEC) cells also adhere, invade cells, and move from cell-to-cell, but do not produce toxin. The pathogenicity of EIEC resembles infection caused by Shigellas pp. And the predominant symptom is dysentery. The EIEC serogroups are O28, O29, O112, O124, O136, O143, O144, O152, O159, O164, and O167.

The diffusely adhering *E. coli* (DAEC) cells adhere to epithelial cells, but they neither invade nor produce toxin.

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معجنات الاغذية كمصدر محتمل للاصابة بميكروبات الايشريشيا كولاي.

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الملخص العربي

تم جمع 90 عينة عشوائية من البيتزا وفطائر اللحم و المكرونة الباشاميل من اماكن مختلفة تجهيز الوجبات السريعة بالقليوبية بواقع 30 عينة من كل نوع وقد تم نقل هذة العينات علي وجة السرعة و تحت ظروف صحية مشددة الي المعمل لمعرفة الحالة البكتيريا لها من حيث نسبة الايشيرشيا كولاي و قد اوضحت النتائج ان متوسط نسبة ميكروب الايشيريشيا كولاي المعزولة من العينات المختبرة هو 20% و26.76%و 36.76 % من البيتزا وفطائر اللحم و المكرونة الباشميل علي التوالي.وقد وجد ان عترة انتيروهيموجينك ايشرشيا كولاي والتي تم عزلها من البيتزا النمط المصلي O26 , H4 : O111بنسبة 3.33 %و 6.67 % علي التوالي.بينما وجد ان نفس العترة والتي تم عزلها من فطائر اللحم ولكن النمط المصلي H4 : O111بنسبة 10 % .ونفس العترة بالنمط المصلي O26 , H4 : O111 توجد بنسبة 10 % و 6.67 % في عينات المكرونة الباشاميل علي التوالي.وقد وجد ان عترة الانتيروباثوجنيك ايشرشيا كولاي والتى تم عزلها من البيتزا النمط المصلي O44, H18توجد بنسبة 3.33 % .بينما وجدت هذة العترة النمط المصلي H6 : O119 و H21 : O114 وH7 : O55 بنسبة 3.33 % في عينات فطائر اللحم.ونفس هذة العترة النمط المصليH21 : O114 وH6 : O119 في عينات المكرونة الباشاميل بنسبة حدوث 3.33 % . وهناك عترة الانتيروتوكسوجينك ايشيرشيا كولاي و التي تم عزلها من العينات المختبرة كالاتي عينات البيتزا النمط المصلي H6 : O127بنسبة 6.67% بينما من العينات المكرونة البشاميل بنسبة 10% نفس النمط المصلي.و عترة الانتيروانفازيف ايشيرشيا كولاي O114 وجدت بنسبة 6.67% في عينات فطائر اللحم و بنسبة 3.33% في المكارونا البشاميل.و قد اوضحت النتائج انة تم عزل ميكروب الايشيرشيا كولاي الممرضة بنسب مختلفة من العينات المختبرة و تم تصنيف العترات التي تم عزلها كالاتي H4 : O111 ، H7 : O55 ، H6 : O127 ، O44, H18 ، O26 ، O114 : H21، H6 : O119 و O114. وقد تم دراسة ومناقشة الاهمية الصحية للميكروبات التي تم عزلها وكذلك الشروط الصحية الواجب توافرها اثناء اعداد و تقديم هذة الوجبات لتجنب خطر هذة الميكروبات.

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