

Detection of *E. coli*, as a contaminant of minced meat in certain locations of Baqubah city-Diyala Province-Iraq

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Abstract

This survey investigated the prevalence of *Escherichia coli* (*E.coli*) in raw minced beef meat collected from different areas of Baqubah city in Diyala Province for detection of contamination with *E.coli*. A(90) samples were collected at (10) samples from each area in Baqubah city. and different dilutions were made for each one (10^{-1} , 10^{-2} , 10^{-3} , 10^{-4}).

These sample were pour onto MacConkey Agar and then streak onto eosin methylene blue agar (EMB) plates. The results was shown presence of high contamination ratio of the microbial isolation from minced beef meat of *E.coli* bacteria in different areas of Bagubah city. with positive (76.85%), while negative (23.1%).

Keywords : *E.coli*, beef minced meat, contamination, microbial

الخلاصة

أجري هذا البحث لغرض التحقق في انتشار بكتريا الإيشيريشيا كولاي في لحم البقر المفروم الخام الذي تم جمعه من مناطق مختلفة من مدينة بعقوبة في محافظة ديالى للكشف عن تلوث اللحوم التي تباع في الاسواق المحلية من قبل محلات القصابية. حيث تم جمع (90) عينة بواقع (10) عينات من كل منطقة واجريت سلسلة من التخفيفات لكل واحدة من هذه العينات (10^{-1} , 10^{-2} , 10^{-3} , 10^{-4}). تم زرع هذه العينات في الوسط (ماكونكي) ثم في الوسط (ايوزين المثيلين الازرق). وقد أظهرت هذه العينات وجود نسبة تلوث عالية للعزل الميكروبي من لحم البقر المفروم لبكتريا الإيشيريشيا كولاي في مناطق مختلفة من مدينة بعقوبة حيث كانت بنسبة التلوث تقدر ب (76.85%) بينما كانت نسبة الغير ملوث تقدر ب (23.1%).

1. Introduction

A Gram negative rod (bacillus) is *Escherichia coli* belongs to Enterobacteriaceae. in the intestinal tract originate normal commensals of the majority *E. coli*. this organism which Pathogenic strains are famous from normal flora by their control of virulence factors like exotoxins[1]

Escherichia coli can reason serious sickness in humans. Either infectivity may be caused by consumption polluted water or food, by get in touch with an infected person or by get in touch with the surrounds the animals or with animals. [2]

four most important pathogens that have commonly been related with meat products and meat as well as *Listeria monocytogenes*, *Campylobacter* spp *Salmonella* spp and *Escherichia coli*. These organisms leads to a numeral of human disease [3]

mainly significant food-borne pathogens that has gained enlarged interest in latest years is *E. coli*. Typical disease caused by *E. coli* infectivity can be life destructive, and appears as collection of symptoms including thrombotic thrombocytopenic

purpura, hemolytic-uremic and hemolytic colitis [4].

Main natural reservoirs regarded are ruminants. Outbreaks and random cases of diseases in human caused by *E. coli* wick related to water, unpasteurized fruit juices, vegetables, dairy products, raw milk, ground beef and meat [5,6]

Infections may be occur by direct get in touch with person-to-person extend and by animals [4,7]

E.coli and *Salmonella* could be present in the gastrointestinal tract of farm animals then discard in faeces [8]

Beef carcasses can be converted into polluted with these pathogens when faecal substance from the gastrointestinal tract or hide is transmited to the carcass through evisceration and hide removal [9].

The surfaces of raw meat can be contaminated After the slaughtering process, with *Salmonella* and VTEC as well as further pathogens like *Clostridium perfringens*, *Staphylococcus aureus* , *Listeria monocytogene* sand *Campylobacter*, [10] .

Generally microbial contamination present on the uncovered surface of meat that whole cuts, while minced meat, microorganisms wick are on the shell of the meat be converted into mixed all through. therefor should be cooked carefully about minced meat .. Cooking minced beef to a core temperature of 75°C or equal (e.g. 70°C for two minutes) is recommended [11,12].

Studies going to the quality of microbiology of food apperthat minced meat is a good medium in nutrients necessary for the pathogenic microorganisms growth [13,14]

Microbiological value of meat is resolute by the quality of hygien. High levels of microorganisms counting pathogens are disturbing on the quality of hygien for meat. Loss of hygienic food handling lead to pathogenic contamination of food [15].

Microorganisms introduced as of decreases of sanitation in equipment, slaughtering location , operators' hands and environmental exposure contaminate the meat product[16,17]

Customers of food also include a connection in the series of food-borne disease with unsuitable cooking and storage of meat products and meat [16,17,15].

Intoxications and infection by Food-borne can happen due to the found of some bacteria (e.g. *Salmonella*, *S. aureus*, *E. coli* spp. [13,15] .

Therefore, recognition and detection of food pathogenic bacteria also evaluate the general microbial load of the food is of large importance[17].

To evaluate the hygienic quality of foods and the probable presence of pathogens the indicator bacteria is an ordinary practice. *E. coli* find total viable counts, coliform bacteria and total number of bacteria may, reflect the hygienic quality of food. *E. coli* is most frequently the contaminating organism, and is commonly a reliable indicator of fecal contamination in milk, food, water, meat and products [18].

The purpose of this study is to detection of *E. coli*, as a contaminant of minced meat in certain locations of Baqubah city in Diyala Province wick obtained from local butcher shops

2. Material and Method

2.1. Sample collection and preparation

From November 2015 to May 2016 ninty minced meat samples were purchased collected from randomly selected from various butcher shops from different areas of Bagubah city in Diyala Province, Iraq. and were examined microbiologically to determine their microbiological quality and eligibility for safe consumption.

All the samples were transported to laboratory by placed in separate sterile plastic bags to prevent cross contamination and stored at 4 °C for a maximum of 24 h until they were analyzed.

2.2. Microbiological analysis.

The parameters of study were determination of counts of indicator bacteria (total coliform, *E.coli*) Twenty-five g of each sample were homogenized in 225 ml distal water and Preparation serial dilution (10^{-1} , 10^{-2} , 10^{-3} , 10^{-4}) then transfers (1)ml. or (0.1)ml. of each dilution

into serial plate then. incubated at 37 °C for 24h.

Then the enrichment samples were pour onto MacConkey Agar and then streak onto eosin methylene blue agar(EMB) plates . (HiMedia, India).[19].

Coliforms and *E. coli* were grown on MacConkey agar and incubated under aerobic conditions at 37°C for 24–48 h. Colonies were visualized by using colony counter (Stuart, UK) and counted (pink colony). Identification of the colonies were performed by using selective media to allow growing colony called EMB(eosin methylene blue agar).[20,21].

E.coli appear on EMB agar as (blue-black colony with a greenish metallic sheen) Results were calculated as CFU per gram. Stock cultures were stored at -20 °C until use.

Statistical analysis had been done with Microsoft Excel software package 2013.

3. Results

Table 1 show the prevalence of *coliform bacteria*. isolated from minced beef meat

in different areas of Bagubah city in Diyala Province,Iraq. In this study, 80 of 90 samples (88.88%) positive,(11.12%) negative.

were found to be contaminated with *coliform bacteria*. The highest prevalence of *coliform bacteria*. was found in (**Hay al-mualmeen , 7-nissan, Al-tahreer**) for each area (11.11%), followed by(**Al-mufraq, Al-markas, Al-rahmmah**) for each area (10%),(**Shiftah, Buhris**)for each area (8.88%), and (**Baqubah-al-jadidah**) area (7.77%).

The highest average coliform plate count (CPC)(log CFU/g) when pour onto levine violet red bile agar was found in(**7-nissan**) 125 log CFU/g followed by,(**Hay al-mualmeen**)115 log CFU/g,(**Al-markas**)32 log CFU/g,(**Al-mufraq**)28 log CFU/g,(**Al-tahreer**)25 log CFU/g,(**Al-rahmmah**)23 log CFU/g,(**Shiftah**) 21 log CFU/g,(**Buhris**) 19 log CFU/g, and (**Baqubah-al-jadidah**)14 log CFU/g. respectively.

Table 1. Prevalence Of coliform Bacteria Isolated From Beef Minced Meat.

	Area	No.of samples	No. (%) of positive <i>coliform</i> samples	average coliform plate count (CPC)(log CFU/g)
1	Hay al-mualmeen	10	10(11.11)	115
2	7-nissan	10	10(11.11)	125
3	Al-mufraq	10	9(10)	28
4	Al-markas	10	9(10)	32
5	Al-tahreer	10	10(11.11)	25
6	Al-rahmmah	10	9(10)	23
7	Shiftah	10	8(8.88)	21
8	Baqubah-al-jadidah	10	7(7.77)	14
9	Buhris	10	8(8.88)	19
	Total	90	80(88.88)	

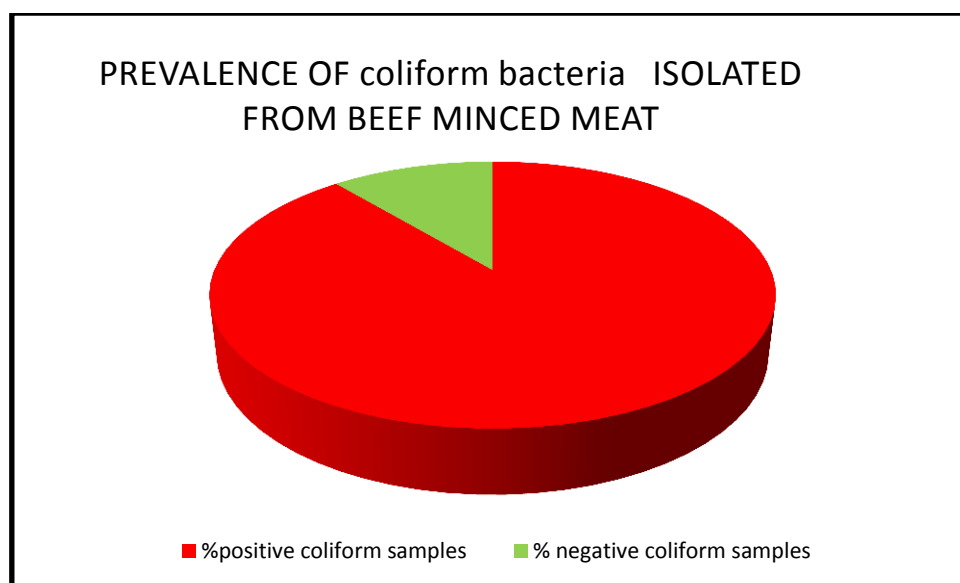


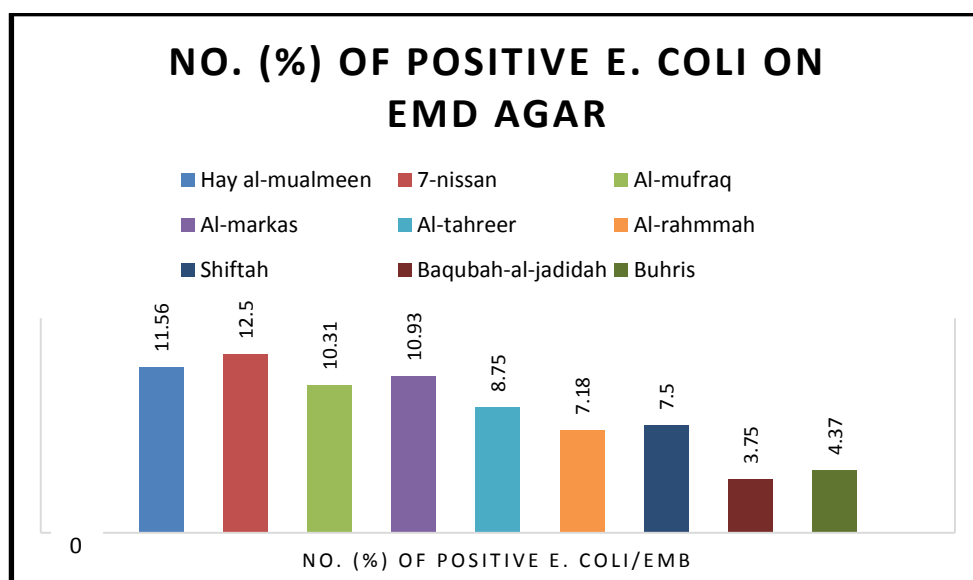
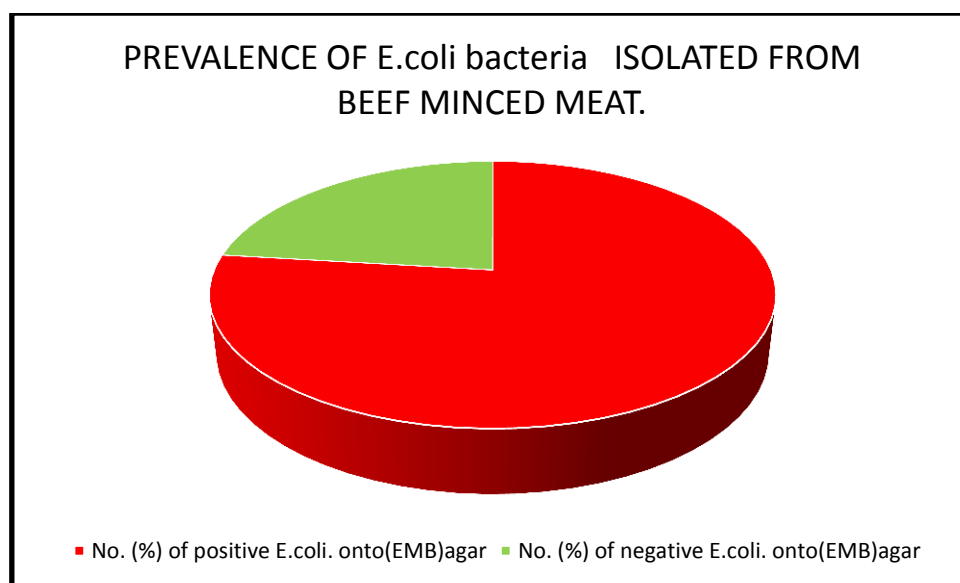
Table 2 show the prevalence of *E.coli*. isolated from minced beef meat in different areas of Bagubah city in Diyala Province,Iraq. In this study 80 contained coliform bacteria 76 contained *Ecoli*, (95%) positive,(5%) negative.

The results was shown presence of high contamination ratio of the microbial isolation from minced beef meat of *E.coli* bacteria in different areas of Bagubah city in Diyala Province,Iraq. with positive (76.85%),while negative (23.1%).

The highest prevalence of *E.coli* bacteria,when streak onto eosin methylene blue agar(EMB) plates with serial dilution (10^{-1} , 10^{-2} , 10^{-3} , 10^{-4}) . was found in (**Hay al-mualmeen**) area(11.56%) followed by (**7-nissan**) area (12.5%),(**Al-mufraq**) (10.31%), (**Al-markas**) (10.93%) ,(**Al-tahreer**) (8.75%), (**Al-rahmmah**) (7.18%),(**Shiftah**) (7.5%),(**Baqubah-al-jadidah**) (3.75%) and(**Buhris**) area (4.37%)respectively.

Table 2. Prevalence Of *E.Coli* Bacteria Isolated From Beef Minced Meat.

	Area	No. of samples	No. (%) of positive <i>E.coli</i> samples	No. (%) of positive <i>E.coli</i> . onto(EMB)agar	No. (%) of negative <i>E.coli</i> . onto(EMB)agar
1	Hay al-mualmeen	10	10(12.5)	37(11.56)	3(0.93)
2	7-nissan	10	10(12.5)	40(12.5)	0(0)
3	Al-mufraq	9	9(11.25)	33(10.31)	3(0.93)
4	Al-markas	9	9(11.25)	35(10.93)	1(0.31)
5	Al-tahreer	10	10(12.5)	28(8.75)	12(3.75)
6	Al-rahmmah	9	9(11.25)	23(7.18)	13(4.06)
7	Shiftah	8	8(10)	24(7.5)	8(2.5)
8	Baqubah-al-jadidah	7	5(6.25)	12(3.75)	16(5)
9	Buhris	8	6(7.5)	14(4.37)	18(5.62)
	Total	80	76(95)	246(76.85)	74(23.1)



4. Discussion

Minced beef meat is an exceptional medium for growth microorganism. normally The microorganisms come across on surface of meat are spread thoroughly and start replicate into the meat product when the situation are approving during packaging, , storing , mixing and grounding, lead to lack of product quality and creating potential health hazards [22,20]

. The variation among the results may be due to the variation of the quality of hygienic situation in the processing plant, climate, the variation in sampling locations, the number of sampling ,analysis methods employed,. presence of an elevated number of bacteria, an indicator of the probable shelf life of meat, raises the probability in a short time occur meat spoilage [23]

In different parts of the world has been broadly reported presence of bacteria in meat [24,25]. Some groups documented the presence of bacteria, especially Gram-negative , as an indication of open-air meat spoilage [26]

High counts of bacteria and the presence of possible pathogens on the floor and walls of shops, meat-processing equipment, , represent their environmental hygiene status. The presence of pathogenic bacteria in meat-processing equipment and linked surfaces may supply to the contamination of meat. it has been confirmed that mincing meat with dirty equipment considerably raise level of contamination level in minced meats as compare to that in complete carcasses [27]

On the other hand, pathogens of food-borne which can distribute from contaminated meat

to such surfaces[28] can extend infectivity in the community.

There for ; in this study The highest average coliform plate count (CPC)(log CFU/g) when pour onto levine violet red bile agar was found in(7-nissan) 125 log CFU/g followed by,(Hay al-mualmeen)115 log CFU/g,(Al-markas)32 log CFU/g,(Al-mufraq)28 log CFU/g,(Al-tahreer)25 log CFU/g,(Al-rahmmah)23 log CFU/g,(Shiftah) 21 log CFU/g,(Buhris) 19 log CFU/g, and (Baqubah-al-jadidah)14 log CFU/g.respectively;

The highest prevalence of *E.coli* bacteria when streak onto eosin methylene blue agar(EMB) plates with serial dilution (10^{-1} , 10^{-2} , 10^{-3} , 10^{-4}). was found in (Hay al-mualmeen) area(11.56%) followed by(7-nissan) area (12.5%),(Al-mufraq) (10.31%),(Al-markas) (10.93%) ,(Al-tahreer) (8.75%), (Al-rahmmah)(7.18%),(Shiftah) (7.5%),(Baqubah-al-jadidah) (3.75%) and(Buhris) area (4.37%)respectively., as an indication of open-air meat spoilage [26]

The results was shown presence of high contamination ratio of the microbial isolation from minced beef meat of *E.coli* bacteria in different areas of Bagubah city in Diyala Province,Iraq. with positive (76.85%),while negative (23.1%) [27,28]

The recommended temperature or storage that used for chilled foods, such as raw meat, is 5oC or less because low temperatures slow down or prevent , the growth of microorganisms that pathogens and spoilage of meat[11,29]

Raw minced beef and should be completely cooked before consuming (to a core temperature of 75oC, or an equal time/temperature combination).is recommended [11,12].

As a result, the most significant practice that should be careful in animal slaughtering are cleaning dirty animals before slaughtering, skinning while being on the rail, separating carcasses from each other and keep away from contact between the carcasses and external surface of the hide. Hygiene procedures must be enough to prevent contamination from clothing, mincing equipments, saws, knives and hands.[30]

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