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Detection of bacterial and fungal contamination unsalted and salated cheese in markets of Samara city-Iraq

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Abstract---This study was conducted on 60 samples of local dairy products from fresh unsalted white sheep cheese, locally manufactured pasteurized and non-pasteurized, as well as salted cheese found in the local markets of Samarra city. For the period from January to March 2021, which microbial tests were carried out, included the total bacterial number, the total count of Coliform, and the total number of molds. Research results showed a high content of unsalted cheese from the Microorganisms that included a number of human pathogens and food poisoning triggers. The results in this study showed the emergence of five different types of bacteria, namely Salmonella, Escherichia coli, Shigella, Brucella and Staphylococcus aureus. The highest numbers, ranging from $(7000) \times 10^{-3}$ in unsalted cheese samples, were significantly higher than salted cheese samples, with the highest percentage $(300) \times 10^{-3}$ for fungi. These species appeared in local products studied and were more often in unsalted and non-pasteurized cheese samples. Also, studied allergic types for Antibiotic.

Keywords---bacterial, fungal contamination, salted cheese, markets.

Introduction

The cheese considered as a food like a medium that is ideal for the growth of many microorganisms. It is suitable for moisture and contains many compounds, and nitrogen, as well as the availability of densities of different millimeters and minerals that make it a viable environment for the bacteria and fungi, which plays a significant role in making living and chemical changes that determine the quality and potency of the cheese [1]. Cheese and cream are one of the most highly sought-after foods in all cities of Iraq. Local mild cheese and local cream are mainly produced by animal breeders or by agricultural and sold directly to the individuals. Therefore, it is expected that the necessary sanitary conditions will

not be applied in production and exposure to various sources of pollution, starting from marketing and storage and then going to the consumer. Because the products are damaging fast, due to being suitable for micro-growth and the increase in spreading infections from bacteria and fungi, which eventually leads to human poison [2].

Microbes cause a lot of economic and health damage. The health damage is due to the food being suitable for the growth of micro-diseases and their proliferation on bacteria and fungi, which eventually lead to human poisoning [3]. The method of examining the cheese qualitatively in microbial terms is based on the determination of the type of cheese, such as cheese made from raw milk, pasteurized, thermally labelled, cooked, etc., because this will determine the method or protocol that is performed detection of germs, as well as, the type of quality table of standard specifications to be used to judge the checked sample. The qualitative macrobiotic value of milk is influenced by the amount of natural fluorine spores, as well as by productive conditions such as pollution after thermal coefficients [4].

A major problem in the manufacture of cheese in general is the use of fresh milk, which has not been treated for high temperature of the pasteurized, as well as the weak side of tools sterilization that used in the manufacturing process. Therefore, the presence of colon bacteria (*coliform*) and *E. coli* spores in milk and milk products is a fecal indicator of milk contamination as a result of weak management and failure to comply with the sanitary requirements of the regions where the cheese is manufactured. The isolation and diagnosis of colon spores in food is one of the most common criteria for assessing and judging food as a substance that allows consumption based on national standards and standards. *Salmonella* and *Shigella* are not fecal contamination spores because of their pathological severity when found in food in general [5].

Cooking temperature plays an important role in determining the quality of cooked cheese. Cooking heat changes randomly distributed Cazine jelly into a homogeneous liquid. It also has a significant impact on the structure of cooked cheese. In cookery cheese, the optimal cooking temperature is (90-95 c) [6]. Raw milk is also the basis for the manufacture of cheese after it has been converted into local white cheese and then into cooked cheese, so the quality of raw milk is affected by the factors in the animal environment and the milking place. Hence, the importance of care about raw milk, which in one way or other results in the contamination of cooked cheese by microscopy, as well as other factors that lead to the contamination of cooked cheese [7].

The demand for dairy has increased and domestic production has become even though its increase is not sufficient to reach the daily needs of the cheese population. The traditional methods of manual milking are still used by some small workshops and rural houses that are engaged in the production of cheese. Most of them are often not subjected to any heat treatment sufficient to eliminate the pathogenic microbiome. So, it's an appropriate medium for microbiome growth and reproduction. There for, cheese is an important source of foods poisoning,as it is used in the manufacture of many foods, which leads to poisoning by many types of microbes, including *listeria monocytogenes*, which

causes meningitis and septicemia and can lead to death. Also, cheese poisoning can develop into severe chronic diseases, such as chronic arthritis, *Brucella melitensis*, which causes ripple fever, or Malta fever, which are severe diseases that can survive for a long time and cause bowel inflammation and *Yersinia enterocolitica*, the fact that local white soft cheese is a food that is highly consumed in Samarra and is marketed in unhealthy ways in shops and on streets [4].

Objective of the study

- Insulation of bacterial and fungal pathogens from salted and unsalted local white cheese and estimation of the percentage of microbes in cheese samples.
- Cheese is better at eating for human health.

Methods

Sample collection

Collected 60 samples of different cheese as shown in table 1. (1-35) samples were of sweet white cheese and (36-50) a sample were of salted soft cheese and from (50-60) a sample of sweetener cheese. Most of samples were taken from local markets and street vendors in Samarra city for three months. (January, December, March) from 2021, taking a note that most of the samples have not been sterilized, pasteurized and have no good material.

Isolation

To prepare the cheese emulsion, we weighed (1) grams of the sample and moved it after cutting it with sterile blade and then placed it in a test tube containing (9) ml of sterile distilled water. A series of decimal assays were then made for the purpose of conducting the total census of bacteria and then transported 0.1ml and implanted into the elected farming community to isolate the contaminated microbes of the specimen [8].

Culture media

The following media as communities were used and prepared by following the company instructions equipped. The media were prepared using Autoclave, executed at 121 c, 1.5 bar pressure for 15 minutes. The molds (fung) were isolated using the medium of potatoes and PDA(potato dextrose agar) using the method of pouring dishes and hugging dishes at 25 C and for five days.

- The medium of Nutrient agar was used to calculate the total number of bacteria.
- *Brucella* agar.
- Amalgamated to calculate the total number of colon bacteria.
- Eosin methylene blue (EMB) agar to diagnose *E. coli*.
- *Salmonella and Shigella*.

Antibiotic disks

Table 1
Shows the antibiotic tablets used in the study

| No | Antibiotic | Symbol | Disc Concentration | Company |
|----|----------------|--------|--------------------|-------------|
| 1 | Penicillin | P | 10 | Bio-analyse |
| 2 | Cefaclor | CEC | 30 | Bio-analyse |
| 3 | Gentamicin | CN | 10 | Bio-analyse |
| 4 | Amlkacin | AK | 30 | Bio-analyse |
| 5 | Cefotoaxime | CTX | 30 | Bio-analyse |
| 6 | Oxyteracycline | T | 30 | Bio-analyse |
| 7 | Vancomycin | VA | 10 | Bio-analyse |

Results and Discussion

Table 2 shows that the total number of bacteria in (60) local cooked cheese samples from different regions and street vendors in Samarra city were ranged from (0-4000) Cell/g. Thus, a number of these samples showed more than one type of negative and positive gram bacteria as in figure (1) (2). The difference in the total numbers of bacteria of local milk is under study, primarily due to the different temperature of cooking when using thermal coefficients, reduces the total number of bacteria [10]. An increase was observed in the total number of bacteria in samples for local non-pasteurized cheese (35) All are generally and especially taken from itinerant vendors if they range from 22-400 x 10, due to the high preparation method in the countryside, as well as the fact that those cheese are marketed in conditions that are far from healthy as well as exposed to air, are exposed on sidewalks and roads, thus increasing the content of the total number of bacteria [11].

The high total number of bacteria is also due to the use of substances such as Starch, which reduces the impact of cooking heat and then makes it difficult to get rid of the bacteria in the cheese, the results are similar to what it found by [12]. Total numbers of colon bacteria between 0-300 cell/g as shown in figure (3) (4) Colon bacteria have appeared for samples (for non-pasteurized sweet cheese) Specimens for some pubescent cheese where this rise in colon bacteria numbers is due to the lack of proper sanitary conditions in terms of the cleanliness of work and workers, as well as the type, cover and life of the packaging, and for some salty cheese samples where colon bacteria appear [13]. If a change in the color of the medium is observed, the McConkey used to grow the *E. coli* bacteria becomes the color of the medium pink, which gives an indication of the *E. coli* colon's characteristic of lactose sugar fermentation [14]. It is also contaminated by manufacture and not in the shops, as well as the lack of access to the appropriate heat specified by Iraqi standard [15].

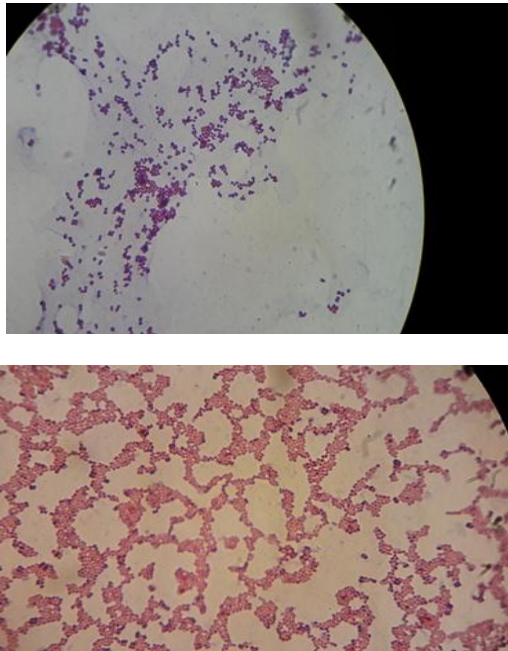


Figure 1. Gram Positive Bacteria

The results were close to those of [16] [17]. Saline cheese samples had adverse results in total coliform and *E. coli*, samples containing salts were recorded. (Salted cheese) The lowest number in the total count of coliform, and *E. coli* in the cheese, and these samples run counter to the paragraphs of the Iraqi standard that stipulated the cooked cheese should be completely free of these bacteria. Colon bacteria used by community health microbiologists are an indicator of fecal contamination of both water and food sources [15].

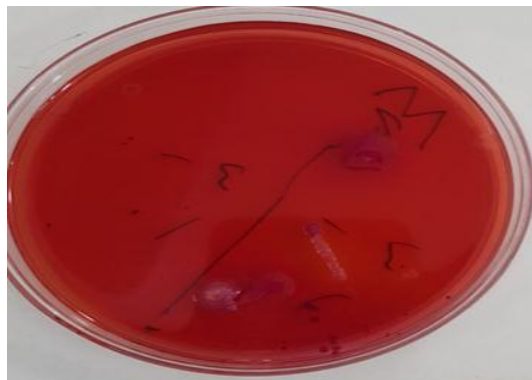
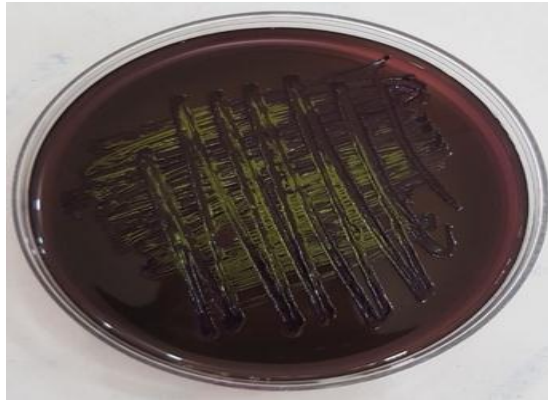


Figure 2. *E. coli* (gram negative bacteria) on McConkey Agar

Figure 3. *E. coli* on EMB

Brusilla was also isolated after incubations ranging from 2-4 days on the middle on the optional brucilla acar at 37 °C. They appeared as small, conical colonies with pale yellow and could become brown after a longer time, with the species belonging to the genus *Brucella* having the advantage of being for a short neurotransmitter ranging in length. (1.5-0.6) micrometers between (0.5 and 0.7) micrometers wide, rarely seen in the form of pairs, short chains or small clusters not coated with a wallet and not composed of spores. The contamination of cheese with may be due, inter alia, to the release of this bacterium in the milk of the infected animal, which causes it to contaminate the cheese made from it, as well as the hands of the workers [18]. There are many studies that emphasize the importance of milk and milk products, particularly white cheese, as one of the main causes of in the spring, especially since it is most common in causing disease [19]. As identified, the *salmonella spp* as well as the *shigella* by choosing its medium matched the epithelial tests for recognition. Also the yeast and molds ranged in numbers from. (0-100) a cell/g, and mostly were found *Alternaria alternate*, *Aspergillus falvius* *Penicillium italicum*, [2].

Table 2

Shows the Numbers of pathogenic microbes isolated from salted and unsalted local cheese samples

| No | Total No of Yeast and Molds | Total No of Colon Bacteria Cell\g 10 ³ - | Bacteria Total Count Cell\g 10 ³ - |
|----|-----------------------------|---|---|
| 1. | 1 | 0 | 230 |
| 2. | 0 | 0 | 220 |
| 3. | 0 | 0 | 620 |
| 4. | 0 | 0 | 640 |
| 5. | 0 | 160 | 600 |
| 6. | 10 | 80 | 420 |
| 7. | 12 | 60 | 350 |
| 8. | 10 | 30 | 330 |

| | | | |
|-----|----|-----|------|
| 9. | 21 | 60 | 220 |
| 10. | 11 | 250 | 3200 |
| 11. | 10 | 300 | 1600 |
| 12. | 31 | 150 | 300 |
| 13. | 27 | 220 | 7220 |
| 14. | 3 | 100 | 600 |
| 15. | 52 | 60 | 1500 |
| 16. | 1 | 80 | 630 |
| 17. | 0 | 20 | 520 |
| 18. | 38 | 130 | 1000 |
| 19. | 7 | 140 | 2000 |
| 20. | 0 | 60 | 220 |
| 21. | 0 | 30 | 1300 |
| 22. | 0 | 20 | 1000 |
| 23. | 2 | 180 | 1500 |
| 24. | 0 | 200 | 700 |
| 25. | 4 | 140 | 880 |
| 26. | 1 | 220 | 470 |
| 27. | 1 | 160 | 580 |
| 28. | 1 | 300 | 650 |
| 29. | 23 | 210 | 3300 |
| 30. | 21 | 130 | 3310 |
| 31. | 2 | 110 | 4000 |
| 32. | 3 | 120 | 2200 |
| 33. | 2 | 140 | 1370 |
| 34. | 3 | 0 | 4000 |
| 35. | 0 | 0 | 2800 |
| 36. | 2 | 20 | 120 |
| 37. | 2 | 20 | 220 |
| 38. | 1 | 0 | 300 |
| 39. | 0 | 0 | 20 |
| 40. | 2 | 0 | 10 |
| 41. | 0 | 0 | 20 |
| 42. | 1 | 0 | 40 |
| 43. | 2 | 0 | 60 |
| 44. | 0 | 30 | 0 |
| 45. | 0 | 40 | 120 |
| 46. | 0 | 0 | 0 |
| 47. | 0 | 0 | 130 |
| 48. | 1 | 0 | 110 |
| 49. | 0 | 0 | 160 |
| 50. | 0 | 60 | 20 |

| | | | |
|-----|---|----|-----|
| 51. | 3 | 20 | 100 |
| 52. | 6 | 30 | 120 |
| 53. | 4 | 20 | 220 |
| 54. | 0 | 0 | 130 |
| 55. | 1 | 0 | 200 |
| 56. | 0 | 0 | 50 |
| 57. | 2 | 0 | 20 |
| 58. | 1 | 0 | 20 |
| 59. | 1 | 0 | 100 |
| 60. | 0 | 0 | 50 |

Antibiotic sensitivity test

Table 3 shows the results of the antibacterial sensitivity test, where cheese isolates showed antibiotic resistance to almost all antibiotics. *Escherichiacoli* recorded antibiotic resistance. (amalcasin) depending on its effectiveness as well as cefotaxin to inhibit bacterial cell wall protein synthesis [20]. When it showed inhibition as in form 5, the *Brucella* resisted all but the antibiotic. (Vancomycin) was also the diameter of the obvious inhibition of the antibiotic Salmonella bacteria (Amlkacin) The rest of the species did not show any inhibition of bacterial species as in figure (5).

Table 3
Sensitivity Test Results

| No | Antibiotic | Symbol | Bacteria | Disc Conc | Inhibition |
|-----|--------------|--------|------------|-----------|------------|
| 1. | Cefaclor | Cef | Brucelle | 30 | 0 |
| 2. | Amikacin | AK | = | 30 | 1mm |
| 3. | Vancomycin | Av | = | 10 | 0 |
| 4. | Ceftaxin | c.tx | = | 30 | 0 |
| 5. | Penicillin | P | Shigella | 10 | 0 |
| 6. | Ceffo taxime | Ctx | E. coli | 30 | 0 |
| 7. | Amlkacin | Ak | E. coli | 30 | 1mm |
| 8. | Amlkacin | Ak | Salmonella | 30 | 2mm |
| 9. | Vancomycin | Va | Shigella | 10 | 0 |
| 10. | Penicillin | P | Salmonella | 10 | 0 |
| 11. | Penicillin | Cn | Shigella | 10 | 0 |
| 12. | Cefaclor | P | Shigella | 10 | 0 |
| 13. | Penicillin | P | E. coli | 10 | 0 |

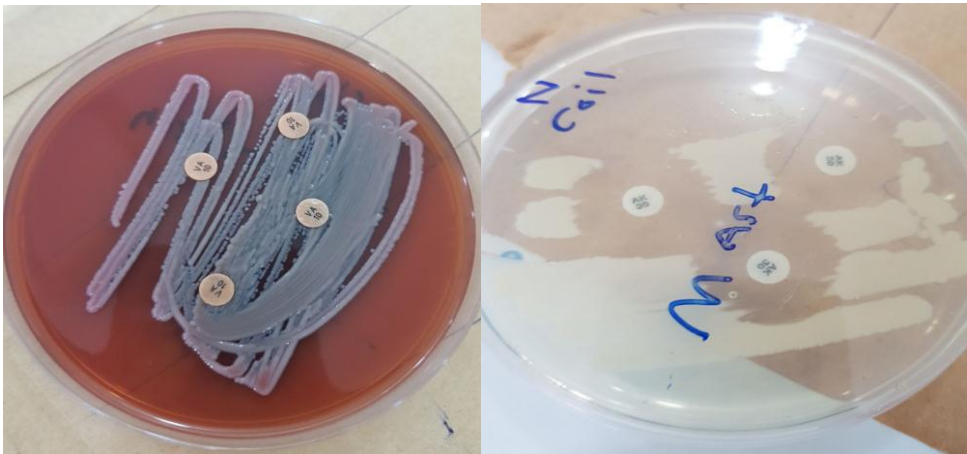


Figure 4. Shows the Inhibition of the Antibiotic on *E.coli*



Figure 5. Bacteria Resistance to Antibiotics

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