

İHSAN EROL ÖZÇİL

**A RESEARCH ON OCCURRENCE OF *SALMONELLA* AND
STAPHYLOCOCCUS AUREUS IN HALLOUMI CHEESE PRODUCED IN
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NEU, 2016

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**A THESIS SUBMITTED TO THE GRADUTE
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OF
NEAR EAST UNIVERSITY**

By

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**In Partial Fulfillment of the Requirements for the
Degree of Master of Science
in
Food Engineering**

NICOSIA, 2016

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ABSTRACT

In this study, existence of enterotoxigenic *Staphylococcus aureus* (*S.aureus*) and *Salmonella* which is common cause of food borne illness are investigated in halloumi cheese that are produced in various plants of Turkish Republic of Northern Cyprus (TRNC). Baird-Parker Agar solid medium has been used for the isolation of *S.aureus* and MacConkey, SS agar solid medium has been used for the isolation of *Salmonella*.

Total 34 halloumi cheese samples produced in different regions were collected from the various markets in Nicosia (Lefkoşa). Whereas no *Salmonella* was observed in any sample, 2 out of 34 samples were containing *Staphylococcus aureus*. However number of *S. aureus* colonies were lower than risk levels according to Turkish Food Codex.

Keywords: *Salmonella*; *Staphylococcus Aureus*; Enterotoxin; Halloumi Cheese; Northern Cyprus

ÖZET

Bu çalışmada, Kuzey Kıbrıs Türk Cumhuriyetinde farklı işletmelerde üretilip satışa sunulan çeşitli hellim peynirlerinde *Staphylococcus aureus* (*S.aureus*) ve *Salmonella* varlığı araştırılmıştır. *S.aureus* izolasyonu için Braid-Parker (BPA) katı besiyeri, *Salmonella* izalasyonu için ise Mc Conkey ve SS agar katı besiyeri kullanılmıştır.

K.K.T.C’de farklı işletmelerde üretilen ve Lefkoşa bölgesi marketlerinde satışa sunulan 34 adet hellim peyniri ürünü örneği analiz edilmiştir. İncelenen 34 hellim peyniri örneğinde *Salmonella* bulunmadığı; 34 adet hellim peynirinin iki tanesinde saptanan *S.aureus* miktarının Türk Gıda Kodeksi Mikrobiyolojik kriterler tebliğinde belirtilen sınırlar içerisinde olduğu, halk sağlığını riske atmadığı ve diğer 32 örneğin ise *S.aureus* içermediği saptanmıştır.

Anahtar Kelimeler: Salmonella; Stafilokok; Enterotoksin; Hellim Peyniri; Kuzey Kıbrıs

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LIST OF SYMBOLS AND ABBREVIATIONS

AD	Atopic Dermatitis
α	Alfa
ATP	Adenosine Triphosphate
a_w	Water Activity
BPW	Buffered Peptone Water
BPA	Baird Parker Agar
°C	Degrees Celsius
CE	Cementum
CFU	Colony-Forming Units
cm	Centimeter
CN	Cyanide
γ	Delta
DNA	Deoxyribonucleic Acid
DNase	Deoxyribonuclease
DT	Definitive Type
E.Coli	Escherichia coli
°F	Degrees Fahrenheit
FDA	Food and Drug Administration
g	Gram
g/L	Gram/Liter
G-C	Guanine and Cytosine
h	Hour
H antigen	The Antigen That Occurs In The FLAGELLA of Motile Bacteria
HIV	Human Immunodeficiency Virus
HPLC	High-Performance Liquid Chromatography
IDF	International Dairy Federation
kcal	Kilo Calorie
kg	Kilogram
kob/g	Colony/Gram
kob/L	Colony/Liter
L	Liter

LPL	Lipoprotein Lipase
LPS	Lipopolysaccharide
µg	Microgram
mg	Milligram
min	Minute
mL	Milliliter
MLVA	Multiple-Locus Variable
µm	Micrometre
mm	Millimeter
mol.	Mole
MRSA	Methylllysine Resistant Staphylococcus aureus
NaCl	Sodium chloride
Ng	Nano gram
NKTTn	Muller-Kauffmann Tetrathionate / novabiocin
O antigen	The Antigen That Occurs In The Bodies of Bacteria
PCR	Polymerase Chain Reaction
PFGE	<i>Pulsed-Field Gel Electrophoresis</i>
pg	Pico gram
pH	Power of Hydrogen
PS	Psoriasis
RIA	Radioimmunoassay
RNA	Ribonucleic Acid
RVS	Rappaport-Vassiliadis Soya
S.aureus	Staphylococcus Aureus
SE	Staphylococcal enterotoxins
sec	Second
SS agar	Salmonella Shigella Agar
TRNC	Turkish Republic of Northern Cyprus
UHT	Ultra-High Temperature
UV	Ultraviolet
VSAs	Volatile Fatty Acids
WHO	World Health Organization
%	Percent

CHAPTER 1

INTRODUCTION

An unripened semi hard and brined cheese, known especially as Halloumi / Hellim as a Cypriot brand, produced generally through a mixture of milk of goat and sheep or sometimes the cow (Ayto & John, 1990; Gibbs et al., 2004). Due to its special texture formed thanks to its high melting point, this cheese can either be grilled or fried. Helloumi is differentiated to other kinds of cheese with the preparation method that includes rennin, consisting either no acid or bacterium that produces acid (Ayto & John, 1990; Gibbs et al., 2004). *Halloumi* cheese is the traditional cheese of Cyprus, holding the traditional title of Cyprus, Halloumi is also consumed widely in countries like Jordan, Lebanon and also Turkey (Allen & Gary, 2007). Originally found during the Medieval Byzantine era (AD 395-1191) then later became a popular product within the Middle East. With no produced pigmented colonies *Salmonella* spp. is a non sporing rods, gram negative, catalase negative and oxidase negative (Quinlan et al., 2000). It is a pathogen with one of the most repetitive frequency rate in the industry of food. Being one of the mostly encountered base for the foodborne diseases, especially in dairy products, the historical roots to this microorganism reaches a century. Accordingly most of the species found within are pathogenic (Kasrazadeh & Genigeorgis, 1994; ICMSF, 2006; De W Blackburn & McClure, 2009). Annual cases for thyfoid fever reaches upto 16 million as gastroenteritis cases reaches up to 1.3 billion and finally *Salmonella* death rate in the world reached upto 3 million deaths (Bhunias, 2008). In short, *Salmonella* can be described as a bacterium sized approximately 2-3 x 0.4-0.6 μm and which is rod shaped gram negative flagrated facultative anaerobe (Montville & Matthews, 2008). Futhermore the *Staphylococcus aureus* is a oxidase negative, catalase positive and Gram-positive cocci. It forms clusters showing pigmented colonies when grown in nutrient agar. Clumps, rather characteristic, can be seen bunches of grapes (Quinlan et al., 2000). *Staphylococcal* food poisoning also known as *Staphylococcal* intoxication is due to the ingestion of enterotoxins formed when *S. aureus* growth in foods. Human origin consists of Enterotoxin production as the most common *S. Aureus* which holds sound correlation to the enzyme coagulase production (Sutherland & Varnam, 2002).

1.1 Historical Background of Halloumi Cheese

According to the historical legends, the halloumi cheese supposedly was first introduced to the Cyprus island by the Syrian and Palestinian mercenaries in the time of Frankish autonomy (AD 1192-1489), eventually becoming the traditional taste of Cyprus. The earliest recorded mentioning of this cheese reaches back to 1554 as the Italian author Florio Bustron describing the manufacturing of cheese made from the mixture of both sheep's and goat's milk (Papademas, 2006). The production of this cheese has accumatively increased initially and notably in 1999 to 4730 tons and 6600 tons by the year 2002. This is thanks to the growing reputation of the halloumi cheese exceeding the borders of Cyprus.

The halloumi cheese has exceeded the export ratio of the highest ranking wine in the years of 2001 and 2002 been accountable for the 27% of the total agricultural exports of Cyprus in 2004 accordingly to Gibbs and Morphitou. Halloumi cheese was export statistics from Cyprus to the UK 972 tons, Greece 601 tons, Germany 430 tons, Kuwait 401 tons, UAE 364 tons and Saudi Arabia 265 tons, in 2003. The statistics for the years of 1999 to 2003 also indicates that total export for the halloumi cheese have increased from 2522 to 3976 tons, this number is expected to increase (Papademas, 2006). An application was made to European Union by the Cypriot authorities to hold the labeling for the halloumi cheese under Protected Designation of Origin (PDO). Thus with this execution the production of the halloumi cheese in other countries than Cyprus will be prohibited. In this respect, no cheese similar to halloumi will be produced or presented to market as halloumi. Despite the fact that Halloumi is not yet designated, it is labeled as product of Cyprus both in USA and EU.

Collective trademarks were granted to Halloumi in USA, Greece and EU in the respective years of 1999, 2000 and 2002. Furthermore, more markets are on the way both in Canada and Brasil (Gibbs et al., 2004; Papademas, 2006).

1.2 Chemical Composition of Halloumi Milk

In gross chemical compositions, there are observable differences in chemicals. The rate of fat, protein and minerals differ between the caprine and bovine milks. (Table 1) shows the chemical composition of various milks used in the manufacturing of the Halloumi cheese.

Table 1.1: Per g/100g Ratio is Used for Different Milks Used in Halloumi Production
(Adapted from Papademas, 2006)

Constituent	Sheep's Milk	Goat's Milk	Cow's Milk
Total solids	16.84	13.22	11.33
Fat	6.20	4.33	3.34
Protein	5.50	3.75	2.86
Calcium	0.18	0.11	0.11
Ash Casein	0.90	0.83	0.77

The different level of carotene content, normally higher, in the cow's milk results in color difference (more yellowish) in halloumi cheese production compared to the one produced with either sheep's or goat's milk, which is evident enough for the consumer to note the difference (Jandal, 1996). The odors are also vary compared to sheep and goat milk to cow's. This is also due to the fatty acid content of each milk type. Caproic (C6:0), Caprylic (C8:0), Capric (C10:0) and Lauric (C12:0) fatty acids are evidently higher in sheep's and goat's milk compared to cow's. These differences in flavours, resulted due to the levels of fatty acids, priorities the consuming selection of ovine and caprine milks (Park et al., 2007).

1.3 Production Stages of Halloumi Cheeses

The Halloumi cheese production vary in cooking time, 30 minutes and 60 minutes, of sheep's milk. The cooking temperature for the cheese curd is around 93–95 °C in non-protein whey (Sheep & Milk, 1983; Galanakis et al., 2014).

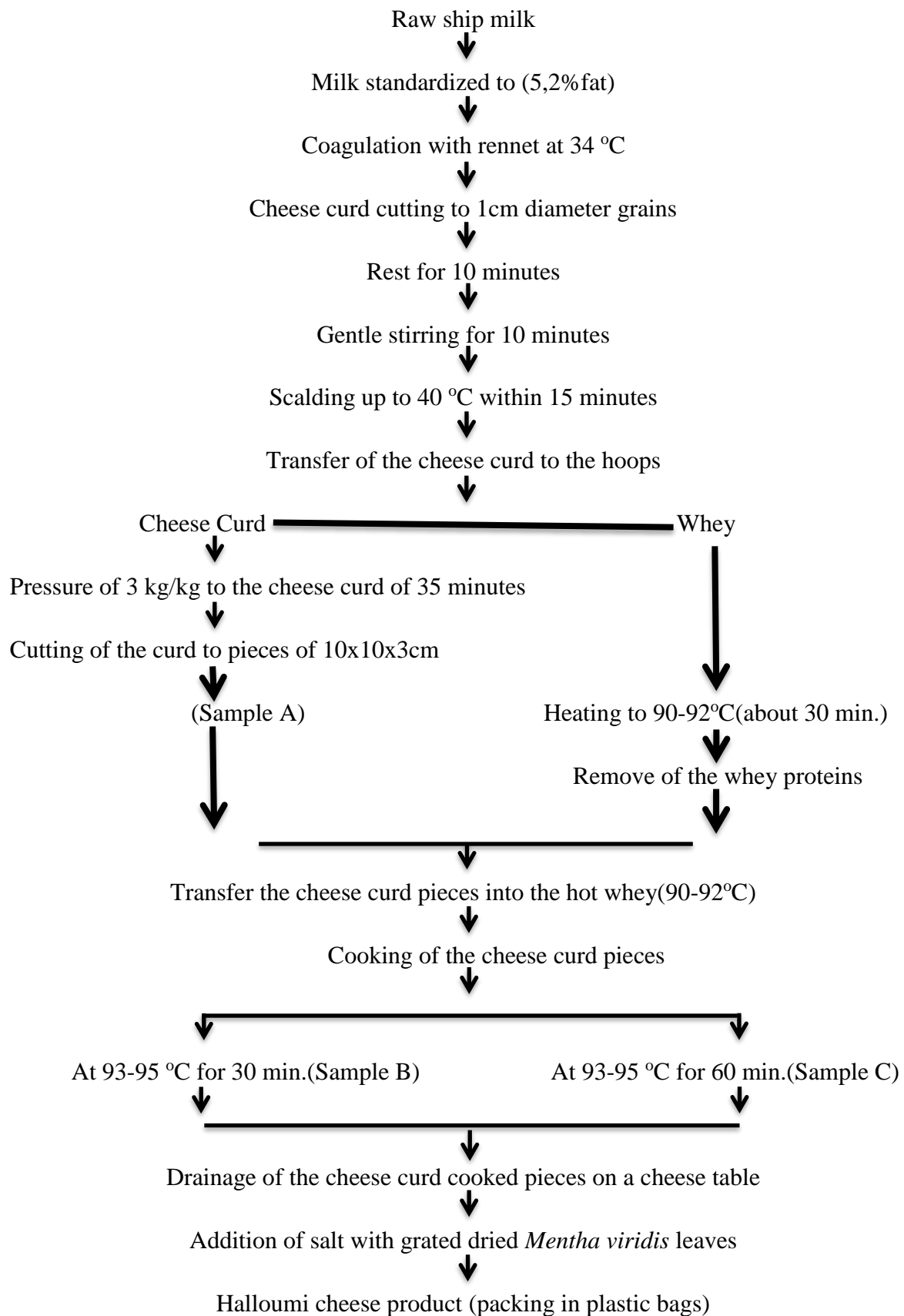


Figure 1.1: Cheese Making Process of Sheep Halloumi Cheese (Sheep & Milk, 1983)

1.4 Composition of Halloumi Cheese

With its compact texture it resists pressure. The cheese has no holes and it is elastic. There may be holes, if so it is of scarce and considered irregular. Generous portions can be sliced from the cheese easily. It's white and yellowish color may vary depending on milk origin from which cheese is produced either ovine or caprine or bovine milk. Different in means of stretch and melting characteristics, the texture of this cheese upon heating is similar to the raw cheese. Evenly melting can be observed on sliced cheese and has the potential to stretch, despite that it's not chewy or tough; similarly reacts like concentrated viscoelectric polymer mixture at melting point (Lelievre et al., 1990; Robinson, 1991; Papademas & Robinson, 1998). The fresh Halloumi cheese can be consumed right after manufacturing.

Having a milky and creamy flavour the fresh halloumi cheese has a distinct aroma to the mature Halloumi cheese which requires 40 days after manufacturing to be sold as it is matured. The texture and taste alters as it matures (Papademas, 2006).

The manufacturing of Halloumi cheese has been described previously (Papademas, 2006). Nevertheless the traditional Halloumi cheese was initially produced using either mixture of ovine and caprine milks or separately, the high demand in the market forced no regulations to be implemented in order to produce this cheese from bovine milk also (Moatsou et al., 2004). A connoisseur can with no difficulty detect the differences of the halloumi cheese and its milk type depending on its taste (Papademas, 2006). Despite this, the real difference is more distinctive as the cheese is more mature (Papademas & Robinson, 2000).

The history of mankind reveals that the surplus milk was made use of through the method of preserving it as a cheese in agrarian economies. There are legions in regards to various types of cheese production. For the sake of Halloumi cheese, it has been part of the Cyprus tradition for centuries. The method of pasteurization is used to prepare the Halloumi cheese to cook it in its own whey through boiling using rennet. Traditionally the bacterias were halted by preserving the cheese in brine. Contemporary Cyprus community today preserves this cheese as its casual taste.

Table 1.2: European Union Rapid Alerts by Cheese Type (European Food Safety Authority Rapid Alert System, 2006)

Heat Treatment	Cheese types	E. coli	<i>Salmonella</i> spp.	<i>Listeria monocytogenes</i>	<i>Brucella</i> spp.	<i>S. aureus</i>	Other	Total
Raw	Raw goat milk cheese			2	1			3
	Raw milk Camembert	1						1
	Raw milk cheese	1		3		1		4
	Roquefort cheese		1					1
	Semi hard raw milk cheese			1				1
Pasteurised	Pasteurised milk cheese	1		1				2
Unknown	Artisanal cheese					1		1
	Camembert/Brie		1	3			2	6
	Curd	2						2
	Emmentaler cheese			1				1
	Sheep milk cheese			1				1
	Feta cheese			1				1
	Fresh cheese		1					1
	Goat cheese		2	2				4
	Gorgonzola			16				16
	Halloumi cheese			2				2
	Hard cheese	1						1
	Maroilles soft cheese			1				1
	Parmesan cheese					1		1
	Raclette cheese							1
	Ricotta cheese		1					1
	Saint Nectaire cheese			1				1
	Smoked milk cheese			1				1
	Soft cheese	1		4				5
	Tallegio cheese			1				1
	White mould cheese							1
	Other cheese	1		11			3	12
TOTAL		8	6	50	1	3	5	73

1.5 Types of Halloumi Cheese

1.5.1 Fresh Halloumi

The fresh Halloumi can be described as fresh maximum twenty four hours before the manufacturing. This cheese is milky and creamy in taste which holds its distinct aroma and mint is added to the layer formed after folding the block of Halloumi cheese to give the cheese its minty flavor as the Halloumi cheese is specified as semi-salty (Papademas, 2006).

1.5.2 Mature Halloumi

40 days following the manufacturing of the cheese is the time lapse necessary for the Halloumi cheese to be regarded as mature. Mature cheese is rather altered in taste and texture. The hardness of the cheese is the most notiable change. The harder body and intense texture is the distinctive element of the mature Halloumi to the fresh one (Papademas, 2006).

1.6 *Salmonella*

Salmonella is one of the most significant pathogens caused via food and are the main elements known for more than a century to be the main reason for many food poisoning cases or moreover typhoid, paratyphoid, septicaemia and bacterameia and other related long-term conditions.

The most encountered clinical manifestation is food poisoning. Antigenic, patterns of sugar fermentation and bacteriophage characteristics are main basis of seperation this genus.

One of the most significant pathogens is the members of the *Salmonella* bacterial genus; they cause infections both on humans and animals.

1.6.1 History

The initial encounter with *Salmonella* was in 1880. Karl Eberth was the first person to visualize this in typhoid patients with Peyer's pathes and spleens (Eberth et al., 1880). The pure culture breed of the pathogen was successfully undertaken by Georg Theodor Gaffky in 1884, four years later (Hardy, 1999). Theobald Smith, after one year, discovered the later known as *Salmonella enterica*. Smith was then taking Office as research assistant in US Department of Agriculture- Veterinary Division.

The veterinary pathologist, Daniel Elmer Salmon was in charge of the department administration (FDA, 2008). "Hog-cholera bacillus" was the name given to the *Salmonella* Choleraesuis by Salmon and Smith, which was thought to be the triggering agent of hog cholera. Not until 1900, right after the proposal of Joseph Leon Lignieres to honor Daniel Salmon, the name *Salmonella* was not in use (Heymann et al., 2006).

1.6.2 Characteristics

Salmonella is classified under the Enterobacteriaceae family (Ewing, 1986; Brenner et al., 2005). This genus specifications are motile, facultative rods and gram-negative. The only carbon source and nitrogen source for the *Salmonellae* are citrate and lysine and also on the production of H₂S in triple sugar agar respectively. Exceptionally to this, species, subspecies and serotypes are defined by these traits (Ewing, 1986).

Two species are the composition of the genus *Salmonella*; these are *Salmonella enterica* and *Salmonella bongori* (Tindall et al., 2005; Judicial Commission of the International Committee on Systematics of Prokaryotes, 2005). *S. Enterica* is subdivided into six:

S. enterica subsp. *enterica*, often called subspecies I;

S. enterica subsp. *salamae*, or subspecies II;

S. enterica subsp. *arizonae*, or subspecies IIIa;

S. enterica subsp. *diarizonae*, or subspecies IIIb;

S. enterica subsp. *houstenae*, or subspecies IV and

S. enterica subsp. *indica*, or subspecies VI.

Genus "Arizona" holds the descriptions of subspecies IIIa and IIIb. Monophasic strains are included in IIIa and diphasic strains in IIIb (Rohde, 1979). Despite their historical similarity the subspecies IIIa and IIIb are different than other entities mentioned (McQuiston et al., 2008; Desai et al., 2013).

High level of genetic diversities are indicated as a result of Genome analysis on *salmonellae*. For example, both in presence or in the absence of whole genes the *S. Enterica* subspecies I of Serotypes are distinct in gene content to about 10% (Edwards et al., 2002; Porwollik & McClelland, 2007).

The *S. Enterica* subspecies I strain recombination fairly contributes to this mentioned diversity (Lan et al., 2009). As of the end of 2013, *Salmonella* strains >500 complete or partial sequences are available as the expansion of the strains characterization whole genome sequence analysis continues. The knowledge relevant to *Salmonella* biology and phylogeny accumulates as the whole genome sequence characterization increases.

Two diverse lineages of *Salmonella* Enterica subspecies were analysed to reveal genome subsequences; thus diversities in the content of gene were notable and thus this will result in advancement in epidemiologic strain typing as the application of analysis of whole genome subsequence via different methods are been applied on single nucleotide polymorphisms, core genes and etc. (Timme et al., 2013).

Salmonella belongs to the Enterobacteriaceae family with its gram negative and rod shape as a facultative anaerobe bacterium. 2-3 x 0.4 – 0.6 µm is the size of this bacterium (Montville & Matthews, 2008). Having been motile by peritrichous flagella these are non-spore with oxidase negative and catalase positive.

The *Salmonella* are eligible to minimize nitrate to nitrite, form carbon source on citrate, produce gas and acid through glucose, generate hydrogen sulfide from triple sugar iron also decarboxylate ornithine and lysine and finally hydrolize urea and indole.

Salmonella strains are distinguishable through agglutination reactions. This reactions are with combination of antigens of each strain and homologous antisera. These are classified in two species, *S. enterica* and *S. bongori*, *S. Enterica* is further divided into six subspecies, *Salmonella enterica* subsp. *arizonae*, *Salmonella enterica* subsp. *diarizonae*, *Salmonella enterica* subsp. *enterica*, *Salmonella enterica* subsp. *houtenae*, *Salmonella enterica* subsp. *indica*, and *Salmonella enterica* subsp. *salamae* (Tindall et al., 2005).

The peak growth of *Salmonella* is apparent at 35-37 °C (mesophilic), despite the fact that it is also able to grow in temperatures higher than this such as 5°C to 46 °C in line with the serotype (El-Gazzar & Marth, 1992). Despite the fact that the maximum growth is apparent in 6.5 and 7.5 pH levels, salmonellae can also grow in pH 4 environment as well, where acidity level is higher. No sodium chloride is required for their growth; they can grow around 0.4 – 4% of sodium chloride.

The *salmonella* can be killed at high temperatures like 70 °C or above; which is the pasteurization heat. Despite the fact that they survive in high water activity conditions they can also manage to hold on dry foods at ($a_w < 0.2$) (Pui et al., 2011).

1.6.3 Food Poisoning by *Salmonella* sp.

Among the different foodborne illnesses, *salmonellosis* is the most critical one both as far as the illness severity, simplicity of spread and trouble in counter measures and control. As indicated by WHO, it speaks to 60-80% of all reported instances of foodborne illnesses. The purposes behind the expansion in event of *salmonellosis* are:

- Increase in multinational foods
- Increase in global exchange of foods
- Higher occurrence of salmonellosis in pets
- Widespread utilization of family cleansers meddling with sewage treatment
- Wide use of prepared foods
- Widespread consumption food of containing antibiotics that leads to drug-resistant salmonellae and their potential transmission to individuals.

It is vital to take in the scientific classification and morphological elements of the creature, development qualities, survival attributes, stores, food borne flare-ups, malady manifestations, irresistible measurements, brooding period and death rate, pathogenicity and harmfulness components, lab finding.

1.6.4 Importance To The Consumer

People contaminated with *Salmonella* create loose bowels, fever, queasiness, stomach issues, cerebral pains, and spewing 12–72 h after disease. Side effects normally most recent 4–7 days and the lion's share of those contaminated by *Salmonella* recuperate without treatment. In a few people, in any case, the looseness of the bowels might be severe to the point that they should be hospitalized. More serious systemic diseases may happen in people who are more helpless, for example, the exceptionally youthful, the elderly, and the immunocompromised. Sometimes, these contaminations can have all the more long haul outcomes prompting unending conditions, for example, joint pain or endocarditis.

1.6.5 Analysis of *Salmonella*

Depending on previous history of contamination methods of isolation and detection is developed for various foods. For sufficient results, it may necessitate 4-6 days through the conventional methods of identification.

The screening of foods may be applied through various methods; lasts 1-2 days. Assays based on DNA or RNA are methods used for this purpose. Most of the time, for regulatory reasons, cultural methods are required to confirm the *Salmonella* presence.

1.6.6 The Factors Affecting Growth of *Salmonella*

Physiology

A scope of natural conditions influences the development, demise, or survival of *Salmonellae*, shaping the premise for control and conservation measures in the sustenance handling industry. These incorporate temperature, pH, and water activity (a_w), and blends thereof.

Temperature

Salmonellae can develop inside the reach 2–54 °C, despite the fact that development beneath 7 °C to a great extent has been watched just in bacteriological media, not in sustenances, while development above 48 °C is restricted to mutants or tempered strains.

The ideal temperature for development is 37 °C, which is not astounding given that the normal environment of most *Salmonella* strains of worry to general wellbeing is the gastrointestinal tract of warm-blooded creatures. Over the most extreme development temperature, *salmonellae* kick the bucket rapidly and, all in all, are promptly demolished by mellow warmth procedures, for example, purification. Vulnerability changes with strain, be that as it may. Investigations of numerous strains in model frameworks have exhibited mean D-values at 57 and 60 °C of 1.3 and 0.4–0.6 min, separately, and z estimations of 4–5 °C.

Salmonella Senftenberg 775W, the most warmth safe strain of *Salmonella* up to this point recognized, has D-values at the same temperatures of 31 °C and 4–6 min. Introduction to unfavorable conditions, including presentation to sublethal temperatures and extremes of pH, expands resistance. Nourishments high in solids content, especially protein or fat, and low in dampness (and a_w) are exceedingly defensive, with survival in sustenances, for example, chocolate or nutty spread measured in hours somewhere around 70 and 80 °C.

Expanded warmth resistance is less stamped when solutes, for example, NaCl instead of sugars are utilized to lessen water movement. *Salmonellae* survive entirely well at low temperatures. Despite the fact that the time shifts with substrate and the impact of such elements as pH and a_w , strains may make due for quite a long time to weeks at chill temperatures. During solidifying, a population of *salmonellae* will be lessened conversely to the rate of solidifying, further affected by the level of assurance managed by the lattice in which the living being is held and the physiological status of the cells, with log-stage cells being more vulnerable to harm (D'Aoust & Maurer, 2007; Porwollik, 2011).

Subsequent to solidifying, a populace of *Salmonella* experiences a moderate decrease, and the rate of decay is contrarily corresponding to the capacity temperature. In a defensive framework, and under business solidifying conditions, salmonellae may make due for a considerable length of time or years.

pH

The ideal pH for development of *Salmonella* is in a range of 6.5–7.5 between, with strains developing at pH values up to 9.5, and down to 4.05, despite the fact that the base fluctuates impressively with the acidulant used to lessen pH. Despite the fact that development happens down to or near the base pH with nonvolatile natural acids, for example, citrus extract, or mineral acids, for example, hydrochloric corrosive, development stops at higher pH values when unpredictable unsaturated fats (VFAs) are utilized (e.g., pH 5.4 within the sight of acidic corrosive).

The inhibitory impact of VFAs is conversely corresponding to bind length and to increments under anaerobic conditions, probably because of a diminishing in accessible vitality (adenosine triphosphate (ATP) and a resulting diminish in capacity to expel the acids from the intracellular environment. Expanding temperature builds affectability to low pH, as does the presence of nourishment added substances, for example, salt or nitrite. Affectability to pH is a noteworthy additive element in nourishments to which acidulants are included, for example, mayonnaise, or in which acids are delivered by maturation, for example, salami or cheddar. The impact of acidulant is exemplified by the expanded survival of *Salmonella* Enteritidis in mayonnaise made with lemon juice (citrus extract) as opposed to vinegar (acidic corrosive). Resilience or adjustment to low pH is huge as for harmfulness, improving the probability of surviving gastric sharpness, or the acidic intracellular environment of phagocytic cells.

Salmonellae for the most part display three particular reactions to causticity, a general pH-autonomous, RpoS-interceded stress reaction, and a pH-subordinate reaction, both enacted in the stationary stage, and in addition a pH-subordinate log-stage reaction (D'Aoust & Maurer, 2007; Porwollik, 2011).

Water Activity (a_w)

Salmonellae develop at (a_w) values somewhere around 0.999 and 0.945 in research facility media, down to 0.93 in sustenances, with an ideal of 0.995. In spite of the fact that there is no development underneath 0.93, *Salmonella* survives, and the season of survival increments as a_w reductions. For example, pasta, nutty spread, and chocolate, survival is measured in months. Salt (NaCl), utilized as a solute to lower water activity and as an additive in nourishments, is inhibitory toward *Salmonellae* at centralizations of 3–4%, with resilience expanding at temperature somewhere around 10 and 30 °C (D'Aoust & Maurer, 2007; Porwollik, 2011).

1.6.7 Types of *Salmonella*

Salmonella Typhimurium

S. Typhimurium is a pathogen usually flexible in adapting itself to varieties of hosts such as horses, sheep, cattle, poultry, rodents and as well humans. Over 200 varieties of this pathogen are found and identified to adapt themselves principally to niches in the environment and intestines of various animal species (Winfield & Groisman, 2003). Despite that various fitness factor encoding mobile genetic elements and also phage combinations are present (Brüssow et al., 2004), the content of genome *S. Typhimurium* strains is significantly similar (Jarvik et al., 2010). *S. Typhimurium* was used widely for the determination of *Salmonella* pathogenicity and vaccine development (Curtiss et al., 2009).

Salmonella Enterica

According to US data, the most commonly known cause of bacterial food-borne illness is *Salmonella Enterica* (Mead et al. 1999; Anonymous, 2011). Strains of *Salmonella* have been reported which persisted environment for long years, stress and nutrient depletion (Parker et al., 2010). This bacterium uses various vertebrate sources to develop like wildlife, poultry, livestock and herds of animals (Mandrell et al., 2009).

Some outbreaks of high-profile have occurred due to fresh produce incidents as a vector for *Salmonella* infection (Sivapalasingam et al., 2004; CDC, 2008; Hanning et al., 2009). Usually fresh produce preharvest contamination for human enteric pathogens are due to contact with contaminated environment or water, animals, manure, farm equipment, dust and human carriers. Previously, surveys to describe the measure of incidence and amount of this mentioned contamination (*Salmonella*) have been reported (Haley et al., 2009, Wacheck et al., 2010).

Salmonella Bongori

Belongs to genus *Salmonella*, the *Salmonella bongori* is pathogenic bacterium, previously known as *Salmonella* subspecies V (or *S. enterica* subsp. *bongori* or *S. choleraesuis* subsp. *Bongori*). *Salmonellosis*, a gastrointestinal disease caused by a Gram negative rod shaped bacterium called bacillus is characterized through cramping and diarrhoea. Despite its similarity to other genera, it is differently regarded as a microbe of cold blooded animals; mostly mentioned with reptiles.

The Discovery was made from a lizard found in Chad's city of Bongor in 1966, happened to allow the derive of a unique name *bongori* (Le Minor et al., 1969). Following the controversy that lasted for decades on *Salmonella* nomenclature, it was granted the species status by 2005 (Agbaje et al., 2011).

1.6.8 Control of *Salmonella*

Control of *Salmonella* with specific respect to foodborne infection is dangerous, given the nearby connections between the surroundings, nourishes, sustenance creatures, and people and, extensively, requires watchfulness at two levels, in sustenance generation and sustenance handling. A scope of administration techniques have been created or conceived to control *salmonellae* in nourishment generation situations, especially that for the generation of creatures, a noteworthy vehicle of transmission of *Salmonella*.

These systems incorporate the arrangement of sans salmonella stock and encourage, stringent biocontrol, especially of rodents, immunization with lessened *Salmonella* strains, and utilization of probiotic arrangements.

Maybe the most essential control measure in nourishment preparing includes instruction, first of business sustenance handlers in the regions of individual and nourishment cleanliness, especially in the nourishment administration part of the nourishment business, and second of buyers, who are the sustenance handlers required in sustenance administration at the residential level. In spite of the fact that *Salmonella* may never be disposed of totally, noteworthy lessening ought to be accomplished through the use of suitable control methodologies inside a very much created and actualized peril examination and critical control point– based nourishment wellbeing arrangement from the initiation of generation through to utilization (D'Aoust & Maurer, 2007; Porwollik, 2011).

1.6.9 Salmonellosis

Clinical signs and manifestations of average human *Salmonellosis*, which might be foodborne, incorporate intense onset of fever, stomach torment, gastroenteritis, sickness, and heaving. The hatching time frame is for the most part 12–72 h, generally 12–36 h, with a normal length of 2–7 days. The infection is generally self-restricting, with patients recuperating uneventfully (without anti-toxins) inside a week.

Anti-microbial treatment is important in under 2% of clinical cases, where serious drying out happens, particularly in the elderly (>50 years), youthful youngsters (<5 years), or the immunocompromised, who may represent up to 60% of all advised cases and may contribute fundamentally to the general low death rate of 0.1–0.2%. A few patients (<1%) create complexities or long haul impacts (sequelae), which may incorporate joint inflammation, osteoarthritis, an infected appendix, endocarditis, pericarditis, meningitis, peritonitis, and urinary tract contaminations.

Taking after clinical sickness, there additionally might be a time of discontinuous fecal shedding, enduring from days to years, with a medium term of 5 weeks, and <1% getting to be perpetual transporters. Regardless of advances in the treatment of irresistible illnesses, pathogenic microorganisms, including *Salmonella*, are an imperative risk to wellbeing around the world (Wren, 2000). *Salmonella* enterica serovars taint various has and cause a few distinct illnesses through complex connections with host cells.

Salmonella enterica serovar Typhi (*S. typhi*) is a human-limited pathogen and the causative operator of the systemic febrile ailment typhoid or enteric fever. Non-typhoidal *Salmonellae*, for example, *Salmonella enterica* serovar Typhimurium (*S. typhimurium*), contaminate different has and cause gastroenteritis in people, cows, steeds, and other substantial creatures however cause a systemic disease in vulnerable ingrained mice that looks like typhoid fever. *Salmonellae* are typically orally ingested in polluted sustenance or water and survive the gastric environment to achieve the small digestion tracts. *Salmonella enterica* is an enteric bacterium that causes an assortment of sicknesses in people, running from gentle gastroenteritis to serious systemic contaminations (Ohl & Miller, 2001; Rabsch et al., 2001). *S. enterica* subspecies *enterica* incorporates the majority of the serovars that cause ailment in people and domesticated animals creatures. Pathogens, for example, *S. enterica* serovars Typhimurium (*S. Typhimurium*), Enteritidis (*S. Enteritidis*) (in both created and creating nations) and Typhi (*S. Typhi*) (in creating nations) are often disengaged from people, where they are connected with an expected 115 million clinical cases for every year around the world (Rabsch et al., 2001; Crump et al., 2004; Bhan et al., 2005; Majowicz et al., 2010). Contingent upon the serovar and the host, *Salmonella* diseases have diverse results, and can be grouped into three general sorts that are seen in immunocompetent people: (i) Gastroenteritis.

A self-restricted contamination of the terminal ileum and colon, prompting looseness of the bowels and irritation, which contains bacterial scattering past the intestinal submucosa. (ii) Typhoid fever. A systemic disease starting in the terminal ileum from where microscopic organisms disperse through the lymphatic framework and through transport of the pathogen inside phagocytes. Spread grants colonization of inner organs, for example, the liver, the spleen, the bone marrow and the nerve bladder. (iii) Chronic carriage. A small amount of people recuperating from typhoid fever get to be asymptomatic, long lasting bearers of *S. Typhi*.

Non-typhoidal *Salmonella* serovars can additionally cause determined diseases, either connected with cholecystitis or asymptomatic, in spite of the fact that the span of carriage is typically restricted to a while. Like people, domesticated animals and wild creatures can be asymptomatic transporters of non-typhoidal *Salmonella* serovars ready to contaminate people, in this manner going about as repositories for human disease.

Salmonella are enteric microscopic organisms that are a noteworthy reason for irresistible maladies all through the world. These microbes taint both people and different creatures and are a typical reason for zoonotic infection. The sort *Salmonella* consolidates Gram-negative, facultative anaerobic pole molded bacilli that are delegated individuals from the family Enterobacteriaceae. This sort, which is evaluated to have separated from *Escherichia coli* roughly 100–150 million years prior, is hereditarily various and has adjusted to colonize a wide range of corners and has.

For instance, *Salmonella* microscopic organisms can be discovered both as commensal and pathogen in a scope of warm and merciless creatures and they are equipped for surviving free in the earth for broadened timeframes. In created nations, quickly *Salmonella* is all the more generally connected with intense, gut-related, non-systemic gastroenteritis; in any case, particular serovars of *Salmonella* (e.g. typhi, paratyphi and so forth.) are truly imperative as causative specialists of the human systemic disease typhoid fever (enteric fever) that is still a typical ailment in numerous creating nations. The assorted qualities of *Salmonella* is an essential thought while checking on resistance, as individual bacterial disconnects can possibly vary fundamentally regarding antigenic sythesis, host inclination, and destructiveness potential.

Therefore, thinks about in model frameworks will be essentially reliant on the specific *Salmonella* seclude under study. The microbiology ought not be overlooked by the immunologist and visa versa.

1.6.10 Significance To The Food Industry

Sources

This bacterium uses various vertebrate sources to develop like wildlife, poultry, livestock and her of animals; thus widely spread into nature. The transition of contamination is through fecal-oral and via contaminated water. (Certain protozoa may act like a reservoir for an organism). This may cause the contamination of meat and farm irrigation water and also soil, factory, equipment, hands, kitchen surfaces, etc. Untreated sewage seems to be the only source of the outbreak of *S. Typhi* and *S. Paratyphi A* as these are only hosted by humans through transiting from organisms found in the environment like drinking water or irrigation water. In an environment of endemic organism are present it is significantly recommended to use portable water and also pre-cooked vegetables.

Food Sources

Illnesses caused by *Salmonella* serotypes are known as *salmonellosis*. Disease is started by utilization of crude or undercooked spoiled food creature sustenance (basic origin, source of contamination for people) or water containing fecal material. Notwithstanding transmission by nourishment, *Salmonella* can likewise be spread by means of nature in a number of ways, by implication contaminating people and different creatures, as it can make due for drawn out stretches of time under both wet and dry conditions. The result of this disease to a great extent relies on upon the serotype and kind of host. Serotypes Typhimurium and Enteritidis can bring about malady in people, cows, poultry, sheep, pigs, stallion and wild rodents. They are two clinical kind of human *salmonellosis*: enteric fever (a serious, life-debilitating illness) taking after contamination with *S. typhi* or paratyphi and the more regular nourishment borne illness disorder brought on by nontyphoid *Salmonella* species. In both cases, the capable microorganisms enter the body through the oral course.

Salmonellosis for the most part is a self-restricting intense gastroenteritis like intestinal flu, and is accepted to be horribly underreported. Onset of non-typhoidal *salmonellosis* regularly happens 12 to 36 hours after ingestion of the polluted nourishment portrayed by sickness and heaving; manifestations which have a tendency to die down inside a couple of hours. Indications incorporate queasiness, spewing, chills, fever, stomach agony or spasms, and cerebral pain, is normally trailed by loose bowels (El-Gazzar & Marth, 1992). The disease more often than not keeps going 4 to 7 days and most people recuperate without anti-microbial treatment. The elderly, newborn children and those with fundamental constant illness or immuno-bargained people will probably have a serious disease. *S. typhi* is in charge of bacteremia-related enteric fever alluded to as typhoid fever. Onset normally happens inside 8 to 15 days, and some of the time the length of 30 to 35 days.

Manifestations incorporate fever, migraine, discomfort, anorexia, and blockage of the mucous films, particularly of the upper respiratory tract. The high death rate of *S. typhi* (10%) contrasted with other *Salmonella spp.* can be diminished with the brief organization of anti-infection agents.

Sources of Salmonella

Either dead or living, mostly biological entities act as sources of *Salmonella*, thus causing an outbreaks of foodborne diseases. Knowing that the natural habitat of this bacteria, *Salmonellae* is significantly in the tract of human gastrointestinal, it is common to encounter the transmission of this illness through animal derived foods. It is therefore significant to acknowledge that animals and animals fed with infected plants are important persistence of *Salmonellae*. The poultry industry is a significant example of controlling the spread of the infection by controlling the feed of the animals, thus lowering the carriage rate in this sector. Depending on the geographic location, water is held its significance of being an important vehicle of transmission either through consumption or via the usage of water in the processed food industry. Therefore vigilance is important to undertake both in food production and processing. Particularly for the poultry industry, various measures of detection strategies are developed to control the spread of *Salmonella*. As mentioned previously, plants are also significant in terms of acting as means of transmission. Strategies like *Salmonella*-free stock and feed, use of probiotic preparations, stringent biocontrol; specifically of rodents may be applied. Furthermore the most significant control measure against the spread of the illness is through education to those who are working in food processing sector as first handlers of the food in terms of hygiene and also in food service sector as food handlers.

Despite the fact that *Salmonella* is not possibly be able to eliminated permanently, through appropriate measures, high level of reduction is possible with appropriate hazard analysis and sound safety plans through production to consumption levels (D'Aoust & Maurer, 2007; Porwollik, 2011).

1.6.11 Cross Contamination

This type of contamination occurs when *salmonella* contaminated food or handler (human) or animal gets into contact with other food to spread the infection. This usually occurs when contaminated food, raw meat, seafood, eggs, poultry are kept in same place with non-contaminated foods during preparation or through non-hygenic handler or equipments and/or utensils. Regardless of where, the cross contamination has the potential to spread at any period of the food process, wheather in factory, equipment surfaces, kitchen or utensils. Wildlife pets such as frogs or turtles may spread *salmonellosis* outbreak when cross contacting with food or utensils; even the legs of culinary frogs have contaminated *salmonellosis*.

1.6.12 Typhoidal *Salmonella*

Typhoid fever is brought on by *Salmonella* serotypes which are entirely adjusted to people or higher primates—these incorporate *Salmonella* Typhi, Paratyphi A, Paratyphi B and Paratyphi C. In the systemic type of the ailment, *salmonellae* go through the lymphatic arrangement of the digestive system into Increase in communal feeding:

- Increase in universal exchange of human nourishment
- Higher rate of salmonellosis in animals
- Widespread utilization of family unit cleansers meddling with sewage treatment
- Wide appropriation of arranged nourishments
- Widespread utilization of creature encourages containing antimicrobial medications that support drug-safe *salmonellae* and their potential transmission to people.

It is critical to take in the scientific categorization and morphological components of the life form, development attributes, survival qualities, supplies, nourishment borne episodes, sickness side effects, irresistible dosage, hatching period and death rate, pathogenicity and harmfulness variables, lab finding.

1.6.13 Concepts in Detection

Salmonella has an effect on culturability, when in a physiological state. It is much feasible to isolate the *salmonellae* when found as clinical specimen through direct planting, as it is apparent with higher numbers here and in total vegetative state. The opposite for the most part is valid for a sustenance test, in that *salmonellae*, if present, will be in low numbers and regularly in a poor physiological state, enduring harm because of such procedures as chilling, solidifying, warming, or extremes of pH. All things considered, such cells are still fit for recuperation after ingestion, conceivably bringing on illness, and subsequently shall be recognized. To help recuperation of *salmonellae* and encourage the identification procedure, sustenance sample sare subjected to nonselective fluid preenrichment (resuscitation). This is trailed by specific fluid enhancement, allowing further development of the now-vegetative *salmonellae*, while smothering the foundation verdure that creates during resuscitation. At long last, the specific improvements are plated and any separates are portrayed. A few sustenances additionally may impact the recuperation of *Salmonella*.

Numerous flavors demonstrate inhibitory toward *salmonellae* in culture, due much of the time to the antimicrobial action of vital oils associated with smell and flavor, whereas the anthocyanins in chocolate and other cocoa-based items additionally repress development. These must be killed to encourage recuperation, utilizing such methodologies as dissolved, expansion of killing operators, or use of an elective improvement medium. Most *salmonellae* display a typical example of biochemical responses and physiological characteristics, a significant number of which are misused in cultural techniques for recognition. A few strains, notwithstanding, may show one or once in a while more atypical responses or attributes, including maturation of disaccharides, for example, lactose and sucrose, inability to deliver hydrogen sulfide, absence of lysine decarboxylation, or absence of motility. On account of such strains, social discovery may fall flat. Atypical strains are uncommon in connection to the numerous thousands disconnected every year, happening at a rate of under 0.1% for any given attribute.

The frequency of atypical strains in connection to a particular nourishment framework might be much higher, in any case, on account of specific weight, with an illustration being lactose-positive strains in dairy items. Identification of *salmonellae*, including numerous atypical strains, can be performed utilizing a scope of noncultural strategies, normally tailing some type of improvement.

These incorporate serological systems, for example, latex agglutination and enzyme-linked immunosorbent measure, and nucleic corrosive procedures, for example, PCR; these methods are the subjects of resulting sections (D'Aoust & Maurer, 2007; Porwollik, 2011).

1.6.14 Detecting *Salmonella* Sp.

Cultural techniques, Polymerase Chain Reaction (PCR) and in addition immunology-based strategies are the most well-known techniques utilized for pathogens location including *Salmonella* discovery. They include numbering of microscopic organisms, DNA examination and antigen-counter acting agent connections, individually. These techniques are frequently joined together to yield more reliable results. Recognition of *salmonella* in nourishments by routine cultural techniques comprise of four stages 1) non specific culture, 2) particular enrichment in various media, 3) plating on specific and characteristic media, and 4) confirmation through biochemical and serological tests. It requires 4-6 days.

1.6.15 Diagnosis

Despite the fact that around 100 types of *Salmonella* serotypes can be identified through pure culture they are yet over 2400 more other types that can only be identified through the traditional method of serotyping.

1.6.16 *Salmonella* Serotypes

Salmonella serotyping is a subtyping strategy in view of the immunologic portrayal of three surface structures: O antigen, which is the external most parcel of the LPS layer that covers the bacterial cell; H antigen, which is the fiber part of the bacterial flagella; and Vi antigen, which is a capsular polysaccharide present in specific serotypes. Serotyping of *Salmonella* is usually performed to encourage general wellbeing reconnaissance for *Salmonella* diseases and to help in the acknowledgment of flare-ups.

While sub-atomic techniques, for example, PFGE and MLVA have become the foundation of general wellbeing subtyping (Gerner-Smidt et al., 2006; Nadon et al., 2013), serotyping remains an imperative device. The serotype of a seclude regularly corresponds with a specific disease syndrome or nourishment vehicle, making serotype information particularly useful in recognizing cases and characterizing outburst.

1.6.17 Global Monitoring

In Germany, nourishment harming diseases must be accounted for. Somewhere around 1990 and 2005, the quantity of formally recorded cases diminished from around 200,000 to around 50,000 cases. In the United States, around 50,000 instances of *Salmonella* disease are accounted for every year. A World Health Organization study assessed that 21,650,974 instances of typhoid fever happened in 2000, 216,510 of which brought about death, alongside 5,412,744 instances of paratyphoid fever (Crump et al., 2004).

1.6.18 Incidence

Around the world, foodborne bacterial contamination connected with *Salmonella* is thought to be second just to that including *Campylobacter*, with the frequency of *Salmonella* disease apparently expanding. To some degree, the expansion might be credited to better reporting and observation, as opposed to a real increase in infection. In any case, a critical extent of there ported cases speaks to a real increment.

The case rate for human *salmonellosis* shifts massively, from <1 to >300 per 100 000 population and is significantly affected by geographic, demographic, financial, meteorological, and natural variables. Concerning particular serovars, the prevailing sort connected with food borne disease for a long time in manyparts of the world was the pervasive Typhimurium.

From the mid 1980s, a noteworthy general wellbeing issue started to emerge, including strains of serovar Enteritidis equipped for systemic colonization of poultry prompting broad nourishment borne malady connected with utilization of debased eggs and crude or gently cooked food containing them. Since the mid 1990s, a particular sort of *Salmonella* Typhimurium known as complete sort (DT) 104 has turned into a majör problem in the United Kingdom and Western Europe and now likewise in the United States. Strains of *S.* Typhimurium DT104 are to a great degree intrusive, and numerous contain vast plasmids, giving imperviousness to a scope of anti-infection agents, including ampicillin, chloramphenicol, streptomycin, sulfonamides, and antibiotic medication. Still more up to date strains have been observed to be safe totrimethoprim and ciprofloxacin. Distinctive locales of the world experience issues with particular serovars every once in a while. Utilizing Australia for instance, more than 80% of human diseases with serovar Virchow happen in the condition of Queensland, though those including serovar Mississippi are generally limited to Tasmania. (D'Aoust & Maurer, 2007; Porwollik, 2011).

1.6.19 Target Populations

With no certain or exact age scope, the potential of becoming infected with this bacterium of *Salmonella* is possible. Ofcourse, the possibility of becoming infected is greater to those with non-sound immune systems; children and elderly people. Furthermore thos who are on cancer treatment and are on chemotherapy for cancer or using drugs of immunosupportive types; especially for arthiritis treatment are more likely to become infected. People who are diagnosed with HIV are likely to have 20 times more possibility to become infected.

1.6.20 Clinical Significance

Strains of *Salmonella* are arranged as typhoidal and nontyphoidal, relating to the illness disorder with whichthey are related. Strains of nontyphoidal *Salmonella* usuallycause intestinal contaminations (joined by looseness of the bowels, fever, and stomach issues) that regularly most recent 1 week orlonger (Acheson & Hohmann, 2001).

Less generally, nontyphoidal *Salmonella* can cause extraintestinal diseases (e.g., bacteremia, urinary tract contamination, or osteomyelitis), particularly in immunocompromised people. People of any age are influenced, however the incidence is most astounding in newborn children and youthful youngsters. *Salmonellosis* universal in creature populaces, and human illness typically connected to nourishments. *Salmonellosis* is additionally transmitted by direct contact with creatures, by water, and once in a while by human contact. Every year, an expected 1 million cases of disease and 378 passings are brought about by nontyphoidal *salmonellosis* the United States (Scallan et al., 2011).

1.6.21 Control Measures

- Keep hot food hot and frozen food frozen. Try not to keep food in the temperature peril zone (i.e. at or underneath 5 or more 60°C) any more than would normally be appropriate
- Reheat food to steaming hot before serving (at least 75°C)
- Cook food appropriately, warmth to no less than 75°C
- Keep raw and cooked food separate
- Keep kitchen and utensils clean
- Wash and dry your hands appropriately
- Avoid taking care of food when you are sick

1.6.22 Evaluation, Interpretation and Reporting of Results

A preparatory report can be issued when a hypothetical distinguishing proof of *Salmonella* is gotten. Much of the time, a hypothetical distinguishing proof depends on phenotypic attributes decided either by customary or business frameworks, or by reactivity with *Salmonella* O gathering antisera.

An affirmed distinguishing proof requires both phenotypic ID and O bunch or serotype assurance. Since national reconnaissance frameworks rely on upon the receipt of serotype data for *Salmonella* strains disengaged in the nation, research centers ought to take after the methodology prescribed by their state wellbeing divisions for submitting *Salmonella* disconnects for further portrayal, including complete serotyping.

The antimicrobial powerlessness of typhoidal *Salmonella* strains and strains from typically sterile destinations ought to be resolved, and the strains ought to be sent to a reference or general wellbeing research center for complete phenotypic recognizable proof and serotyping.

1.7 *Staphylococcus Aureus*

Staphylococcus aureus is a gram-positive coccal bacterium that is an individual from the Firmicutes, and is habitually found in the nose, respiratory tract, and on the skin. It is frequently positive for catalase and nitrate decrease. Despite the fact that *S. aureus* is not generally pathogenic, it is a typical reason for skin contaminations, for example, abscesses, respiratory diseases, for example, sinusitis, and sustenance harming.

Pathogenic strains frequently advance contaminations by delivering strong protein poisons, and communicating cell-surface proteins that quardary and inactivate antibodies. The rise of anti-infection safe strains of *S. aureus*, for example, methicillin-safe *S. aureus* (MRSA) is an overall issue in clinical pharmaceutical.

Staphylococci are the primary driver for diseases, for example, folliculitis, endocarditis, furunculosis and *staphylococcal* singed skin disorder (SSS) found in both people and creatures, and the *staphylococcal* inebriations found in people. *Staphylococcus* diseases can be brought on by both coagulase positive and coagulase negative staphylococci. In any case, the overwhelming wellspring of *staphylococcal* nourishment inebriation is the *S.aureus* (Oliver et al., 2005; Tang et al., 2007).

The principle toxicosis really brought about by *S.aureus* are the inebriations started by the utilization of enterotoxin sullied sustenance. Inside a brief span, for example, couple of hours taking after utilization, disease begins to advance with beginning sickness, trailed by spewing and the runs. The dangerous stun disorder known since 1978 is overwhelmingly created by strains delivering the TSST-1 poison. The dermatitis exfoliativa is brought about by *staphylococci* which produce exfoliatin. The disease episodes as broad scaling of the skin, frequently joined by tingling and male pattern baldness (Tunail, 2009).

As indicated by Wieneke and his partners, sustenance poisonings brought on by *S.aureus* and different sorts of *staphylococci* are among the main nourishment harming cases as far as significance (Niskanen et al., 1978; Wieneke et al., 1993).

Nourishment harming is created by the admission of poisons or concentrated measures of microscopic organisms. *Staphylococcal* sustenance harming is brought on by the admission of poisons produced inside the foodstuff by these microscopic organisms through the digestive framework.

While *S.aureus* was at first kept in charge of all *Staphylococcal* sustenance poisonings, later research uncovered that different *staphylococcus* sorts with coagulase positive properties, for example, *S.intermedius*, *S.hycius* and *S.delphini*, likewise brought on nourishment harming. *S.aureus* is a wellspring of bacterial disease as well as causes sustenance harming through the enterotoxins it produces. It was additionally uncovered that while some coagulase negative *staphylococcus*, for example, *S.epidermis*, can likewise create enterotoxin, not all coagulase positive sorts produce enterotoxins (Diatek, 2012).

The digestive framework is an essential passageway door for contaminations. Numerous operators of disease are disposed of through the stool or pee of contaminated creatures, patients, or the general population that come into contact with them. From that point, they can achieve the digestive arrangement of defenseless individuals through grimy hands, water and nourishment. Nourishment gained from tainted creatures and also the pollution of sustenance by encompassing microorganisms can likewise prompt contaminations through the digestive framework (Öksüz, 2008).

As an results of the prior discoveries, the possible recognizable proof of *staphylococci* in foodstuffs can't be kept constrained to *S.aureus*. In like manner, the new Microbiological Criteria Regulation of the Ministry of Food, Agriculture and Livestock of the Republic of Turkey incorporates the general reference of coagulase positive *staphylococci* (Diatek, 2012).

1.7.1 History

Despite the fact that *Staphylococci* were initially watched and distinguished in 1878 by Pasteur and Robert Koch, point by point research on *staphylococci* was done in the years 1880 and 1881 by Ogston, and afterward in 1884 by Rosenbach. Ogston portrayed micrococci as microorganisms creating suppurative skin irritation when their action and spreading region are low, and bringing on septicaemia and pyemia when they have the ability to spread, focusing on that they have pathogen properties. In 1884 Rosenberg created unadulterated *staphylococci* societies interestingly and recognized them through biochemical investigations.

He considered *staphylococci* a family and watched that they shaped white and orange hued states. He has recognized two distinct staphylococci sub-sorts from this family as per their pigmentation properties.

Rosenberg named the microorganisms framing orange hued provinces as *Staphylococcus pyogenes aureus*, and the microorganisms shaping white shaded states as *Staphylococcus pyogenes albus*. Thereafter, *Staphylococcus pyogenes citreus*, shaping yellow shaded provinces was likewise recognized and included. Winslow incorporated the *staphylococci* to the Micrococaceae family in 1920. In 1957, Evans decided their capacity to anaerobically fermentate glucose and ordered this family as *Staphylococcus*. A while after, Winslow distinguished a second sort known as *Staphylococcus epidermidis*. Until 1972, *S.aureus* was the main known sort. Its principle contrast from *Staphylococcus epidermidis* is its capacity to produce coagulase.

The third sort, in particular the *S. saprophyticus* was added to the *staphylococci* in 1974. The quantity of recognized sorts was 13 in 1980 and rose to 20 in 1984. Aside from *S.intermedius* and *S.hyicus*, the greater part of the newfound sorts are coagulase negative (Şardan, 2000; Tang et al., 2007).

1.7.2 Characteristics

The term *Staphylococcus* was initially utilized by Scottish specialist, Alexander Ogston, in light of their trademark group like appearance under magnifying instrument, and was gotten from the words "staphyle" which implies grape cluster and "coccus" which implies round piece in Ancient Greek.

Staphylococci are recorded as individuals from the *Staphyloccaceae* family in the last release of Bergey's Manual of Systematic Bacteriology (Tang et al., 2007).

However as per more seasoned sources, *Staphylococcus* species are individuals from Micrococaceae family which are gram-positive, cocci formed, still microbes which don't deliver spores and have distances across somewhere around 0.5 and 1.5 µm, furthermore have facultative anaerobic, catalase positive properties (Tükel & Doğan, 2000).

Staphylococcus class are gram-positive, facultative anaerobic, non-spore creating, still and catalase positive microscopic organisms. This variety incorporates no less than 28 species (strands) and 32 sub-species (sub-strands) (Milci & Yaygın, 2006).

Staphylococci contain guanine and cytosine (G-C) in low rates (30-39% mol). Nonetheless, their Micrococcus sort individuals contain G-C at levels achieving 68-74% mole. *Staphylococcal* cell divider is thick (30-60 nm) and has a run of the mill gram-positive microorganism structure.

The cell mass of *S.aureus* has a thickness of 120 nm and is made out of peptidoglycan, teichoic corrosive and proteins. Among these parts, proteins contain fibronectin, fibrinogen, laminin and collagen which are critical for official to eukaryotic cells and attachment. With the official of bond proteins, bacterial sticking to tissues happens. The best antigenic protein will be Protein A which is available in 90-98% of *S.aureus* strains (Zipp & Aktas, 2006).

Staphylococcal catalysts can be recorded as; coagulase, hyaluronidase, Fibrinolysin and Deoxyribonuclease (DNase) created by *S.aureus*; catalase and penicillinase delivered by all *staphylococci*; and lipase delivered by *S.aureus* and also some coagulase negative *staphylococci*.

Coagulase changes over fibrinogen to fibrin and accommodates the bunching of microbes. Hyaluronidase accommodates the spreading of microorganisms all through the tissue.

Staphylococci are mesophyll life forms. They can produce (replicate) at temperatures between 20-40 °C (Tunail, 2009). *S.aureus* is a circle or oval molded (0.5-1.5 µm measurement), gram-positive, still, sans spore, more often than not without container, facultative anaerobic, catalase positive, oxidase negative microorganism which ideally produces at 37°C'. The base and greatest temperature essential for them to create poisons is marginally higher and is between 10 to 48°C. Cells frame exclusively or in couples or in grape pack like bunches (Madigan et al., 2009).

Staphylococcus aureus, which has a place with the family, is profoundly touchy to all applications utilized for the lessening of microorganisms, including and particularly, to warmth applications (Milci & Yaygin, 2006).

Staphylococci are a standout amongst the most safe microorganisms to encompassing conditions and disinfectants among the without spore microbes. They can be safeguarded for 2 to 3 months at a temperature of 4°C, and for 3 to 6 months at a temperature of 20°C in society structure. At a temperature of 60°C they can keep going for 30 minutes of preparing (Kloos & Bannerman, 1995; Sagdic et al., 2008).

The warmth resistance of *S.aureus* differs as indicated by the surface and attributes of the sustenance item it territories. For instance, its warmth resistance in milk is resolved to be 3,1 to 3,4 minutes at 60 °C (Tunail, 2009). They make imperviousness to anti-infection agents quickly. They dispose of the impact of penicillin as an results of their penicillinase property (Kloos & Bannerman, 1995; Sagdic et al., 2008).

Staphylococci produce heat stable enterotoxins which cause nourishment harming in people. *Staphylococcal* enterotoxins (SE) are single-strand proteins which contain a lot of lysine, tyrosine, aspartic and glutamic corrosive and which have a sub-atomic weight shifting somewhere around 26900 and 29600 Da. (Milci & Yaygın, 2006).

Staphylococcal enterotoxins are single-strand proteins with low sub-atomic weight (26-34 kDa) which can be created during all periods of expansion yet are for the most part delivered in the center or toward the end of the exponential stage.

They are impervious to proteolytic compounds, for example, pepsin, trypsin, chymotrypsin, rennin and papain, and are moderately impervious to warm (Balaban & Rasooly, 2000).

They require temperatures between 10°C to 48°C to create poisons. With a specific end goal to deliver poisons in sustenance, their base pH necessity is somewhat over their vegetative multiplication prerequisites (9-5.1 pH). A comparative circumstance is available for water action esteem also. The base water movement esteem for oxygen consuming multiplication is 0,83-0,86; and the base water action esteem for anaerobic proliferation is 0,90. Nonetheless, they require higher water movement qualities to create poisons (Tunail, 2009).

The enterotoxin known not the vast majority of nourishment poisonings is the SEA (Tsai & Li, 2009). *Staphylococcal* enterotoxins have an amazing imperviousness to gamma radiation. *Staphylococcal* enterotoxin based nourishment poisonings get to be symptomatic inside 30 minutes to 8 hours taking after inebriation. The side effects are sickness, the runs, heaving, stomach issues and weariness (Gomes et al., 2007).

There are different strands separated from *S.aureus* that produce enterotoxin. These are *S.haemolyticus*, *S.xlosus*, *S.equorum*, *S.lentus*, *S.capitis* and *S.intermedius*.

Staphylococci proliferation on fluid media frame a deposit and haziness. They can without much of a stretch replicate in oxygen consuming and anaerobic conditions on strong media. Gram positive staphylococci can indicate gram negative properties in experienced societies. *S.aureus* structure a brilliant settlement on blood agar medium and consequently the name of its strand sort was gotten from the Latin word aureus which implies gold. *S.aureus* frame level, glossy, roundabout curved molded states on nonselective medium. *S.aureus* show imperviousness to a few chemicals, for example, telluride, mercury, chloride, sodium azide and a few anti-infection agents, for example, neomycin and polymyxin.

The property isolating *S.aureus* from different sorts is their capacity to fermentate glucose and produce α -poison in anaerobic conditions.

The human body is really the characteristic hotspot for *S.aureus* strands, which cause numerous contaminations, (for example, skin and tissue diseases, bacteraemia, poisonous stun disorder, and endocarditis) and sustenance harming (Demir et al., 2003).

S.aureus is a saprophytic microorganisms which can be distinguished all in all at a rate of 50-30% and at a rate of 20% in nostrils. (Tomi et al., 2005).

While it is prevalently a nosocomial microscopic organisms, *S.aureus* can likewise exist commensally in human skin verdure (Tunail, 2009).

As far as overall population wellbeing, enterotoxin creating staphylococci are the most vital subject of study. The explanation behind this is the way that entero-toxigenic *staphylococci* cause poison based nourishment harming. Sustenance harming is brought about by poisons created by microscopic organisms and, as this synthetic action does not bring about any detectible changes in foodstuffs, purchasers don't understand that they are eating tainted nourishment. *S.aureus* and coagulase positive *S.hyicus* and *S.intermedius* are prevalently in charge of most *staphylococcal* nourishment poisonings. In any case, the way that the enterotoxin incorporating capacity of the other two sorts are much lower than *S.aureus*, this microorganism emerges among other staphylococci regarding their part in sustenance poisonings.

Notwithstanding nourishment harming, *staphylococci* cause different ailments in both people and creatures. They can be the reason for sicknesses, for example, contaminated injuries on skin, maternity fever, meningitis and septicaemia. Additionally, it is assessed that half of the mastitis cases recognized in creatures are at present brought about by *staphylococci*.

This microorganisms can infiltrate into milk and accordingly to dairy items if the fundamental cleanliness measures are not taken at the draining regions. They can bring about nourishment harming, queasiness, retching and looseness of the bowels as a consequence of the vast measure of enterotoxin they deliver.

Creature carers and draining faculty can likewise specifically exchange these microscopic organisms to different people. *S.aureus* is an especially essential microorganism in dairy part (Tunail & Köşker, 1989).

S.aureus can likewise be exchanged to sustenance by cross pollution and can stay alive even after warmth treatment. In the event that enterotoxins have as of now been produced in nourishment, inactivation of these poisons won't not be conceivable through applications, for example, heat treatment. While it is at first conceivable to dispense with microbes in dairy items by purifying, it is likewise conceivable that microorganisms infiltrate the item during alternate periods of generation through cross defilement. Cross tainting can happen through human hands, devices and apparatus, air, added substances, water, and so on. A while later, if the appropriate conditions get to be available, the microorganisms begin producing enterotoxins. Specifically, *S.aureus* strands which enter dairy items after purification are known not and begin producing poisons much quicker.

1.7.3 Food Poisoning by *Staphylococcus Aureus*

Food poisoning has been depicted by the World Health Organization as an illness which happen as a consequence of expending polluted sustenance or water. 250 unique sorts of harming have been accounted for nourishment harming which is recorded among the most critical sicknesses influencing the world when all is said in done. It has additionally been accounted for that 2/3 of these diverse sorts of harming have been brought about by microscopic organisms. *Staphylococcal* sustenance harming is realized by the presence of *staphylococcal* enterotoxins in nourishment (Le Loir et al., 2003; Kumar et al., 2009). More than 60-90% of nourishment poisonings are brought about by microscopic organisms present in nature. Bacterial sustenance poisonings are isolated into two gatherings on the premise of how they influence human living being. The two gatherings are called toxi-contamination sort nourishment poisonings and dangerous sort sustenance poisonings.

The microbes which incite nourishment harming through inebriation, as such those which cause sustenance harming through multiplying and emitting exotoxin inside nourishment are *C. Botulinum* and *S.aureus*.

Moreover, the microscopic organisms whose own mass thickness, at the end of the day, whose own presence and their endotoxins, cause food poisonings advancing with indications of gastroenteritis are *C. perfringens*, *B. cereus*. The microscopic organisms which cause sustenance harming through disease are *Salmonella* spp.

Shigella spp., and *E. coli*. Furthermore, there is *Proteus* spp. furthermore, *Pseudomonas* spp. which are known not nourishment harming yet their etiology has not yet been clarified (Belitz et al., 2009).

The expansion and poison era of *S.aureus* strains are especially quick particularly on the off chance that they are exchanged to sanitized dairy and dairy items after purification. The explanation behind this is the microorganisms which contend with *S.aureus* microscopic organisms are as of now pulverized during purification, totally leaving the medium to *S.aureus* strains which have been exchanged after sanitization.

Different staphylococcus sorts and numerous biotype *S.aureus* strains are exchanged to dairy from the creature during draining. Specifically, dairy obtained from creatures with mastitis constitute a vital hotspot for enteropathogenic *S.aureus* strains. While the fundamental wellspring of exchange are people and creatures, the microorganisms can likewise be exchanged from soil, water sources, septic water, vegetable surface, clean and air, where these microorganisms exist. Moreover, the exchange from defiled creatures to solid ones through the draining gear is unavoidable (Demiret, 2000).

As a rule, crude dairy as of now contains toxigenic *staphylococcus* impervious to penicillin and if cheddar is delivered under any good conditions to their multiplication, poison emission begins. At the point when milk is warmed to 25-30 °C during cheddar creation, a perfect medium is set up for *staphylococcal* poisonous impact if the microbes in the starter society don't get to be dynamic on time and begin delivering acids. The harmful impact turns out to be considerably quicker with low rate of mugginess misfortune (Ünlütürk et al., 1991).

The low pH benefit of coagulating milk additionally accommodates the *staphylococcus* to multiply inside a brief period. In this appreciation, deficiently purified milk is perilous. Inferable from their various properties diverse cheddar sorts have contrasting inclinations as far as *staphylococcal* development.

The issue of creation security turns out to be much more imperative as the expansion of toxigenic *staphylococcus* in the milk utilized for cheddar generation can without much of a stretch cause nourishment harming. All studies went for upgrading creation wellbeing for cheddar depend on the standard of satisfactory sanitization of milk. While this technique prompts moderate development of cheddar, it counteracts bacterial tainting. At first purification qualities were kept low to ensure the essence of milk. Notwithstanding, it has been resolved that such sanitization was insufficient for slaughtering all pathogens.

As a result, fifteen seconds of purification at 66 °C is adequate for staphylococcus to kick the bucket. Notwithstanding, for an ensured result it is typically prompted that purification is connected for 15 seconds at 68 °C. In addition, in a few nations 15 seconds at 71 °C is adjusted as common routine of sanitization. This purification standard is, truth be told, adequate to slaughter *Streptococcus pyogenes*, *Salmonella*, *Brucella abortus* and *S.aureus*. Nonetheless, *Streptococcus faecalis* may stay alive at these warmth treatment values. Aside from *Streptococcus pyogenes*, all the above referred to microscopic organisms can proceed with their movement for various periods in different cheddar sorts.

As the sanitization of milk at low temperature and for brief periods upgrade profitability, these standards are favored in most dairy ranches.

Staphylococcal poisonings exuding from cheddar utilization are seen every once in a while and the examination of these cases demonstrate that *S.aureus* multiplication and era of poison happens during the assembling phases of cheddar. The moderate movement rate of the starter society added to dairy during cheddar creation and the subsequent low corrosiveness (0,4% or lower) readies an appropriate medium for *S.aureus* development.

The multiplication of the microorganism keeps during the sifting of curd (4-5 hours or more) and its number can achieve sums adequate for poison era, (for example, 10⁷ CFU/g).

Notwithstanding, the presence of oxygen inside the medium hinders poison era. Regardless, the way that low *S.aureus* sum is distinguished in a cheddar does not as a matter of course mean the nonattendance of poison.

S.aureus which multiplies to numbers adequate for poison era during assembling, can in time diminish in number inside the host cheddar. With a specific end goal to keep *staphylococcal* harming under control, adequate warmth treatment, guaranteeing ordinary starter action and the execution of a decent sanitation system is essential (Ünlütürk et al., 1991).

In Turkey, dairy and dairy items are generally delivered in little foundations and dairy ranches without any observation or control components. This circumstance definitely expands the danger of contamination and nourishment harming exuding from devouring milk and dairy items. In the TRNC the same expanded danger is available for the very same reasons. It has been controlled by numerous studies that *S.aureus* is all the time present in milk and dairy items delivered in Turkey.

In a study completed on sanitized milk sold in Ankara, coagulase positive *staphylococcus* presence at numbers above 1,30 log CFU/ml level were distinguished in 44% of the specimens. It ought to be noted, in this connection, that inadmissible keeping conditions convey genuine dangers regarding general wellbeing as they may bring about the increase of microorganisms (Demiret, 2000).

S.aureus ought to ideally be not present or be available in little sums in cheddar and other milk items. In Turkey, all phases of generation, from the creature giving milk to the dispersion and protection of the item ought to be kept under control with the important measures in place, keeping in mind the end goal to maintain a strategic distance from the financial harm, including the loss of labor, experienced *S.aureus* poisonings brought on by milk and dairy items (Demiret, 2000). The same circumstance and the same needs apply to the TRNC.

S.aureus, is a microbes not welcome in sustenance. Its presence in straight forwardly expended sustenance items, for example, cheddar ought not really be acknowledged or endured. Be that as it may, its presence in low sums in cheddar is permitted as a general worldwide standard in accordance with assembling innovation conventions. As needs be, both TS 591 and Turkish Food Codex (Anonymous 2011), refer to n c M values as 5; 2; 1,0x10² CFU/g; 1,0x10³ CFU/g separately.

1.7.4 Importance To The Consumer

In *Staphylococcal* nourishment harming cases manifestations as a rule advance quickly. The indications set off inside 1-6 hours taking after inebriation. Overall side effects progress inside three hours. The transcendent and genuine manifestation is retching and it takes after extreme sickness. Different indications are stomach torment, fever, unsteadiness, shuddering, migraine, stomach issues and looseness of the bowels. Indications as a rule vanish inside 1-2 days of determination and in spite of the fact that demise rate is amazingly low, cases coming about with death have been accounted for (İşeri & Erol, 2009)

It has been accounted for that, when all is said in done, the presence of no less than 1µg poison in 100g nourishment is fundamental for sustenance harming to happen as a consequence of devouring tainted sustenance.

Notwithstanding, it has additionally been accounted for in a sustenance harming case brought on by chocolate milk amongst school youngsters that exclusive 0.2 µg SEA set off the harming (Le Loir et al., 2003).

1.7.5 Analysis of *Staphylococcus Aureus*

Exploration to grow quicker and more touchy techniques for microbiological investigation of foodstuffs is a constant exertion. While the hindrance of aggressive greenery at the most elevated amount is focused for expanding affectability, it is a prerequisite to a specific degree that the microscopic organisms is not hurt through this procedure. In like manner, the distinguishing proof of the microorganism using different chromogenic or fluorogenic substrate is additionally connected notwithstanding repressing focused greenery (Baron et al., 1995; Anonymous, 2011)

1.7.6 The Factors That Affect The *Staphylococcus Aureus*

Contamination Level

In polluted sustenance, *S.aureus* produced poisons at a sum under 1.0 µg is adequate to set off the manifestations of staphylococcal inebriation.

The adequate measure of poison for bringing about nourishment harming is a subject of discourse and the base sum fluctuates as indicated by poison sort. On account of *S.aureus*, it is accounted for that the era of adequate poison sum for inebriation is achieved when the *S.aureus* tally is more than 100.000 CFU/g-mL. As such, if the *S.aureus* number is 5x10⁵ CFU/g-mL inside a nourishment item, it is certainly unsafe. In any case, a low *S.aureus* tally in a nourishment item does not as a matter of course imply that it is sheltered (Tükel & Doğan, 2000).

pH and NaCl

The ideal pH esteem for the era of enterotoxins is between pH 6.0-7.0. In contrast with SEB era, the era of SEA is more tolerant to pH varieties. In contrast with salt free conditions, 5% NaCl fixations increment *S.aureus* expansion rate. Then again, NaCL focuses at 7.5% and 10% levels are known not development rate to a specific amplify (Erol & İşeri, 2004).

Temperature

The ideal temperature for *S.aureus* development is 37°C, while the ideal temperature for enterotoxin era changes between 40-45°C (Erol & Işeri, 2004).

Competitive Property

Staphylococci are effectively restrained by different microorganisms in blended societies. Enterococcus, Lactococcus and Leuconostoc are known not vital hindering microscopic organisms. What's more, E. coli, Pseudomonas, Serratia and Aerobacter additionally have a hindering impact on *S.aureus* development (Sagdic et al., 2008).

1.7.7 Methods of Distinguishing of *Staphylococcal* Enterotoxins

The distinguishing proof of enterotoxins in foodstuffs is corresponding to the measure of enterotoxin vital for bringing on sickness in people. This dosage is 100-200 ng (FDA, 2001). Different strategies for immunologic and serologic examination have been created for the distinguishing proof of *staphylococcal* enterotoxins in nourishments. The immunologic examination strategies are touchy and depend on the particular distinguishing proof of enterotoxins. Be that as it may, the ID of certain uncharacterized *staphylococcal* enterotoxins is done on the premise of creating the extraordinary emetic exercises discovered just in monkeys. It has been accounted for that in half of youthful Rhesus monkeys, 5-20 µg poison sum created emetic response (List Biological, 1999).

Table 1.3: Examples of Intoxications Where Only Enterotoxin was Detected Because the Cheese was Heat-Treated (Wieneke et al., 1993)

Product	<i>Staphylococcal</i> Cells and Enterotoxin Detected.	Reference
Soft Raw Milk Cheese	From one batch of cheese; $>1.8 \times 10^7$ CFU g ⁻¹ of CPS was detected in cheese sampled from one outbreak, while $<10^2$ CFU g ⁻¹ was detected in another outbreak where the cheese had been cooked before eating. SEE was detected in samples from both outbreaks.	(Ostyn et al. 2010)
Halloumi Cheese	SEA was detected in cheese and brine, but <i>S. aureus</i> could not be found. During manufacture, Halloumi has a cook step following the pressing of curds which would kill staphylococcal cells.	(Wieneke et al. 1993)
Cooked (non dairy) Produce from Food Outlets	Eight outbreaks associated with food outlets tested positive for <i>staphylococcal</i> enterotoxins, with <10 CFU g ⁻¹ CPS detected.	(Wong, 1996)

1.7.8 Immunologic Strategies

Radioimmunoassay (RIA), is a broadly utilized technique for enterotoxin distinguishing proof as a part of society filtrates and sustenance extricates. This strategy depends on the contending of an unlabelled poison in the specimen with a standard radioactive named poison to the grip parts of counter acting agent atoms. This strategy can for the most part distinguish fast (3-4 hours) poisons under 1-10 ng/g level.

Its detriments can be recorded as non-particular responses, the requirement for exceptionally decontaminated enterotoxins, unfriendly impacts that may emerge during the marking of immune response epitopes, the short half-life time of radioisotopes, the hurting impacts of radionuclides to human wellbeing and the requirement for costly recognizable proof hardware (Brett, 2006).

1.7.9 Control Measures

- Keep hot food hot and cool food chill. Try not to keep food in the temperature peril zone (i.e. at or underneath 5 or more 60°C) any more than would normally be appropriate
- Keep raw and cooked food separate
- Keep kitchen and utensils clean
- Wash and dry hands appropriately
- Cover any cuts or bruises when taking care of food

CHAPTER 2

RELATED RESEARCH

Halloumi is the Cypriot traditional cheese and also famous in the regional countries such as Lebanon and the most popular in its category. The popularity of this cheese has increased in the recent years (Papademas, 2006). The Halloumi cheese can either be consumed fresh or after matured in whey brine. Salt is one of the most significant actor in the production of this cheese as it holds a role for the taste and the preservation of the cheese. The customer satisfaction is well achieved as the salt reduces the level of bitterness in the cheese and increases the level of other flavours (Kilcast & Angus, 2007). In order to concentrate and preserve the milk, humans have produced cheese for long time as this fact makes cheese one of the most historical food manufactured.

Which its level of production and export have increased significantly, contributing to agricultural exports. The production of this cheese has accumulatively increased initially and notably in 1999 to 4730 tons and 6600 tons by the year 2002 (Papademas, 2006).

The cheese is white in color and holds a rather distinctive texture; like mozzarella, also is salty in flavour. The Halloumi cheese can be stored at frozen conditions of below -18°C (0°F) and defrosted to $+4^{\circ}\text{C}$ (39°F) as it is preserved in salty water (natural juices) up to a year. A traditional garnishing method is placing a mint leave between the layer of cheese as it is believed to enrich the cheese with more flavoursome taste and keeps it more fresh. Thus, the producers place mint leaves to the surface of the cheese within the packages.

Approximately the traditional halloumi cheese is a large wheel in size with a semicircular shape that weighs around 220–270 grams. The cheese contains approximately 17% protein and 25% fat when wet and 47% when dry. The cheese squeaks when chewed due to its firm texture. As mentioned before the unpasteurised form of sheep and goat milk is used to produce the traditional halloumi. Differently to the cheese found in the West, the aged halloumi, which is mostly favoured, is much drier and saltier in taste; as kept in brine.

As the halloumi cheese matures the level of saltiness and acidic flavour increases and the texture becomes hard. The cognoscenti do mostly appreciate this variation of the halloumi cheese more (Papademas & Robinson, 2000).

Approximately the size of exports from Cyprus for the halloumi cheese have increased upto 2500 metric tonnes as the popularity of the cheese has exceeded the borders of Cyprus to an international level (Papademas & Robinson, 2001; Papademas & Robinson, 2002).

This increased demand is met by the allowance of Cyprus to the manufacturer to produce the Halloumi cheese from bovine milk (Papademas & Robinson, 2001).

Considering the fact that the drained curd obtained in the scald of 90-95 degree as the ground water, plays a distinctive role in halloumi in contrast to other cheeses. For a long period of time the mixtures of milk from various kinds of cheese has been detected as subject of interest. Various techniques of analysis such as chromatographic techniques (de Frutos et al., 1991), electrophoretic techniques (Amigo et al., 1992), DNA-based techniques (Plath et al., 1997; Klotz & Einspanier, 2001) or Western blotting techniques (Molina et al., 1996) were used for the description of the qualitative assessments of ewe's milk cheeses containing minimal levels of cow's milk; were described mostly as methods in the literature. A method using the isoelectric focusing γ -CN following the plasminolysis (European Commission, 1996) as a reference, for the detection of bovine caseins in cheeses, have also been used in cow's milk found in the Halloumi cheese (Kandarakis et al., 1996). 1% of cow's milk was detected in the haloumi prduced from the ovine milk subecuent to the analysis of γ -CN post plasminolysis by HPLC also, through the analysis of α S1-CN have detected cow's milk in the ovine. Despite this, there is no apparent method to detect goat's milk in the Halloumi cheese. Isoelectric focusing on whey proteins (Amigo et al., 1989; Molina et al., 1996) or methods such as immunological through the usage of polyclonal (Rodríguez et al., 1994) or monoclonol antibodies (Haza et al., 1999) to goat proteins were exercised to detect goat's milk in cheese which were produced apart from caprine. A method of capillary electrophrosis is being used over the last years to decompose and analyse the fractions of casein compositions of cow's, goat's and ewe's milk. Following the statistical results gathered, binary and ternary milk compositions were identified and quantitatively determined thus allowing the observation of differences occured between the CE patters of casein (Molina et al., 2000).

In 1985 the Standards Committee of the Ministry of Commerce and Industry of Cyprus had defined the standards of the Halloumi cheese. This cheese reaches approximately a tonne in total export annually (Papademas & Robinson, 2000). Despite the fact that the initial production of the halloumi cheese was of small ruminants' milk, due to the increase in the cow's milk production this was altered to a mixture of mostly cow's milk and either sheep's or goat's milk.

Counter to the halloumi made from the sheep's milk, the color is more yellowish and the taste is rather different from the original sheep's milk halloumi (Kaminarides et al., 2000).

The halloumi has no rind and is semi-hard as it is preserved in brine. Further description to the manufacturing of the halloumi is made by (Kaminarides et al., 1995).

Distinctively to the manufacturing of other forms of cheeses, the Halloumi cheese is different in taste and texture due to its method of being cooked in curd blocks approximately 30 cm³ using whey with non-protein elements and cooked for 30 minutes, at least at 93-95 °C (Papademas & Robinson, 2000). Alkaline phosphatase and lipoprotein lipase (LPL) are destroyed due to the method of cooking the cheese curd thus eliminates the rennet and in large scale, formed in microbial flora, this otherwise contributes the cheese to ripen (Kaminarides et al., 1995).

In a study emphasized on the differences in sensory and textural characteristics of cheese curd being cooked in separate time lapse, 30 and 60 minutes at 93-95 °C. This is considered to be less economic process.

An experiment was conducted with a Chios breed 40 sheep's milk undertaken in Agricultural University of Athens. These sheep were mainly had fed on 74% maize, 7% sunflower, 5% rice bran, 6% molasse and 3% soybean, and also alfalfa hay. Presents the process of making Halloumi cheese from sheep's milk. The cheese curds, while uncooked, were cut into four blocks; quarter of a kilo each, these blocks of cheese curd are resembled as sample A. Two subsequent blocks of cheese were divided.

Sample B consists of 38 blocks all cooked for 30 minutes under 93–95°C in non-protein whey as the other 38 blocks were cooked under same conditions but for 60 minutes; Sample C. Four trials were undertaken for the production.

CHAPTER 3

MATERIAL AND METHOD

3.1 Halloumi Sampling of *Salmonella*

In this study, 34 tests of different day by day halloumi items collected from business sectors in the Nicosia territory in the TRNC have been utilized as material. Halloumi items have been gathered randomly specifically from the business sector stands, transported to lab environment by means of unbroken cool chain and have been broke down inside a brief span for *Salmonella* presence.

3.1.1 Media and Test Kids Utilized For The Isolation and Identification of Bacteria

Peptone Water (BPW, Oxoid CM 0509) It has been utilized for dissolved during the separation stage.

Segments

- In sterile stomacher packs 500's
- Buffered Peptone Water 225 ml
- pH 7.2±0.2

25 gr of sample was weighed and homogenized with 225 ml of pH-adjusted pepton water(BPW, Oxoid CM 0509), then transfered into stomacher bag and incubated at 37 °C for 24 hours.

Mc Conkey Agar

It is the particular medium utilized for the identification of *Salmonella*.

Segments

Rappaport-Vassiliadis Soya Peptone Broth (RVS, Oxoid CM 0866) 10 ml

Muller-Kauffmann Tetrathionate/novobiocin Broth (MKTTn, Oxoid CM 1048) 10 ml

Strategy

Initial Step: 25 gr of cheese sample was homogenised with 225 ml of sterilized pepton water (BPW, Oxoid CM 0509) for 1 minute and then incubated at 37 °C for 24 hours in a stomacher bag.

Second Step: This 0.1 ml 10 ml of pre-improvement fluid Rappaport-Vassiliadis soya peptone juices (RVS, Oxoid CM 0866) to tubes containing 1 ml in 10 ml of Muller-Kauffmann tetrathionate/novabioic Broth (mkttn, Oxoid CM 1048) which will be immunized into tubes.

Third Step: RVS tube at 42°C, while mkttn tubes in the wake of being brooded for 24 hours at 37 °C after hatching time end McConkey from every tube and SS agar exchanging on loaded with a circle with a round-finished circle, 37 °C, will be left for 24 hours brooding.

Last Step: Then McConkey agar yellow in shading and dark in light of SS agar, *Salmonella* suspect states will be recognized utilizing the BD Phoenix gadget.

Evaluation of Test Results

It was reasoned that from the halloumi tests, no decided defilement rate was seen Salmonella.

3.3 Halloumi Sampling of *S.aureus*

In this study, 34 halloumi test samples on different days which were gathered from markets sectors in the Nicosia zone in the TRNC have been utilized as material. Halloumi items have been taken randomly straightforward from the business sector stands, transported to research laboratory environment by means of unbroken cold chain and have been broke down inside a brief span for *S.aureus* presence.

3.3.1 Media and Test Kids Utilized For The Isolation and Identification of Bacteria

Peptone Water (Oxoid CM0009)

It has been utilized for dissolved during the detachment stage.

Parts

- Peptone 10.0 g
- Sodium Chloride 5g
- Distilled Water 1000 ml
- pH 7.2±0.2

15 g of prepared mix from the medium has been dissolved in 1000 mL refined water and set at pH 7.2±0.2. The readied medium base has been appropriated to 9 mL tubes (second dissolved) and 90 mL bottles (first dilution) and has been left to cool after autoclave cleansing at 121°C for 15 minutes.

Baird-Parker (BPA) Agar Base (Oxoid CM0275)

It is the specific medium utilized for the ID of *Staphylococcus aureus*. (FDA/BAM, 2001).

Parts

- Tryptone 10.0 g
- Lab-Lemco powder 5.0 g
- Yeast Extract 1.0 g
- Sodium Pyruvate 10.0 g
- Glycine 12.0 g
- Lithium Chloride 5.0 g
- Agar 20.0 g
- Distilled Water 1000 ml
- pH 6.8±0.2

Egg Yolk-Tellurite Emulsion (OXOID SR0054 C)

50 ml Egg Yolk Tellurite Emulsion contains approximately 3 ml 3.5% potassium tellurite.

Staphytest Plus (OXOID DR 0850) Latex Agglutination Test Kit

Parts

DR851M Staphytest Plus Test Reagent (5,6 ml)

Blue latex particles covered with both porcine, fibrinogen and rabbit IgG together with particular polyclonal antibodies raised against capsular polysaccharide of *S.aureus*. Every container contains adequate reagent for 100 tests.

DR852M Staphytest in addition to Control Reagent (5,6 ml) Blue unsensitised latex particles. Every container contains adequate reagents for 100 tests.

DR500G Reaction Cards. There are 35 expendable response cards.

Strategy

Initial Step: Bring the latex reagents to room temperature. Ensure that the latex reagent is blended by incredible shaking and remove any latex from the dropper pipette for complete blending.

Second Step: Dispense 1 drop of test latex onto one of the circles on the response card and 1 drop of control latex onto another circle.

Third Step: Using a circle, get and spread what might as well be called 5 normal measured suspect *Staphylococcal* provinces (identical to 2-3 mm breadth of development) onto a circle from a society media plate and blend this in the Control Latex reagent. Spread to cover the circle. Dispose of the circle properly.

Fourth Step: Using a different circle continue similarly with the Test Latex.

Last Step: Pick up and shake the card for up to 20 seconds and watch for agglutination under typical lighting conditions. Try not to utilize an amplifying glass.

Evaluation of Latex Test

An outcome is sure if agglutination of the blue test latex particles happens and a smooth blue suspension stays following 20 seconds. This hypothetically distinguishes the strain as *S.aureus*.

A negative result is acquired if no agglutination happens and a smooth blue suspension stays following 20 seconds in the test circle. This possibly distinguishes the strain as non *S.aureus*.

Slight graininess of the test latex joined by no adjustment in the presence of the control latex ought to be translated as an obscure result. Strains ought to be re-tried after subculture onto non-specific media.

The test is uninterpretable if the control reagent demonstrates agglutination. This shows the way of life causes auto agglutination.

Constraint of The Procedure

The propensity of detached settlements to bring about auto agglutination increments with brooding times past the prescribed 36 hour duration.

The immune response utilized as a part of Staphytect Plus has been upgraded to dodge potential cross-responses with shared antigens from coagulase negative *staphylococci*.

Planning

63 g prepared blend from the BPA medium has been dissolved in 1000 mL refined water, set at pH 6.8 ± 0.2 and totally broke down in high temp water shower. After autoclave disinfection at 121°C for 15 minutes, it has been cooled to 50°C and after that 50 mL Egg-Yolk Telluride Emulsion (Oxoid SR 0054, it contains potassium tellurite and is in ready 100 mL fluid structure) has been included and blended. The readied arrangement is poured onto clean petri dishes and left to cool at room temperature.

3.4. Isolation and Identification of *S.aureus*

3.4.1 Cultivation in Strong Medium Base and Assessment of Settlements

The distinguishing proof of coagulase positive *staphylococci* by exemplary system has been done as per FDA/BAM (2001). 10 g test has been taken in a way to speak to the entire of the specimen item and put in stomacher pack. At that point, 90 mL peptone water has been included and homogenized in stomacher for 60 seconds. 1 mL arrangement was taken by pipette from the homogenate (10 - 1) and similarly conveyed and developed on pre-arranged BPA medium plates by spread plate system. The petri dishes were, then, left for hatching in anaerobe environment at $35 \pm 1^{\circ}\text{C}$ for 24-48 hours. The same strategy and technique has been rehashed for each of the 34 tests.

Toward the end of the hatching time frame, the sparkling dark dim settlements with a measurement of 2-3 mm and hovered by limited, curved molded, smooth, gleaming zones were considered as plausible *S.aureus* states and checked.

3.4.2 Confirmation

Latex affirmation test was connected to likely *S.aureus* strains which multiplied as an results of BPA medium development. Latex reagent has been conveyed to room temperature and homogenized by shaking. A short time later, one drop of test reagent has been poured on each test curls on the response card; five suspicious settlements were taken from the petri dish with sterile circle; and were spread over the test loop whereby they blended with the test reagent. The response card has been shaken in roundabout developments for 20 seconds and watched for agglutination.

As agglutination was seen by the testing of all settlements, the outcome was viewed as positive. We continued to the second period of affirmation and rehashed all the previously mentioned stages with a control reagent.

The testing of the investigated colonies brought about agglutination with latex test reagent, however no agglutination was seen with the control reagent. It was, hence, affirmed that the confined *S.aureus* states were, truth be told, coagulase positive strains.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Result of *Salmonella*

As a result of the analyses of 34 halloumi product samples produced in different manufacturing facilities in the TRNC and collected from markets in the Nicosia area, it has been no detected of the 34 Hellim Cheese samples for *Salmonella*.

Table 4.1: BD Phoenix Device Test and Culture Results of the Halloumi Products Produced in TRNC and Sold in Nicosia Area.

Label Sequenc e No.	<i>S.aureus</i> kob ^(iv) /g	<i>E.coli</i> kob ^(iv) /g	Coliform Bacteria kob ^(iv) /g	Yeast and Mold kob ^(iv) /g	<i>Salmonell</i> <i>a</i> spp. (25 g.)	<i>Listeria</i> monocytoge nes (25 g.)	<i>E.coli</i> 0157(25 g.)
	Limit ⁽ⁱ⁾ :0/g Limit ⁽ⁱⁱⁱ⁾ :1 0 ³ /g	Limit ⁽ⁱ⁾ : 0/g	Limit ⁽ⁱ⁾ : 10 ³ /g	Limit ⁽ⁱ⁾ :1 0 ³ /g	Limit ⁽ⁱ⁾⁽ⁱⁱ⁾ : 0/25g	Limit ⁽ⁱⁱ⁾ :0/2 5g	Limit ⁽ⁱⁱⁱ⁾ :0/ 25g
1	0	0	0	0	Not Detected	Not Detected	Not Detected
2	0	93	93	5x10 ²	Not Detected	Not Detected	Not Detected
3	0	$\frac{\geq 2.4 \times 10^3}{3}$	$\geq 2.4 \times 10^3$	1x10 ²	Not Detected	Not Detected	Not Detected
4	7x10 ³	$\frac{\geq 2.4 \times 10^3}{3}$	1100	5x10 ⁴	Not Detected	Not Detected	Not Detected
5	0	0	0	0	Not Detected	Not Detected	Not Detected
6	0	0	0	4.5x10 ²	Not Detected	Not Detected	Not Detected
7	0	0	0	0	Not Detected	Not Detected	Not Detected
8	0	0	0	0	Not Detected	Not Detected	Not Detected

4.2 Result of *S.aureus*

As a result of the analyses of 34 halloumi product samples produced in different manufacturing facilities in the TRNC and collected from markets in the Nicosia area, it has been determined that; the *S.aureus* amount detected in 2 sample (6%) was within the accepted limits cited in the Turkish Food Codex Regulation on Microbiological Criteria and did not endanger public health and 32 (94%) samples did not contain *S aureus*. *S aureus* colonies were isolated from 2(6%) of the 34 Hellim Cheese samples.

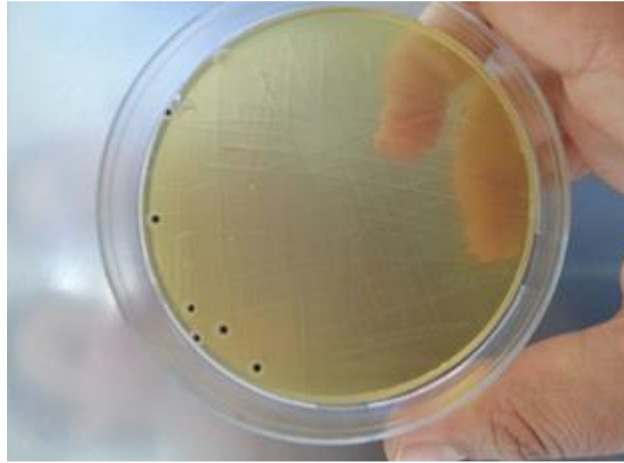


Figure 4.1: Below limit *S.aureus* colonies isolated from Hellim Cheese sample and counted in BPA medium base.

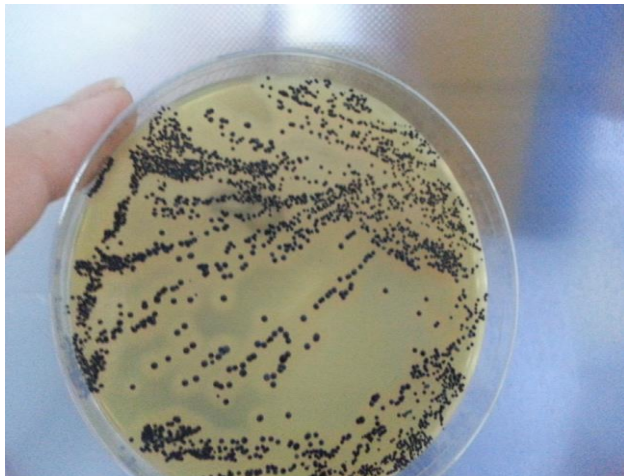


Figure 4.2: Above limit *S.aureus* colonies isolated from Hellim Cheese sample.

4.2.1 Confirmation

It was confirmed by Oxoid Staphytect Plus type latex test that all the suspected colonies from the BPA medium were in fact *S.aureus* colonies. The results of the latex agglutination test are summarised in Table 4.2 according to sample distributions.

Table 4.2: Latex Agglutination Test and Culture Results of the Halloumi Products Produced in TRNC and Sold in Nicosia Area.

[illegible]

4.3 Discussion

The microorganism contamination of foodstuffs can as often as possible happen for various reasons during the planning, manufacturing, packaging, transportation and stocking stages. Accordingly, not only the nature of the food item is undermined creating financial revenue, but also various who consumes these food clinical indications and also food waste can occur in individuals expending the item. Dairy and dairy products constitute an exceptionally appropriate development medium for microorganisms. Microorganisms exchanged to dairy from various sources can increase quickly (Demiret, 2000).

The multiplication and poison production of *S.aureus* strains are especially quick particularly in the event that they are exchanged to purified dairy and dairy items after sanitation of dairy and dairy products. The purpose behind this is the microorganisms which contend with *S.aureus* microbes are now wrecked during sanitation, totally leaving the medium to *S.aureus* strains which have been exchanged after purification.

Microorganism entrance as an result of cross contamination during the preparation, manufacturing, packaging transportation and stocking of food products is a broadly seen case.

S.aureus, is one of the main sources of mastitis in milk delivering creatures. As needs be, *S.aureus* is as often as possible present in the milk of creatures with mastitis. What's more, *S.aureus* can be exchanged to dairy from the draining staff.

It has been resolved that people are the principle wellspring of tainting exchanging *S.aureus* to nourishment in sustenance based inebriations. People go about as a transporter and exchange this microorganisms to different people and nourishments. Be that as it may, the way that this microscopic organisms can without much of a stretch be segregated from air, dust, sewage and water, appears there are a wide range of wellsprings of defilement undermining foodstuffs.

The meat and chicken items instant for utilization; smoked or heat treated items described as shop, for example, ham and salami; cooked egg and products containing cooked egg; baked goods loaded down with meat and meat items, dairy items and, specifically, frozen yogurt and sweet cream; prepared foods, for example, rice puddings, and green vegetable mixtures with mayonnaise; have been recognized as the moving food items in *staphylococcal* intoxication.

The normal properties of these nourishment items are that they are for the most part cooked, arranged by hand and safeguarded in cooler until utilization. Meat and meat items and milk items go about as vital moving nourishment items in *staphylococcal* inebrinations.

In their study completed with 26 tests of Erzincan Tulum Cheese, Özalp and partners (1978), recognized presence of *staphylococcal* enterotoxin, despite the fact that they didn't distinguish presence of *staphylococci* in any of the examples.

In their study completed with custom made cheddar in Chile, Castro and associates (1986), confined 103 *S.aureus* from their specimens. They reported that the coagulase and thermonuclease movement of all specimens were sure, yet no enterotoxin presence was identified in any of the examples.

As a consequence of his study on *S.aureus* defilement in different foodstuffs, Ewald (1987) reported that 20 of the detaches were coagulase negative, and that one of these separates which was distinguished as *S. haemolyticus* could deliver C and D sort enterotoxins in the meantime. He additionally reported that 38 of the 150 disengages distinguished as *S.aureus* were enterotoxic and could deliver it is possible that one and/or two or three A, B, C, D, E sort enterotoxins, and that C sort enterotoxin was the for the most part created poison.

In their study completed with goat cheddar delivered in England, Bone and partners (1989) decided SEA presence, in spite of the fact that they didn't locate any live pathogen microscopic organisms in any of the examples.

Wieneke and associates (1993), decided 359 sustenance harming cases brought on by *S.aureus* enterotoxins between the years 1969 and 1990 in the city London. The investigation they completed on the sustenance items devoured by the said patients decided the normal *S.aureus* number in the examined nourishment tests as 3.0×10^7 CFU/g. Besides, they confined *staphylococcal* enterotoxin from the cheddar tests devoured by two patients who were hospitalized for sustenance harming, albeit no presence of live *S.aureus* microorganisms were distinguished in any of the cheddar tests.

Mutluer and associates (1993) examined the capacity of enterotoxigenic *S.aureus* strains to multiply and deliver poisons in White Turkish Cheese. They investigated white cheddar delivered from purified dairy and white cheddar created from crude milk sullied with *S.aureus* strains (at 10^5 CFU/mL sullying level) creating A, B, C, D sort poisons.

Subsequently, they discovered that the *S.aureus* check in cheddar created from crude milk were higher than the number in cheddar produced using sanitized milk during all periods of generation and developing. Enterotoxin An incitement was seen at the primary day of development in cheddar created from crude milk.

In their study completed in Ankara, Kisa and partners (1996) recognized coagulase positive *staphylococci* presence; in 73.3% of the plain cream cake tests at a normal level of 6.3×10^2 CFU/g, and in the majority of the cacao and organic product enhanced cream cake tests at a normal level of 1.7×10^3 CFU/g. Additionally they distinguished coagulase (+) *staphylococci* consider 105 CFU/g which is at danger level for enterotoxin era. They additionally verified that the coagulase (+) *staphylococci* confined from an aggregate of 25 (26 %) cream cake tests could deliver enterotoxin. Four (36.4%) of the said tests were plain cream cake tests; 12 (22.6%) were cacao seasoned cream cake tests; and 9 (28.1%) were natural product enhanced cream cake tests.

Rosec and partners (1997), confined 213 *S.aureus* strains from 121 nourishment tests they broke down in France. They reported that 15.9% of the *S.aureus* strains separated structure cheddar tests created from crude milk fortified enterotoxin, and that 43% of the *S.aureus* strains disengaged from different nourishments had enterotoxigenic property.

In their study did with frozen yogurt advertised at Bursa commonplace focus, Özcan and Kurdal (1997) decided *Staphylococcus aureus* presence at a normal level of $10,11 \times 10^3$ CFU/g in lemon seasoned dessert; at a normal level of $8,01 \times 10^3$ CFU/g in harsh cherry enhanced frozen yogurt; and at a normal level of $11,46 \times 10^3$ CFU/g in strawberry enhanced dessert.

Erol and associates (1998), completed the microbiological investigation of 100 frozen yogurt tests showcased in Ankara. Accordingly, they decided coagulase positive *staphylococci* presence in 20-30,8% of the specimens at 10^2 - 10^4 CFU/g level, and secluded *E. coli* and *Salmonella* microscopic organisms from 2% of the specimens.

Evrensel and Güneş (1998), did a microbiological study on plain frozen yogurt promoted in Bursa. Thus, they recognized coagulase positive *S.aureus* presence at 1.0×10^2 - 6.3×10^5 CFU/g level.

Rasooly and Rasooly (1998), did a microbiological examination study on different nutrition classes (mushroom, milk, potato plate of mixed greens, frozen yogurt and meat items) sold in the United States of America. They reported that they recognized SEA presence in the specimens at least level of 100 pg/mL.

Toklu and Yaygın (2000) completed a study with 69 dessert tests showcased in the Antalya region and confirmed that coliform was available in 88.40%; *E. coli* was available in 69.5%, and *S.aureus* was available in 49.27% of the specimens. Yücel and Çıtak (2000) did a study with 30 frozen yogurt tests showcased in Ankara and decided the most minimal and largest amount of *S.aureus* presence in the specimens as 1.0×10^2 and 3.0×10^3 CFU/mL individually.

Akineden and partners (2001) completed a study with 103 milk tests promoted in Germany and disengaged *S.aureus* provinces from the greater part of the specimens. They reported that 17 tests contained SEI; 21 tests contained SEG and SI; 21 tests contained SED and SEJ; 15 tests contained SEC+SEG+SEI and TSST-1; and 1 test contained SEA+SEC and TSST-1.

Bostan and Akin (2002) reported that as an results of the study they did in Istanbul, they didn't identify *Salmonella spp.*, *S.aureus* and *E. coli* settlements in any of the 300 bundled frozen yogurt tests they broke down.

Cowel and partners (2002), completed a microbiological investigation of the nourishments devoured by 42 individuals who were hospitalized in Queensland, Australia, for looseness of the bowels and spewing. The underlying examination uncovered SE presence. The further investigations of the chicken, cakes and servings of mixed greens devoured by the patients verified that the *S.aureus* level was $>2.5 \times 10^6$ CFU/g.

Mukan and Evliya (2002) did not separate any *S.aureus* provinces from any of the frozen yogurt tests they broke down in Adana. Be that as it may, they separated coagulase negative *staphylococci* from the greater part of the examples. They recognized the disconnects as *S. epidermidis* provinces and reported the normal microorganisms consider 2.4×10^5 CFU/g.

Alişarlı and partners (2002), completed a study on the capacity of enterotoxigenic *S.aureus* strains to multiply and create poison in cream cakes. Subsequently, they confirmed that Enterotoxin-A creating *S.aureus* strains could invigorate poison even at low temperatures.

Küplülü and partners (2002), completed a study on SE presence in sanitized milk promoted in Ankara and distinguished SEA presence in two of the 250 tried specimens at a level >0.1 ng/mL.

Alişarlı and associates (2003), did a microbiological study with pudding and cream cakes and separated *S.aureus* in 10% of 100 pudding sorts and 27% of the cream cakes. A sort enterotoxin was identified in 7 tests, C sort enterotoxin was distinguished in 2 tests, and A/B blended enterotoxin was recognized in 2 tests. It has additionally been accounted for that the greater part of the enterotoxin delivering *S.aureus* likewise had thermonuclease movement.

Becker and colleagues (2003), carried out a study with patients carrying *S.aureus* bacteria at the Munster University Hospital in Germany. They determined that 50.8% of the *S.aureus* colonies isolated from 429 patients contained exfoliative toxin, whereas 73% contained *staphylococcal* enterotoxin.

Günşen and Büyükyörük (2003), broke down 125 examples of vacuum bundled youthful cheddar advertised in Bursa. Thus, they distinguished *S.aureus* presence in four examples and reported most astounding *S.aureus* consider 1.8×10^3 CFU/g and least *S.aureus* consider 1.0×10^2 CFU/g.

In their study completed in Bursa, Evrensel and associates (2003), distinguished coagulase(+) *staphylococcus* in crude milk tests and decided the microbes consider 1.9×10^4 CFU/mL, recognized *staphylococcus*/micrococcus tally in sanitized milk tests and decided the microorganisms consider 103-104 CFU/mL, and decided coagulase(+) *staphylococcus* consider in salt water 5.0×10^4 CFU/g.

As an results of the microbiological examination of 75 dessert tests promoted in Van, Ağaoğlu and Alemdar (2004), segregated coagulase (+) *S.aureus* in 13.3% of the examples.

Normanno and partners (2005), completed a study on different nutrition types (meat, UHT milk, cheddar, dessert, cake, egg, fish) sold in a business sector in Italy. Subsequently, they recognized 541 coagulase positive *staphylococcus* settlements, 537 of which was distinguished as *S.aureus*. They confirmed that 298 of the *S.aureus* states (%55.5 of the aggregate) contained one or more enterotoxin, and reported the enterotoxin sort circulation as 33.9% SEC, 26.5% SEA, 20.5% SEA+SED, 13.4% SED, 2.7% SEB, 1.7% SEA+SEB, 7% SEC+SED, 0.3% SEA+SEC and SEB+SEC.

Fueyo and partners (2005), detached 269 *S.aureus* settlements from high quality foodstuffs (cheddar, cream, dessert) showcased in Spain, and reported that 57 confines contained no less than one of the four enterotoxins, recorded as SEA, SEB, SEC and SED.

Korel and partners (2005), did a microbiological study with 85 dessert tests promoted in Manisa. They didn't distinguish *S.aureus* in any of the 15 bundled and 70 unpackaged frozen yogurt tests they tried.

In their study on recognizing *S.aureus* on the skin and in the nostrils of patients with Atopic Dermatitis (AD), Psoriasis (PS), erythroderma, skin contaminations and sepsitis, where a solid control gathering was likewise included, Tomi and associates (2005), disengaged *S.aureus* from the sore regions of the skin of 22 patients from an aggregate of 25 patients with AD, and from 15 of 25 patients with PS.

Fujikawa and Morozumi (2006), demonstrated that the measure of poison present in milk expanded proportionately with expanding *S.aureus* number, after its level achieved 106.5 CFU/mL, furthermore established that poison era expanded somewhere around 14 and 32°C.

In their study did with the milk of substantial and little dairy creatures in Italy, Cremonesi and partners (2006), decided coagulase positive *S.aureus* in the majority of the 111 tried specimens. 95 (%86) of the specimens contained no less than one sort of enterotoxin. 58 (%79) of the 73 expansive dairy creature tests and 37 (%97) of the 38 little journal creature tests contained enterotoxin.

In their study did in Kahramanmaraş, Erdoğan and partners (2006), did not decide *S.aureus* presence in any of the mayonnaise, colza, skimmed yogurt and bulgur tests they dissected.

In their study did in the Isparta region with whey cheddar (tort cheddar) produced using dairy animals, sheep and goat white cheddar whey, Şimşek and Sağdıç (2006), did not decide *S.aureus* presence in any of the specimens they examined.

Soejima and associates (2007), deliberately polluted skimmed milk with *S.aureus* and taking after stirring left the milk to brood at 35 °C. Subsequently, they reported that these conditions expanded the rate of *S.aureus* multiplication and SEA era.

In their study did with plain frozen yogurt tests taken from 55 distinctive offering focuses in Istanbul, Keskin and partners (2007), verified that 12.7% of the examples were not appropriate regarding *S.aureus* presence as per the control on microbiological criteria for foodstuffs. They reported that none of the specimens contained poison.

In a study did by Gülbandılar (2009) at Kütahya Public Health Laboratory, nose societies were assumed control over a time of right around one year (May 2006-June 2007) from an aggregate of 3048 individuals who desired different examinations however whose employment either included nourishment taking care of (cook, dough puncher, and so on.) or required direct contact with overall population (hair stylist, coiffeur, and so on.). *S.aureus* provinces were detached from 217 examples. 37 (17.05 %) of these disconnects originated from tests procured from female givers and 180 (82.9%) originated from tests gained from male contributors.

Kumar and associates (2009), decided *S.aureus* presence in one sample out of 10 milk test samples they dissected in India and reported the *S.aureus* consider 4.5×10^1 CFU/g.

In a study did at Soxony Hospital in Germany, Monecke and associates (2009), decided the presence of enterotoxin in 45.8% of the 155 *S.aureus* defiled specimens they broke down, and distinguished 17.42% of these enterotoxins as SEA.

In a study completed with 207 specimens at 3 distinct healing facilities in Bronx, United States, Varshney and associates (2009), distinguished 19 unique sorts of *staphylococcal* enterotoxins, specifically SEA, SEB, SEC, SED, SEE, SEG, SEH, SEI, SEJ, SEK, SEL, SEM, SEN, SEO, SEP, SEQ, SER, SEV, and TSST.

Garcia and partners (2009), reported that they decided presence of *S.aureus* in the greater part of the 75 milk tests gathered from different dairy ranches in Spain.

Önganer and Kırbağ (2009), completed a microbiological study in Diyarbakır with 30 Cokelek Cheese tests. They identified *S.aureus* in the majority of the specimens and reported the least *S.aureus* consider 6×10^6 CFU/g and the most noteworthy consider 10.28×10^6 CFU/g.

In their study completed in Kayseri region with 60 crude milk tests, Yılmaz and Gönülalan (2010), decided *S.aureus* presence in 30 tests and SE presence on 37 tests.

As far as possible for the presence of coagulase positive *staphylococcus* in foodstuffs prepared for utilization is given as ≤ 1.103 in Annex 3-Pathogen Microorganism Limits of the Turkish Food Codex Regulation on Microbiological Criteria (Anonymous, 2011).

In a study they completed in Ankara, Küplülü and partners (2002), identified *staphylococcal* enterotoxins, despite the fact that they didn't decide presence of *S.aureus* provinces in the purified milk tests they broke down. Thus, Wieneke and associates (1993), reported that in their examination of the cheddar tests devoured by two patients hospitalized for sustenance harming, they didn't recognize live *S.aureus* microorganisms however segregated *staphylococcal* enterotoxin in the specimens.

Staphylococci are the microorganisms which frame the common fora present in the upper respiratory framework and skin of individuals. It is hence that the sustenance business faculty are determined as the most imperative danger element for *staphylococcal* sullyng of foodstuffs. Deficient purifying conditions and sullyng after purification are likewise refered to as critical elements in the *staphylococcal* tainting of sanitized milk.

Staphylococci are greatly delicate to warmth treatment and are totally disposed of inside 30 minutes of warmth treatment at 60°C. Interestingly, *staphylococcal* enterotoxins are impervious to warmth treatment (Erol and Işeri, 2004). Therefore, if *S.aureus* colonization happened and animated enterotoxin era in foodstuffs before warmth treatment, *staphylococcal* enterotoxin can be distinguished after UHT and purification albeit no live *S.aureus* are available. Tragically, the warmth treatment techniques used in the sustenance business miss the mark concerning inactivating *staphylococcal* enterotoxins at a satisfactory level.

Utilization of cheddar polluted with the specified pathogens can prompt genuine wellbeing issues which can incidentally be lethal for buyers. In the most recent couple of years, a few episodes of malady because of utilization of polluted dairy items have been accounted for. The microbiological attributes of this cheddar rely on upon the nature of crude dairy, the systems and the states of generation, the work force and the capacity conditions. After the aging time frame, some unsafe nourishment pathogens, for example, *Salmonella spp.* may in any case cause genuine nourishment security issues for shoppers, regardless of included salt, the antimicrobial metabolites, the presence of low pH and dampness levels (Oksuztepe et al., 2005).

In their microbiological studies, Özalp and associates (1978), reported that they identified *staphylococcal* enterotoxin in Tulum Cheese tests albeit no *staphylococci* presence was resolved in any of the 26 tests;

Bone and partners (1989), reported that albeit no live pathogen was distinguished, *staphylococcal* enterotoxin was resolved in the sheep milk cheddar tests they examined. Then again, Cremonesi and partners (2006), decided *S.aureus* presence in the majority of the 111 milk tests they broke down and recognized enterotoxins in 95 tests.

S.aureus is a main source of clinical or sub-clinical mastitis. It is along these lines as often as possible present in and can be segregated from crude milk. In the light of the way that *staphylococcal* enterotoxins are thermostable and not crushed at purification temperatures, it is evident that *S.aureus* sullied milk convey potential wellbeing dangers even after sanitization. (Küplülü et al., 2002).

In a study went for deciding the conceivable presence of coagulase positive *staphylococcus* in 140 specimens of different dairy items (23 of which were Turkish White Cheese), the presence of this microscopic organisms was recognized in 18.6% of the examples as an results of the investigation completed with Baird-Parker Agar medium (Baştepe, 1977). In another concentrate additionally completed on Turkish White Cheese, coagulase positive *staphylococcus* was distinguished in precisely 1/3 of the 60 tests. Baird-Parker Agar medium was used as the principle specific medium in this study too (Aşkın, 1983). In another study, BPA comes about gave the *S.aureus* consider 7 log CFU/g or above for 3 (7.5%) of the 40 examined cheddar tests altogether. This outcome is extremely huge as far as nourishment wellbeing as poison era can be normal at this level of defilement (Ünlütürk et al., 1991).

In a study did with Civil cheddar tests gathered from Ankara market, *staphylococcal* presence was observed to be under identifiable level (<100 CFU/g), despite the fact that the microbiological nature of the investigated tests was assessed to be low, (Polat, 2001).

(Altın & Tekinşen, 2002), completed a microbiological study on the nature of white salted cheeses sold in Konya zone and its encompassing settlements. Subsequently, oxygen consuming mesophyll microorganism, coliform microscopic organisms, *Staphylococcus spp.*, *Lactobacillus spp.*, yeast and mold considers were individually decided 1,60x10⁸ CFU/g, 1,75x10⁵ CFU/g, 1,69x10³ CFU/g, 2,68x10⁷ CFU/g and 2,46x10⁵ CFU/g. It was additionally reported that 60% of the cheddar tests were not comparable to white cheddar principles as far as coliform microbes presence, while 66 % of the specimens were not keeping pace with white cheddar guidelines as far as yeast and mold presence.

It was, along these lines, presumed that a standard strategy was not utilized for creation of white cured cheddar in Konya and its encompassing settlements, and/or powerful quality control was not connected during their generation and promoting stages.

Usca and Erol (1998), completed a microbiological study on 50 Hellim cheddar tests delivered and showcased in the TRNC. They decided presence of live aerobe mesophyll in 96% (104 CFU/g) of the specimens; presence of enterobacter in 64% (104 CFU/g) of the examples; presence of coliform in 26% (103 CFU/g) of the specimens; presence of enterococcus in 52% (103 CFU/g) of the examples; and presence of yeast/mold in 66 % (103 CFU/g) of the examples. They likewise distinguished 103 CFU/g coagulase (+) *staphylococcus* presence in 26% and 103 CFU/g *E.coli* presence in 12% of the examples.

As opposed to our present study, Usca and Erol (1998), did not report *S.aureus* presence at levels posturing danger to general wellbeing. Notwithstanding, the examination of the consequences of the two studies can likewise be translated as a lessening from 26% to 6% in the general *S.aureus* presence in Hellim Cheese in 16 years. Regardless, it is not by any means conceivable or dependable to specifically connect the consequences of the two free studies, and our taking after assessment and remarks were not taking into account such an examination.

The fare of dairy items, as a rule, and Hellim Cheese specifically, overwhelmingly expanded in the course of the most recent ten years. Appropriately, the makers began to create and modernize their generation offices (with European Union guide) keeping in mind the end goal to meet the microbiological investigation criteria requested by the bringing in nations.

The EU upheld training programs gave to the concerned gatherings on sustenance generation security, made the producers more cognizant and delicate about this issue.

Likewise, the skilled powers expanded the supervisions and approvals went for expanding sustenance wellbeing all in all.

Conceivably the best accessible choice for little labs is a latex agglutination unit, however the time required for an outcome is 20-24 h. The Oxoid unit has been appeared to identify poison (sort unreported) in Halloumi cheddar (Berry et al., 1987) despite the fact that the same paper additionally reported a non-particular autoagglutination in sheep's milk cheddar.

An investigation of foodborne ailment in the USA presumed that more than 185,000 instances of *staphylococcal* nourishment harming happen every year, involving 1.3% of the aggregate number of foodborne sicknesses (Mead et al., 1999). The illness contributed 2.9% of foodborne hospitalisations and 0.1% of the passings.

A comparable, yet later, ponder evaluated more than 241,000 locally obtained instances of staphylococcal inebriation in the USA every year (Scallan et al., 2011). Given an expected 6.4% hospitalization rate for research facility affirmed cases, a mean 1,064 cases were hospitalized, and 6 passings came about.

An audit of cheddar related flare-ups in the USA from 1973 to 1992 presumed that episodes connected with this nourishment were uncommon (Altekruse et al., 1998). Of the 11 episodes distinguished one and only was brought on by *S. aureus*, in spite of the fact that the extent of cases hospitalized (68%) was high. The flare-up was brought about through inappropriate sanitization. A later investigation reasoned that 1-9% (mean 4.8%) of all *S. aureus* flare-ups in Europe could be credited to dairy and dairy items (Scientific advisory group on veterinary measures identifying with general wellbeing 2003), in spite of the fact that attribution of the extent of cases to these sustenance sorts was not endeavored. A late survey records reports of *staphylococcal* sustenance poisonings connected with milk and cheddar utilization (Cretenet et al., 2011).

CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

The increasing number of food safety problems occurring worldwide in recent years has heightened consumers' food safety awareness and caused public distrust of the increasingly complex and globalized food production and trading system. The frequency of *Salmonellosis* should not be disregarded because of the staggering impacts on human health. Awareness about *Salmonella* and its development is vital to guarantee food security and safety. Intercession methodologies are consequently essential to control *Salmonella* and *Staphylococcus aureus* from ranch to fork.

Total 34 halloumi cheese samples produced in different regions were collected from the various markets in Nicosia (Lefkoşa). Whereas no *Salmonella* was observed in any sample, 2 out of 34 samples were containing *Staphylococcus aureus*. However these samples of *S. aureus* colonies were lower than risk levels according to Turkish Food Codex.

We reach to a conclusion that, the levels of contamination in the samples are below the minimum infected dose according to Turkish Food Codex Committee on Microbiological Criteria and they may not cause any hazard for public wellbeing.

The results of the investigations halloumi samples obtained from various organizations of the TRNC and also from business sectors in the Nicosia (Lefkoşa). Showed that Hellim cheeses are quite clean regarding *Salmonella*.

However the use of good quality of raw and the utilization of the basic production techniques are not adequate for assuring microbiological quality of dairy products. Microbiological contamination from air-dust, apparatus and hardware, staff and different sources is in a high probability. Contamination after sanitation is of specific significance as far as product quality and customers' health and wellbeing.

Good quality of crude material satisfactory production methods and other accompanying criteria are critical so as to assure and sustain bacterial quality of the dairy products in the TRNC and for general wellbeing of public. Our recommendation for these purposes are as following:

- Processing of the raw material day ought to be completed at the same,
- Personnel ought to have routine medical checks,
- Personnel ought to sufficiently be prepared on microbiological contaminations,
- Personnel ought to be trained on individual hygiene,
- Temperature which are of specific significance for microbial development at storage and production sites ought to be entirely controlled particularly in hot summer days in the TRNC,
- Necessary legislative warning and actions ought to be taken for every single culpable condition.
- Organizations and producers ought to be educated about good hygiene practices which deliver this sort of food.

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