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TECNICAL-LEGAL FRAMEWORK AND EXPERIMENTAL DETERMINATION OF THE SHELF LIFE OF FRUIT SALADS IN THE PRODUCE SECTION: APPLICATION TO LARGE-SCALE RETAIL CHANNELS

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ABSTRACT
This thesis explores the technical-legal framework and the experimental determination of the shelf life of fruit salads in large-scale retail distribution. The first objective was to analyse the current national and European regulations regarding food safety, labelling, and storage requirements for packaged fresh products. Subsequently, a series of experiments were conducted to evaluate the shelf life of fruit salads, considering factors such as storage temperature and ph. The results obtained allowed for the identification of best practices to extend the freshness and quality of the product while reducing the risk of microbial contamination. Finally, operational recommendations were formulated for large-scale retail channels to improve the management of the fruit salad supply chain and ensure a safe and high-quality product for the consumer.

PURPOSE OF THE THESIS

The duration for which a product is available for sale corresponds to the period during which the product is suitable for human consumption and meets all organoleptic standards. Each type of food can be contaminated by specific bacteria, depending on the characteristics of its food matrix. These microorganisms and their metabolic activities, along with the enzymatic activities of the product itself, determine the shelf life of food products. Regarding the food matrix, factors such as water activity, acidity, pH, and temperature primarily influence microbial loads. These factors, in turn, vary depending on the product in question. In this case, the product under examination are fruit salads, a fresh, ready-to-eat item prepared by the operators of the fruit and vegetable department of UNICOMM Group stores, a prominent entity in large-scale organized distribution in northern Italy. The objective of storage tests is to ascertain the most accurate shelf life of these fruit salads, thereby optimizing the sales period post-production and minimizing food waste.

1. INTRODUCTION

Large-scale organized distribution (GDO) has transformed the way people buy and consume food. With supermarkets and hypermarkets, GDO has made a large variety of food products available, usually at competitive prices. This shift has significantly changed eating habits, leading to increased consumption of processed and ready-to-eat foods, often to the detriment of local and fresh products. The convenience of access to a wide range of products, including non-seasonal foods, has expanded consumer choice but has also contributed to increased dependence on industrial foods and a decline in traditional local food practices.

The GDO does not only sell local products but provides goods from various suppliers, offering customers a wide variety of non-native and international options. This various supply chain allows consumers to access a broad selection of foods that cannot be locally available, improving their choices and variety. Furthermore, the GDO is not only a retailer of products made by other companies; it also produces its own food stuff. Starting with raw materials, the GDO processes and manufactures food products under its own brands, which are then sold in supermarkets together with other goods.

As an entity that sell and produce food products, the GDO has to delineate and follow a specific HACCP (Hazard Analysis and Critical Control Points) plan, that has to be followed from the arrival of raw materials to the retail moment, including processing, packaging and storage.

Following the HACCP plan ensure that all the stages of the process are safe and compliant with strict and safety quality standards, reducing every possible contamination so as to propose safe food for consumers.

The food operators are responsible for preparing on-site food products, including bread, pastries, cakes, pizzas, cooked meet, fruit salads etc. During these food's production is very important to vigorously follow the HACCP guidelines to make sure that the final product meet all the health and safety standards before the purchase.

It is important for the GDO to give precise information to consumers about the proper storage conditions and the shelf life of ecery product, helping consumers to maintain quality and safety food from the moment it is purchased until the moment when it is consumed.

To achieve this, the GDO must include detailed labelling on packaging, which specifies critical storage instructions, like recommended temperature ranges and handling procedures. Additionally, expiration dates or "use by" dates must be clearly indicated to inform consumers about the period during which the product is assured to be safe and fresh. By offering these details, the GDO complies with food safety regulations and also helps consumers make informed decisions, thus enhancing food safety and minimizing the risk of food spoilage or food contamination.

It is crucial and essential for the GDO to accurately determine the shelf life of each food stuff to guarantee it is sold within its optimal timeframe.

Understanding and managing the shelf life of a product involves some important considerations:

- The GDO have to conduct comprehensive evaluations to establish the precise shelf life of each product. This involves evaluating how long the product remains safe, nutritious, and of high quality under given storage conditions. Factors like the product’s composition, packaging, and storage environment play an important role in determining its shelf life.
- Accurate shelf life information is necessary conform to food safety standards and regulations. Proper labelling and compliance to expiry dates are legally required to safeguard consumer health and avoid potential responsibilities.
- Providing products with precise shelf life information helps build consumer trust. When customers are confident that they are buying products that are within their safe consumption period, it enhances their confidence in the GDO’s dedication to quality and safety.
- Proper shelf life management helps to reduce food waste. By ensuring that products are sold within their optimal time frame, the GDO can minimize

the amount of unsold, expired foods that ultimately result as waste, therefore supporting sustainability efforts.

To sum up, accurately determining and managing the shelf life of products is essential for the GDO to maintain high standards of food safety, comply with regulations, optimize sales and improve consumer trust, all while being sustainable by reducing waste.

According to Article 14 of Regulation (EC) No 178/2002, foods that pose a risk to human health or are considered unfit for human consumption must not be sold.

- Hazardous foods are foods containing microorganisms, their toxins, their metabolism's products, residues of chamical compounds with quantities higher than tolerable by human's organism. Not fit for human consumption foods are foods with nutritional and sensorial characteristics not compliant with the standards. (27)
- Foods that are considered safe contain pathogens or residues of chemical compounds in quantities still tolerable by a human being and have their typical sensorial and/or nutritional characteristics within the storage conditions foreseen by the manufacturer. (27)

The GDO is not required to supply information about the shelf life, since this is usually indicated by its own producer. However, the GDO must provide scientific evidence about the durability of the food products that it produces and sells. This responsibility extends to both the products sold directly and those that are fractioned and sold after the original packaging has been opened. In these cases, even if the expiry date is initially provided by the manufacturer, it can change once the packaging has been opened or modified.

For fresh and microbiologically perishable foods, it is especially important to scientifically determine the shelf life. This process ensures food safety and maintains the quality of the product during its shelf life and during the period of sale and consumption.

2. SHELF LIFE

The Istituto Zooprofilattico delle Tre Venezie (32) defines shelf life as the “commercial life” of a product, intending the period between production and consumption during which the food remains safe for the consumer. The term “shelf life” literally translates to “life on the shelf” and is used in the context of food safety to indicate the duration for which a product can be stored and consumed without presenting health risks to the consumer.

The commercial lifespan of a food product begins when it is introduced to the market and lasts until it is either consumed or exceeds the expiration date or minimum durability date specified by the producer (1).

In latest years, several definitions of shelf life have been presented.

Daun (2) describes shelf life as “the maximum period during which a food product retains its predetermined quality characteristics”, though without detailing specific timeframes.

Tauokis (3) offer a more precise definition, asserting that shelf life is “the period of time, specific to each food product, that starts after production and during which the product can maintain an acceptable level of sensory quality and hygienic safety under defined storage conditions”.

The above mentioned se authors acknowledge that minor changes in sensory and hygienic attributes can occur, but not to the extent that the total quality of the product is compromised.

For fishery products, Waterman (4) defines shelf life as “the duration during which a fish of high initial freshness can be preserved under specific conditions before it loses a significant portion of its nutritional value or becomes unsuitable for human consumption “. As a result, the product's durability is closely linked to the initial condition of the raw material.

Fu and Labuza (5) describe shelf life as” the timeframe from the completion of a product's production until it becomes unsuitable for consumption from a sensory, hygienic, or nutritional perspective “.

They also note that “it is virtually impossible to create a universal definition of shelf life for a food product”.

The meaning of a product's commercial life is particularly increased for those items sold while still alive, like certain fishery products (like eels, trout and bivalve molluscs) and plants that continue to live even after being harvested or uprooted.

When establishing a food's shelf life, it is necessary to consider that plants continue to breathe. This is particularly important when they are packaged in plastic film bags, where the inability to exchange gases with the external environment can lead to spoilage.

Regulation (EC) No 178/2002 (33) of the European Parliament and of the Council, of 28 January 2002, establishes the general principles and requirements of food law, creates the European Food Safety Authority, and dictates procedures in matters of food safety. The regulation stipulates the following food safety requirements:

- Food shall not be placed on the market if it is unsafe.
- Food shall be deemed unsafe if it is considered to be: (a) Injurious to health; (b) Unfit for human consumption.

Given the critical importance of shelf life, it is important to everyone associated in the entire food chain. There is a rising awareness that high standards of food safety and quality can only be reached through a comprehensive and integrated approach that includes the entire food chain, "from farm to fork".

A lot of factors can influence the shelf life of food, including the hygiene of work environments, the choice of materials that interact with food, the personal hygiene of the staff handling food and beverages, the chemical composition of the food, the intrinsic physical characteristics of the food, like its pH and water activity, the bacterial microflora present on the raw materials and the environmental microflora that interact with the food and the technologies utilized for food production and preservation.

Consumers themselves also play an important role in ensuring the correct preservation of food, for example, by maintaining it at the proper temperatures and consuming it according to the manufacturer's instructions within the labelled expiration dates.

Regulation (EC) No 852/2004 (28) of the European Parliament and of the Council, dated 29 April 2004, on the hygiene of foodstuffs, requires food business operators to make sure that all stages of production, processing, and distribution under their control meet all the hygiene requirements present in the regulation. This regulatory framework permits operators to maintain high hygiene standards throughout all phases of the production and distribution chain, making sure that the food products are as safe as possible for consumption.

Additionally, the regulation helps to preserve the shelf life of food products by preventing contamination during production, handling, transportation, storage, and sale.

Regulation (EC) No 2073/2005 (29) of 15 November 2005 on microbiological criteria for foodstuffs defines the term "shelf life" in the Article 2, Section f, as: "the period preceding the use-by date or the minimum durability date, as defined respectively in Articles 9 and 10 of Directive 2000/13/EC (30)." This definition determines the timeframe within which a food product remains safe and suitable for human consumption under the specified storage conditions, before reaching either its use-by date or its minimum durability date.

Directive 2000/13/EC was repealed by European Regulation (EU) No 1169/2011 (31) of 25 October 2011 on the provision of food information to consumers. This regulation establishes the framework to guarantee a high level of consumer protection in relation to food information, presenting general principles, requirements, and responsibilities that govern the provision of food information. These principles apply to food business operators at all stages of the food chain when their activities involve giving information about food to consumers. The regulation applies to all food intended for the final consumer.

Regulation (EU) No 1169/2011 defines the" minimum durability date" like "the date until which the product retains its specific properties under appropriate storage conditions".

It also describes the principles governing mandatory food information, such as shelf life, storage conditions, and safe use.

3. STORAGE TEST

Storage tests play a crucial role in determining the shelf life of food products. These tests evaluate how environmental factors such as temperature, light exposure, humidity and packaging materials affect the quality and safety of food over time.

The ultimate goal of the storage test is to ensure that foods remain safe and retain their sensory, nutritional, and hygienic properties during their commercial life.

The primary function of storage tests is to establish the timeframe during which a product remains safe for human consumption, while maintaining its intended quality characteristics. Such testing also helps manufacturers meet international standards, such as the European Regulation (EC) No. 178/2002, which outlines the general food safety requirements (6).

Storage tests pay attention to a number of essential variables:

- Chemical Composition: Monitoring the presence of proteins, carbohydrates and lipids helps identify degradation processes such as hydrolysis or oxidation, that can alter the flavour, texture, or safety of the product (7).
- Physical Properties: Factors like water activity (aw) and ph are crucial in determining microbial stability. For example, foods with low water activity limit microbial growth, extending shelf life. Regular measurement of these parameters allows food producers to evaluate the potential for spoilage (6, 7).
- Microbial Load: The presence of both intrinsic and extrinsic microorganisms is measured during storage tests. This is particularly critical for ready-to-eat foods or minimally processed products that are inclined to contamination (7).
- Packaging Technology: The packaging of food products is vital for prolonging shelf life. Modern techniques such as active packaging, which interacts with the product environment, or intelligent packaging, which monitors spoilage indicators, are increasingly used to improved product preservation (7).

There are two main types of storage tests:

1. Real-Time Testing: Food products are stored under the conditions they are likely to encounter in real-world settings, with periodic sampling to monitor

changes. Although time-consuming, this method provides accurate results reflective of actual product lifespan (7).

2. Accelerated Testing: In this method, products are exposed to more extreme storage conditions (e.g., higher temperatures or increased humidity) to accelerate the degradation process. These tests offer quicker insights into shelf life but require careful interpretation to avoid overestimating durability (6).

In this thesis, we will exclusively examine the physical properties and microbial load of the fruit salads under investigation, utilizing a real-time storage test.

For food manufacturers, understanding the outcomes of storage tests is essential for improving product life span, reducing waste, and enhancing sustainability. By determining the precise shelf life of a product, companies can ensure that consumers buy food in optimal conditions while complying with health and safety regulations (7).

Storage tests are essential in ensuring that food products encounter safety and quality standards throughout their shelf life. By combining advanced monitoring techniques with improved packaging solutions, the food industry can extend shelf life while safeguarding consumer health. The results of these tests also provide essential insights for manufacturers regarding the correct storage and handling instructions needed to maintain product quality throughout the supply chain (6, 7).

4. A BIT OF SUSTAINABILITY

Significant losses occur throughout the food supply chain, particularly in fruits and vegetables, from the time they are harvested until they arrive to the consumer. An important portion of food produced never reaches the consumer cause of inefficiencies and losses in the supply chain.

The post-harvest loss segment is estimated to account for around 25-30% of the total production. Globally, approximately 14% of food is lost between harvest and retail, while an additional 17% is wasted at the retail and consumer levels.

In line with Agenda 2030 for Sustainable Development, Target 12.3 aims to halve per capita global food waste at the retail and consumer levels, while also reducing food losses across production and supply chains, including post-harvest losses.

Nowadays, there is a growing recognition of the synergy between the need to preserve the quality of perishable products and the advancement of global food markets. Reducing post-harvest losses is an essential measure that must be immediately addressed. It is estimated that 1.3 billion tons of food are lost annually, representing 33% of global food production, according to the FAO (8).

This important amount of waste highlights the need for a collaborative effort from all stakeholders in various sectors of the food supply chain to minimize losses and improve the preservation of horticultural products post-harvest.

The shelf life of food products is generally defined as the period during which a food product remains microbiologically safe and retains its desired sensory, physicochemical, and nutritional quality. Maintaining these qualities throughout the food's lifespan is key to reducing waste and ensuring that food remains safe and nutritious for consumers.

By extending shelf life and reducing post-harvest losses, significant progresses can be made toward achieving global food security and sustainability goals (9).

Consumer’s education is very important in reducing food waste and maximizing food shelf life.

Managing and preserving food products are crucial aspects of reducing waste and maximizing the shelf life of foods after purchase. Consumer education plays an crucial role in this process, that can significantly influence purchasing, storage, and consumption practices.

Food waste is a global issue with significant economic and environmental implications. According to a report by the Food and Agriculture Organization (FAO), approximately one-third of the food produced for human consumption is lost or wasted each year (14).

The education of consumers and, as a consequence, consumers choices abd practice is essential in reducing the impact the amount of food that ends up in the trash.

A study conducted by Brandenburg et al. In 2022 demonstrated that educating consumers about date labels and optimal consumption can significantly reduce food waste (15).

Labels such as” best before“ and ”expiry date“ are often misunderstood by consumers, contributing to excessive discarding of still-edible foods. Educational programs that clarify these terms and provide guidance on proper food storage can contribute in reducing food waste.

The shelf life of a food product after sale is influenced by various factors, including the type of product, storage conditions, and handling practices. According to a study published in Critical Reviews in Food Science and Nutrition, proper storage and handling of food can significantly extend its useful life (16). Consumers need to be informed about maintaining foods under optimal conditions to preserve freshness and safety.

Additionally, Gonzalez *et al*. (2021) highlight the importance of educating consumers on preservation techniques such as freezing, using airtight containers, and maintaining appropriate temperatures to prevent microbial growth (17).

Greater awareness about managing food stocks, such as using inventory systems like FIFO (First In, First Out), can reduce the risk of spoilage and extend the consumption period of foods.

By understanding concepts such as expiration and optimal consumption, and adopting proper storage practices, it is possible to reduce food waste and improve food safety. Investing in educational and informational programs can lead to significant benefits both at the individual and global levels, contributing to a more sustainable and efficient food system.

5. READY TO EAT

The term” ready-to-eat food“(RTE) refers to food products that can be consumed exactly how they are sold, without additional preparation.

This market portion has experienced significant growth in recent years, driven by lifestyle changes and innovations in food technology.

The ready-to-eat food market has seen important expansion. According to a report by Grand View Research, the global market for ready-to-eat food is expected to grow at a compound annual growth rate (CAGR) of 5.2% from 2023 to 2030 (10).

This increase is assigned to the rising demand for convenience among consumers and the dynamic lifestyle in urban areas.

Technological innovations have played a crucial role in the development of RTE products. The adoption of innovative packaging techniques, like modified atmosphere and high-pressure pasteurization, has improved the shelf life of foods without compromising quality (11).

In addition, rapid cooking technologies, such as microwave ovens have made quick preparation possible without reducing taste and nutrition.

The principal benefits of RTE foods are convenience and speed of preparation. On the other hand, it is also very important to consider nutritional aspects. Many RTE products can contain high levels of salts, sugars, and preservatives, which can effectively have negative health effects if consumed in excess (12).

Consequently, it is important for consumers to read labels carefully and make informed choices.

Another important aspect is the environmental impact of RTE products. Packaging and production of these foods can include significant resource consumption and generate waste. According to a study published in Environmental Science & Technology, the ecological footprint of RTE foods is generally higher than fresh foods prepared at home (13). Companies have to work to improve sustainability through more eco-friendly production practices and recyclable packaging.

The ready-to-eat food market is continuously evolving, addressing the needs of modern consumers looking for quick and convenient solutions. However, it is essential to balance convenience with health considerations and take in consideration that the environmental impact associated with these products. With ongoing innovation and increasing environmental awareness, the RTE food sector may evolve to offer healthier and more sustainable solutions.

The ready-to-eat fruit salads sold in the supermarkets of the Unicomm Group are a prime example of convenience food. Given the freshness of the products, they serve as a natural source of vitamins and minerals due to the fruit being cut on the same day.

6. NON-THERMAL BACTERIAL INACTIVATION TECHNIQUES

It is crucial to eliminate pathogenic bacteria from food before products are made available for sale. Traditional food processing methods such as pasteurization and sterilization rely on direct heat application. While effective in eliminating pathogens, these methods often fall short in preserving the micronutrients naturally present in food. This has led to increased interest in non-thermal food processing methods, which inactivate microbes without the use of heat or with minimal heat application. These methods are becoming more popular due to evolving consumer demands for fresh, nutrient-rich foods with extended shelf lives.

Non-thermal food processing technologies can be categorized into physical and chemical processes. Physical methods include Pulsed Electric Fields (PEF), High-Pressure Processing (HPP), Pulsed Light (PL), and ultrasound technology, while chemical methods involve ozone treatment and non-thermal plasma (NTP) technology. (34)

- Pulsed Electric Fields (PEF)

PEF technology utilizes high-voltage electric fields applied in short pulses to inactivate microbes. The process involves placing food between two electrodes and applying short bursts of electric fields. This leads to electroporation, where pores form in the cell membranes of pathogens, causing the cytoplasmic contents to leak out, ultimately leading to cell death. Studies show that PEF is highly effective in microbial inactivation without compromising the nutritional and sensory qualities of food.

- High-Pressure Processing (HPP)

HPP is a technique that applies pressures ranging from 100 to 800 MPa, often at temperatures below 20°C, for a few seconds to a few minutes. The intense pressure inactivates spoilage-causing bacteria, thus extending the shelf life of the food. This process is particularly valued because it preserves the sensory and nutritional qualities of the food while achieving food safety standards.

- Ultrasound Technology

Ultrasound technology, also known as sonication, uses ultrasonic vibrations to generate reactive oxygen species, which target and kill microorganisms. The high-energy sound waves disrupt microbial cell structures, leading to cell death. This method has shown promise for improving food safety without damaging the food’s physical or chemical properties.

- Pulsed Light (PL)

PL technology uses short bursts of intense light from a broad spectrum to inactivate microbes on food surfaces. The high-energy pulses break down microbial DNA, preventing reproduction and ensuring food safety. This method is particularly useful for decontaminating food surfaces without affecting the product's quality.

- Ozone Treatment

Ozone, classified as a Generally Recognized As Safe (GRAS) substance, is a powerful antimicrobial agent used in food processing. It works by destroying the protein structures of microbial membranes, leading to microbial death. Ozone is particularly effective in surface decontamination and can be applied to food products without leaving chemical residues.

- Non-Thermal Plasma (NTP)

Non-thermal plasma is an ionized gas created when a gas is exposed to a strong electric field. The reactive species generated in plasma, such as electrons and ions, attack and destroy microbial cells. This method is effective at inactivating bacteria and viruses on food surfaces, and it can be applied to packaged foods.

Advantages of Non-Thermal Technologies

The main advantage of non-thermal technologies is their ability to inactivate pathogens while preserving the nutritional, physical, chemical, and sensory properties of food. Additionally, these processes are more energy-efficient and environmentally friendly compared to thermal methods. Some of these technologies can even be applied to already-packaged food, further enhancing their versatility.

Despite their potential, the widespread adoption of non-thermal food processing technologies faces several challenges.

High initial investment costs for equipment and maintenance are significant barriers. Additionally, some bacteria may develop resistance to non-thermal treatments by forming spores, which could pose public health risks.

Worker safety during the operation of certain technologies, such as high-pressure processing, is also a concern. Furthermore, consumer acceptance of some of these methods remains limited due to perceptions about food safety and quality.

In conclusion, while non-thermal food processing technologies appear very promising to enhance food safety to extend shelf life, their commercial application requires overcoming technical, economic, and regulatory challenges. Also research and innovation are needed to improve these processes, making them more accessible and acceptable to both industry and consumers.

7. BIOLOGICAL PARAMETER

The deterioration of the sensory qualities of food is primarily powered by the excessive proliferation of *Specific Spoilage Organisms* (SSOs) (35, 36).

These microorganisms are responsible for food spoilage, and accurately estimating a product’s shelf life requires identifying and quantifying the specific SSOs associated with that product (35, 36).

1. Aerobic Microorganisms at 30°C: This parameter indicates the total count of aerobic microorganisms in a food product and provides a general sense of its hygienic condition and preservation status. These microorganisms grow in the presence of oxygen at temperatures between 20°C and 45°C, with an optimal range of 30–37°C. Although no specific legal limit is set for this type of microorganism, general reference values are based on industry guidelines and experience.

2. Enterobacteriaceae: Enterobacteriaceae are mesophilic bacteria that can multiply at temperatures above 6–7°C, although certain species, such as *Yersinia* spp., are psychrotrophic and can thrive at lower temperatures (35, 36). These bacteria are commonly found in the environment but originate primarily from the intestinal tracts of animals. Enterobacteriaceae are capable of degrading proteins, carbohydrates, and lipids, resulting in the degradation of the food's sensory qualities (38). They are considered indicators of hygiene and are often linked to the sanitation and operational conditions of food handling and processing (37).

3. Lactobacilli: Mainly anaerobic, lactobacilli thrive in vacuum-packed or modified atmosphere packaging (38, 36). They are categorized into:

    - Homofermentative: These strains mainly produce lactic acid from sugar metabolism, which helps preserve food by lowering the pH (37).

    - Heterofermentative: These strains generate a range of organic acids (e.g., lactic and acetic acids), ethanol, CO_2, aldehydes, and ketones, potentially leading to undesirable flavours, surface slime (ropiness), and package swelling in vacuum-sealed products (38, 36).

4. Pseudomonadaceae: Pseudomonads are Gram-negative, aerobic, and psychrotrophic bacteria, meaning they can grow at refrigeration temperatures (35, 36). Known for their proteolytic, saccharolytic, and lipolytic activities, pseudomonads produce substances like hydrogen sulphide and pigments that contribute to off-odours (38). They are often associated with the spoilage of protein-rich foods, such as meat, poultry, and fish, especially under aerobic and refrigerated conditions.

5. *Escherichia coli* (β-glucuronidase-positive): This microorganism is characteristic of the intestinal tract of humans and animals, and its presence in food, especially meat products, signals contamination, which may result from inadequate hygiene during raw material handling, food processing, or environmental contamination. Some strains of *E. coli* are pathogenic and can cause severe gastrointestinal illnesses (36).

6. Coagulase-positive Staphylococci: These bacteria, including *Staphylococcus aureus*, are commonly found on human and animal skin and in the respiratory tract. Contamination can occur through the use of raw materials or surfaces contaminated with staphylococci, or through improper hygiene practices during food handling. *Staphylococcus aureus* is particularly warrying as it can produce heat-stable enterotoxins that cause gastrointestinal issues (38).

7. Molds: molds are primarily aerobic, though some species can grow in anaerobic conditions (35, 36). They are adaptable and thrive in a variety of environments due to their ability to metabolize different substrates (38). Molds are highly resistant, tolerating low water activity, pH, and temperature, which makes them common contaminants in many food products.

8. Yeasts: Yeasts are resilient microorganisms that grow in acidic environments ($pH < 4.5$) and under conditions of low water activity ($aw < 0.900$) (38). Commonly found in products like fruit preserves, juices, and mayonnaise, yeasts can spoil food by producing off-flavours and visible surface films through gas production. Both yeasts and molds can produce secondary metabolites, like mycotoxins, under certain conditions, which can contribute to food spoilage and pose health risks to humans (38).

9. Sensory Analysis (appearance, odour, colour, texture): sensory analysis is crucial for products where sensory attributes significantly influence consumer choices. Throughout a product's shelf life, characteristics such as odour, colour, and texture may change due to microbial or chemical degradation, which can result in the product losing its quality and appeal to consumers. Thus, maintaining consistent sensory qualities throughout the commercial life of a product is essential (38).

The growth of SSOs is influenced by environmental factors such as water activity, temperature, pH, and the presence of preservatives like salts or sugars (38, 36). Low water activity or the addition of salts and sugars can trigger microbial defence mechanisms, increasing resistance and enabling the growth of certain microorganisms under challenging conditions. Microbial homeostasis, or the internal balance within microorganisms, is a critical factor in their ability to grow and cause spoilage (37). When homeostasis is disrupted, microbial growth can be delayed or halted, but SSOs typically proliferate during a food's shelf life, often outcompeting the natural microflora and producing spoilage-related compounds (35).

Understanding the specific SSOs present in different food products is essential for accurately estimating shelf life and ensuring food safety (35, 36).

While some microorganisms are harmless or even beneficial, others can cause significant sensory degradation or pose serious health risks (38).

Therefore, effective microbiological control is crucial in preventing spoilage and minimizing the risk of foodborne illnesses in the food industry.

8. THE EVOLUTION AND IMPACT OF ORGANIZED LARGE-SCALE RETAIL IN ITALY

Organized Large-Scale Retail (GDO in italian language) represents a form of retail sales across extensive areas, yet it remains a part of the broader retail sector. It encompasses large groups operating on a national and international scale.

The term" Grande Distribuzione"(GD) refers to centralized structures managed by a single owner, which control multiple directly operated retail points. In contrast," Distribuzione Organizzata"(DO) involves smaller entities that unite to enhance competitiveness and negotiate better supply terms with multinational companies.

Historically, DO benefited from favourable legislation, providing it with a competitive edge over GD. However, recent shifts in GD strategies have enabled it to surpass DO's dominance (18).

A key challenge for GD lies in the heterogeneity of retail formats, which complicates centralized control and management.

Currently, Italy faces a competitive disadvantage compared to international multinationals, largely because no Italian group has achieved widespread national coverage (19).

The GDO sector includes various types of retail formats:

- Hypermarket: Retail area exceeding 2,500 m^2.
- Superstore: Retail area ranging from 1,500 m^2 to 3,500 m^2.
- Supermarket: Retail area between 400 m^2 and 2,500 m^2.
- Self-Service: Retail area from 100 m^2 to 400 m^2.
- Discount: Retail outlets that do not carry branded products.
- Cash & Carry: Wholesale retail area.
- Traditional: Retail area less than 100 m^2.
- Self-Service Specialists: Focus on home and personal care products (20).

The origins of GDO date back to 1830 in France with the Magasins de Nouveautés, marking the transition from traditional retail to the large department store model.

While Le Bon Marché, which opened in Paris in 1852, is often cited as the first large department store, Macy's in New York, established in the following decade, is considered by many to be the first due to its broader merchandise range (21).

The first supermarket, King Kullen, was founded in the United States in 1930 (22).

In Italy, the GDO history began with the opening of the first large department store in Milan in 1877 by the Bocconi brothers, Luigi and Ferdinando, under the name Aux Villes d'Italie.

The first Italian supermarket chain, Esselunga, was established in 1957. Gros Market Lombardini opened the first Cash & Carry in 1964, followed by the first hypermarket, Carrefour, in 1972, and the first discount store, Lidl, in 1992 (23).

Today, GDO accounts for approximately two-thirds of retail food sales in Italy. My thesis analyzes this sector through a collaboration with a prominent player in the GDO market, UNICOMM Srl, which has significantly expanded its operations over the past 55 years (24).

Established in 1969 by Marcello and Mario Cestaro, UNICOMM opened its first Super A&O store in Vicenza the following year. The company launched its distribution center in Malo in 1971 and, following its expansion, transformed into a joint-stock company. As of now, the UNICOMM Group employs over 7,000 people and operates more than 270 direct retail outlets. These are located in Friuli Venezia Giulia, Veneto, Emilia Romagna, Tuscany, Umbria, Marche, and Lazio, in addition to numerous affiliated stores.

UNICOMM Srl manages several brands, including:

- Svelto A&O (superettes),
- Super A&O (supermarkets),
- Famila and Famila Superstore (integrated supermarkets),
- Emisfero (hypermarkets),
- Cash and Carry (25).

UNICOMM Srl is also a major member of the Selex commercial group, one of the leading players in the national distribution landscape, with a market share of 8.7% (26). Selex-branded products number over 1,900.

9. MATERIALS AND METHODS

Determination of the Shelf Life of Fruit Salads

This thesis presents to the management of UNICOMM srl, located at Via E. Mattei 50, Dueville (VI), the results of a series of storage tests designed and conducted to assess the shelf life of fruit salads produced directly at the UNICOMM srl retail outlets for sale to the final consumer on-site.

The ready-to-eat fruit salads are retailed as pre-packaged products and displayed in refrigerated counters maintained at a temperature of 4°C until purchased by the customer.

The storage of fruit salads prepared according to proper hygiene practices and maintained at 4°C prevents the growth of microorganisms responsible for product spoilage. Among these, some may result from contamination during the cutting and packaging stages carried out by department staff.
The purpose of these storage tests is to evaluate the shelf life of the fruit salads entirely prepared by the produce department staff of UNICOMM srl, which are then placed directly for sale on-site as ready-to-eat food kept at 4°C until purchased.
The results presented pertain to the fruit salads subject to these storage tests, produced in accordance with the preparation procedures established in the HACCP plan of UNICOMM srl.

Storage Test Setup
We have decided to schedule our storage tests to verify the shelf life of the fruit salads of four types:

• Golden apple, oranges, pineapple, kiwi, strawberries

Image 1 from Unicomm Srl archive

• Pineapple, Brazilian melon, kiwi, strawberries

Image 2 from Unicomm Srl archive

• Pineapple, strawberries, blueberries, raspberries

Image 3 from Unicomm Srl archive

• Pineapple, melon, kiwi, strawberries, white grapes, black grapes, blueberries, raspberries

Image 4 from Unicomm Srl archive

The storage tests were conducted in six different UNICOMM srl retail locations:

- Emisfero Perugia
- Emisfero Monfalcone
- Emisfero Fuimicino
- Emisfero Scorzè
- Emisfero Bassano
- Emisfero Vicenza

As this study focused on the shelf life of the samples, the analyses were carried out over a specific timeframe of 3 to 4 days. Upon receipt at the laboratory, the sample units were stored in a refrigerated chamber set to 4°C, where they remained for at least 2 days. Over the next two days, some samples continued to be kept at 4°C, while the rest were exposed to a temperature abuse test at 8°C. The analyses were conducted across the 4-day period, with tests being performed every 24 hours. Each sample unit consisted of a plastic cup (PE) sealed with transparent film in an air-filled environment.

The samples were examined on days 1, 2, 3, and 4 after production, and each one underwent a thorough microbiological analysis, which included the quantitative determination of:

• Total microbial count at 30°C (cfu/g);

• Enterobacteriaceae (cfu/g);

• Beta-glucuronidase-positive Escherichia coli (cfu/g);

• Coagulase-positive staphylococci (Staphylococcus aureus and related species) (cfu/g);

• Pseudomonas spp. (cfu/g);

• Mesophilic lactic acid bacteria (cfu/g);

• Yeasts (cfu/g);

• Molds (cfu/g);

Additionally, qualitative tests were conducted to detect the presence of the hepatitis A virus.

All the individual determinations mentioned above were conducted using a specific ISO (39) standard when applicable, or alternatively, with an internally validated test method from the laboratory in cases where no ISO standard existed. The ISO standards applied are outlined in the table below.

Microbiological Parameter	ISO Norm	Year of publication	Title
Total microbial count at 30°C	UNI EN ISO 4833-2	2022	Microbiology of the food chain - Horizontal method for the enumeration of microorganisms - Part 2: Colony count at 30°C using the surface inoculation technique
Enterobacteriaceae	UNI EN ISO 21528-2	2017	Microbiology of the food chain - Horizontal method for the detection and enumeration of Enterobacteriaceae - Part 2: Method for colony count
Escherichia coli beta-glucuronidasi positive	ISO 16649-2	2001	Microbiology of food and animal feed - Horizontal method for the enumeration of beta-glucuronidase-positive Escherichia coli - Part 2: Colony count technique at 44°C using 5-bromo-4-chloro-3-indolyl beta-D-glucuronide
Stafilococchi coagulase-positive	UNI EN ISO 6888-2	2023	Microbiology of the food chain - Horizontal method for the enumeration of coagulase-positive staphylococci (Staphylococcus aureus and other species) - Part 2: Method using rabbit plasma and

			fibrinogen agar medium
Pseudomonas spp.	Metodo di prova interno		Internal test method, EPTA NORD MI025 (Pseudomonas spp. plate count)
Mesophilic lactic acid bacteria	ISO 15214	1998	Microbiology of food and animal feed - Horizontal method for the enumeration of mesophilic lactic acid bacteria - Colony count technique at 30°C
Yeasts and molds	ISO 21527-1	2008	Microbiology of food and animal feed - Horizontal method for the enumeration of yeasts and molds - Part 1: Colony count technique in products with water activity greater than 0.95
Hepatitis A virus	ISO 1526-2	2019	Microbiology of the food chain - Horizontal method for the detection of hepatitis A virus and norovirus using real-time RT-PCR technique - Part 2: Method for detection

Table 1

Beyond the individual differences that characterize each of the above-mentioned analytical determinations, we can outline the common steps shared by all the analytical methods used:

1. The sample was appropriately diluted in a sterile buffered saline solution at a 1:10 ratio, using the correct weight and volume of diluent.

2. The sample was homogenized by compression in a Stomacher 400 Circulator until complete fragmentation and homogenization were achieved.

3. Serial decimal dilutions were prepared from the master cultures of each sample to properly quantify each specific microbial load.

4. The most suitable dilutions were chosen, and a specific volume of the suspension (0.1 ml or 1 ml) was plated onto different selective culture media, specific to each determination.

5. Plating was carried out using a sterile pipette or an automatic Gielson pipette with a sterile tip, depending on the ISO method used. The inoculation was performed either by spreading the suspension across the agar surface or using the pour plate method, as in the case of total microbial count.

For the detection of the hepatitis A virus genome, ISO 1526-2:2019 was applied, preparing the sample for further examination using real-time PCR.

In the following Table , I summarize the specific selective media used for the quantitative determination of each parameter in the microbiological examination mentioned above.

Microbiological Parameter	**Selective culture medium**	**Temperature and incubation time**
Total microbial count at 30°C	plate count agar (PCA)	38°C for 48-72 h
Enterobacteriaceae	violet red bile glucose agar medium (VRBG)	37°C for 24 h
Escherichia coli beta-glucuronidase positive	tryptone-bile-glucoronide medium (TBX)	44°C for 24-48 h

Stafilococchi coagulase-positive	rabbit plasma fribinogen agar medium	37°C for 24-48 h
Pseudomonas spp.	cetrinide fucidina cefalosporina agar (CFC Agar) con supplemento antibiotato	30°C for 48 h
Mesophilic lactic acid bacteria	MRS medium (de Man, Rogosa and Sharpe) at pH 5.7	30°C for 72 h
Yeasts and molds	dichloran-rose bengal chloramphenicol agar (DRBC)	25°C for 5 days

Table 2

After the scheduled incubation periods, the inoculated plates were examined and assessed for both colony morphology and colony count. For the quantitative determination of specific microbial loads, only plates with colony counts between 30 and 300 were selected, in accordance with the general ISO guidelines for microbiological testing of food for human consumption. The resulting colony counts were recorded on the appropriate data sheet for further analysis.

10. RESULTS

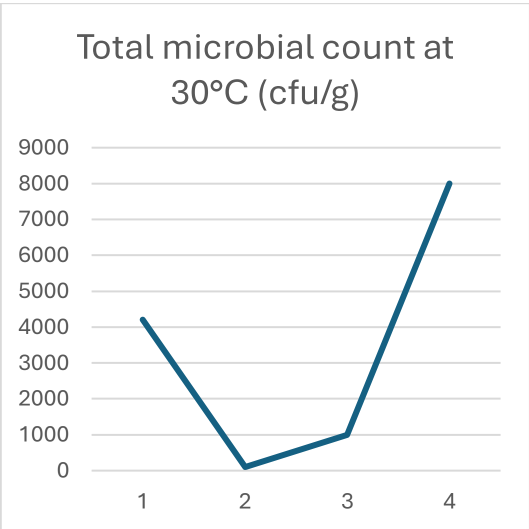
The results obtained from these storage test trials are detailed in the following tables and charts.

OBSERVATIONS EMISFERO PERUGIA

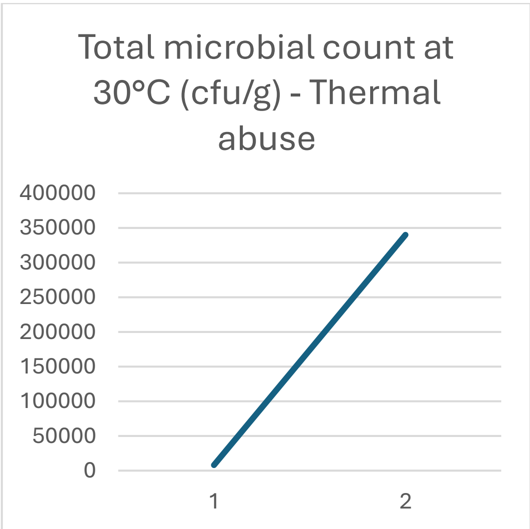
➔ FRUIT SALAD (GOLDEN APPLES, ORANGES, PINEAPPLE, KIWI, STRAWBERRIES)

	STORAGE AT +4°C				STORAGE AT +4°C FOR 48H, THEN AT +8°C	
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T5 - 96H FROM PRODUCTION-24LB0036139	T4 - 72H FROM PRODUCTION-24LB0036140	T5 - 96H FROM PRODUCTION-24LB0036141
Ph	3,61	3,60	-	-	-	-
Wa	0,986	0.988	-	-	-	-
Acidity	0,93	0,95	-	-	-	-
Total microbial count at 30°C (cfu/g)	4.200	100	1.000	8.000	8.000	340.000
Enterobacteriaceae (cfu/g)	170	40	<10	40	40	10.000
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	<10	<10	<10	<10	<10	<10
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10	<10	<10
Yeasts (cfu/g)	<10	240	670	310	210	170.000
Molds (cfu/g)	<10	<10	<10	110	860	<10
Hepatitis A virus	Not detected	-	-	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant	-	-

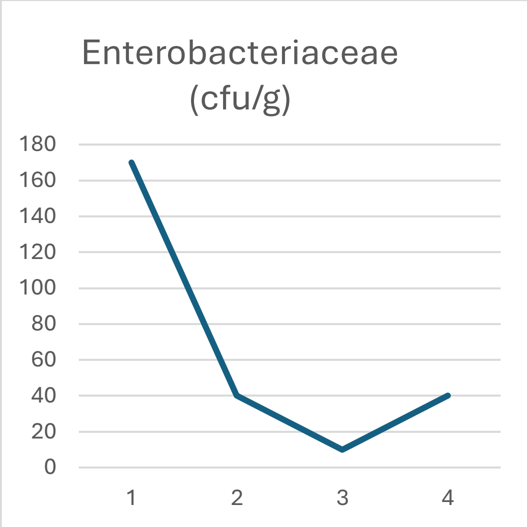
Table 3

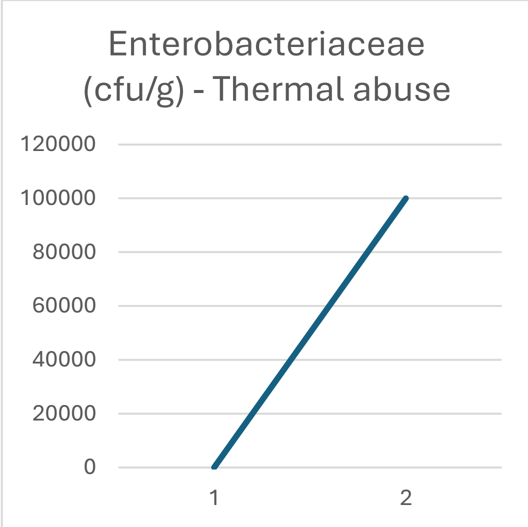


Graph 1



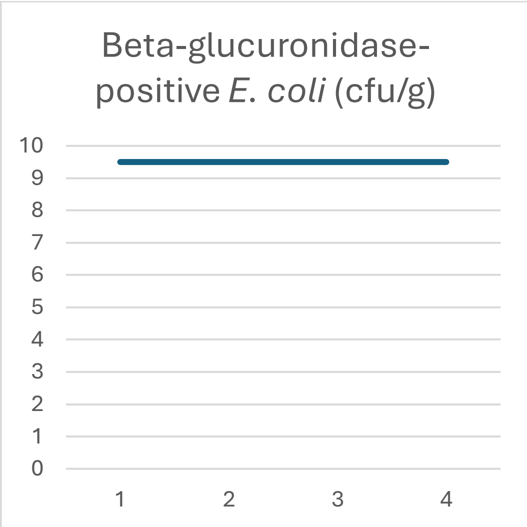
Graph 2

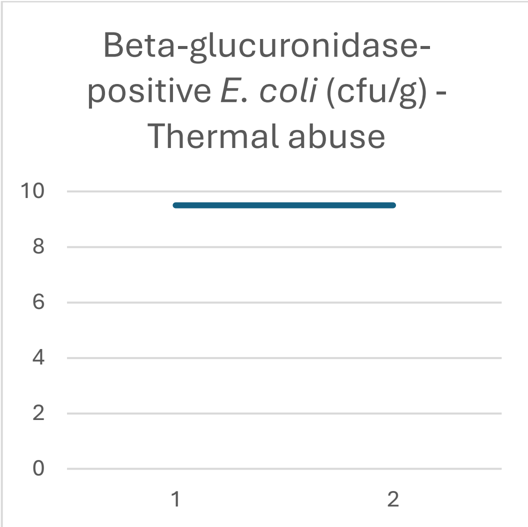




Graph 3

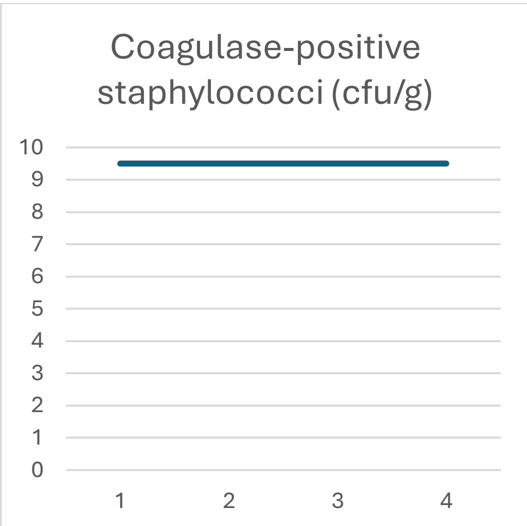
Graph 4

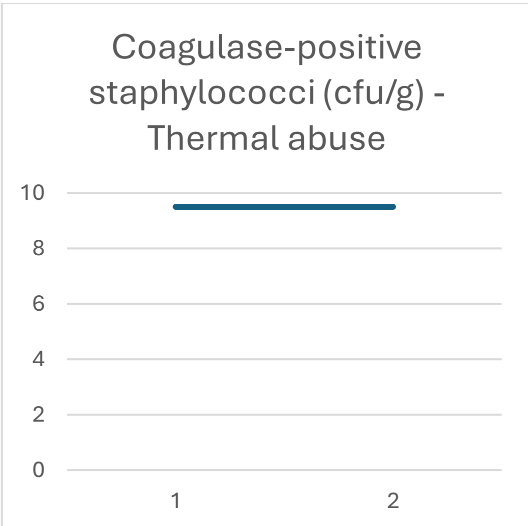




Graph 5

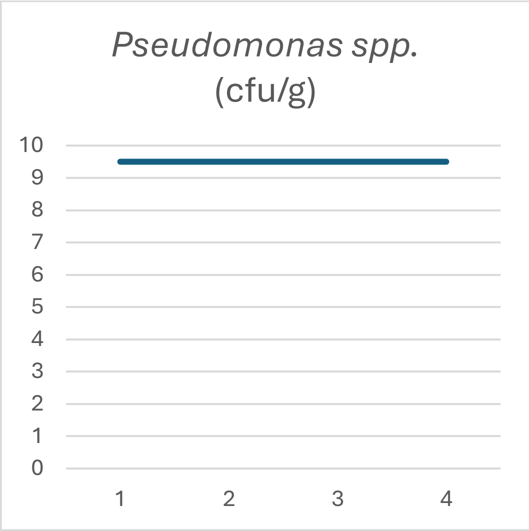
Graph 6





Graph 7

Graph 8



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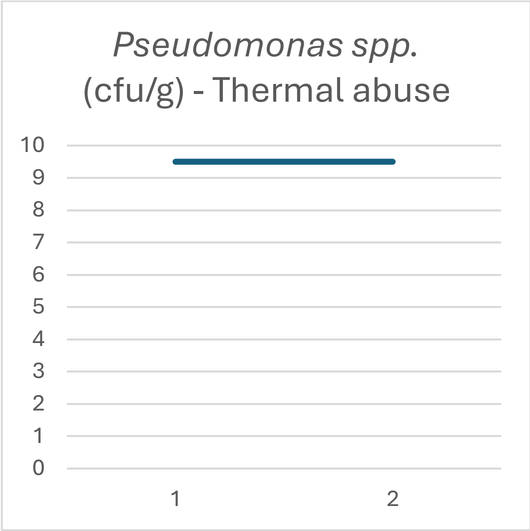
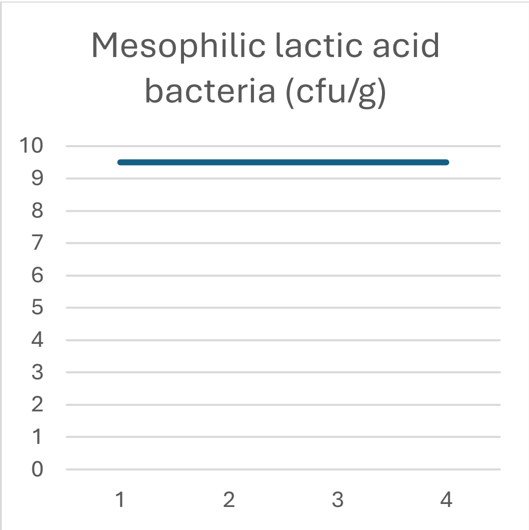
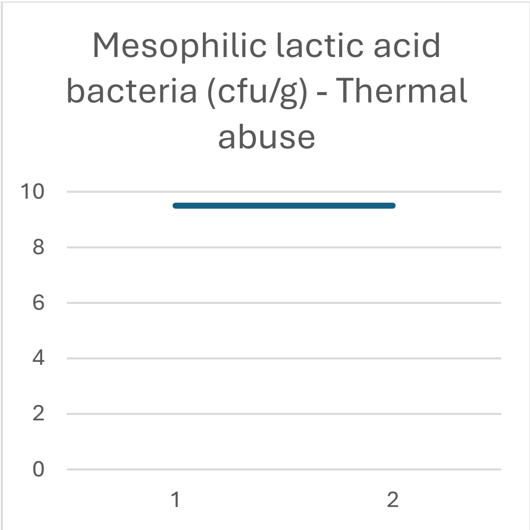


Graph 10



Graph 11



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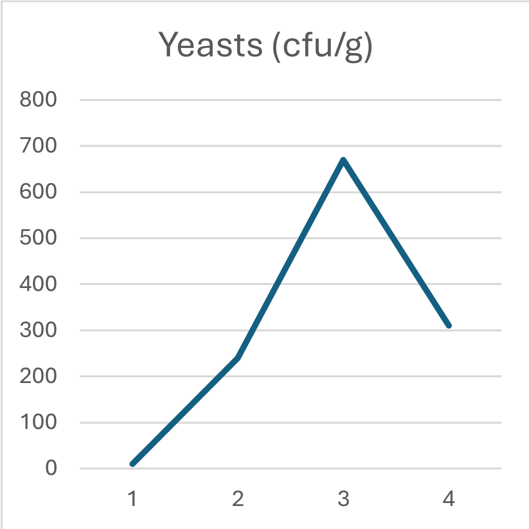
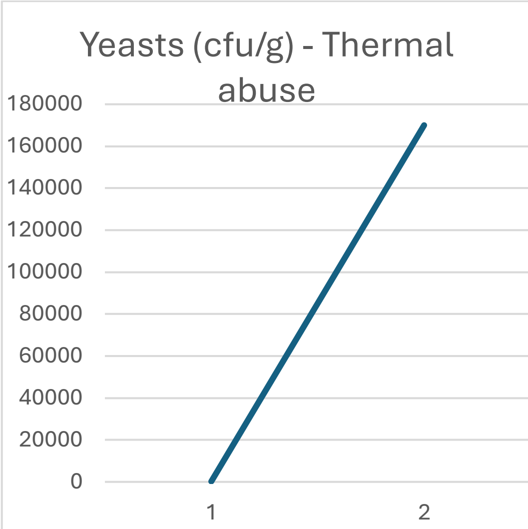
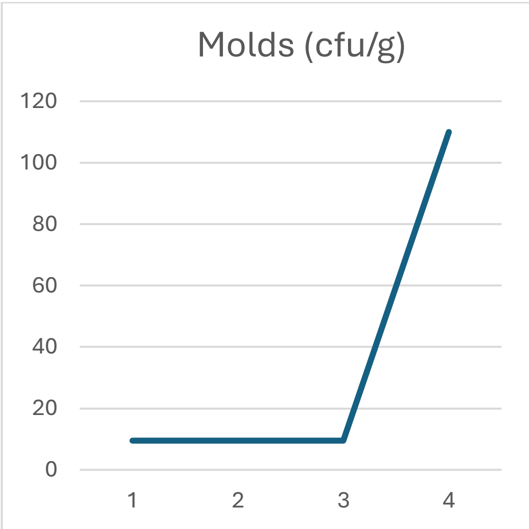


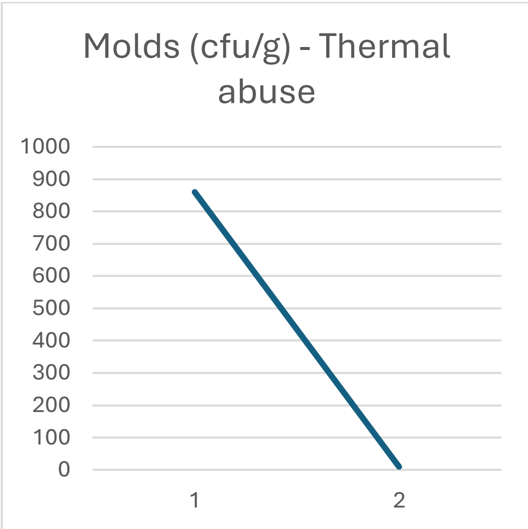
Graph 13



Graph 14



Graph 15

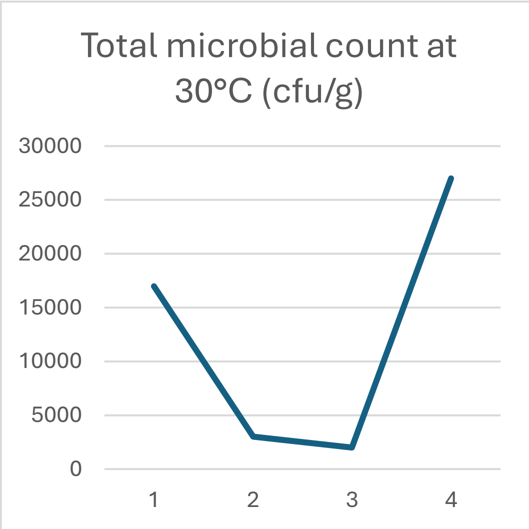


Graph 16

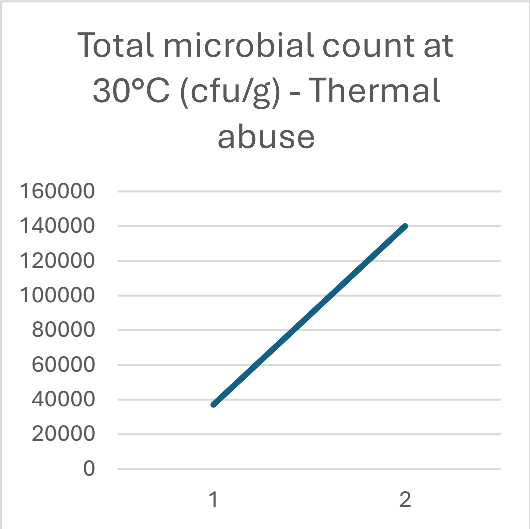
➔ FRUIT SALAD (PINEAPPLE, BRAZILIAN MELON, KIWI, STRAWBERRIES)

	STORAGE AT +4°C				STORAGE AT +4°C FOR 48H, THEN AT +8°C	
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T5 - 96H FROM PRODUCTION-24LB0036139	T4 - 72H FROM PRODUCTION-24LB0036140	T5 - 96H FROM PRODUCTION-24LB0036141
Ph	3,64	3,57	-	-	-	-
Wa	0,990	0.990	-	-	-	-
Acidity	0,66	0,70	-	-	-	-
Total microbial count at 30°C (cfu/g)	17.000	3.000	2.000	27.000	37.000	140.000
Enterobacteriaceae (cfu/g)	2.700	80	80	940	70	7.000
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	<10	<10	<10	3.200	1.400	<10
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10	<10	<10
Yeasts (cfu/g)	250	180	370	310	720	19.000
Molds (cfu/g)	90	<10	1.200	960	100	<10
Hepatitis A virus	Not detected	-	-	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant	-	-

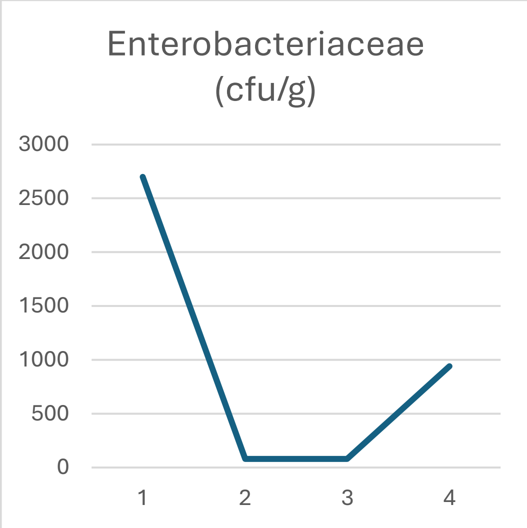
Table 4

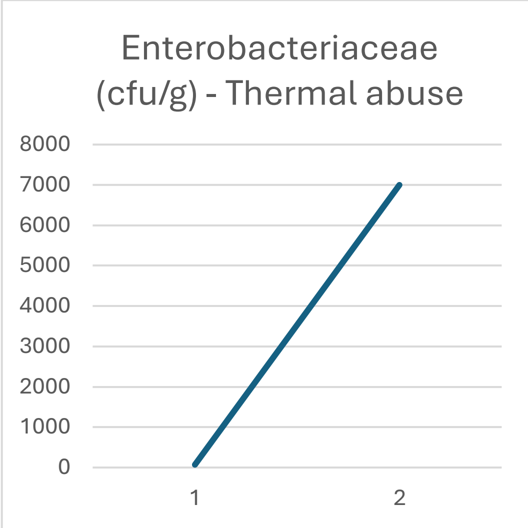


Graph 17



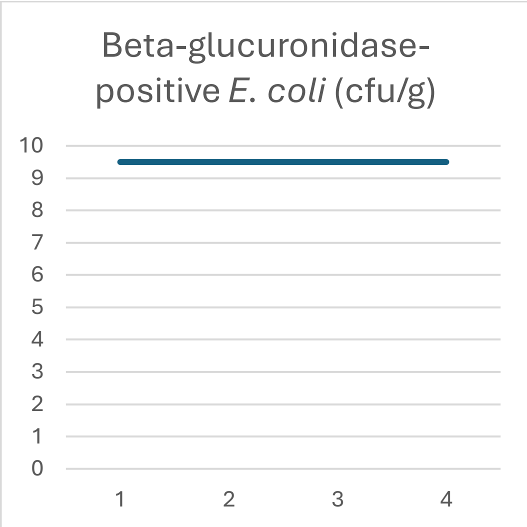
Graph 18

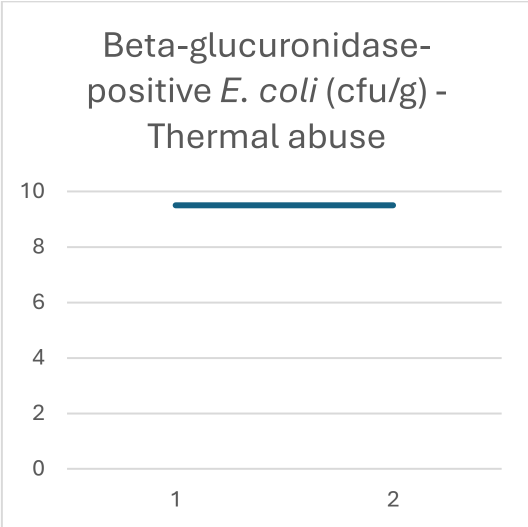




Graph 19

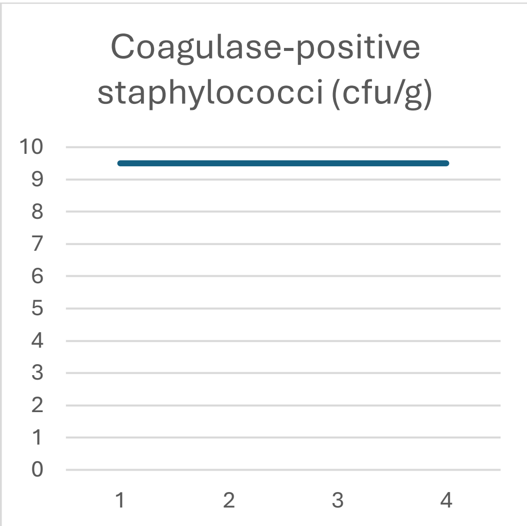
Graph 20

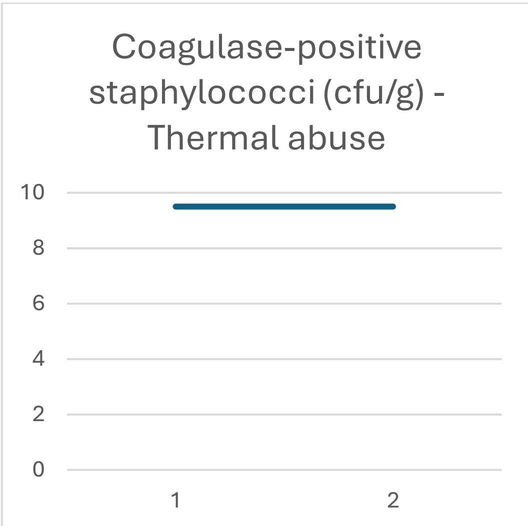




Graph 21

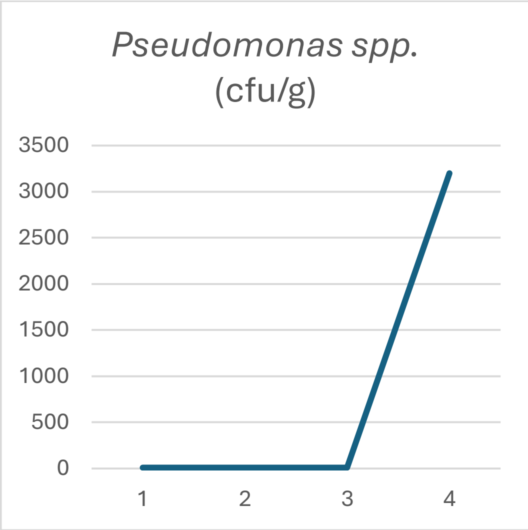
Graph 22





Graph 23

Graph 24



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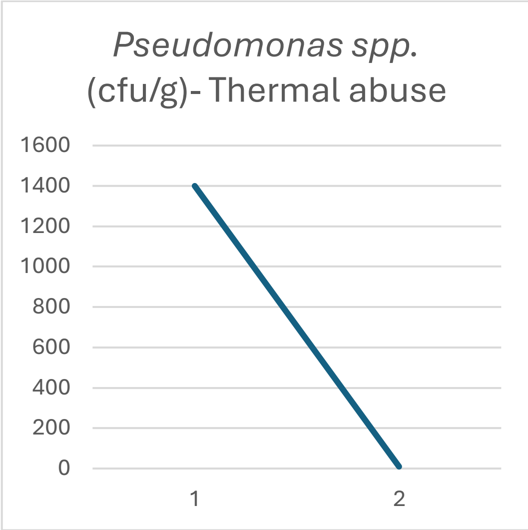
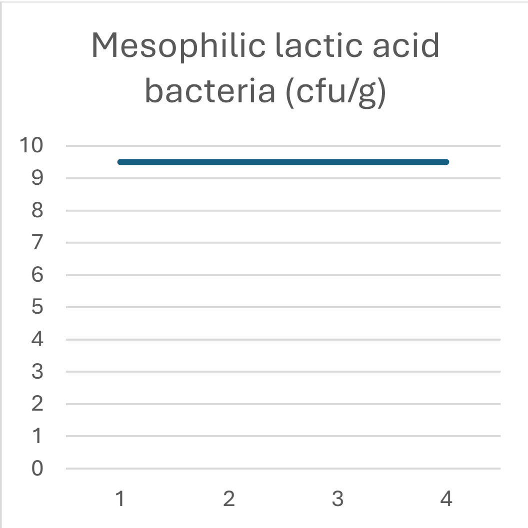


Graph 26



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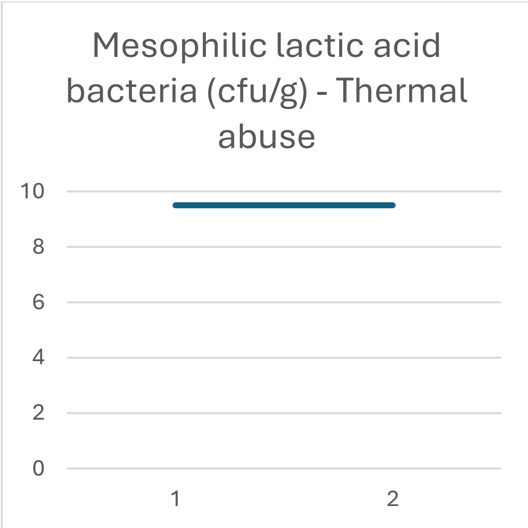

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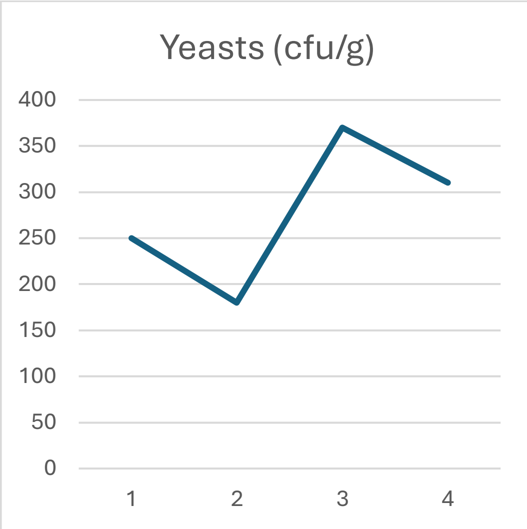
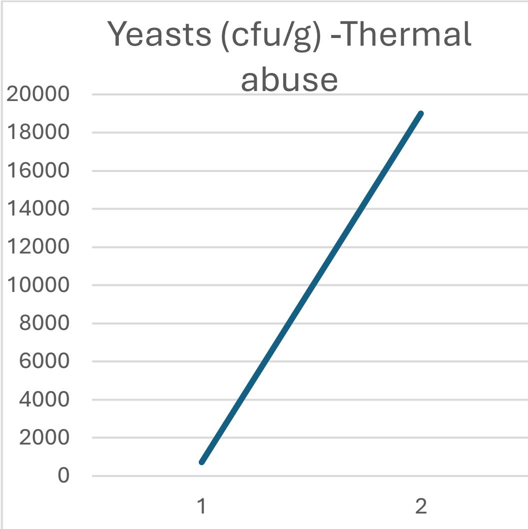
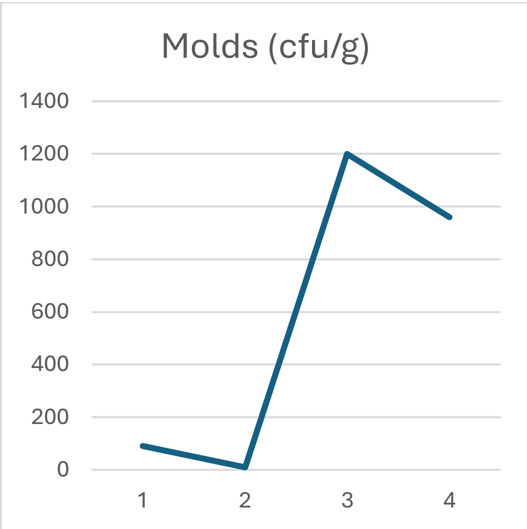
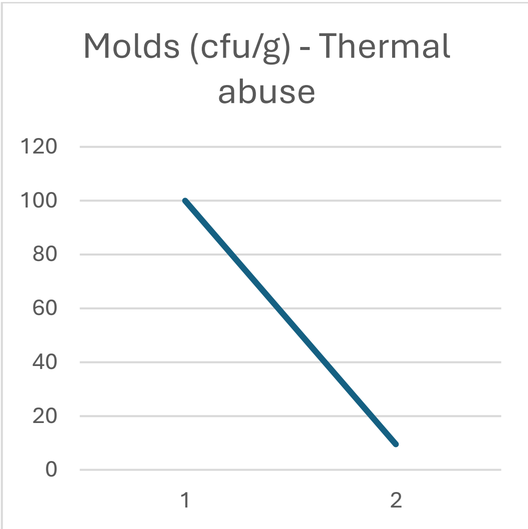




Graph 29

Graph 30





Graph 31

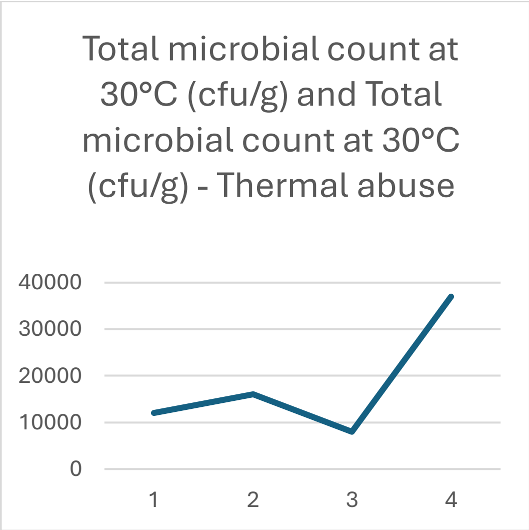
Graph 32

OBSERVATIONS EMISFERO MONFALCONE

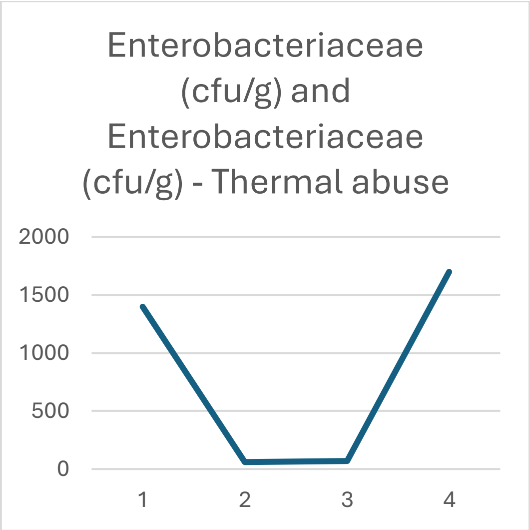
➔ FRUIT SALAD (GOLDEN APPLES, ORANGES, PINEAPPLE, KIWI, STRAWBERRIES)

	STORAGE AT +4°C			STORAGE AT +4°C FOR 48H, THEN AT +8°C
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T4 - 72H FROM PRODUCTION-24LB0036140
Ph	3,71	3,81	-	-
Wa	0,990	0.990	-	-
Acidity	0,77	0,80	-	-
Total microbial count at 30°C (cfu/g)	12.000	16.000	8.000	37.000
Enterobacteriaceae (cfu/g)	1.400	60	70	1.700
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	1.200	200	900	2.300
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10
Yeasts (cfu/g)	100	840	90	870
Molds (cfu/g)	220	<10	260	500
Hepatitis A virus	Not detected	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	-

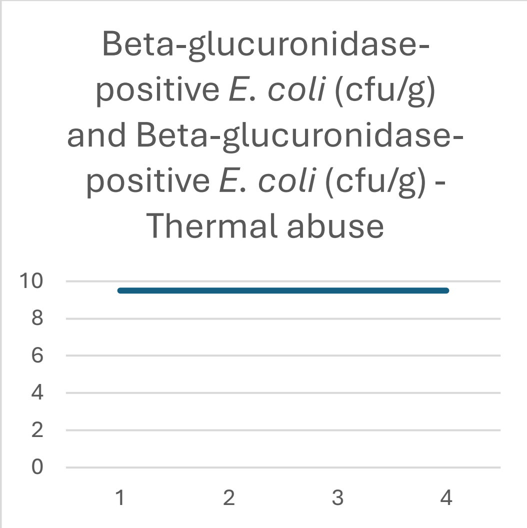
Table 5

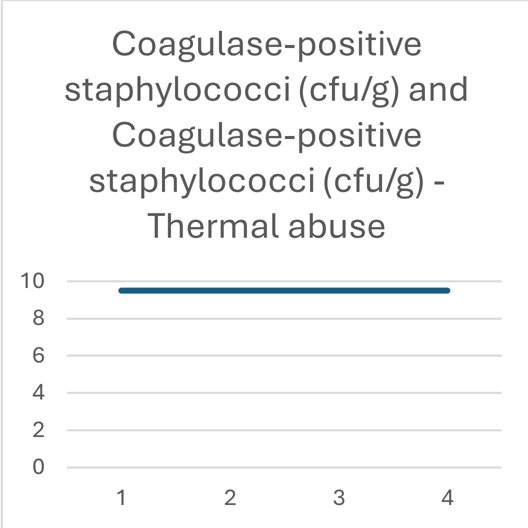


Graph 33



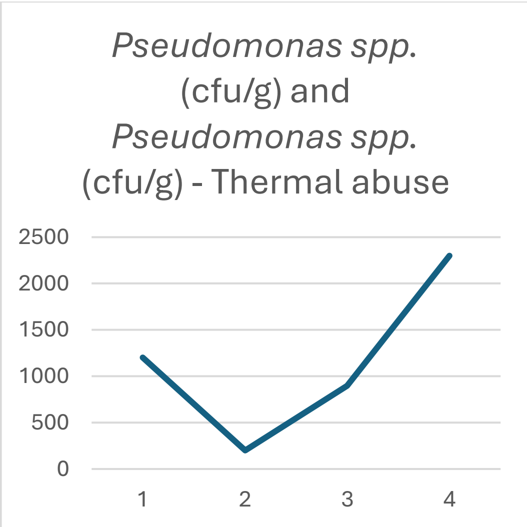
Graph 34

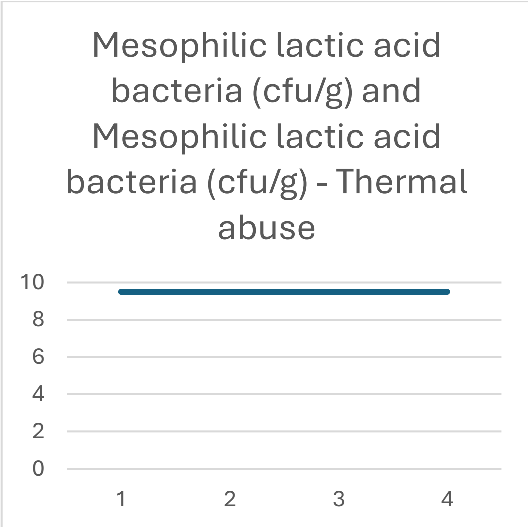




Graph 35

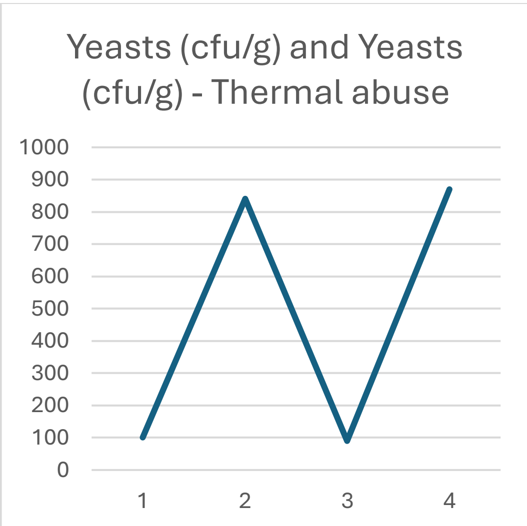
Graph 36

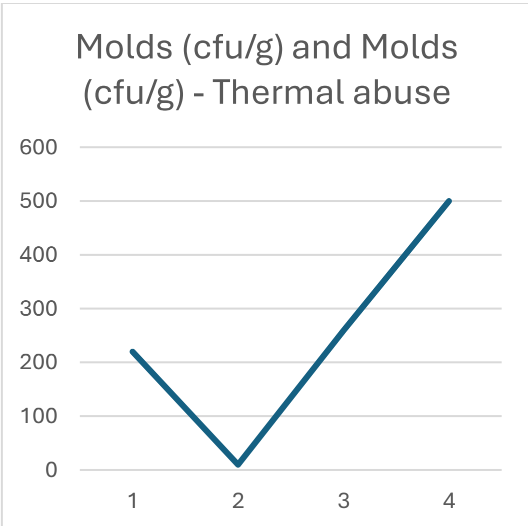




Graph 37

Graph 38





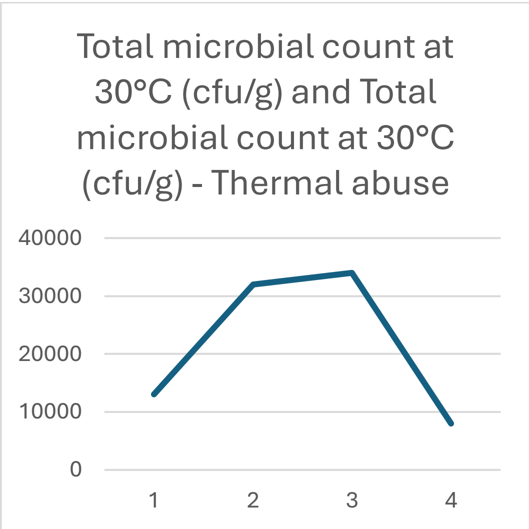
Graph 39

Graph 40

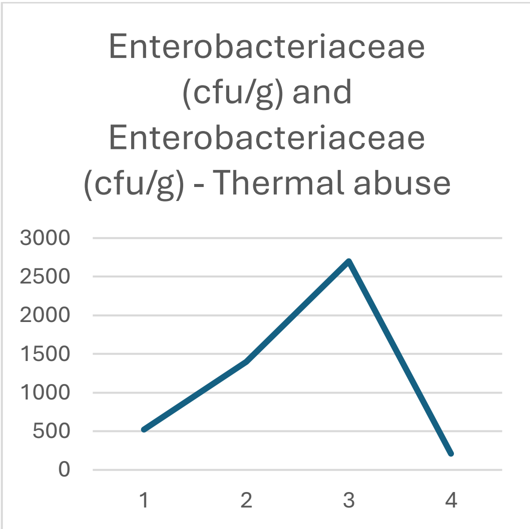
➔ FRUIT SALAD (PINEAPPLE, BRAZILIAN MELON, KIWI, STRAWBERRIES)

	STORAGE AT +4°C			STORAGE AT +4°C FOR 48H, THEN AT +8°C
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T4 - 72H FROM PRODUCTION-24LB0036140
Ph	3,64	3,74	-	-
Wa	0,998	0.989	-	-
Acidity	0,54	0,56	-	-
Total microbial count at 30°C (cfu/g)	13.000	32.000	34.000	8.000
Enterobacteriaceae (cfu/g)	520	1.400	2.700	210
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	800	2.600	2.100	100
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10
Yeasts (cfu/g)	520	900	350	960
Molds (cfu/g)	40	330	1.500	1.100
Hepatitis A virus	Not detected	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	-

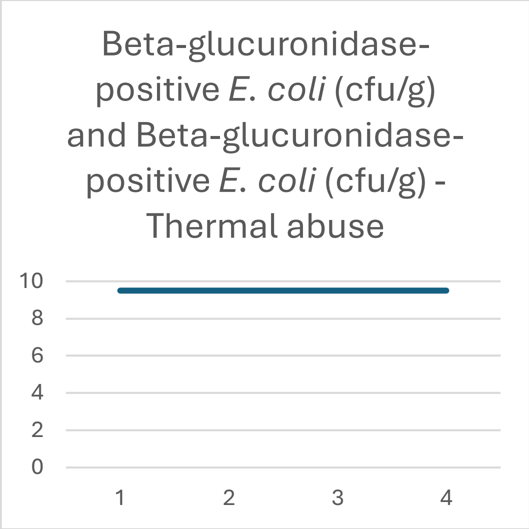
Table 6



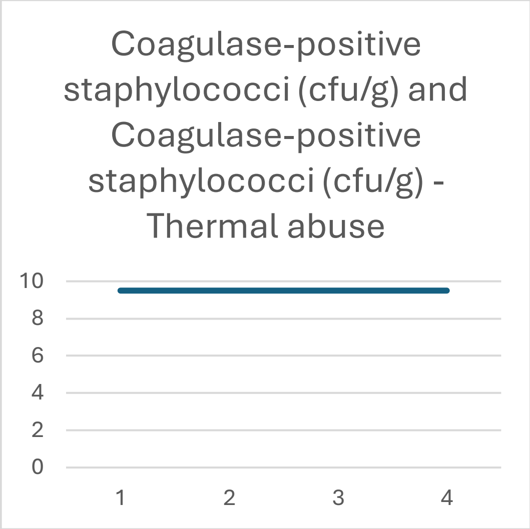
Graph 41



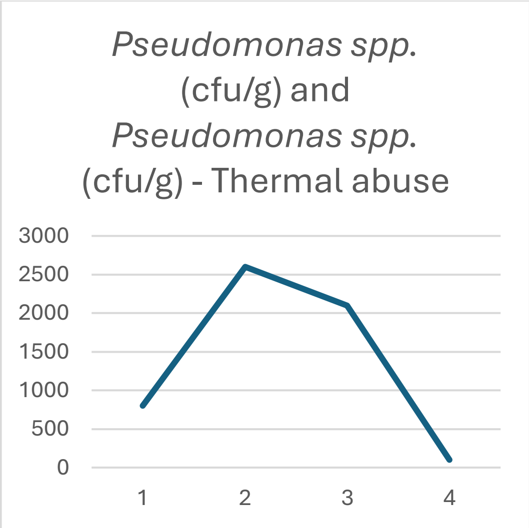
Graph 42



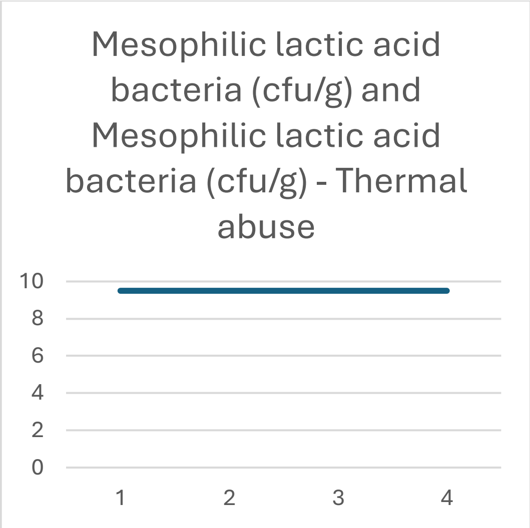
Graph 43



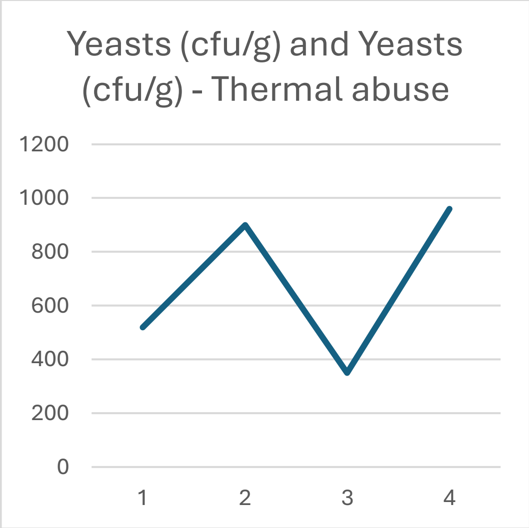
Graph 44



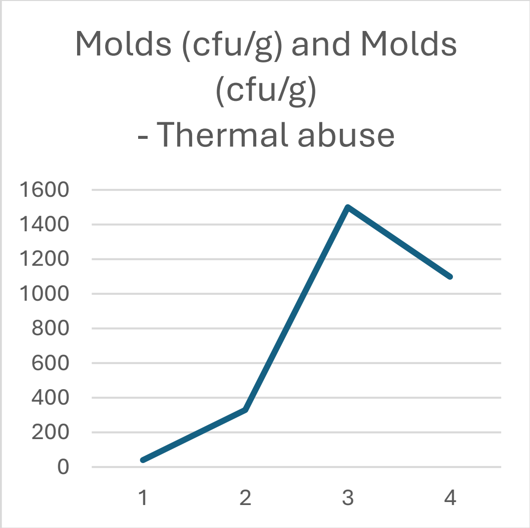
Graph 45



Graph 46



Graph 47



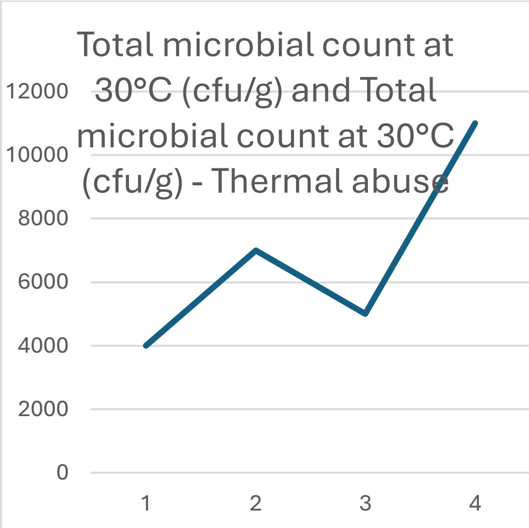
Graph 48

OBSERVATIONS EMISFERO FIUMICINO

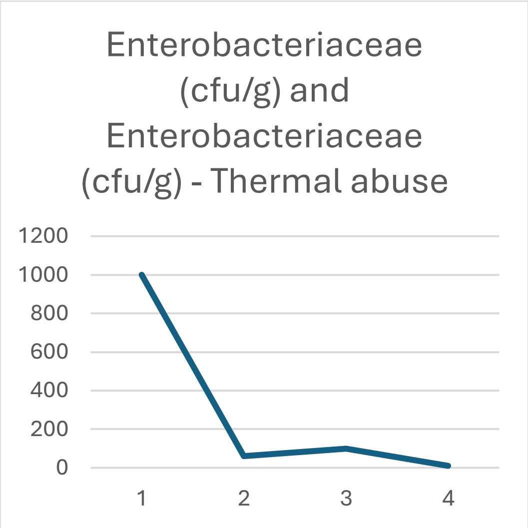
➔ FRUIT SALAD (GOLDEN APPLES, ORANGES, PINEAPPLE, KIWI, STRAWBERRIES)

	STORAGE AT +4°C			STORAGE AT +4°C FOR 48H, THEN AT +8°C
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T4 - 72H FROM PRODUCTION-24LB0036140
Ph	3,58	3,55	-	-
Wa	0,981	0.980	-	-
Acidity	0,71	0,85	-	-
Total microbial count at 30°C (cfu/g)	4.000	7.000	5.000	11.000
Enterobacteriaceae (cfu/g)	1.000	60	100	<10
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	<10	<10	<10	<10
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10
Yeasts (cfu/g)	860	800	2.200	410
Molds (cfu/g)	<10	40	<10	<10
Hepatitis A virus	Not detected	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant

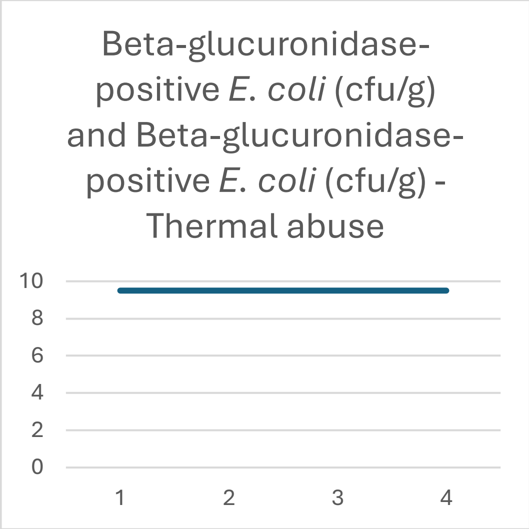
Table 7



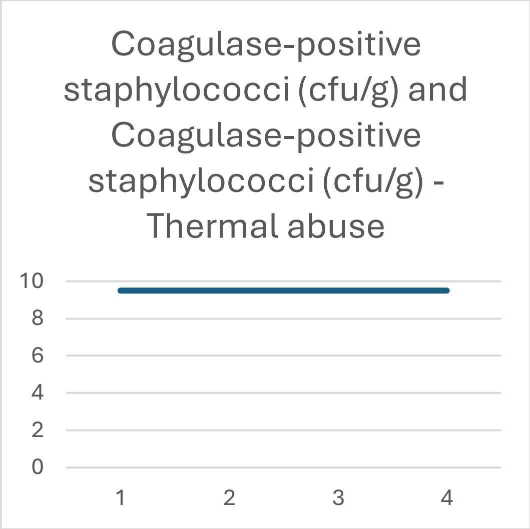
Graph 49



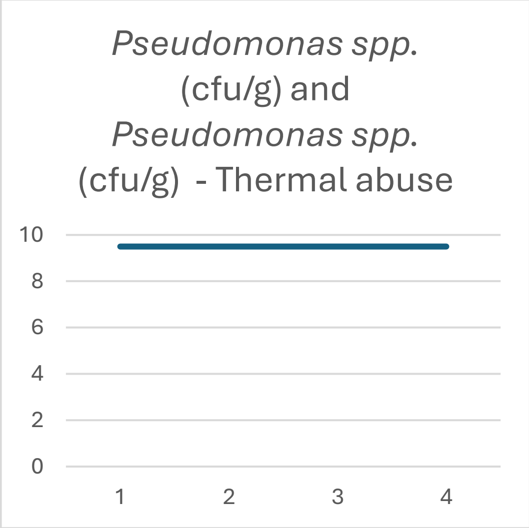
Graph 50



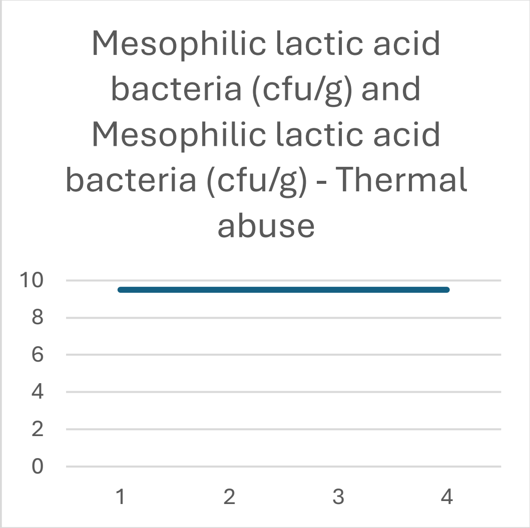
Graph 51



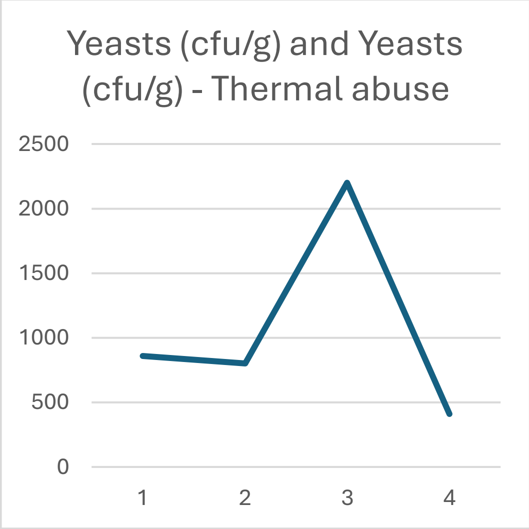
Graph 52



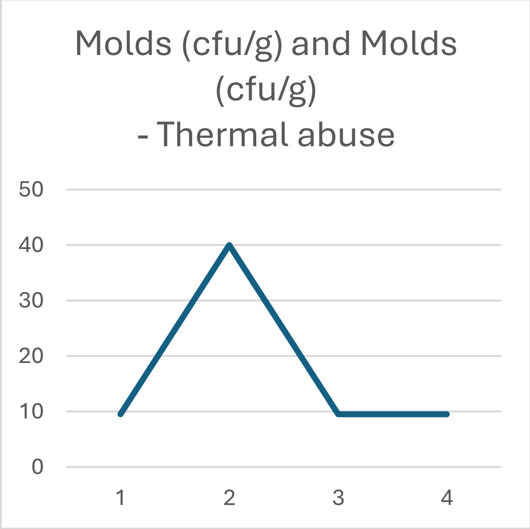
Graph 53



Graph 54



Graph 55

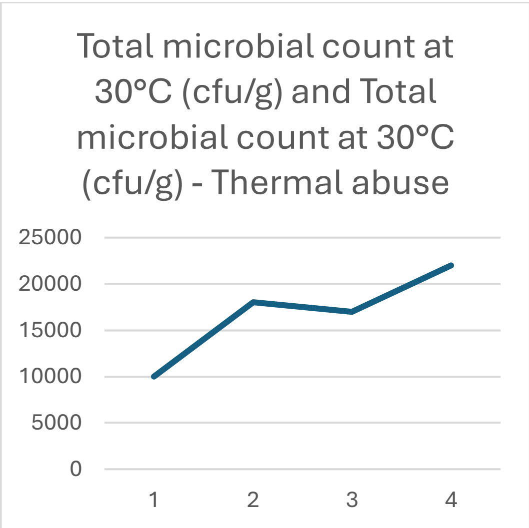


Graph 56

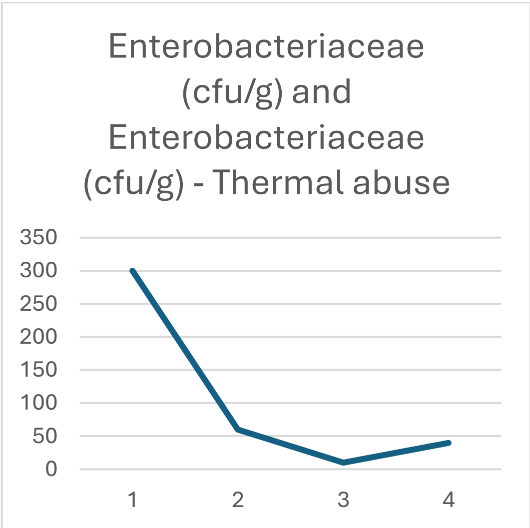
→ FRUIT SALAD (PINEAPPLE, BRAZILIAN MELON, KIWI, STRAWBERRIES)

	STORAGE AT +4°C			STORAGE AT +4°C FOR 48H, THEN AT +8°C
	T1 -DAY OF PRODUCTION- 24LB0036136	T3 - 48H FROM PRODUCTION- 24LB0036137	T4 - 72H FROM PRODUCTION- 24LB0036138	T4 - 72H FROM PRODUCTION- 24LB0036140
Ph	3,65	3,68	-	-
Wa	0,979	0.989	-	-
Acidity	0,56	0,58	-	-
Total microbial count at 30°C (cfu/g)	10.000	18.000	17.000	22.000
Enterobacteriaceae (cfu/g)	300	60	<10	40
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	800	2.600	2.100	100
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10
Yeasts (cfu/g)	1.800	500	960	7.500
Molds (cfu/g)	<10	<10	<10	40
Hepatitis A virus	Not detected	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	-

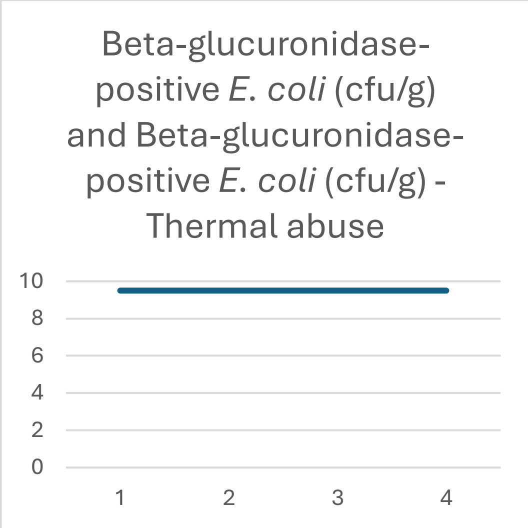
Table 8



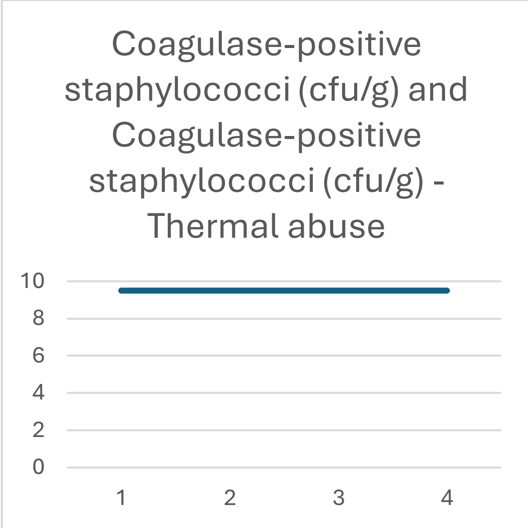
Graph 57



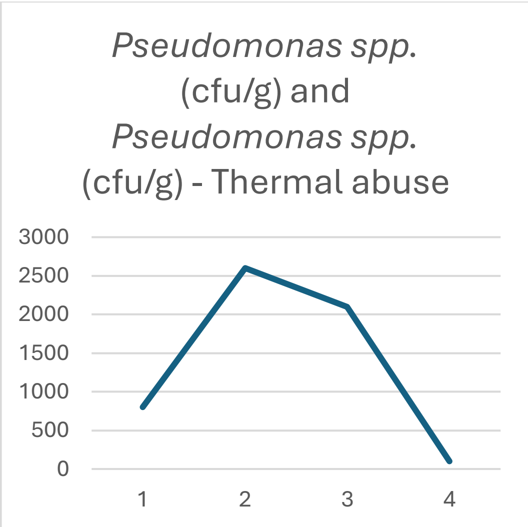
Graph 58



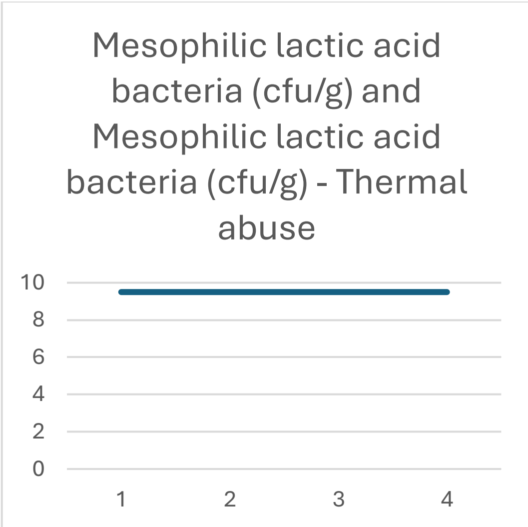
Graph 59



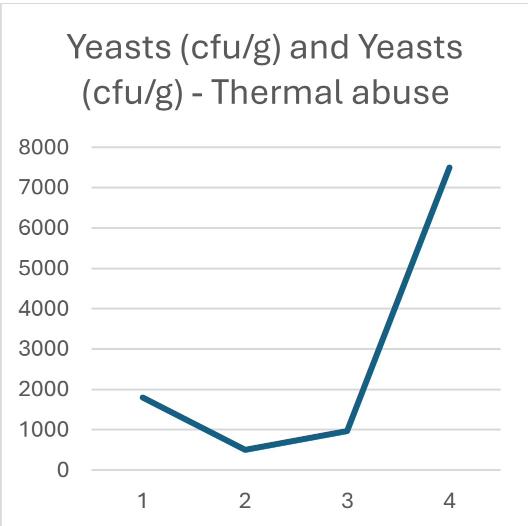
Graph 60



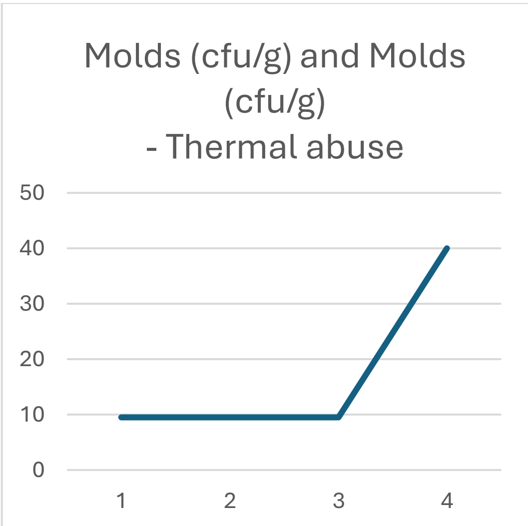
Graph 61



Graph 62



Graph 63



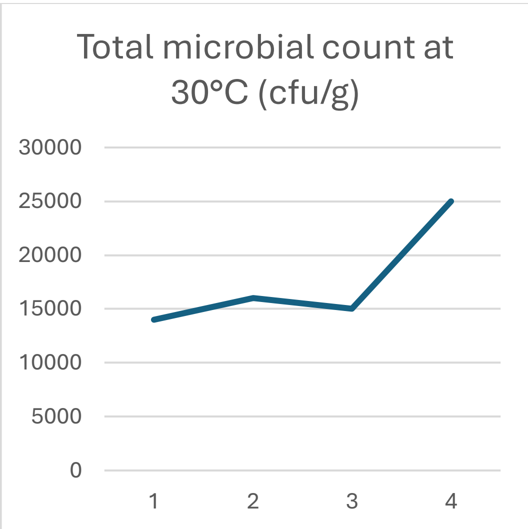
Graph 64

<u>OBSERVATIONS EMISFERO SCORZE'</u>

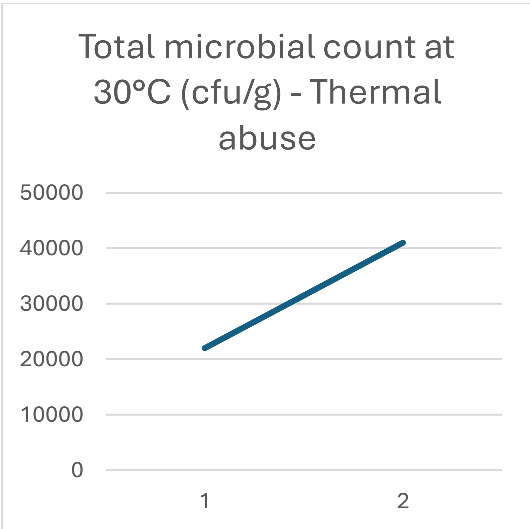
➔ FRUIT SALAD (PINEAPPLE, STRAWBERRIES, BLUEBERRIES, RASPBERRIES)

	STORAGE AT +4°C				STORAGE AT +4°C FOR 48H, THEN AT +8°C	
	T1 -DAY OF PRODUCTION- 24LB0036136	T3 - 48H FROM PRODUCTION- 24LB0036137	T4 - 72H FROM PRODUCTION- 24LB0036138	T5 - 96H FROM PRODUCTION- 24LB0036139	T4 - 72H FROM PRODUCTION- 24LB0036140	T5 - 96H FROM PRODUCTION- 24LB0036141
Ph	3,56	3,63	-	-	-	-
Wa	0,992	0.989	-	-	-	-
Acidity	0,92	0,98	-	-	-	-
Total microbial count at 30°C (cfu/g)	14.000	16.000	15.000	25.000	22.000	41.000
Enterobacteriaceae (cfu/g)	<10	<10	<10	<10	<10	<10
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	<10	<10	<10	<10	<10	<10
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10	<10	<10
Yeasts (cfu/g)	700	1.000	1.800	1.300	2.900	5.100
Molds (cfu/g)	700	1.800	1.400	1.500	2.000	5.300
Hepatitis A virus	Not detected	-	-	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant	-	-

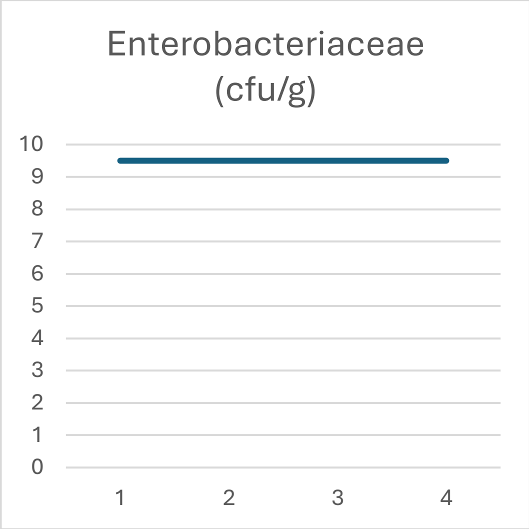
Table 9



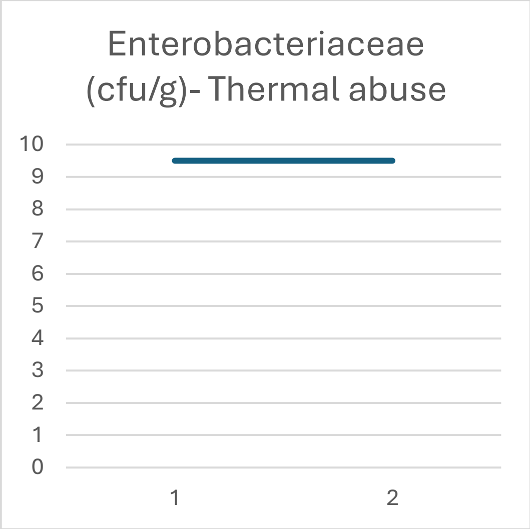
Graph 65



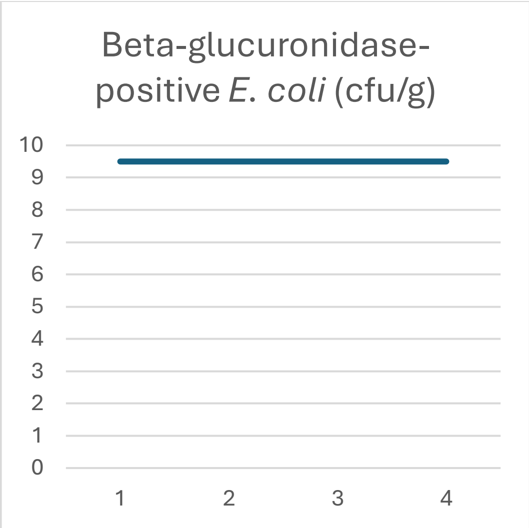
Graph 66



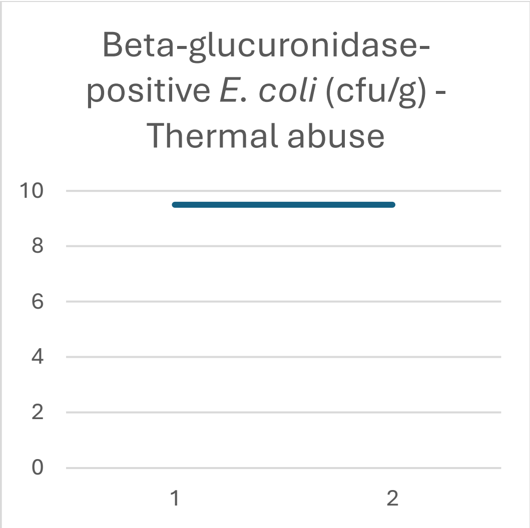
Graph 67



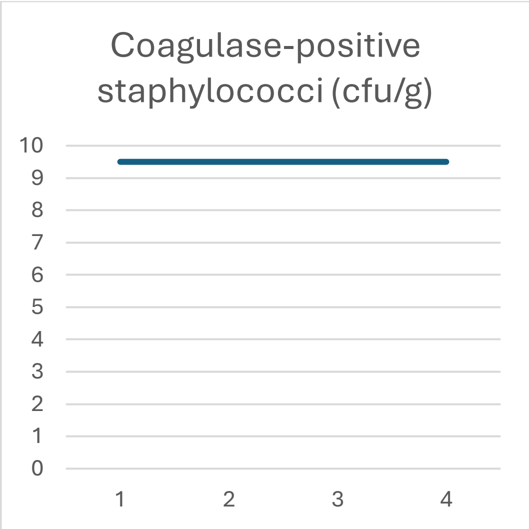
Graph 68



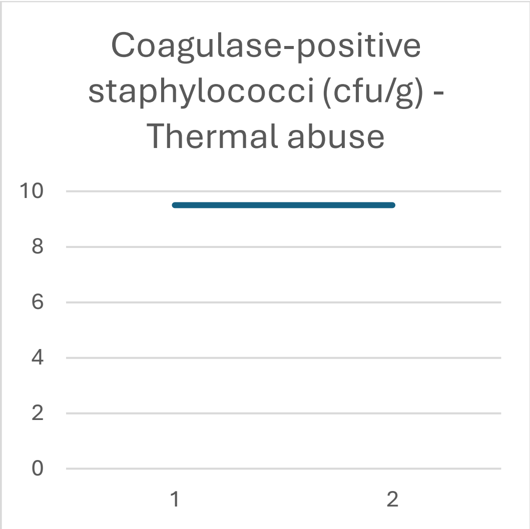
Graph 69



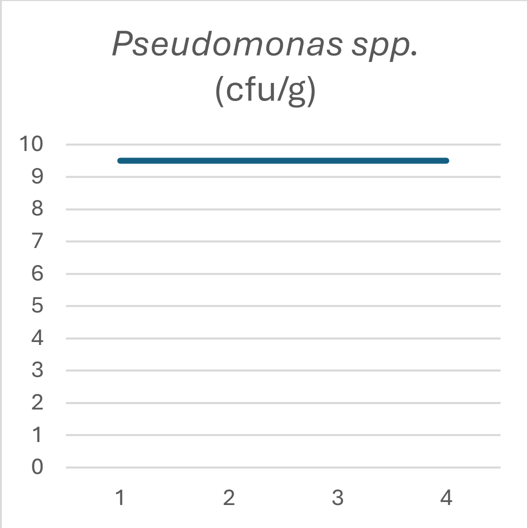
Graph 70



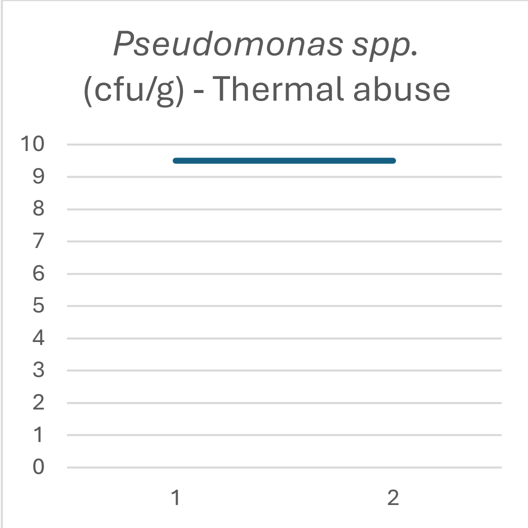
Graph 71



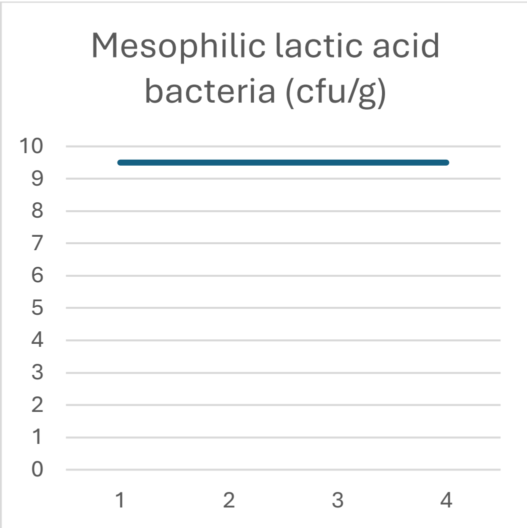
Graph 72



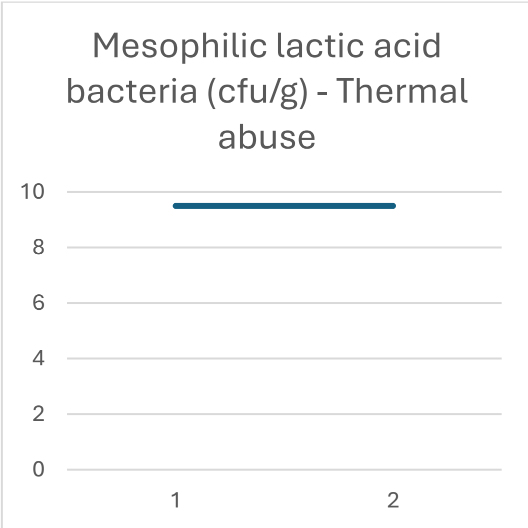
Graph 73



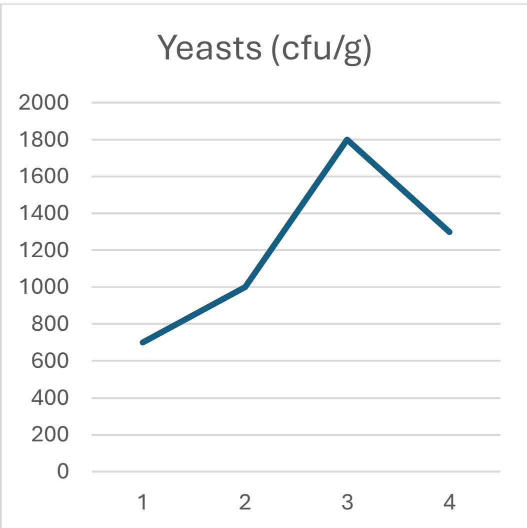
Graph 74



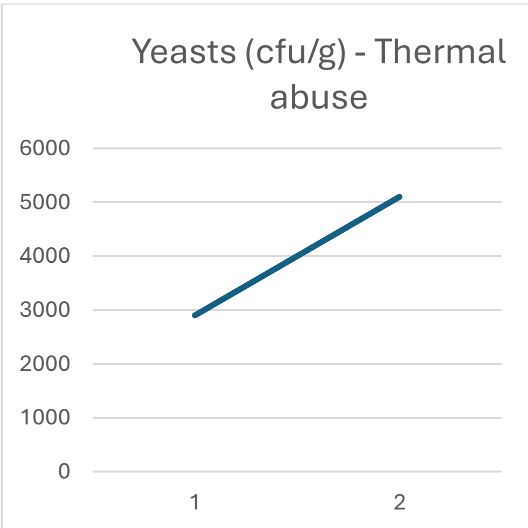
Graph 75



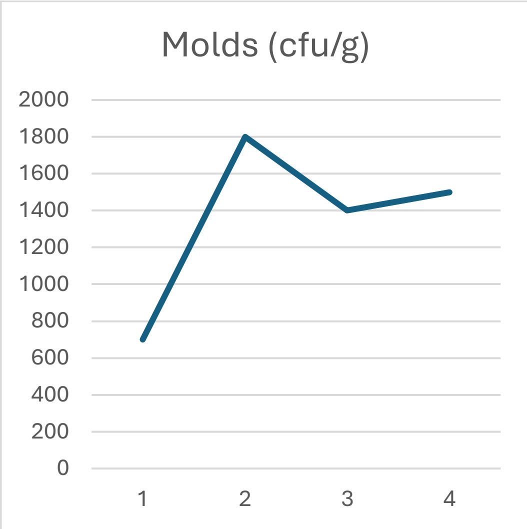
Graph 76



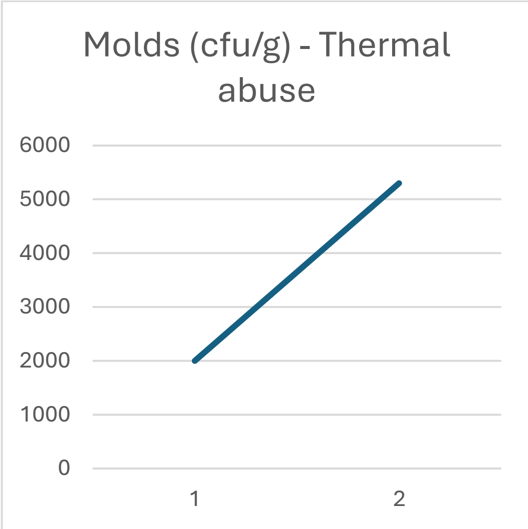
Graph 77



Graph 78



Graph 79

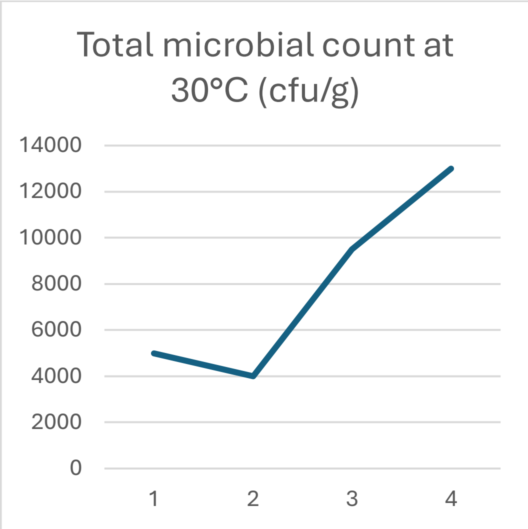


Graph 80

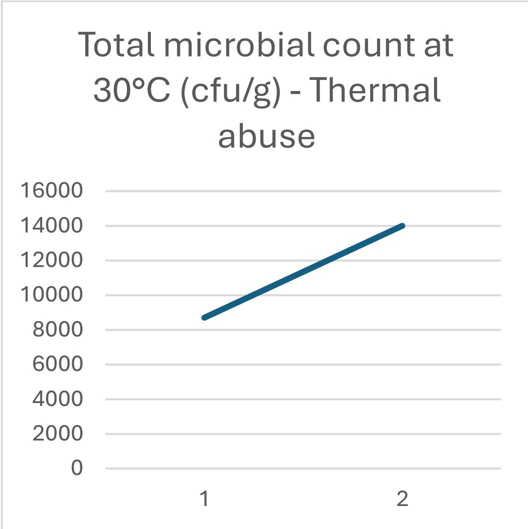
➔ PINEAPPLE, MELON, KIWI, STRAWBERRY, WHITE GRAPES, BLACK GRAPES, BLUEBERRIES, RASPBERRIES

	STORAGE AT +4°C				STORAGE AT +4°C FOR 48H, THEN AT +8°C	
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T5 - 96H FROM PRODUCTION-24LB0036139	T4 - 72H FROM PRODUCTION-24LB0036140	T5 - 96H FROM PRODUCTION-24LB0036141
Ph	3,62	3,48	-	-	-	-
Wa	0,99	0.992	-	-	-	-
Acidity	0,81	0,83	-	-	-	-
Total microbial count at 30°C (cfu/g)	5.000	4.000	9.500	13.000	8.700	14.000
Enterobacteriaceae (cfu/g)	<10	40	40	40	60	70
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	<10	<10	<10	<10	<10	<10
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10	<10	<10
Yeasts (cfu/g)	970	1.100	1.000	1.000	1.800	3.800
Molds (cfu/g)	680	2.400	6.000	3.500	1.600	2.800
Hepatitis A virus	Not detected	-	-	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant	-	-

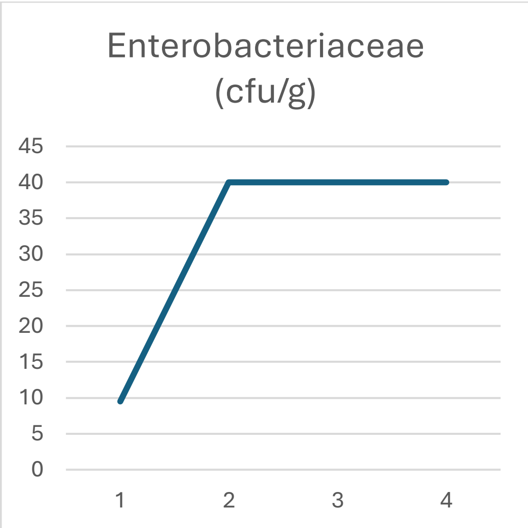
Table 10



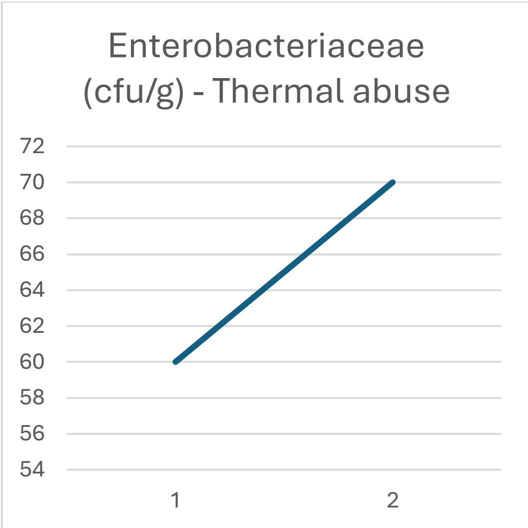
Graph 81



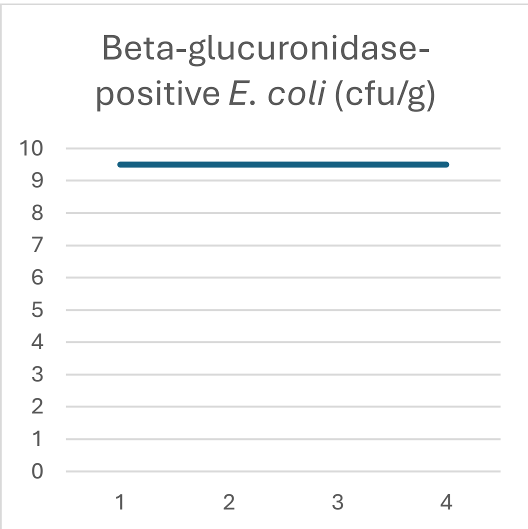
Graph 82



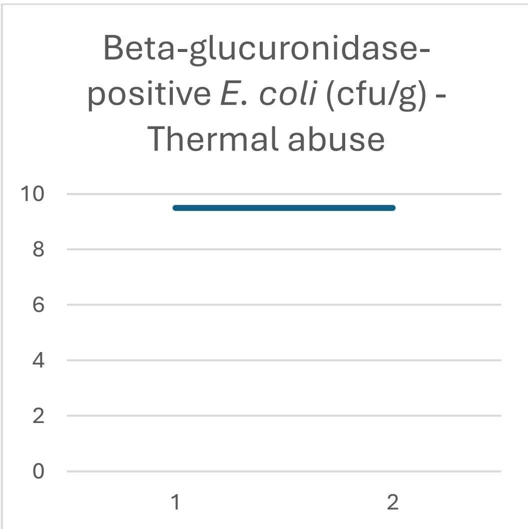
Graph 83



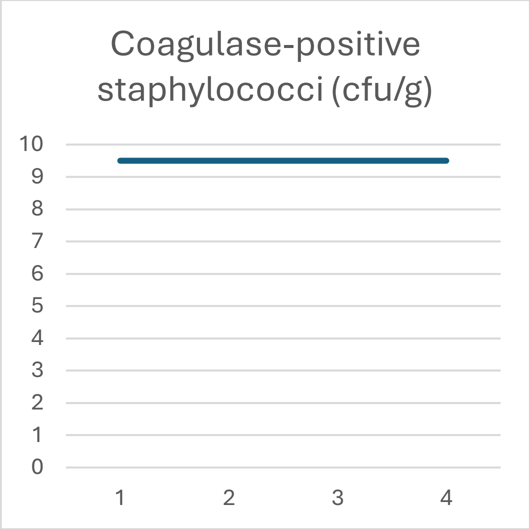
Graph 84



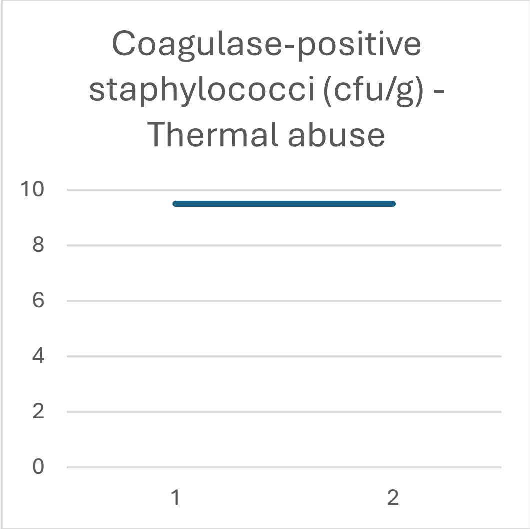
Graph 85



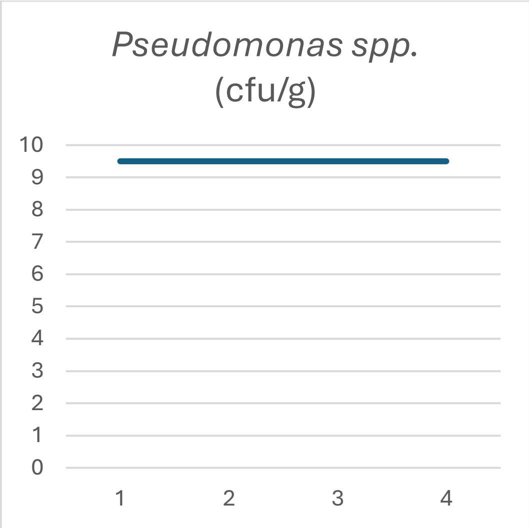
Graph 86



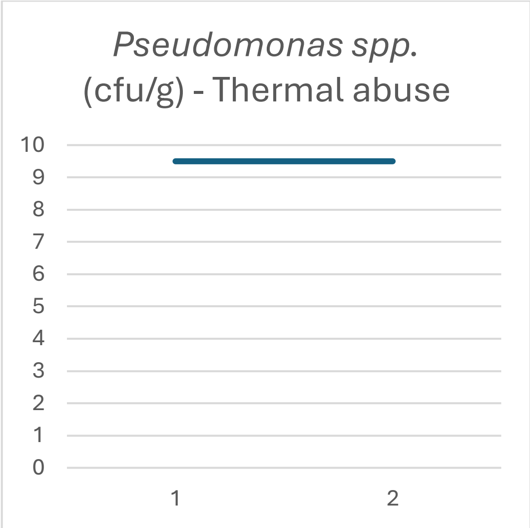
Graph 87



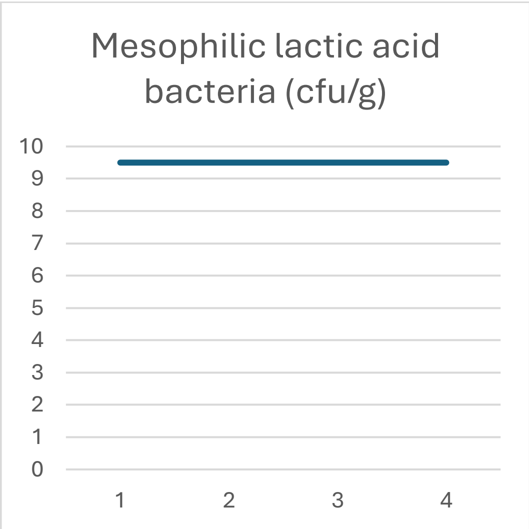
Graph 88



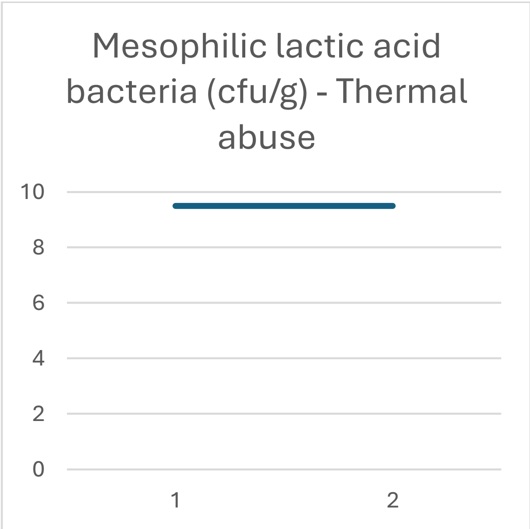
Graph 89



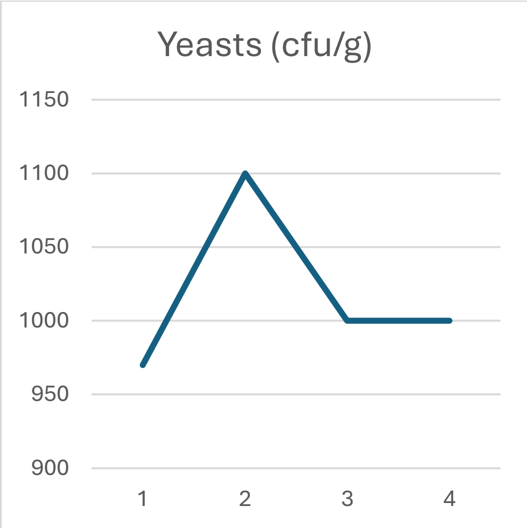
Graph 90



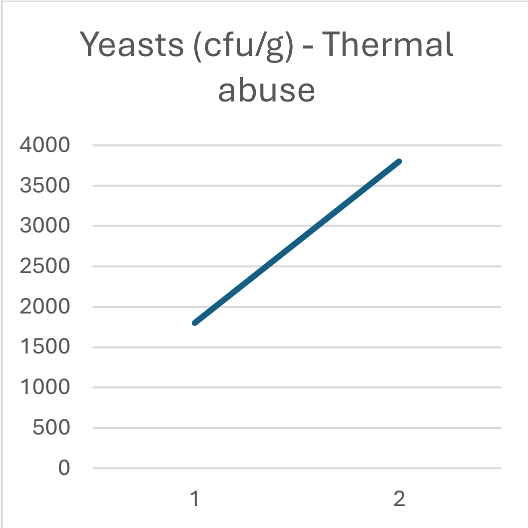
Graph 91



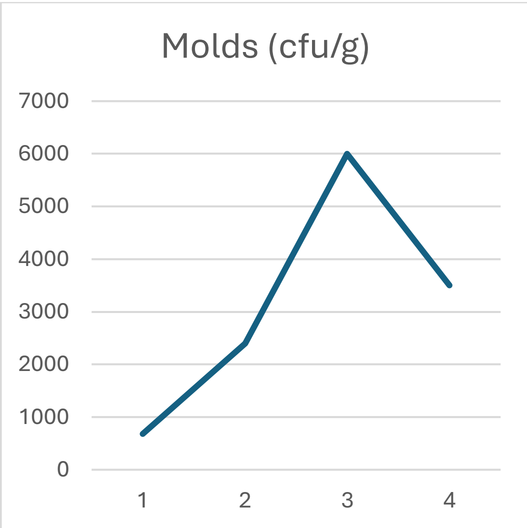
Graph 92



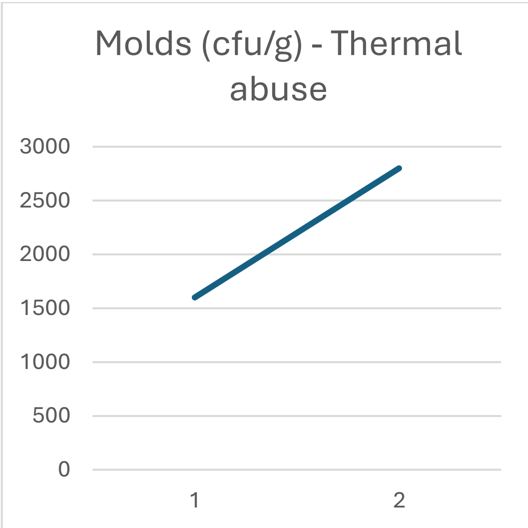
Graph 93



Graph 94



Graph 95



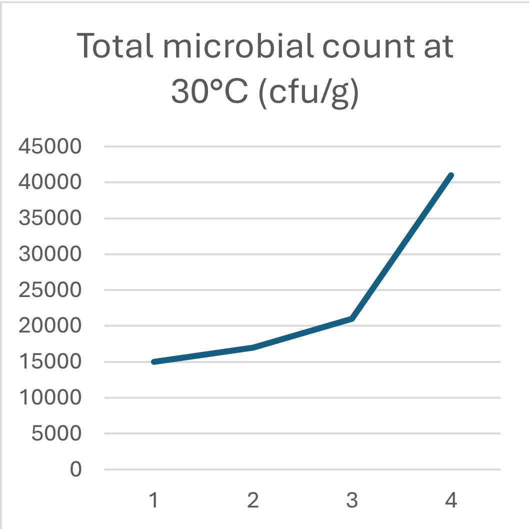
Graph 96

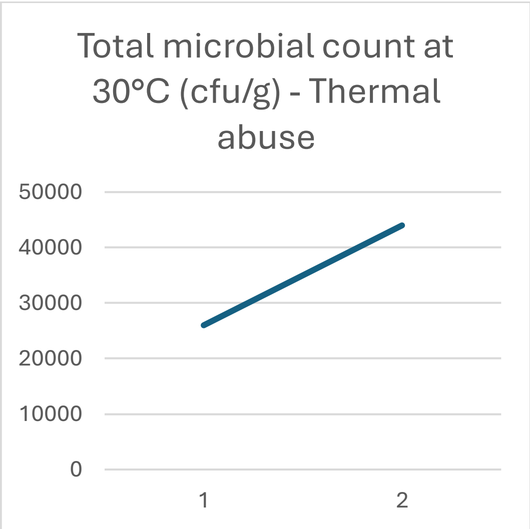
<u>OBSERVATIONS EMISFERO BASSANO</u>

➔ PINEAPPLE, STRAWBERRIES, BLUEBERRIES, RASPBERRIES

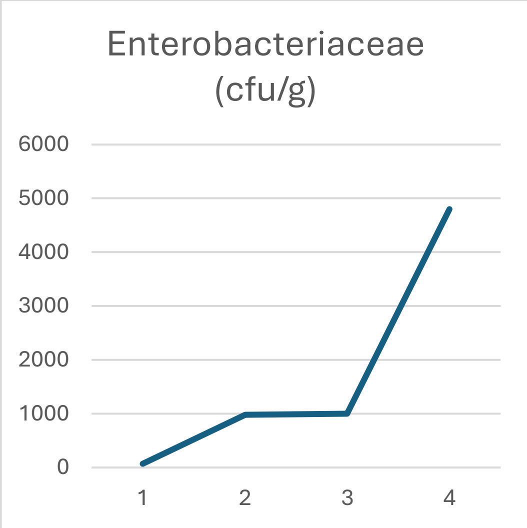
	STORAGE AT +4°C				STORAGE AT +4°C FOR 48H, THEN AT +8°C	
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T5 - 96H FROM PRODUCTION-24LB0036139	T4 - 72H FROM PRODUCTION-24LB0036140	T5 - 96H FROM PRODUCTION-24LB0036141
Ph	3,84	3,53	-	-	-	-
Wa	0,989	0.998	-	-	-	-
Acidity	0,80	0,86	-	-	-	-
Total microbial count at 30°C (cfu/g)	15.000	17.000	21.000	41.000	26.000	44.000
Enterobacteriaceae (cfu/g)	70	980	1.000	4.800	920	3.200
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	<10	400	250	5.300	1.400	6.000
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10	<10	<10
Yeasts (cfu/g)	1.500	4.000	6.800	3.900	8.000	6.000
Molds (cfu/g)	1.100	3.200	5.400	6.000	6.400	2.500
Hepatitis A virus	Not detected	-	-	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant	-	-

Table 11

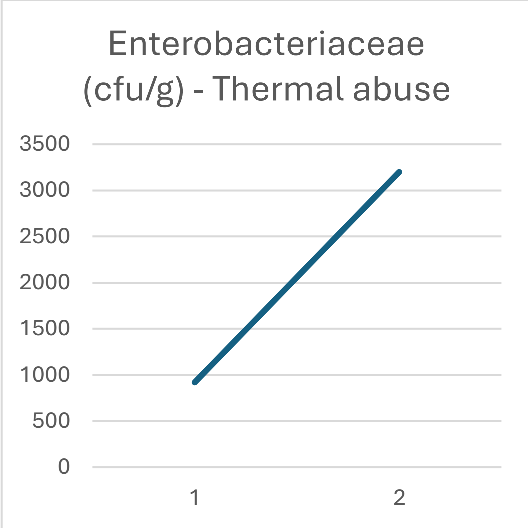




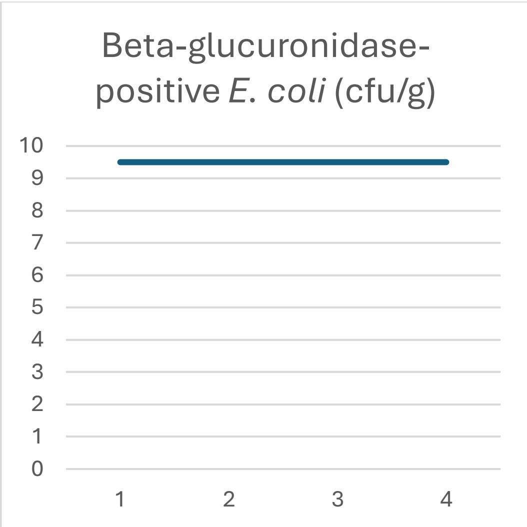
Graph 97 **Graph 98**



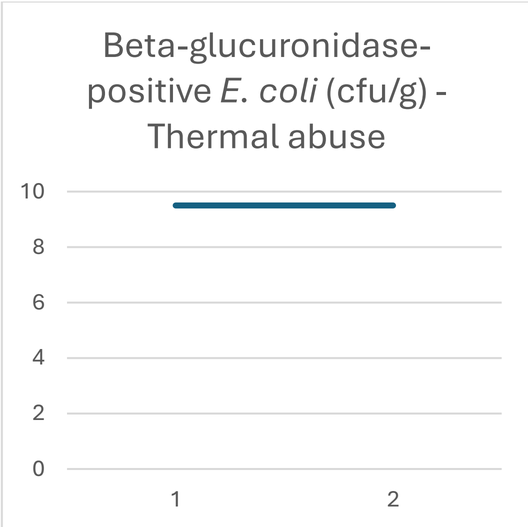
Graph 99



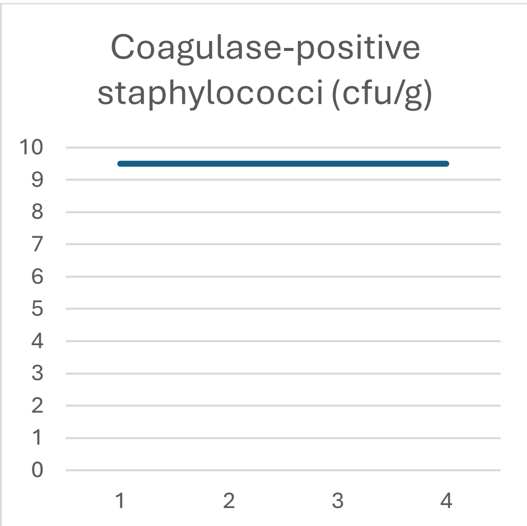
Graph 100



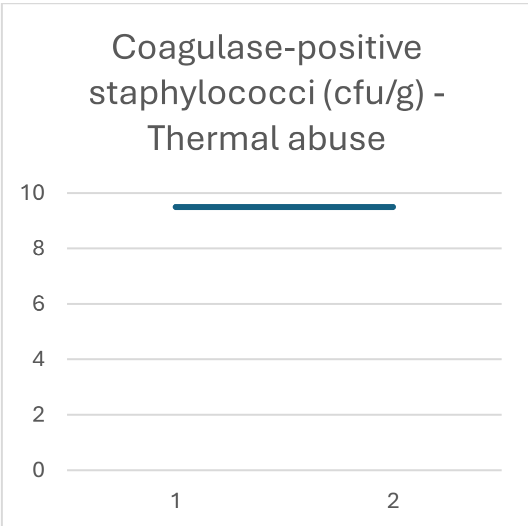
Graph 101



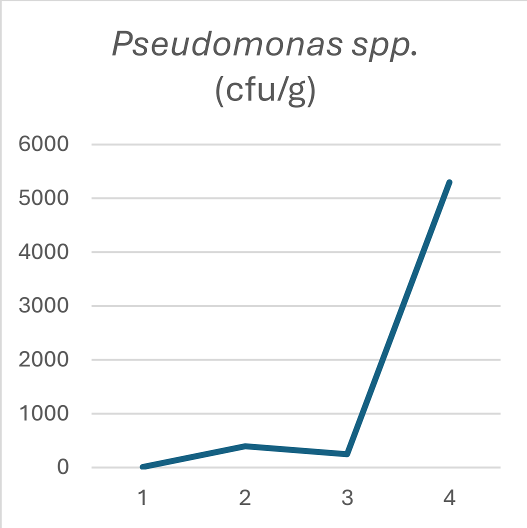
Graph 102



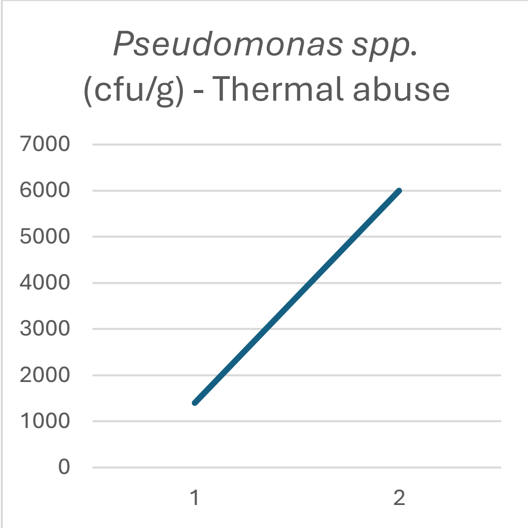
Graph 103



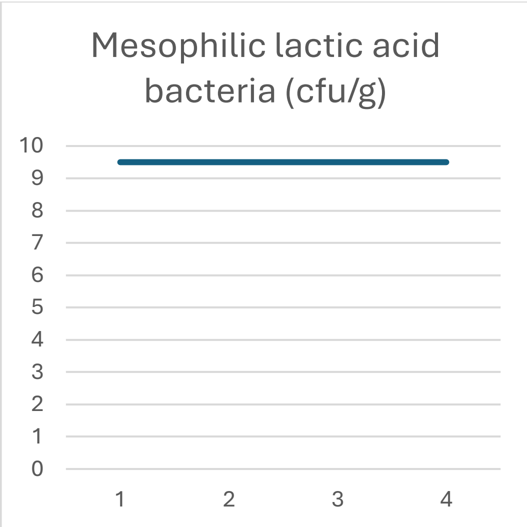
Graph 104



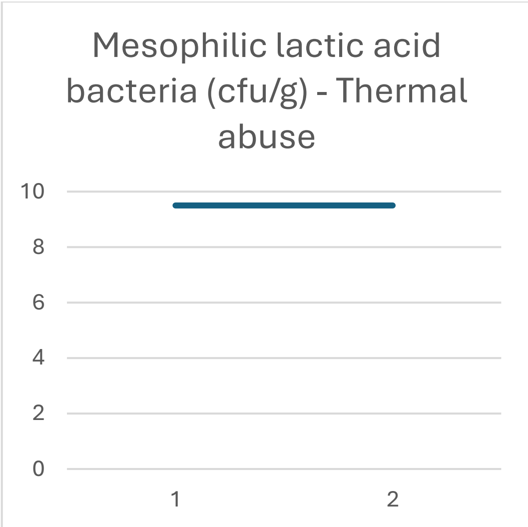
Graph 105



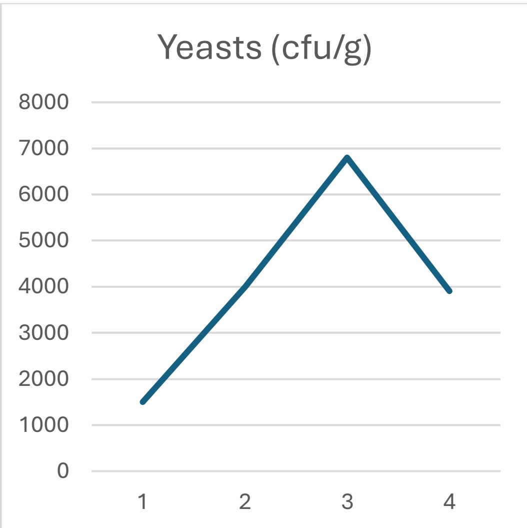
Graph 106



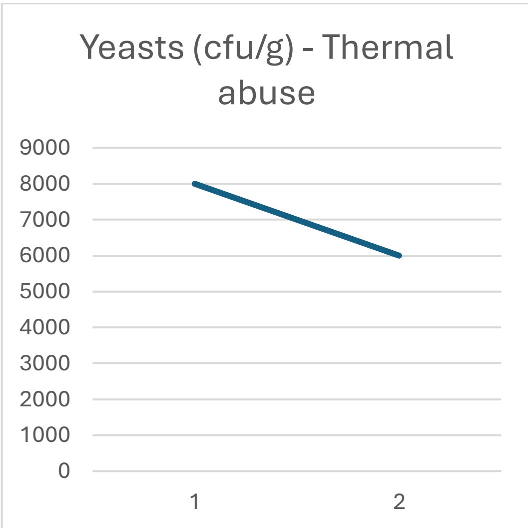
Graph 107



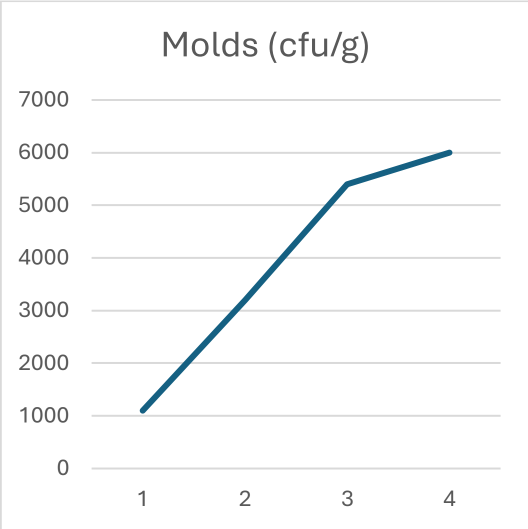
Graph 108

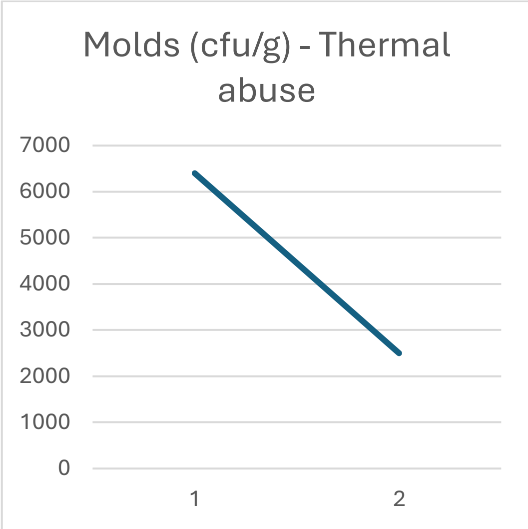


Graph 109



Graph 110



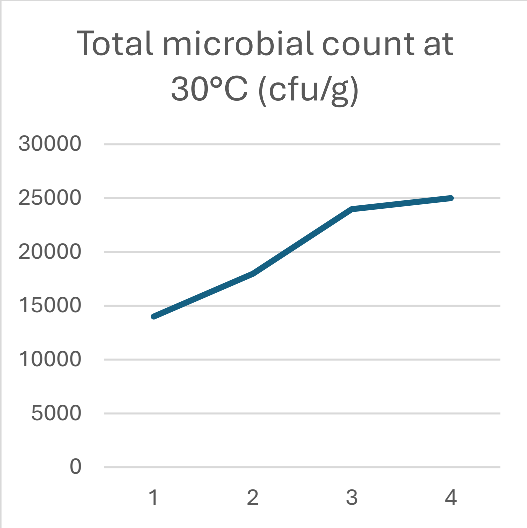


Graph 111 **Graph 112**

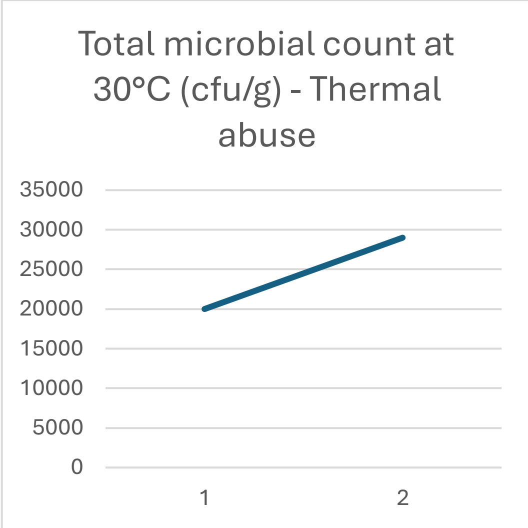
➔ PINEAPPLE, MELON, KIWI, STRAWBERRY, WHITE GRAPES, BLACK GRAPES, BLUEBERRIES, RASPBERRIES

	STORAGE AT +4°C				STORAGE AT +4°C FOR 48H, THEN AT +8°C	
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T5 - 96H FROM PRODUCTION-24LB0036139	T4 - 72H FROM PRODUCTION-24LB0036140	T5 - 96H FROM PRODUCTION-24LB0036141
Ph	3,88	3,48	-	-	-	-
Wa	0,993	0.995	-	-	-	-
Acidity	0,80	0,86	-	-	-	-
Total microbial count at 30°C (cfu/g)	14.000	18.000	24.000	25.000	20.000	29.000
Enterobacteriaceae (cfu/g)	40	40	<10	<10	40	<10
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	2.100	7.000	5.000	3.000	11.000	8.000
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10	<10	<10
Yeasts (cfu/g)	1.100	6.400	3.100	7.000	3.000	1.200
Molds (cfu/g)	100	210	900	2.500	2.200	3.000
Hepatitis A virus	Not detected	-	-	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant	-	-

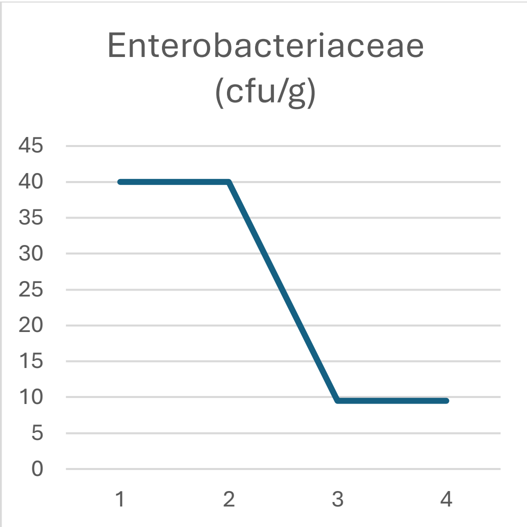
Table 12



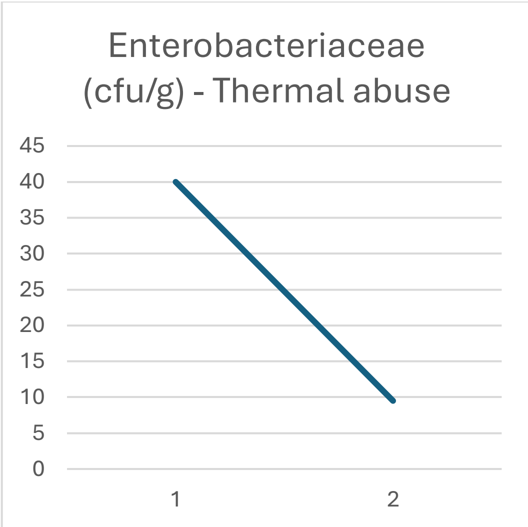
Graph 113



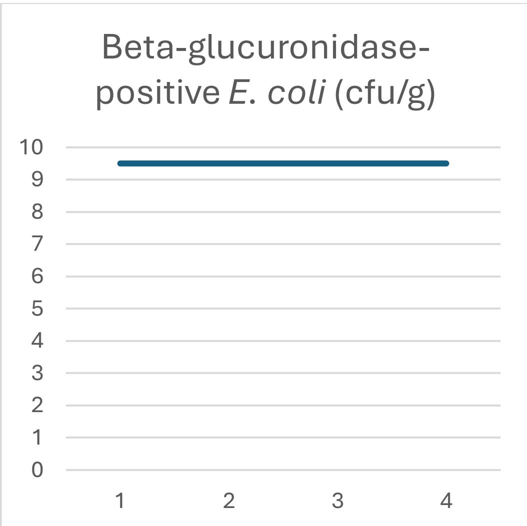
Graph 114



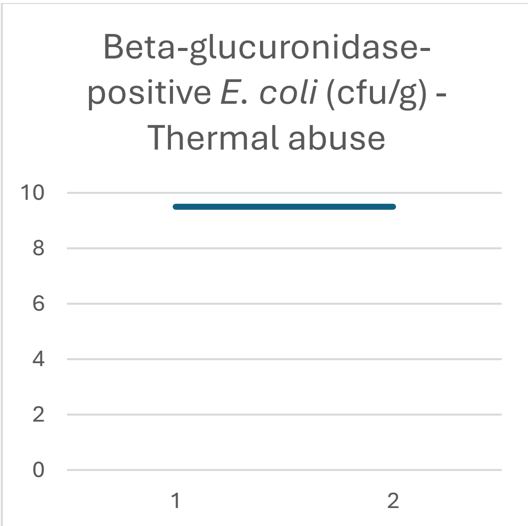
Graph 115



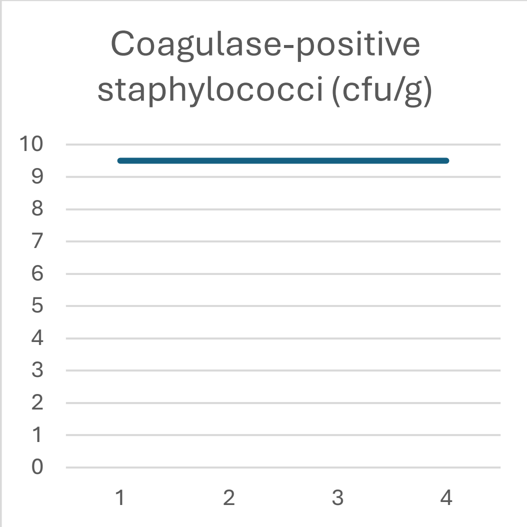
Graph 116



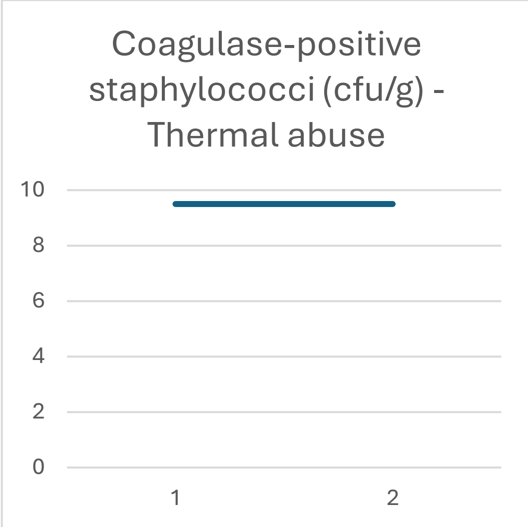
Graph 117



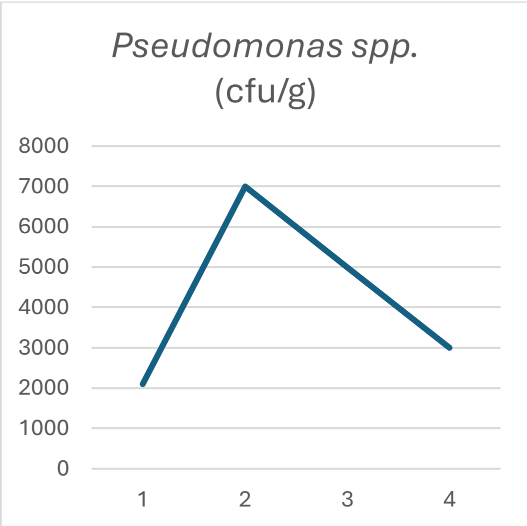
Graph 118



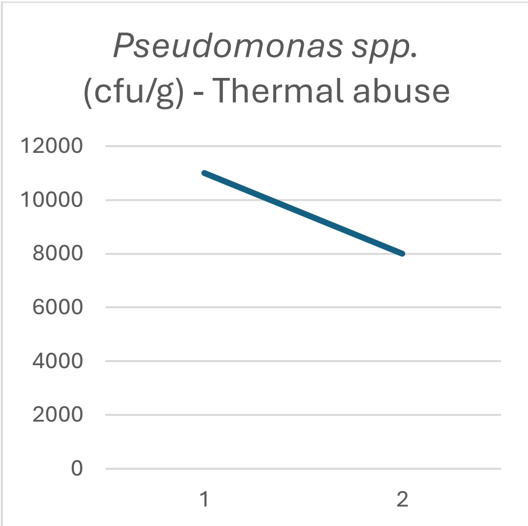
Graph 119



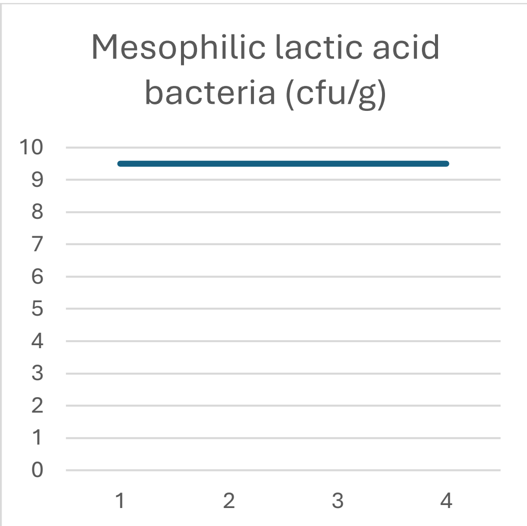
Graph 120



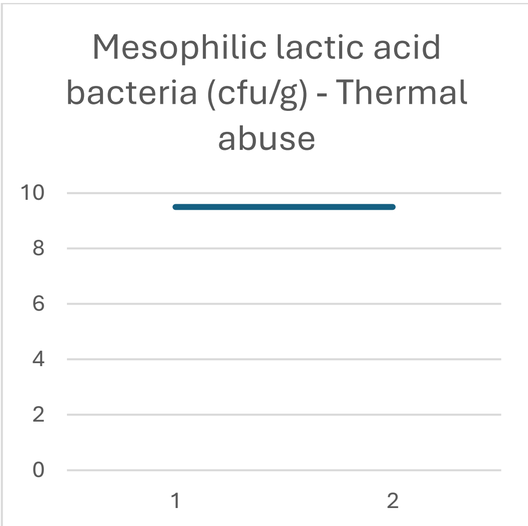
Graph 121



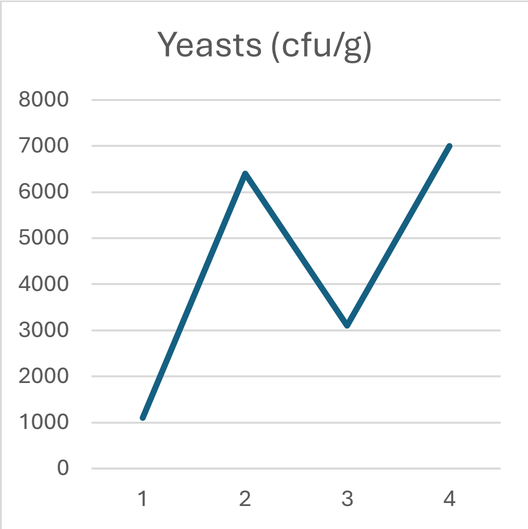
Graph 122



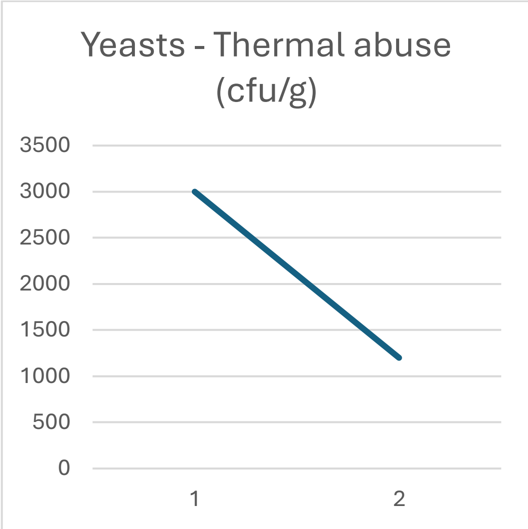
Graph 123



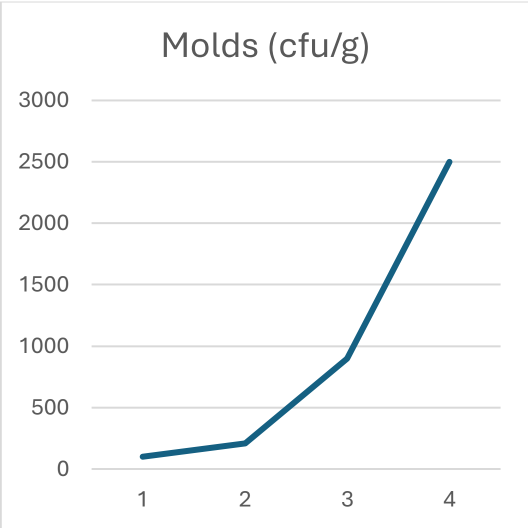
Graph 124



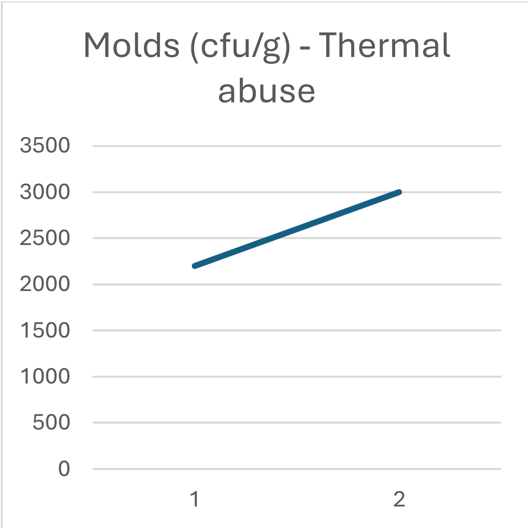
Graph 125



Graph 126



Graph 127



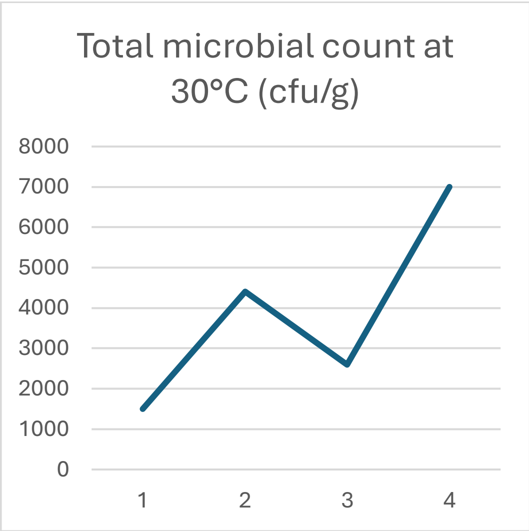
Graph 128

OBSERVATIONS EMISFERO VICENZA

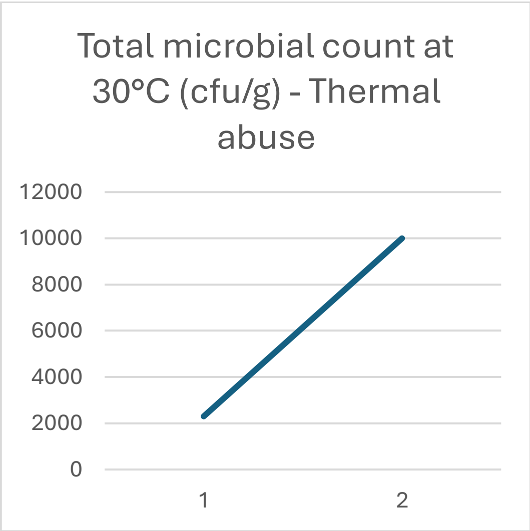
➔ PINEAPPLE, STRAWBERRIES, BLUEBERRIES, RASPBERRIES

	STORAGE AT +4°C				STORAGE AT +4°C FOR 48H, THEN AT +8°C	
	T1 -DAY OF PRODUCTION- 24LB0036136	T3 - 48H FROM PRODUCTION- 24LB0036137	T4 - 72H FROM PRODUCTION- 24LB0036138	T5 - 96H FROM PRODUCTION- 24LB0036139	T4 - 72H FROM PRODUCTION- 24LB0036140	T5 - 96H FROM PRODUCTION- 24LB0036141
Ph	3,42	3,82	-	-	-	-
Wa	0,994	0.993	-	-	-	-
Acidity	0,75	0,80	-	-	-	-
Total microbial count at 30°C (cfu/g)	1.500	4.400	2.600	7.000	2.300	10.000
Enterobacteriaceae (cfu/g)	50	<10	<10	<10	<10	<10
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	<10	<10	<10	940	<10	700
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10	<10	<10
Yeasts (cfu/g)	400	1.000	260	920	360	160
Molds (cfu/g)	500	960	140	570	960	3.400
Hepatitis A virus	Not detected	-	-	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant	-	-

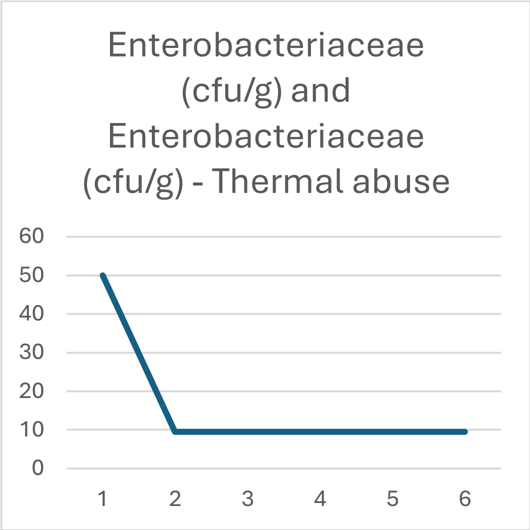
Table 13



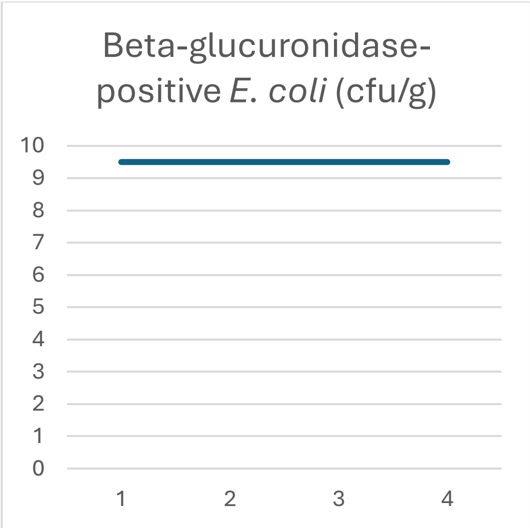
Graph 129



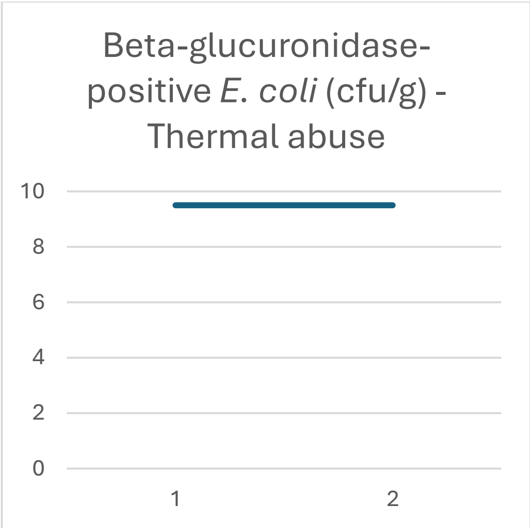
Graph 130



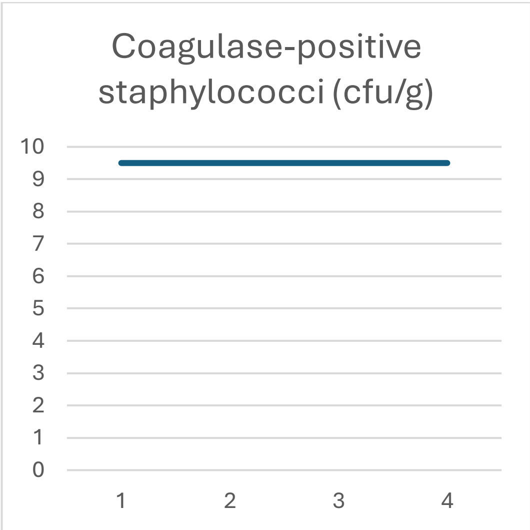
Graph 131



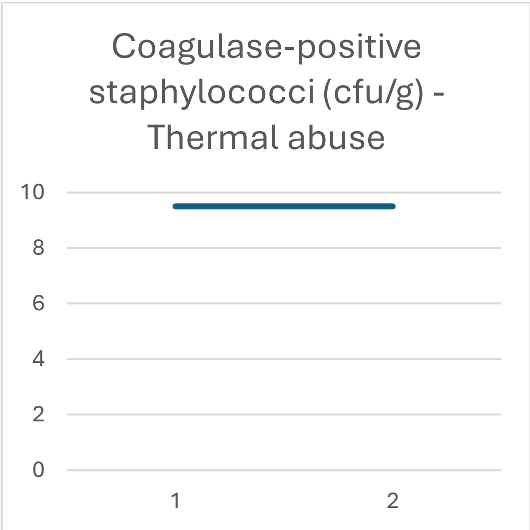
Graph 132



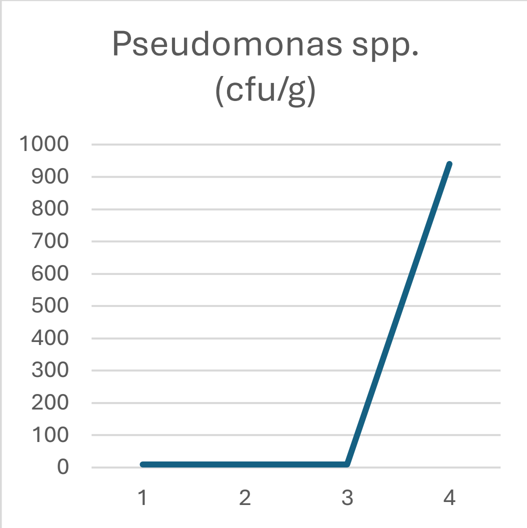
Graph 133



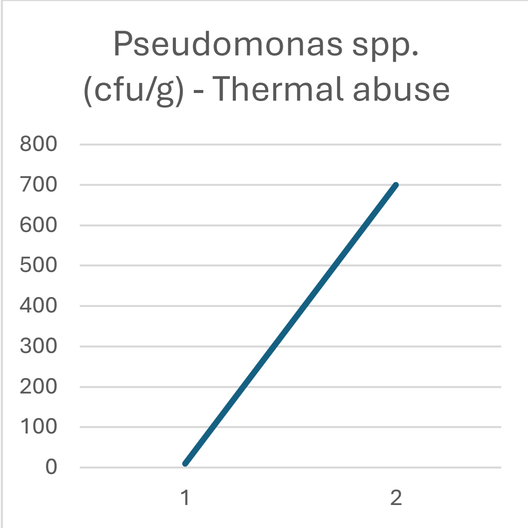
Graph 134



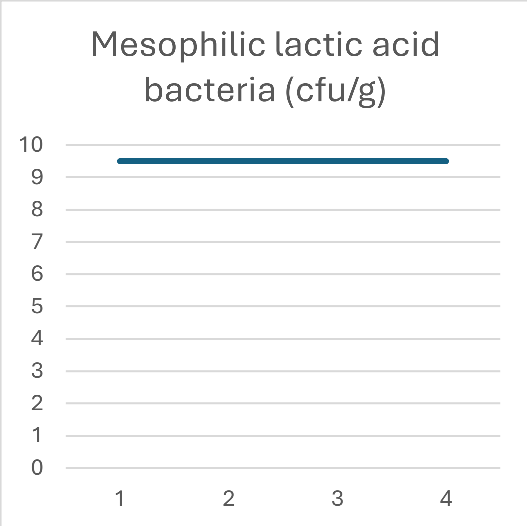
Graph 135



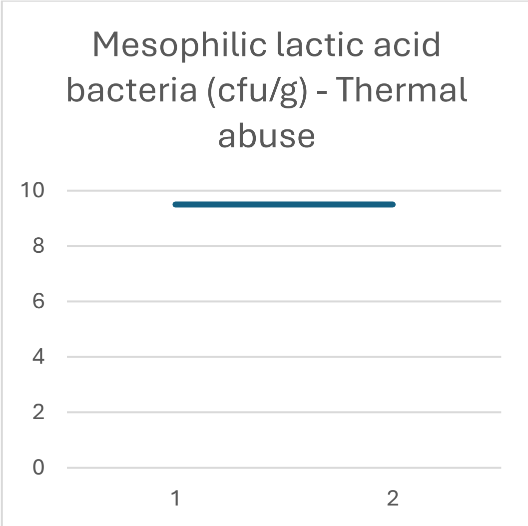
Graph 136



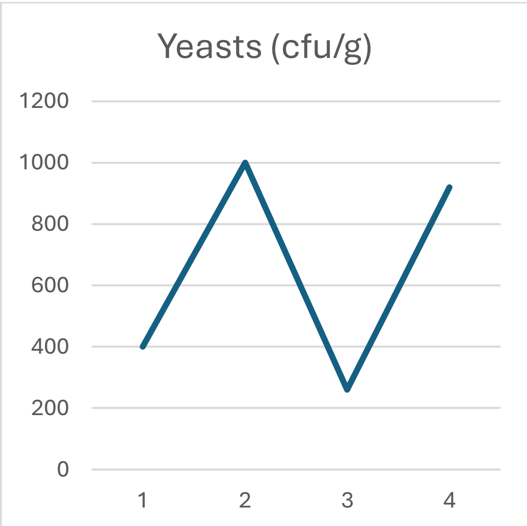
Graph 137



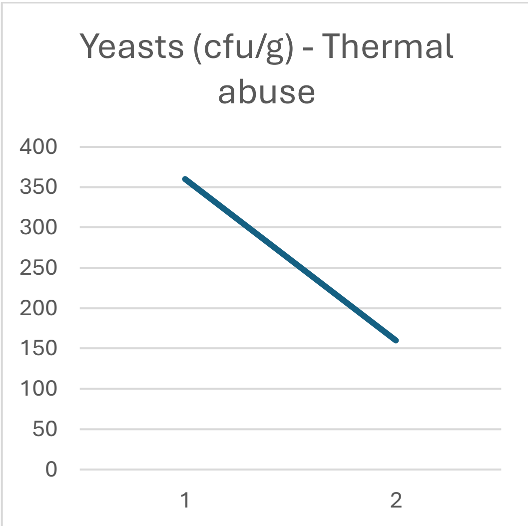
Graph 138



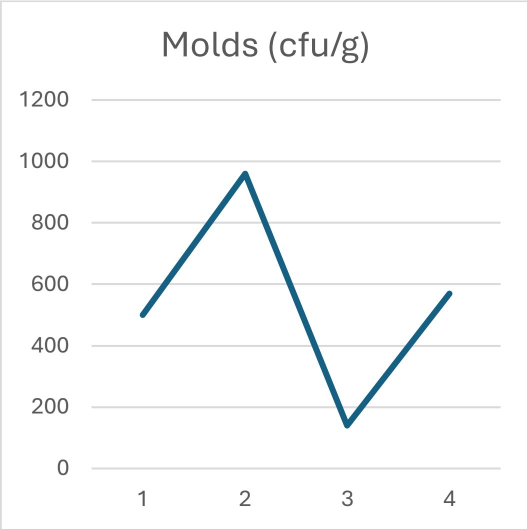
Graph 139

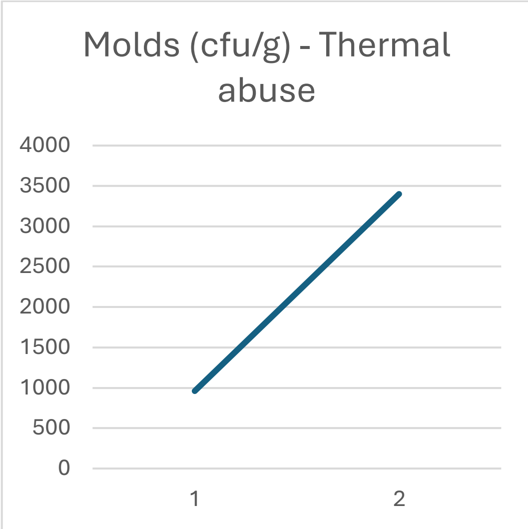


Graph 140



Graph 141



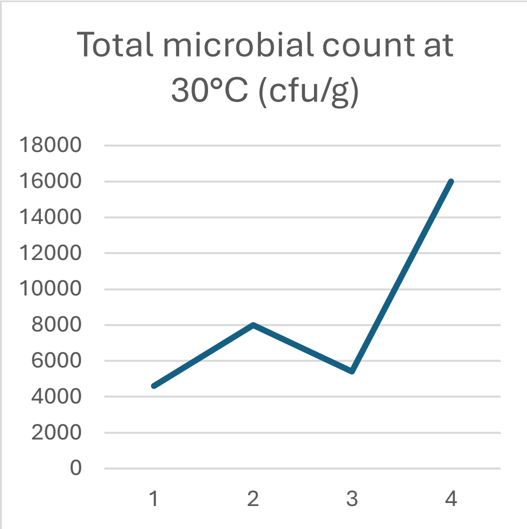


Graph 142 **Graph 14**

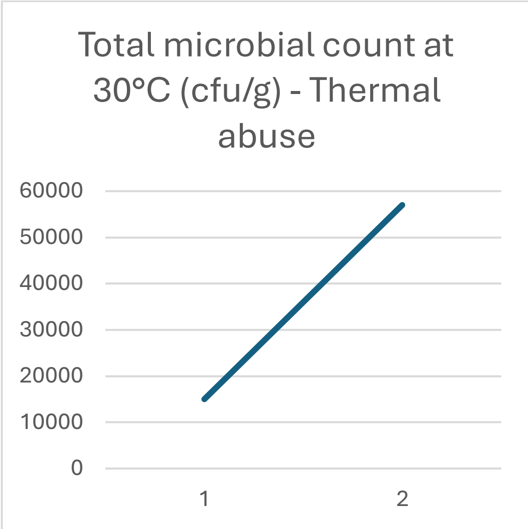
➔ PINEAPPLE, MELON, KIWI, STRAWBERRY, WHITE GRAPES, BLACK GRAPES, BLUEBERRIES, RASPBERRIES

	STORAGE AT +4°C				STORAGE AT +4°C FOR 48H, THEN AT +8°C	
	T1 -DAY OF PRODUCTION-24LB0036136	T3 - 48H FROM PRODUCTION-24LB0036137	T4 - 72H FROM PRODUCTION-24LB0036138	T5 - 96H FROM PRODUCTION-24LB0036139	T4 - 72H FROM PRODUCTION-24LB0036140	T5 - 96H FROM PRODUCTION-24LB0036141
Ph	3,72	3,72	-	-	-	-
Wa	0,989	0.994	-	-	-	-
Acidity	0,92	0,93	-	-	-	-
Total microbial count at 30°C (cfu/g)	4.600	8.000	5.400	16.000	15.000	57.000
Enterobacteriaceae (cfu/g)	<10	<10	<10	40	<10	<10
Beta-glucuronidase-positive *E. coli* (cfu/g)	<10	<10	<10	<10	<10	<10
Coagulase-positive staphylococci (cfu/g)	<10	<10	<10	<10	<10	<10
Pseudomonas spp. (cfu/g)	1.000	2.000	5.300	9.600	6.500	10.000
Mesophilic lactic acid bacteria (cfu/g)	<10	<10	<10	<10	<10	<10
Yeasts (cfu/g)	1100	480	560	9.200	3.000	1.200
Molds (cfu/g)	880	700	510	350	1400	330
Hepatitis A virus	Not detected	-	-	-	-	-
Organoleptic test	Compliant	Compliant	Compliant	Compliant	-	-

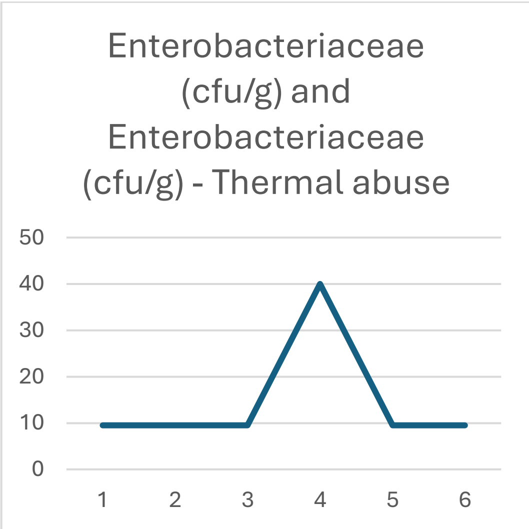
Table 14



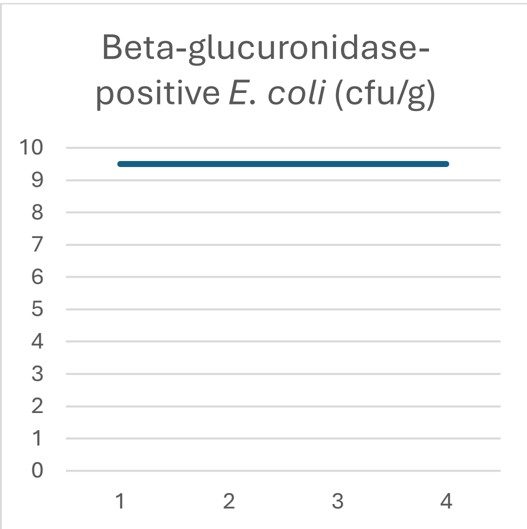
Graph 144



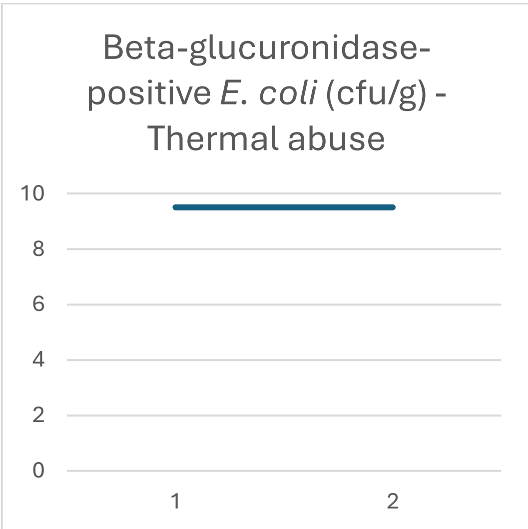
Graph 145



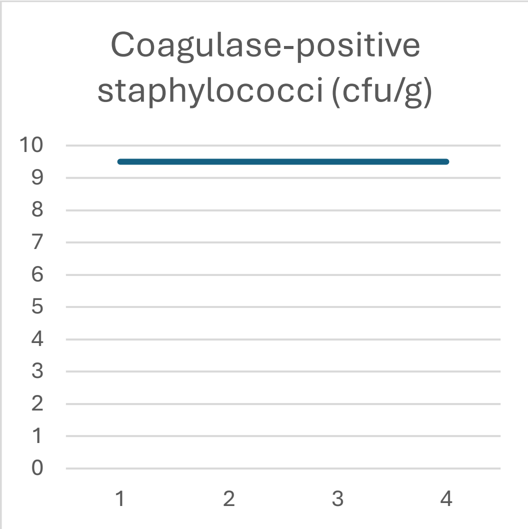
Graph 146



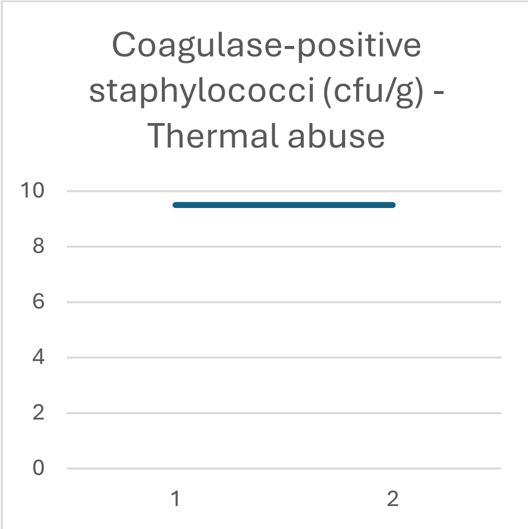
Graph 147



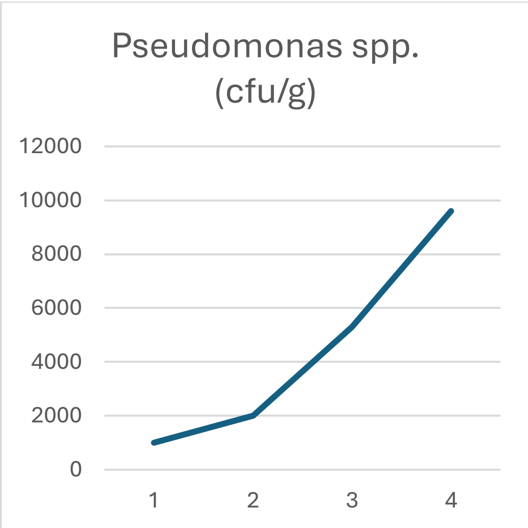
Graph 148



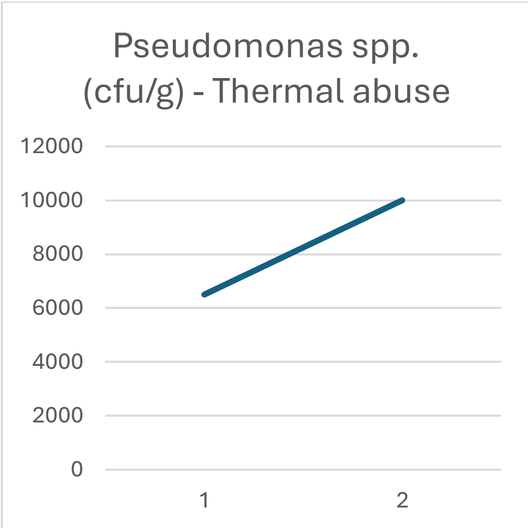
Graph 149



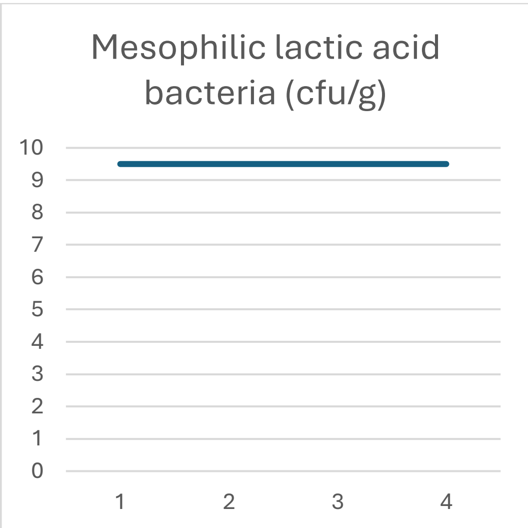
Graph 150



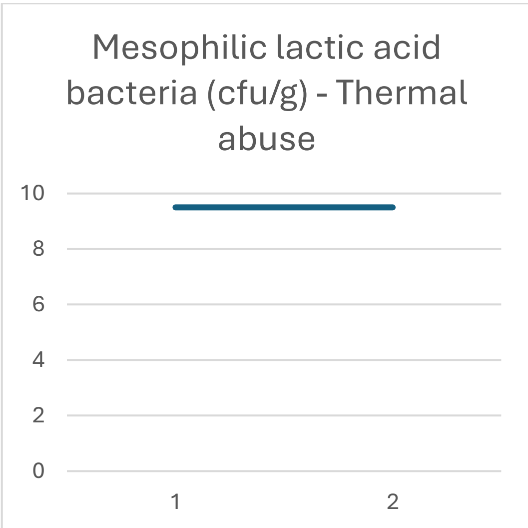
Graph 151



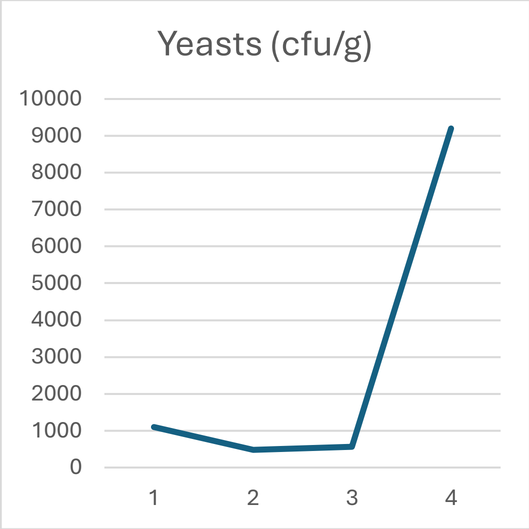
Graph 152

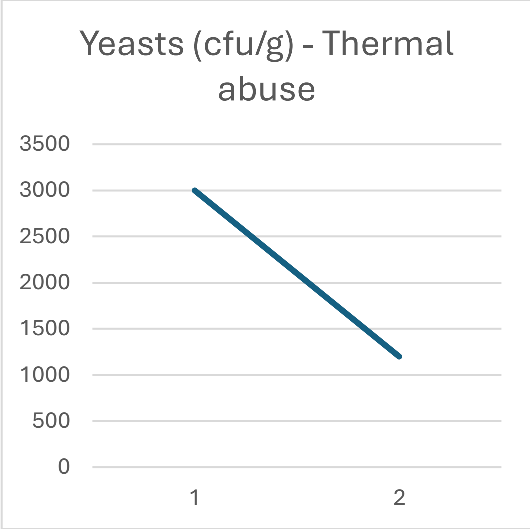


Graph 153



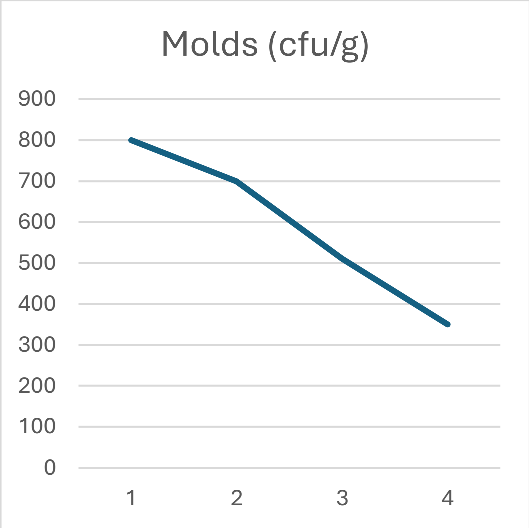
Graph 154

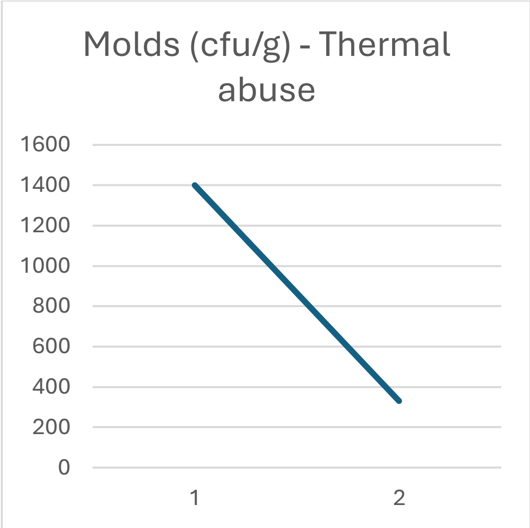




Graph 155

Graph 156





Graph 157

Graph 158

11. DISCUSSION OF THE RESULTS AND CONCLUSIONS

Over the course of this study, we observed that every measured day (up to 72 or 96 hours depending on the fruit salad considered), the measured values of all the microbiological parameters for both types of fruit salads, across all the sales points where tests were conducted, met the strict safety requirements for human consumption.

All the parameters were in full accordance with the values underlined in Regulation (EC) No 2073/2005 (29).

The fruit salads that were subjected to temperature abuse tests, designed to simulate non-optimal storage conditions assessing the durability of the product, maintained acceptable safety standards.

The abuse test is important to provide understanding of how the product performs when maintained in conditions not completely suitable for optimal preservation, that is a crucial consideration for both retailers and food industries.

The storage tests have been conducted in 4 types of fruit salads, produced and sold in retail shops by UNICOMM srl, to be precise, these samples has been taken from six different Emisfero:

- Emisfero Perugia
- Emisfero Monfalcone
- Emisfero Fuimicino
- Emisfero Scorzè
- Emisfero Bassano
- Emisfero Vicenza

The planned tests had the purpose of evaluating the microbiological and sensorial duration of the fruit salads over a period of time. Has been included some thermal abuse tests to ensure that also in under standardized preservation conditions the results were reliable.

It is very important to conduct thermal abuse tests because they emulate the path followed by the fruit salads after retail, like the time during the transportation from the shop to the house and the time spent outside the consumer's fridge before consumption.

How it can be deduced from the numbers reported on the tables, the fruit salads result fit for human consumption for 72 hours after the production, that means that for three full days from the production, the fruit salads respect the safety limits set by the law. Over this period of time the product doesn’t represent a risk for the consumers.

These results are important for both consumers and retailers because now is known that the fruit salad maintain its freshness and safety during its now well-determined shelf life.

Retailers can now maintain in the shop and sell the product more consciously, giving to consumers more awareness about the time they have to consume the product after retail.

From the experiences I have been able to gain from carrying out this thesis of mine, I think I can finally deduce that it is particularly important to ensure that all foods (and particularly those that are more perishable from a microbiological point of view)of proper product handling, storage, and adherence to food safety regulations, ensuring that the final product meets the high standards expected by consumers in the retail food market.

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