THESIS

IMPACT OF FROZEN STORAGE ON GROUND BEEF QUALITY AND ARTIFICIAL INTELLIGENCE STRATEGIES FOR ENERGY OPTIMIZATION IN MEAT COLD STORAGE

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ABSTRACT

IMPACT OF FROZEN STORAGE ON GROUND BEEF QUALITY AND ARTIFICIAL INTELLIGENCE STRATEGIES FOR ENERGY OPTIMIZATION IN MEAT COLD STORAGE

Two independent studies were conducted to evaluate the effectiveness of temperature intensive management in frozen food storage. The first study was designed to elucidate the impact of experimental frozen storage conditions on the microbial quality and lipid oxidation in ground beef products. The second study aimed to measure energy usage in response to Artificial Intelligence (AI) temperature management in cold storage facilities. As global meat supply chains face increased pressure to reduce environmental impacts, cold storage facilities are among the largest energy consumers, highlighting their pivotal role in promoting sustainability initiatives. Sustainability initiatives outlined by private sector companies align with the targets established by the United Nations Net Zero Coalition, aiming to achieve carbon neutrality by 2050. Therefore, the effective management within cold storage facilities, in both refrigeration and frozen conditions, is essential for optimizing performance in the evolving global market.

Given the energy-intensive nature of frozen storage in meat supply chains, innovation is essential to meet sustainability goals without compromising food safety or product quality. The first experiment conducted aimed to evaluate the effects of highly controlled frozen storage temperatures (-20.6°C, -15.0°C, and -9.4°C) on the microbial viability and extent of lipid oxidation in vacuum-packaged ground beef samples held over a 30 d period. Ground beef was inoculated with a mixture of six common meat spoilage organisms to achieve a known

concentration level of 4 log CFU/g. Microbial quality was evaluated using aerobic plate counts (APC), recorded as log CFU/g, while lipid oxidation was quantified using the thiobarbituric acid reactive substances assay (TBARS), and recorded as mg MDA/kg.

Results of the study indicated no statistically significant differences ($P \geq 0.05$) in meat spoilage indicators across the different temperature treatments. The key finding was that ground beef stored at -9.4°C, under precise temperature control (±0.01°C) systems, does not negatively impact meat quality over the 30 d frozen storage period.

The second case study examined energy savings in industrial cold storage facilities when managing temperature variations with AI-controlled systems. For this study, we included two facility locations that were similar in size, ambient temperatures, and operational procedures: one in Colorado and the other in Illinois. The energy utilization in (kW) was recorded from both facilities, before and after implementation of energy-AI software (Crossnokaye's ATLAS) over a 94 d period. We analyzed the data using a two-way fixed effects ANOVA model. Results indicated a 32% reduction in overall power consumptions when using AI ($P < 0.0001$). The reduction of power usage over the 94 d period equated to an estimated cost saving of approximately $42,258 (USD) per facility. The reductions in energy could result in an overall decrease of Scope-II greenhouse gas emissions, thereby achieving a step towards carbon neutrality.

Findings from these studies are relevant to discussions regarding adjustments to the frozen storage temperature setpoints in facilities. Raising temperature setpoints have the potential to reduce energy consumption, lower operational costs, and lower greenhouse gas emissions, all while maintaining the safety and quality of temperature-sensitive meat products. When facilities implement AI temperature management systems, they enable real-time adjustments to dynamic

conditions. This management strategy provides a scalable pathway for the enhancement of operational efficiency and environmental sustainability within meat cold storage supply chains.

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REVIEW OF LITERATURE

1.0 Introduction

Historically, the global food industry has depended on cold storage systems and freezing technologies, as they are a vital part of the maintenance of food safety and quality (Coombs et al., 2017; Nastasijević et al., 2017; Zhao et al., 2021). The growing reliance on freezing technology is in result of the push towards food waste reduction, increased shelf life, and expanding product accessibility (Gummalla et al., 2024; Ramanathan et al., 2022; Zhang et al., 2023a). The increased reliance on freezing technology is essential for promoting environmental and socio-economical sustainability throughout the food system. With amplified challenges in the meat industry related to energy consumption, cost, as well as initiatives to reduce emissions, customers urgently demand more sustainable products and solutions to feed an estimated 9.1 billion individuals by the year 2050 (Asem-Hiablie et al., 2019; Dong and Miller, 2021; FAO, 2009; Fu et al., 2024a; Li and Kallas, 2021). The 2009 report published by the Food and Agriculture Organization (FAO), they predicted that to meet the growing population's expected demand in 2050, food production must increase by 70% (FAO, 2009).

In the contemporary context, the global meat industry is experiencing strong growth, with forecasts from the FAO indicating that global meat production will reach 374 million tonnes by 2024 (FAO, 2024). Of this total, the global beef sector accounts for 78 million tonnes, which accounts for approximately 21% of all meat production (FAO, 2024). Key players in beef production are Brazil, Australia, China, India, Argentina, European Union, and the United States. Each of these countries plays a role towards the enhancement of the global beef industry supply. However, production and environmental efficiencies vary across countries.

Despite cyclical variations in live cattle production data and changing consumer protein preferences, such as increasing market prices prompting a shift towards alternative protein sources, overall beef production has demonstrated remarkable resilience over time (Godfray et al., 2018). Beef production is currently functioning well, despite the historically low herd size (Knight, 2019; Shahbandeh, 2024). The ongoing decline in the live cattle population in the United States, driven by drought conditions, highlights the importance of utilizing cold storage technologies (Knight, 2019). This is vital for innovation, as these technologies are among the key solutions to minimize waste and extend product shelf life. As consumer preferences evolve, the rising quality standards for frozen products further highlight the importance of necessary advancements in cold chain technology to preserve nutritional integrity and reduce waste (Research, 2024).

Frozen storage techniques have influenced modern food preservation, especially in the meat sector. Reduced storage temperatures below refrigeration (4°C) help extend a product's shelf life and maintain quality at a level acceptable to consumers (Coombs et al., 2017; Dobbins et al., 2024; Kitinoja, 2013; Leygonie et al., 2012; Wang et al., 2020). This literature review provides a comprehensive overview of the evolution of cold storage supply chains; the historical and contemporary applications of freezing technologies; the impact of freezing on lipid oxidation and microbiological spoilage affecting meat quality; the sustainability challenges associated with cold storage supply chains; and future considerations.

1.1 Cold Storage Systems

The introduction and subsequent utilization of cold storage supply chains have meaningfully transformed the global trade landscape (Nastasijević et al., 2017). By enabling the long-distance transport of perishable goods, such as meat, while maintaining quality and safety standards (Mustafa et al., 2024a; Nychas et al., 2008). In the past, consumers' access to diverse foods was often limited to local markets due to environmental factors, with short time frames for mobilizing products before they became unsuitable for consumption. Making it almost impossible to trade temperature-sensitive items. Historically, individuals located in Arctic climates were, initially, the only ones able to participate in this frozen food preservation practice (Lawrie, 1979).

The advent of mechanical refrigeration in the late 19th century revolutionized food transportation and logistics by enabling packers to store and move goods over extended periods without spoilage (Goodwin et al., 2002). Clarence Birdseye, a former fur trader and pioneer in freezing technology, documented the first account of flash-freezing methodology (Archer, 2004). This innovation greatly improved the texture and shelf life of frozen foods, establishing the foundation for commercial food freezing processes that are widely used today (Archer, 2004).

By the early 20th century, refrigeration technology was integrated into rail, sea, and road transportation, enabling the distribution of fresh and frozen goods to global markets, greatly expanding market access and reducing food waste (James and James, 2014). Additionally, the development of container cold storage shipping by sea throughout the mid 20th century further bolstered international trade by facilitating the transportation of these products across continents (Heap, 2006; James and James, 2014, 2010).

However, in the 1940s, following World War II, the frozen industry experienced a notable decline in consumer acceptance due to issues such as poor color, off-flavor, inedible products, and mold growth. This reduced acceptability led to an 87% reduction in frozen food production (Ginsberg, 2002). In response to this rapid decline, the United States Department of Agriculture (USDA) established the USDA Western Regional Research Center (WRRC) under the leadership of Helmut C. Diehl (Burr and Elliott, 1960). During this period, Diehl set the standards for the time-temperature tolerance of frozen foods, which are critical for ensuring food safety and quality (McHugh, 2015). Diehl advised that frozen temperatures of -18°C be maintained for maximum shelf life; otherwise, the products were more likely to rapidly develop off flavors and discoloration (Diehl and Warner, 1945).

Cold storage supply chains currently serve as critical infrastructure for supporting international trade of temperature-sensitive products, including seafood, dairy, pharmaceuticals, produce, and notably, meat, with the aim of maintaining high product quality during transit (Neusel and Hirzel, 2022; Zanoni and Zavanella, 2012). The supply chain for beef products is extensive, connecting live animals to consumers (Figure 1.1). It starts with transportation of animals from live animal facilities (i.e., farms and feedlots) to abattoirs, where animals are processed from whole carcass into primals, sub-primals, and retail cuts. These products can be sent straight from processing plants to retail and quick-service locations. However, a more common practice is to store products at cold storage facilities, which act as distribution centers for food service, retail, and international customers before they ultimately reach the end consumer. (Nastasijević et al., 2017).

Cold storage technology has enabled year round access for products that were historically considered seasonal only. This advancement allowed the meat industry to satisfy consumer demand regardless of geographical or seasonal limitations. Moreover, it has greatly bolstered food security by providing a consistent supply of essential goods, especially in newly urbanized regions. The FAO predicts that by 2050, approximately 70% of the global population will reside in urban areas (FAO, 2009). With the ongoing rise in urbanization worldwide, the reliance on supply chains is expected to grow. Consequently, the ability to transport frozen and refrigerated goods, both domestically and internationally, is likely to drive economic growth in regions reliant on agricultural outsourcing. Countries with robust cold storage systems can more efficiently participate in international trade, thereby boosting their competitiveness in global markets and strengthening their food security (James and James, 2010).

As global food trade continues to increase, innovations in cold storage logistics become even more essential for the sustainable distribution of perishable goods across international markets. Utilizing advanced technologies such as AI and the Internet of Things (IoT) is capable of facility temperature monitoring, product inventory management, and improved energy efficiency in refrigeration systems, all of which greatly enhance the overall efficiency and sustainability of cold storage supply chains (Mustafa et al., 2024b; Nastasijević et al., 2017). Advancements in technology, including machine learning (ML), the Internet of Things (IoT), and artificial intelligence (AI), facilitate real-time monitoring and optimization of facility temperatures based on energy demand and electricity prices; consequently, reducing temperature oscillation is now more attainable than ever (Fu et al., 2024a; Kamble et al., 2019; Salgado et al., 2024). Utilization of more precisely managed systems that employ real-time data not only diminishes temperature oscillations but also stimulates discussion on expanded managerial practices, such as increasing frozen storage temperatures. By elevating frozen storage temperatures, it is possible to mitigate the environmental impact associated with the frozen cold chain and reduce the consumption of non-renewable energy (Marchi and Zanoni, 2022). This change ultimately improves efficiency and lowers costs. Utilizing advanced energy AI technologies in conjunction with real-time facility management can substantially reduce power consumption, benefiting the supply chain and lowering greenhouse gas (GHG) emissions (Amath et al., 2021; Dong and Miller, 2021). These initiatives are consistent with the objectives of the meat industry and address consumer expectations for enhanced sustainability throughout the food supply chain (Grunert et al., 2014).

The United States Environmental Protection Agency (EPA) have set guidelines to classify the type of emissions produced into three categories: Scope-I, Scope-II, and Scope-III. Scope-I is classified as the greenhouse gas (GHG) emissions produced from sources owned or operated by an organization. Scope-II emissions are categorized as emissions produced through operations on purchased energy, including electricity, steam, heat, and cooling (US EPA, 2020). Finally, Scope-III emissions are defined as the emissions produced from owned and non-owned assets along the value chain (US EPA, 2016). Such innovations could improve cost-efficiency within the cold supply chain, mitigate GHG emissions, and reduce the overall carbon footprint, thereby fostering significant reductions in Scope-II environmental impacts associated with meat distribution (Dong and Miller, 2021; Garnett, 2011; Nozari et al., 2025; US EPA, 2020).

As cold storage facilities and meat processors work to attain sustainability objectives, such as achieving net-zero emissions by 2050, it is feasible to maintain consistent control over frozen storage temperatures while concurrently reducing energy consumption, provided that food safety and quality are not compromised (United Nations, 2023). These developments aim to extend the shelf life of perishable items while minimizing both direct and indirect environmental impacts, reinforcing the importance of cold storage in global food distribution (Diaz Sanchez et al., 2021).

1.2 Freezing Technology

A fundamental factor facilitating the production and global distribution of beef is the use of frozen storage technologies. Freezing as a method of preservation necessitates considering both the benefits and consequences to the overall product. While freezing does extend product shelf life and minimize microbial growth, a downside of this technology is increased drip loss (e.g., purge) during thawing. Approximately 75% of meat consists of water, which freezes at -5°C (Dave and

Ghaly, 2011). Therefore, ice crystal formation is important, particularly as it relates to quality attributes of concern in beef.

1.2.1 Ice Crystallization

Ice crystals play a critical role in determining the texture, stability, and longevity of frozen products. The crystallization process occurs when water molecules, within an enclosed system, undergo a transition from a liquid to a solid state after reaching the energy threshold required for ice formation (Banerjee and Maheswarappa, 2019; Campañone et al., 2006; Kitinoja, 2013; Leygonie et al., 2012). Ice crystallization consists of four essential phases: supercooling, nucleation, growth, and recrystallization. During the supercooling phase, the temperature of a liquid in a confined system falls below the freezing point of water, typically 0°C; however, for meat products, the freezing point is generally regarded to be between -4°C and -10°C (James and James, 2024). The nucleation phase involves surpassing the energy barrier necessary for crystal formation to establish the initial stable nucleus that all other crystals will form off of (Jia et al., 2022). After nucleation, the crystallization growth process progresses as more water molecules bond with the existing crystal infrastructure (Jia et al., 2022). In the recrystallization stage, the crystal attains its final form and dimensions, subject to the influence of external environmental factors (Banerjee and Maheswarappa, 2019).

Size and distribution of ice crystals in muscle tissues are influenced by product freezing rate, nucleation temperature, and the maintenance of storage conditions. Slower freezing rates and temperature instability favor the formation of larger ice crystals that can harm intracellular structures, exposing nutrient sources to previously dormant microbial populations that can begin to metabolize energy during thawing (Kitinoja, 2013; Leygonie et al., 2012; Vieira et al., 2009).

In contrast, quick freezing and reduced freezing rates are more effective in decreasing the size of the ice crystal, protecting the integrity of intracellular structures while keeping the product's overall size, shape, and quality intact, leading to fewer ultrastructural muscle cellular defects (Campañone et al., 2006; Hanenian and Mittal, 2004; Reid, 1997).

1.2.2 The -18 Degrees Celsius Level

Research in the mid 1900s investigated how the freezing rate, duration, and temperature consistency during frozen storage influenced food quality. Experts widely concur that maintaining a stable freezing temperature with minimal oscillations throughout the duration under cold conditions is essential for optimal product quality and prolonged shelf life (Anese et al., 2012; Kropf, 1971; Reid, 1997). However, there is controversy over the optimal frozen storage temperature for meat products.

A major breakthrough in the frozen meat industry was the initial designation of -18°C as a ‘standard freezing temperature for meat products’, set by the Association of Food and Drug Officials of the United States in 1959 (“Code of Federal Regulations: Frozen Food Industry, 16 C.F.R.,” 1959; Dave and Ghaly, 2011; McHugh, 2015; Zhao et al., 2021). Studies have shown that at -18°C, microbial activity decreases, allowing food to maintain nutritional value and structural integrity (Burr and Elliott, 1960; Vieira et al., 2009).

Subsequent studies further explored microbial quality patterns on meat while under various frozen storage conditions. A study by Adam et al. (2010) indicated that frozen storage temperatures below -5°C effectively prevent microbial spoilage in meat products by impeding microbial growth (Adam et al., 2010; Coombs et al., 2017). Additionally, a study by Winger (1984) showed that lamb products stored at -10°C or lower under frozen conditions exhibited no adverse flavor development when held for at least 2 years.

However, the proper temperature for frozen meat is said to be at -40°C, as this temperature fully inactivates bound water and thus substantially restricts microbial growth(Estévez, 2011; Evans, 2008; Zhang et al., 2023a). Nonetheless, the adoption of -18°C remains the industry standard set point for ensuring the safety and quality of muscle food products throughout the global food supply chain, as temperatures below -18°C provide no additional benefits (Coombs et al., 2017). Recently, there has been a movement within meat logistics companies to increase the static frozen storage temperature set point of facilities by 3°C to reduce energy consumption, in line with sustainable practices for the food supply chain (Allouche et al., 2023).

Understanding cold storage supply chain infrastructure, the ice crystallization process, and the significance of the -18°C temperature standard is essential to optimize frozen storage efficiency and reduce food degradation. Given the developing global demand for frozen meat products, advancements in frozen storage and transportation technologies that encourage sustainable practices will be crucial for maintaining quality and meeting consumer standards throughout the supply chain.

1.2.3 Freezing Methods

Methods for freezing foods are diverse and include conventional technologies such as blast freezing and plate/contact freezing, as well as innovative methods like cryogenic freezing with immersion/spray methods (Graham, 1996). Contact freezing, also known as plate freezing, involves pressing products directly against a metal freezing plate to transfer heat through conduction (Becker and Fricke, 2003). Conduction freezing occurs when heat is transferred directly through a substance without additional movement of the product. Contact freezing is highly effective, enabling faster cooling of the product than traditional blast freezing. This method is most commonly used in the seafood, mechanically separated beef, and lean finely textured beef sectors; however, there are opportunities to expand its use with other types of protein (Vijayakumaran et al., 2019). A major disadvantage of this technique is that widespread industry implementation requires more capital than existing air blast freezing systems (Graham, 1996).

More intensive freezing technologies, such as cryogenic freezing, are expected to enhance product quality. Cryogenic freezing encompasses products passing through refrigerants, such as liquid nitrogen or carbon dioxide, that aid in absorption of heat from the surface of meat products by sublimation (Ikram et al., 2025a). This processing method forms an outer crust layer on the product that safeguards and continues to cool the internal temperature at a rate that preserves internal muscular ultra structure (Ikram et al., 2025a). Cryogenic freezing is most commonly used for individually quick frozen (IQF) products, which are increasingly popular among ready to prepare foods, the food service sector, and cook from frozen products (Joy and Vanapalli, 2023). Of these technologies, blast freezers are the most widely utilized among commercial freezing operations.

Blast freezing is a straightforward method that utilizes a fan within the unit to circulate air around the refrigerated coil inside an insulated space, promoting effective cooling (Becker and Fricke, 2003). A notable disadvantage of this methodology is the inability to transfer heat from the product surface to the surrounding environment, which could result in the product exhibiting a darker appearance when compared to alternative freezing systems (Kropf, 1971). The most common blast freezers within the commercial industry setting are the use of vapor compression refrigeration systems.

1.2.4 Vapor Compression Refrigeration System Overview

The Vapor-Compression Refrigeration (VCR) system commands a dominant position within the cold supply chain, accounting for 80% of market share across industrial, commercial, domestic, and refrigerated transportation sectors (Fu et al., 2024b). Use of VCR systems alone account for 15% of the world's electricity consumption and contribute to 10% of global greenhouse gas emissions (Fu et al., 2024b).

Performance of the system is primarily evaluated based on its coefficient of performance (COP), which measures the efficiency of the refrigeration system. The COP is defined as the ratio of the amount of heat expelled from the refrigerator (Q_c) to the work input into the system (W_{in}), which together determine the overall cooling efficiency ($K_{cooling}$) (Donev et al., 2024; Redko et al., 2020; Zajac, 2016).

$$K_{cooling} = \frac{Q_c}{W_{in}}$$

Higher COP values imply more energy-efficient systems.

Commercial VCR systems entail primary components: evaporators, compressors, expansion valves, and condensers (Fenton et al., 2019; Rodriguez and Rasmussen, 2017) (Figure 1.2). These components ultimately consume electrical energy to produce low-grade thermal energy, which, depending on the gradient, results in greater COP values (Fu et al., 2024b).

The system begins with a refrigerant, either synthetic or natural, which progresses through the compressor. This process involves applying mechanical energy (electricity) to increase the refrigerant’s circulating pressure and temperature within the system, which subsequently flows to the condenser (Fenton et al., 2019). At the condenser, cooled air flows over the tubes carrying the hot refrigerant, facilitating heat transfer (Redko et al., 2020). This process results in heat being transferred from the refrigerant to the surrounding air as the refrigerant cools and transitions from a gas to a liquid (Coker, 2015; Redko et al., 2020). The heat absorbed by the refrigerant is expelled into the outside environment, and the lukewarm, high-pressure liquid refrigerant proceeds toward the expansion valve (Berk, 2018).

At the expansion valve, restricted airflow causes the warm liquid to turn into a cold, low-pressure fluid that flows toward the evaporators ultimately controlling the rate at which the system operates (Fenton et al., 2019). At the evaporators, warm air passes over tubing filled with cold, low-pressure refrigerant, facilitating heat exchange that causes the refrigerant to change phase, with some turning back into vapor (Redko et al., 2020). After vaporization and movement through the system, the refrigerant cools the space (Berk, 2018). When the vapor and liquid return to their initial states at the expansion valve prior to entering the evaporator, the low pressure directs them back to the compressor, continuing the cycle (Berk, 2018; Coker, 2015).

1.3 Meat Quality and Defects

While meat preservation by use of freezing technologies is not entirely preventative of chemical and microbial spoilage, it is known to effectively slow down spoilage decay (Dave and Ghaly, 2011). Rejection of meat products occurs among consumers with items that have experienced chemical or microbial spoilage, impacting all three pillars of sustainability: economic, social, and environmental. When products are deemed unacceptable by the customer, they often end up as waste, resulting in lost revenue from sales at the retailer, decreased willingness to purchase the product again by the consumer, and wasted natural and secondary energy resources (Ramanathan et al., 2022).

1.3.1 Chemical Spoilage

Numerous studies have demonstrated that freezing meat adversely affects the overall quality of the product, as quality gradually deteriorates during time spent in storage conditions (Raharjo and Sofos, 1993; Soyer et al., 2010; Vieira et al., 2009). One way product quality declines is through lipid oxidation. Lipid oxidation can occur in any food system containing lipids, with phospholipids in cellular membranes of muscles being particularly vulnerable due to their structural characteristics and higher degrees of unsaturation (Ahn et al., 2009). Among various food systems, meat is among the most susceptible to the detrimental impact of lipid oxidation. When oxidation occurs, it results in product discoloration, the formation of off flavors, degradation of nutrient values, and the formation of potentially harmful compounds (Calligaris et al., 2016; Durand et al., 2025).

1.3.2 Lipid Oxidation

Lipid peroxidation is a key factor in the decline of quality attributes that consumers care about, such as discoloration, texture, and flavor. Lipid peroxidation, an integral process within oxidative stress, progresses through three primary phases: initiation, propagation, and termination. (Aguilar Diaz De Leon and Borges, 2020; Fernández et al., 1997; Min and Anh, 2005). The initiation phase removes the unstable hydrogen ion (H·) from the unsaturated lipid molecule, forming a lipid alkyl radical (L·) (Min and Anh, 2005). Once initiated, the oxidation process will proceed into the propagation phase without interruption when unoxidized lipid substrate is available. If in the presence of oxygen, this alkyl radical will react with molecular oxygen available to form a lipid peroxyl radical (LOO·) (Dave and Ghaly, 2011). Following the binding of molecular oxygen, the lipid peroxyl radical will remove another unstable hydrogen ion (H·) from a different fatty acid chain, resulting in the formation of a new lipid hydroperoxide (LOOH) (Domínguez et al., 2019). However, because lipid hydroperoxides are chemically unstable, this primary product continues in the oxidation cascade until all available substrate has been depleted, leading to the stabilization phase known as termination (Ahn et al., 2009). During the termination phase, as lipid peroxyl radicals exhaust the available unsaturated fatty acids, they start interacting with each other, leading to the formation of secondary non-radical byproducts such as alcohols, ketones, aldehydes, and hydrocarbons (Fernández et al., 1997; Min and Anh, 2005).

Lipid oxidation can be quantitatively assessed through monitoring; oxygen uptake, the depletion of polyunsaturated fatty acids, and the formation of hydroperoxides, as well as the formation of secondary compounds, which involve the development of carbonyls, aldehydes, hydrocarbons, and fluorescent byproducts (Gray and Monahan, 1992). Notably, malondialdehyde is recognized from the aldehyde group as a secondary byproduct commonly associated with the Thiobarbituric Reactive Acid Substances assay (TBARS) (Ahn et al., 2009; Fernández et al., 1997; Gray and Monahan, 1992; Papastergiadis et al., 2012). Furthermore, the rate of oxidation is influenced by various factors, including the product type, processing methods, and storage conditions (Ladikos and Lougovois, 1990).

1.3.3 TBARS

Variations of the TBARS assay are widely recognized as among the most established and reliable methods for assessing the extent of lipid oxidation in meat products (Gray, 1978; Leygonie et al., 2012; Raharjo and Sofos, 1993). However, a limitation of the TBARS assay stems from the method's inherent analytical characteristics. The use of thiobarbituric acid has the potential to interact with reactive substances other than malondialdehyde, which may reduce the assay's level of sensitivity to malondialdehyde. Despite this limitation, the TBARS assay remains widely used due to extensive research supporting reactive substances similar to malondialdehyde on the spectrophotometry standards, as well as the quick processing speeds needed for large sample volumes (Fernández et al., 1997; Raharjo and Sofos, 1993).

Although less prevalent, malondialdehyde can be effectively quantified by the interaction with 2-thiobarbituric acid. Thus, resulting in the formation of a distinct reddish-pink colored complex (Barriuso et al., 2013; Patton and Kurtz, 1951; Raharjo and Sofos, 1993). This unique reaction enables the precise measurement of the extent of lipid peroxidation that has occurred during meat processing and storage. Following this reaction, the absorbance of the resulting complex is measured using spectrophotometry by detecting light absorption at the peak wavelength of 532 nm (Aguilar Diaz De Leon and Borges, 2020; Gray and Monahan, 1992). Variations of this method have been corroborated by numerous studies over the years, further establishing the reliability and efficacy of this analysis in detecting the extent of lipid oxidation within the context of meat science.

1.3.4 Microbial Spoilage

Microbial spoilage is the process by which microorganisms such as bacteria, fungi, and yeasts multiply to a degree at which their metabolic activities degrade the product's quality. Product quality degradation often occurs alongside chemical spoilage. Byproducts of microbial growth include off odors, discoloration, and slimy textures, which ultimately reduce consumer acceptance. These byproducts can also react chemically with the host. Such spoilage can proliferate under conditions during processing and storage that favor growth, including temperature, humidity, uncleanliness, and oxygen availability (De Filippis et al., 2013; Lawrie, 1979; Zhu et al., 2024).

Meat behaves as an ecological niche, providing a rich source of nutrients that promotes the growth of bacterial organisms. Meat is composed of approximately 75% water, and contains various metabolites, including amino acids, nucleotides, peptides, and sugars (Labadie, 1999). However, several factors can influence the type and extent of meat microbial spoilage. The animal's physiology prior to harvest will influence the initial levels of microbial contamination in beef products (Nychas et al., 2008). Preharvest factors include the animal's conditioning, age, and sex prior to harvesting, as well as operational procedures, including cleanliness during procedures and of the facility at harvest, evisceration, fabrication, and the temperature of the processing environment during storage and distribution (Dave and Ghaly, 2011; Gill, 1996).

External factors can delay the onset and growth of microbial spoilage in meat. External factors include storage temperature and the product packaging system (Huis in't Veld, 1996). Among these external factors, storage temperature stands out as the most critical factor for meat product quality (Koutsoumanis et al., 2006). The temperatures at which products are stored in impact the type of organisms that survive and grow; under freezing conditions, organisms enter dormancy with no growth (Coombs et al., 2017; Fernandes, 2009; Zhang et al., 2023b). Organisms that survive will return to an active state during thawing once returned to refrigeration conditions of 0°C to 7°C. When freezing, bacteria are vulnerable to sustaining cellular damage due to the formation of ice crystals in both the extracellular and intracellular spaces (Archer, 2004). Once thawed, dormant species have increased access to bioavailable nutrients, leading to rapid growth. Aerobically packaged products are most vulnerable to microbial spoilage due to the presence and growth of the dominant *Pseudomonas* spp., especially when compared to lactic acid bacteria, which flourish in low-oxygen environments similar to those created in modified atmospheric packaging (MAP) and vacuum packaging (Dave and Ghaly, 2011). Microbial spoilage caused by *Pseudomonas* spp. is considerably reduced under anaerobic conditions (Gill, 1996; Hernández-Macedo et al., 2011). Among these packaging types, vacuum packaging is the most effective at limiting microbial growth, though ongoing research is examining how active packaging can further prevent microbial spoilage (Cutter, 2006; Hernández-Macedo et al., 2011).

In addition to storage temperature, initial microbial loads on products can substantially affect shelf life (Gill, 1996). Although it may vary, the consumer acceptability threshold for microbial spoilage is approximately 7 to 8 log/CFU/g or log/CFU/cm^2 of total organisms, with the development of slime and off-odor formation generally occurring at these levels on meat products (Ayres, 1960; Nychas and Skandamis, 2005). Although it is believed that as many as 2,000 bacterial species can contribute to meat spoilage, the two spoilage bacterial groups that will be further outlined for their impacts on meat quality are *Pseudomonas* spp. and lactic acid bacteria.

1.3.5 Pseudomonas

*S*pecies belonging to the genus *Pseudomonas* are classified as gram-negative, non-spore-forming, rod-shaped microorganisms (Kwaasi, 2003). *Pseudomonas* can leverage their psychrotrophic properties to enable them to be among the most common specific spoilage organisms (SSO) associated with aerobically packaged meat products (Gill, 1996; Hernández-Macedo et al., 2011; Koutsoumanis et al., 2006). Within the diverse *Pseudomonas* genus, the most prominent species associated with spoilage capabilities include *Ps. fragi*, *Ps. fluorescens*, and *Ps. lundensis* (De Filippis et al., 2013; Ellis and Goodacre, 2001; Nychas et al., 2008;

Stanborough et al., 2018). The biological and ecological characteristics of these species enable them to thrive in oxygen-rich environments, facilitated by aerobic metabolism pathways, which allow them to dominate the spoilage process in air-packaged meat.

Pseudomonas spp. are especially prolific in fresh beef products, as they are able to enumerate rapidly under favorable conditions of: temperature, humidity, and atmospheric packaging (Ayres, 1960; Gram et al., 2002; Stanborough et al., 2018). The ideal temperature for *Pseudomonas* spp. favors microbial growth in meat and meat products with the temporal conditions of 1°C to 25°C (Nychas et al., 2008). However, they are also recognized to contribute to the spoilage of frozen beef (Koutsoumanis et al., 2006; Labadie, 1999). Furthermore, it is well documented in the literature that *Pseudomonas* spp. exhibit proteolytic activity that allows for penetration into the meat, thus enabling access to niche areas that harbor abundant nutrients, which can expedite the rate of proliferation (Gill and Penney, 1977).

Regarding discoloration in meat products, the interaction of fluorescence-producing species of *Pseudomonas* and myoglobin results in the appearance of a distinct green pigment known as pyoverdine. Pyoverdine is a side ionophore that binds with ferric iron, aiding in the continuation of *Pseudomonas* degradation in the system (Labadie, 1999). The green discoloration not only affects the visual appearance of spoiled meat but also indicates microbial activity (Smith et al., 2024). The capability of *Pseudomonas* spp. to produce these pigments further exemplifies their role in meat spoilage and emphasizes the need for control measures to manage and extend the overall shelf life (Sun and Holley, 2012). Understanding the mechanisms behind spoilage activity driven by *Pseudomonas* spp. will aid in developing effective preservation strategies to prolong the product’s shelf life.

The growth rate of *Pseudomonas* spp. is notably reduced in anaerobic environments, where conditions promote the growth of lactic acid bacteria (Gill, 1996; Gram et al., 2002; Hernández-Macedo et al., 2011). This underscores the importance of packaging conditions in influencing the management strategies for spoilage organisms in meat.

1.3.6 Lactic Acid Bacteria

Lactic acid bacteria (LAB) are recognized as contributors to the microbial spoilage of meat products. LAB are gram-positive, rod-shaped, and cocci-shaped, facultative anaerobic bacteria that ferment glucose under anaerobic conditions, that produce lactic acid as a byproduct of their metabolism (Mozzi, 2016). LAB are highly prevalent in processing facility settings; however, the transfer of these bacteria to the product is relatively low compared to the microbial counts within the package (Stellato et al., 2016). Under refrigerated aerobic conditions, these bacteria are less dominant than *Pseudomonas* spp., as they are slower growing (Walker, 2003).

The role of LAB in meat spoilage is especially relevant under vacuum packed conditions, as in this environment, LAB will outcompete their aerobic counterparts, *Pseudomonas* spp. (Gill, 1996; Hernández-Macedo et al., 2011). These bacteria are responsible for the development of off-odor compounds, which are byproducts of lactic acid produced during the metabolic process, and are associated with the perceived “souring” of spoiled meat, including fruity, sulfurous, and pungent smells (Pothakos et al., 2015). Lactic acid bacteria are commonly associated with cured and processed meat products, as the acidity in these environments acts as a barrier against the regrowth of undesirable initial microflora, thereby creating optimal conditions for the continued growth of LAB (Borch et al., 1996).

The most common LAB genera associated with meat products include *Lactobacillus*, *Leuconostoc*, and *Carnobacterium*. These genera are often linked to the spoilage of meat under refrigerated vacuum packaged conditions (Gill, 1996). LAB are considered a means to effectively prevent the growth of alternative bacterial populations and therefore increase the shelf life of products {Citation}. When LAB were intentionally applied to the product, while reducing alternative bacterial growth, the negative outcome of accumulated hydrogen sulfide production amongst the non-essential amino acid, cysteine, caused the development of unpleasant odors and transformation of color from ferrous iron (Fe^{2+}) to the ferric state (Fe^{3+}) (Signorini et al., 2006).

In conclusion, given the gaps in the current literature regarding frozen storage conditions and the impacts of advanced technologies in cold storage facilities, this work aimed to understand the impact of experimental temperatures and how they affect microbial quality and lipid oxidation in beef products. Additionally, it sought to explore proactive measures through the implementation of artificial intelligence systems that monitor and manage temperature to reduce facility energy use and costs.

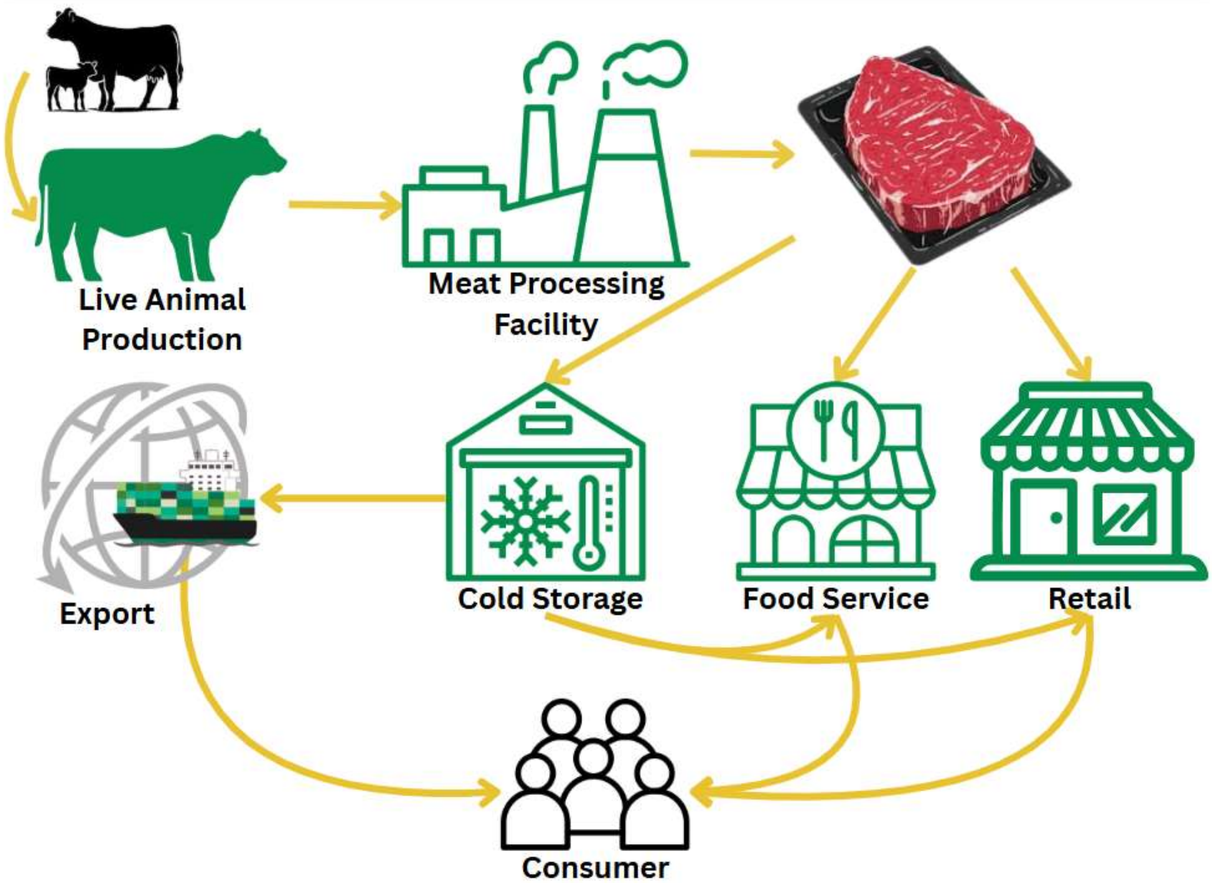

Figure 1.1. Schematic representation of the beef cattle supply chain.

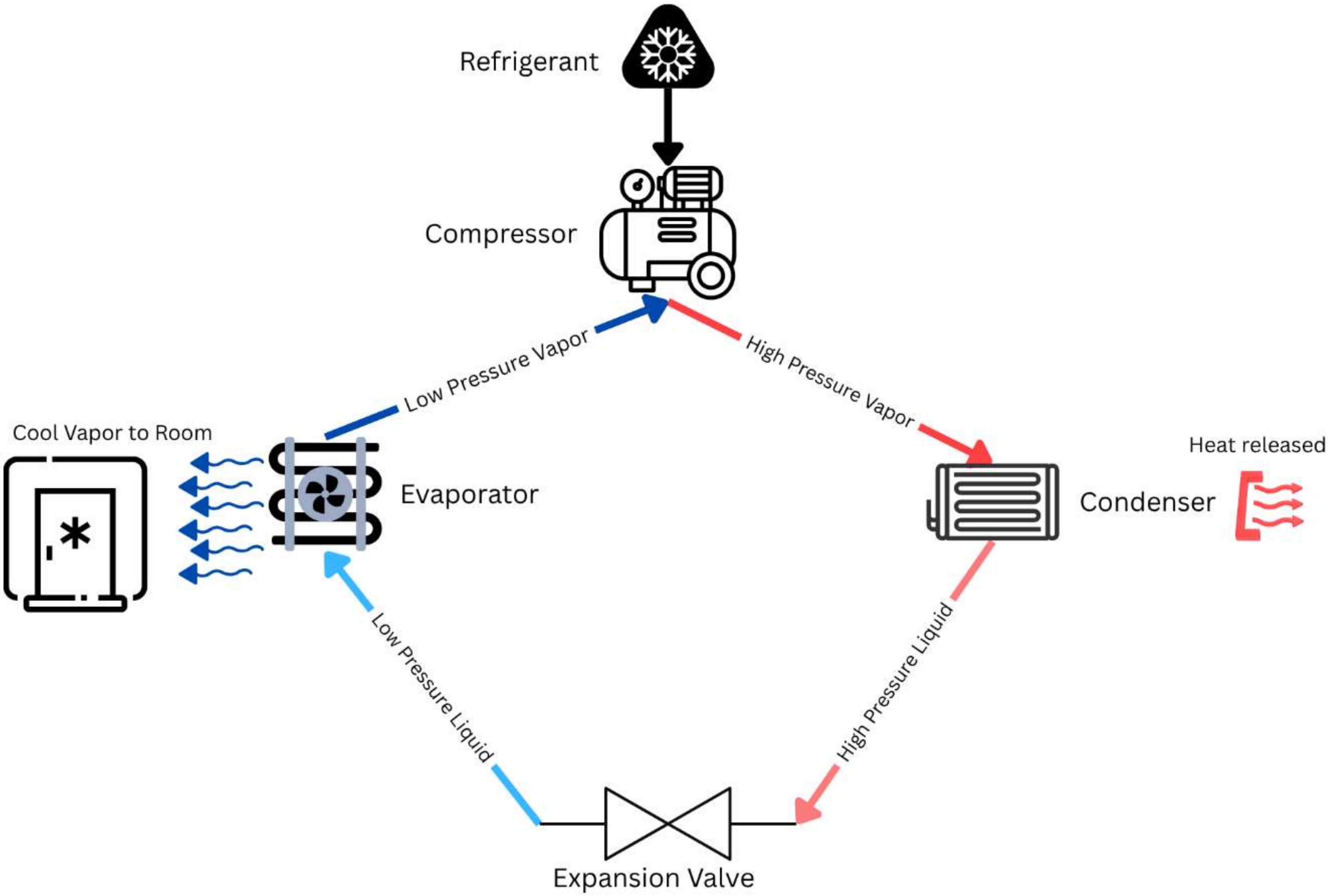

Figure 1.2 – Schematic representation of the vapor-compression refrigeration system

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CHAPTER 2: EFFECTS OF FROZEN STORAGE TEMPERATURE ON THE CHEMICAL AND MICROBIAL SPOILAGE OF GROUND BEEF

2.1 Introduction

Amid rising energy costs, increased consumer awareness of livestock production practices, and the global shift toward climate-smart solutions endorsed by the United Nations Net Zero Coalition with goals of net zero emissions by 2050, (Rotz et al., 2019; United Nations, 2023). This study investigated the effect of three frozen storage temperatures on the survival and growth of microbial populations as well as the lipid oxidation that occurred in ground beef. Frozen storage is an integral component of the meat supply chain. The frozen supply chain is essential infrastructure, supporting product inventory management, international trade, facilitation of domestic product management according to seasonal demand, and overall reduction of food waste for perishable meat products (Neusel and Hirzel, 2022).

The utilization of frozen storage along the meat supply chain is frequently characterized by high energy consumption. This is primarily due to the operational management of the refrigeration systems, which maintain low temperatures at levels typically below freezing (0°C) (Salgado et al., 2024). Maintaining low temperatures during storage is crucial for preserving overall product quality and shelf life, although the specific temperature ranges necessary to achieve these traits are subject to debate in the literature. Winger (1984) proposed that temperatures less than or equal to -10°C are adequate for maintaining product quality standards. Holman et al. (2017, 2018, 2025) determined that continuous cold environments at -12°C and -18°C effectively restricted microbial and chemical spoilage activities related to the degradation of beef products.

This conclusion, stating that microbial activity was halted at -12°C and below, was corroborated by James et al. (2025).

The maintenance of frozen storage systems impacts overall global energy use due to the dependance on non-renewable energy sources (Zanoni and Zavanella, 2012). Electricity is the most widely used energy source for frozen storage operations. Electricity is classified as a secondary energy source as it is generated as a byproduct of primary sources (e.g., coal, natural gas, nuclear energy, solar energy, and wind energy (U.S. EIA, 2024). The International Institute of Refrigeration estimates that for domestic cold storage alone, refrigerators and freezers consume approximately 4% of the world's annual electrical energy consumption (Dupont et al., 2019).

While the meat cold chain industry works to reduce overall energy use, as part of broader Scope-II and III initiatives to meet net zero claims. By means to reduce energy usage cold storage facilities push to increase the static standardized frozen storage temperature by 3°C to lower energy costs in support of cost-saving efforts (Allouche et al., 2023). Nevertheless, it is imperative to ensure that such temperature modifications continue to prevent microbial and chemical spoilage, thereby maintain the product quality at a level acceptable by consumers. Through the enhancement of precision in temperature control during frozen storage durations, this further supports societal sustainability objectives while safeguarding meat safety and quality.

In 2022, the international frozen meat market was valued at $23.2 billion (USD) (Transparency Market Research, 2024), underscoring the economic impact and consequential need for further research to quantify the effect of freezing and frozen storage direct impacts on the sector's financial dynamics. Despite the scale of the frozen meat demand there is limited research available assessing minor incremental changes in temperature and the influence of spoilage dynamics within commercial conditions.

This study aimed to evaluate the impact of various frozen storage temperatures on the microbiological quality and lipid oxidation for ground beef when stored for a period of 30 d, employing it as a model to represent commercially frozen ground beef when maintained under highly controlled storage temperature conditions.

2.2 Materials and Methods

2.2.1 Meat Collection, Processing, and Inoculation

Due to the limitations of the facility, this study was conducted across three experimental setups. A total of six (*n* = 6) USDA Select boneless beef strip loins (*Longissimus lumborum*, IMPS #180) were obtained from a USDA-inspected meat processing facility in Northern Colorado, USA, at three different time points. Two strip loins were collected for each experimental replication. The strip loins were vacuum-packed and transported (~1 h) to Colorado State University, where they were held in dark storage conditions at 2°C for 12 d post-fabrication. Following dark storage, the strip loins were subject to a decontamination process to reduce indigenous microbiota on the meat surface (Dave and Ghaly, 2011; Gill, 1996). For the decontamination process, the strip loins were divided dorsally into smaller portions for ease of handling and submerged in boiling water for 2 min. Using aseptic technique, the heat-exposed surface of the strip loins was then excised. Lean tissue was separated from the fat tissue and cubed to simulate boneless beef trimmings.

To standardize lean to fat ratios across the study's replications, trimmings were weighed in batches of 900 g of lean tissue and 100 g of fat tissue, resulting in approximately a 90% lean-to-10% fat composition. The 1000 g batches of simulated trimmings (*n* = 6 per replicate) were inoculated with 50 mL of a 6 log CFU/mL mixture of meat spoilage bacteria. The bacterial mixture contained six common "ephemeral spoilage organisms" (ESO) that are known to cause meat spoilage (Gill, 1996; Hernández-Macedo et al., 2011; Koutsoumanis et al., 2006; Nychas et al., 2008). Specifically, the spoilage bacteria utilized in this study were previously isolated from spoiled beef steaks. The 6-isolate mixture included three *Pseudomonas* species: *P. fragi* (CMSQ-SB3)*, P. fluorescens* (CMSQ-SB4)*, P. lundensis* (CMSQ-SB5), along with three lactic acid bacteria (LAB): *Carnobacterium divergens* (CMSQ-SB1)*, Leuconostoc gelidum* (CMSQ-SB2)*, Lactobacillus sakei* (CMSQ-SB26). The isolates were maintained at -80°C in tryptic soy broth (TSB; Difco, Becton Dickinson and Company [BD], Sparks, MD) containing 15% glycerol.

For this study, the inoculum was prepared as described by Smith et al. (2024). Briefly, using an inoculation loop, a portion of the frozen culture of each strain was transferred into separate test tubes containing 10 mL of TSB. The inoculated broths were incubated at 25°C for 24 h, after which the cultures were streak-plated onto tryptic soy agar (TSA, Neogen Culture Media, Lansing, MI) plates and incubated at 25°C for 72 h. A single colony from the streak plate of each strain was selected and separately inoculated into 10 mL of fresh TSB and incubated for 24 h (25°C). Subsequently, bacterial strains were sub-cultured by transferring 0.1 mL of the culture into 10 mL of fresh TSB and incubated at 25°C for an additional 24 h.

Following incubation, the cultures of all strains, except *P. fragi,* were diluted tenfold with maximum recovery diluted (MRD, Neogen Culture Media) to a concentration of approximately 8 log CFU/mL. *P. fragi* was excluded from this dilution step based on preliminary findings by Smith et al. (2024), which indicated that this bacterium only reached a concentration of ca. 8 log CFU/mL within 24 h, whereas the remaining five strains reached a concentration of ca. 9 log CFU/mL within the same period of time (Smith et al., 2024). This ca. 8 log CFU/mL cultures of all six strains were combined, and cells were harvested and washed with phosphate-buffered saline (pH 7.4, Sigma-Aldrich, St. Louis, MO) as described by Smith et al. (2024). After the washing steps, the cell pellet was resuspended in 54 mL PBS and then diluted to a concentration of ca. 6 log CFU/mL. This cell suspension was used for the inoculation of the simulated trimmings.

A 50 mL aliquot of the inoculum mixture was added to each batch of 1000 g of simulated boneless beef trimmings. The target inoculation level was ca. 4 log CFU/g. After inoculation, the trimmings were manually mixed for 80 s and maintained at room temperature (ca.22°C) for 20 min to facilitate cell attachment. Subsequently, the inoculated trimmings were processed twice through a meat grinder (LEM, Model 1781, West Chester, OH) fitted with a coarse grind plate (12.7 mm). Ground beef was portioned into 100 g samples, vacuum packaged in 6 x 8.5-inch (3-mil standard barrier) bags (Prime Source Vacuum Pouches, St. Louis, MO), and randomly allocated to temperature test chamber units set at experimental temperatures of -20.6°C, -15.0°C, or -9.4°C (Tenney T2C-A-F4T Temperature Test Chambers, Thermal Product Solutions, New Columbia, PA). The temperatures correspond to the temperatures currently maintained for industry in Fahrenheit, -5°F, as well as the desired experimental temperatures, 5°F and 15°F. Due to equipment limitations, only two chambers were available for use per trial, requiring a designed random replication across the three temperature conditions (see Table 2.1). Sample analyses for microbial counts and lipid oxidation were performed on days 1, 15, and 30 of frozen storage. On each sampling day, samples were removed from frozen storage and allowed to thaw in a refrigerated environment held at 4°C for 24 h before analysis.

2.2.2 Microbiological Analysis

The thawed ground beef samples were analyzed for aerobic plate counts (APC) to determine the effect of frozen storage temperature and duration on the inoculated populations. The vacuum-packaged samples were gently massaged to evenly distribute the outer and inner portions of the product, ensuring all cells exposed to experimental temperatures were included in the microbial analysis. Using aseptic technique, 25 g of ground beef was placed into a 24 oz (710 mL) filter Whirl-Pak bag (Nasco, Pleasant Prairie, WI), along with 75 g of MRD. The mixture was then placed in a paddle blender (Stomacher® 400 Circulator Lab Blender, Seward, West Sussex, UK) and blended at 200 revolutions per minute (rpm) for 2 min. Sample homogenates were then tenfold serially diluted and plated, in duplicate, onto TSA plates. Plates were incubated at 25°C, and colonies were counted after 72 h of incubation.

2.2.3 TBARS Assay

The remaining ground beef samples from the microbial analysis were processed for evaluation of lipid oxidation. Samples were submerged and frozen using cryogenic nitrogen. Once removed from the liquid nitrogen, the samples were blended into a powder using a commercial blender (Waring Products Blender, Model 51BL32, 120V, Torrington, Connecticut), packaged in a 7 oz Whirl-Pak bag (Nasco) to be analyzed.

Frozen powdered meat samples were weighed to approximately 2.5 g (± 0.200 g) and placed into 50 mL conical tubes in duplicate, with the final weight of the conical tubes recorded. An 11% trichloroacetic acid (TCA) solution was prepared within 24 h before the assay by mixing 110 g of TCA (trichloroacetic acid $\geq$ 99.0% (titration); Sigma-Aldrich) with 1 L of deionized water, and then refrigerated at 4.4°C. After chilling, 11.25 mL of the 11% TCA solution was added to each powdered meat sample in the conical tube, and the mixture was homogenized for 30 s. The homogenate was filtered to separate meat particulate from the liquid solution. A 20 mM thiobarbituric acid (TBA) solution (2 thiobarbituric acid, MP Biomedicals, LLC, Solon, OH) was prepared by dissolving 1.44 g of TBA into 500 mL of DI water, then heated at 55°C for approximately 1 h until fully dissolved. The solution was wrapped in aluminum foil to protect it from light exposure and subsequently refrigerated. The 20 mM TBA solution was mixed with the filtration at a 1:1 ratio, in duplicate. Aliquots were held in dark storage at 20°C for 20 h. Subsequently, the mixture was transferred into cuvettes, and absorbance measurements (532 nm) were taken using a spectrophotometer (Shimadzu, Model UV-1800, 120V, Kyoto, Japan).

2.3 Statistical Analysis

For each frozen storage temperature, two trials were conducted with five samples analyzed on each sampling day (n = 10). Temperature conditions were alternated between the two temperature test chambers (A and B) across the three experimental setups, as shown in Table 2.1. During each experimental setup, the samples were randomly assigned to the chambers, and a random 5 samples were pulled during each sampling day.

2.3.1 Microbial Analysis

For statistical analysis of the recovered aerobic plate counts, an ANOVA model was employed, featuring a two-factor fixed-effects factorial design with logarithmic growth ($\log_{10}$ CFU/g) as the dependent variable. Fixed factors included frozen storage temperatures (-20.6°C, -15.0°C, and -9.4°C), storage days (1, 15, 30), and the appropriate interaction. Freezer (A and B) and trial (1 and 2) were treated as random discrete variables to account for any variability caused by freezing conditions and experimental design. Estimated marginal means were calculated for fixed effects and their interactions within each treatment group, and pairwise comparisons were made using Tukey's adjusted pairwise comparisons. Analyses were conducted in R version 4.4.1 (R Core Team, 2024) using the car version 3.1.3 (Fox and Weisberg, 2019), emmeans version 1.10.7 (Lenth, 2025), and broom version 1.0.7 (Robinson et al., 2024). Type I error was set at α = 0.05.

2.3.2 TBARS Analysis

The statistical ANOVA model for evaluating effects of treatment on lipid oxidation utilized a two-way fixed effects factorial design with average absorbance readings, expressed as mg of malondialdehyde (MDA) per kg of beef, as the dependent variable. Fixed effects included 'day' (1, 15, 30) and 'treatment' (-20.6°C, -15.0°C, -9.4°C), along with the appropriate interaction. Trial (1 and 2) was treated as a random discrete covariate to account for possible differences in absorbance readings across trials. All models were tested using Type III F-tests (Fox and Weisberg, 2019). Estimated marginal means for fixed effects and interactions were calculated within each day and compared using Tukey-adjusted pairwise comparisons. Analyses were conducted in R version 4.4.1 (R Core Team, 2024) using the car version 3.1.3 (Fox and Weisberg, 2019), emmeans version 1.10.7 (Lenth, 2025), and broom version 1.0.7 (Robinson et al., 2024). Type I error was established at α = 0.05.

2.4 Results and Discussion

2.4.1 Microbial Populations

Results of the microbiological analysis of the ground beef samples stored at -20.6°C, -15.0°C, and -9.4°C are shown in Table 2.2. For each of the three experimental setups conducted, five inoculated samples were analyzed on day 0 to determine the initial microbial load (i.e., before frozen storage). The aerobic plate counts for these samples were 4.40 ± 0.12, 4.33 ± 0.04, and 4.32 ± 0.03 log CFU/g (data not shown in tables). The recovered plate count values fall within the industry-standard for the initial microbial count range of 10^2 to 10^5 CFU/cm² (Cerveny et al., 2009).

There were no violations of normality, as indicated by the Shapiro-Wilk test ($W = 0.98$, $P = 0.17$). In the microbial quality type II ANOVA model, the day fixed effect on the impact to microbial growth was significant ($P < 0.05$). However, no other fixed effects or random discrete covariates had a significant influence on microbial populations of ground beef ($P \geq 0.05$). Additionally, the interaction between treatment temperatures and day in frozen storage was not significant ($P \geq 0.05$).

Figure 2.1 illustrates the mean APC (log CFU/g) value, with the SE bar representing the variability among replicates. As anticipated, microbial growth did not occur during frozen storage resulted in no viable plate counts, and changes during storage days, regardless of trial repetition ($P \geq 0.05$). These findings are consistent with existing literature, which indicates that frozen storage temperatures below -5°C inhibit the growth of microbial spoilage organisms (Adam et al., 2010; Coombs et al., 2017).

2.4.2 Lipid Oxidation

Effects of frozen storage of ground beef on lipid oxidation at -20.6°C, -15.0°C, and -9.4°C are shown in Table 2.3. For each of the three experimental setups, five samples were collected on day 0 to determine the product's initial lipid oxidation levels. There were no violations of normality, as indicated by the Shapiro-Wilk test (W = 0.99, $P = 0.78$). A significant interaction between day and temperature was observed during frozen storage ($P < 0.05$). The fixed effects of day and trial were not statistically significant ($P \geq 0.05$). However, the effects of temperature trend towards significance ($P = 0.09$). As anticipated, lipid oxidation levels did not impact lipid oxidation levels with changes in frozen storage duration. The lack of lipid oxidation could be attributed towards the packaging style for the product during the experimental frozen conditions, as the lack of oxygen within vacuum packaging conditions limits the extent of lipid peroxidation, therefore potentially affecting our results (Hernández-Macedo et al., 2011; Ikram et al., 2025b; Kenawi, 1994; Nychas et al., 2008).

Conflicting results exist concerning lipid oxidation under frozen conditions, but our findings aligned with existing literature, indicating that short-term frozen storage does not affect the chemical spoilage processes of lipid and protein oxidation (Soyer et al., 2010). However, to acknowledge the conflicting viewpoints within the literature, we must consider the element of storage duration, as our experiment only held the product under frozen conditions for 30 d, which is not the industry standard for the duration of product storage in these conditions (Utrera et al., 2014). To better align with industry standards, additional research should be conducted to examine the long-term effects of the experimental frozen temperatures for a minimum of two years.

Our results indicated that, under extremely controlled experimental frozen storage temperature conditions with minimal variation in temperature over time ± 0.01°C at -20.6°C, -

15.0°C, and -9.4°C, there was no effect on microbial spoilage populations (Table 2.2) or lipid oxidation (Table 2.3) for ground beef products. Whether commercial facilities can sustain such precise temperature control to implement this technology remains uncertain. Within the standard freezing conditions of -18°C, temperature variations of ± 2°C are known to have minimal effect on overall product quality (James and James, 2003; Labadie, 1999). Failure to hold products within the limitations of the temperature thresholds of ± 2°C can accelerate the overall reduction of product quality and shelf life (James and James, 2003; Ladikos and Lougovois, 1990; Raharjo and Sofos, 1993; Soyer et al., 2010; Vieira et al., 2009).

Overall, the cold chain is critical in preserving product quality. The investigation of microbial spoilage and lipid oxidation functions as essential indicators of freshness and shelf life throughout the supply chain (Ayres, 1960; Gill, 1996; Hernández-Macedo et al., 2011; Nychas and Skandamis, 2005). This preliminary study was carried out to attest the need for additional research to corroborate these findings.

2.5 Conclusions

The results of this study indicated that the microbiological growth and extent of lipid oxidation for ground beef when stored for up to 30 d were unaffected by experimental frozen storage temperatures tested. These results offer valuable insights into the impact of storage temperature on microbial and chemical spoilage processes, consequently influencing meat quality. Additionally, the findings established a robust foundation for the development of advanced cold storage technologies aimed to optimize freezer temperature conditions. Nevertheless, additional research is warranted to validate these findings across various meat products, including offal, as well as whole primal and sub-primal cuts, as well as longer frozen storage durations.

Table 2.1 – Experimental design and assignment of samples to two temperature test chambers performed across three experimental setups with 10 replicates per temperature (N = 30).

Tenney T2C-A-F4T Temperature Test Chambers	**Experimental Replication**		
	1	**2**	**3**
A	-9.4ºC	-20.6ºC	-15.0ºC
B	-20.6ºC	-15.0ºC	-9.4ºC

Table 2.2 – Mean APC (log CFU/g ± SE[1]) for inoculated ground beef stored at -9.4°C, -15.0°C, or -20.6°C (n = 10) for up to 30 d.

Storage Days	**Controlled Storage Temperature[1]**		
	-9.4°C	**-15.0°C**	**-20.6°C**
1	4.34±0.02	4.32±0.02	4.33±0.02
15	4.33±0.02	4.27±0.02	4.28±0.02
30	4.30±0.02	4.28±0.02	4.28±0.02

[1] SE = Standard Error

[2] The interaction between storage temperature and storage day was not significant ($P \geq 0.05$); storage temperature and storage day main effects were also not significant ($P \geq 0.05$)

Table 2.3 – Mean malondialdehyde per kg beef (mg MDA/kg) ± SE[1] for beef stored at -9.4°C, -15.0°C, or -20.6°C (*n* = 10) for up to 30 d.

Storage Days	**Controlled Storage Temperature[1]**		
	-9.4°C	**-15.0°C**	**-20.6°C**
1	2.95±0.19	3.38±0.13	2.66±0.08
15	3.20±0.14	3.34±0.15	2.74±0.25
30	2.91±0.19	2.66±0.09	3.13±0.10

[1] SE = Standard Error

[2] The interaction between storage temperature and storage day was not significant ($P \geq 0.05$); storage temperature and storage day main effects were also not significant ($P \geq 0.05$)

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CHAPTER 3: THE IMPACT OF ARTIFICIAL INTELLIGENCE INTERGRATION TO CONTROL FROZEN TEMPERATURES ON ENERGY CONSUMPTION IN MEAT STORAGE FACILITIES

3.1 Introduction

The practice of food preservation through freezing can be traced back to approximately 1,000 BC, when ancient civilizations employed ice and naturally cold environments to extend the shelf life of commodities (Archer, 2004; Diaz Sanchez et al., 2021; Garnett and Jackson, 2007; Lawrie, 1979; Marchi and Zanoni, 2022). Over time, freezing techniques have evolved into a scientific discipline that places products in low-temperature environments with controlled freezing rates, with the aim of reducing microbial activity, enzymatic reactions, and oxidative degradation. Thereby ensuring both product quality and safety during prolonged storage durations (Goodwin et al., 2002).

Cold storage facilities are fundamental components of both international and domestic meat supply chains (Neusel and Hirzel, 2022; Zanoni and Zavanella, 2012). Figure 3.1 shows a schematic layout of the meat cold supply chain. According to the United States Department of Agriculture Economic Research Service (USDA ERS) (2025), the United States exported 1.36 million metric tonnes of beef and veal in 2024 (USDA ERS, 2025). Perishable commodities such as meat require specific temperature-controlled environments to effectively control microbial growth, lipid oxidation, and other degradation processes that could compromise product quality. Innovations such as refrigeration and freezing technologies have significantly improved product preservation and food security, enabling the consistent delivery of quality products across seasonal demands (James and James, 2010). Advancement of technology and the increased utilization of freezing systems entail considerable energy consumption.

The International Institute of Refrigeration (IIR) estimates that domestic cold storage alone accounts for approximately 4% of the world's annual electricity consumption through the cold chain. It is also estimated that approximately 120 million commercial refrigeration systems are operating globally, which consume nearly 20% of the available electricity (Dupont et al., 2019). Electricity is considered a secondary energy resource because it is generated through the conversion of primary energy sources (e.g. coal, natural gas, nuclear energy, solar energy, and wind energy (U.S. EIA, 2024). The total number of systems used worldwide further highlights the widespread nature of these sustainability concerns along the cold storage supply chain. Such energy expenditures raise significant issues related to overall energy consumption, industrial energy efficiency, and the sustainability of the global supply chain. Practical use of cold storage facilities face challenges caused by vulnerabilities within the supply chain, which can lead to errors, primarily resulting in product spoilage, microbial or chemical, that ultimately diminishes consumer acceptance and increases food waste (Nastasijević et al., 2017). The U.N. Food Waste Index Report (2024) estimates that in 2022, approximately 1,052 million metric tons of waste were generated through domestic, food service, and retail distribution systems (United Nations Environment Programme, 2024). The waste accounts for about 19% of all available food in the system. This highlights the urgent need to tackle energy inefficiencies in the frozen supply chain, particularly in cold storage facilities.

In response to the prevailing challenges of reducing energy consumption, adopting more efficient measures may serve as an effective approach to reduce waste and mitigate the environmental impacts associated with the cold chain (Neusel and Hirzel, 2022). Nonetheless, the obstacle to implementing such energy reducing measures remains the need for sufficient financial capital allocated for these investments (Diaz Sanchez et al., 2021; Marchi and Zanoni, 2022).

While society has made substantial technological advancements over the past two decades, the practical application of AI temperature management technology remains limited. When considering modern modifications, such as AI, compared to historical practices and expectations from the mid 20th century, it is important to consider the context of advanced technology and facility capabilities. These advancements are essential when assessing areas for improvement in temperature control for frozen storage conditions.

Advancements in technology, such as the ongoing development and integration of energy demand management profiles, include the Internet of Things (IoT), machine learning (ML), and artificial intelligence (AI) (Salgado et al., 2024). These innovations aim to enhance efficiency and lower energy consumption while effectively meeting operational demands, considering the real-time conditions of the facility (Allouche et al., 2023). The application of AI in energy management offered substantial benefits to cold storage logistics companies by delivering solutions that optimized temperature control, minimized temperature fluctuations, and lowered energy consumption in cold storage environments (Marchi and Zanoni, 2022). By leveraging predictive algorithms based on historical operational data and real time monitoring, the technology was able to predict operational changes throughout the daily cycle, allowing for greater management of temperature deviations within the system. Fewer deviations equated to reduced energy usage by the facility. Through the enablement of AI driven decision making, there was a greater COP for the equipment, such as the compressors, and with a greater COP, a reduced amount of electricity was needed to power the system. The energy was already being maximized during a reduced operating cost while operating during non-peak power rates, creating a more stable internal temperature for the facility.

By leveraging AI for monitoring and temperature control, cold storage logistics operators can enhance storage efficiency, lower operational costs, and reduce their carbon footprint, thereby supporting their corporate sustainability objectives (Fu et al., 2024a; Kamble et al., 2019; Salgado et al., 2024). Historically, the main challenges in manual energy resource scheduling have hindered the ability to adapt to the evolving requirements of dynamic cold storage environments. Variations in ambient temperature during operational periods, the movement of products throughout the facility, and door openings considerably influence the environmental efficiency of these facilities (Wang et al., 2024).

Fluctuations above the temperature set point exceeding ±3.0°C adversely affect the overall quality of the product within the environmental conditions, rendering it susceptible to microbial growth and chemical potential (James and James, 2003; Koutsoumanis et al., 2006). As demand for sustainable food systems among regulatory agencies escalates alongside consumers' financial constraints, the evolution of freezing technologies must continue to balance food safety, waste reduction, and environmental impact (Grunert et al., 2014; United Nations, 2023).

Temperature-controlled supply chains are essential to the global distribution of perishable goods, utilizing advanced refrigeration and monitoring systems to sustain precise temperature ranges that ensure the safety and quality of meat and food products (Nychas et al., 2008). By addressing risks related to temperature fluctuations, these systems contribute to preserving quality and extending the shelf life of perishable products during both international and domestic transportation and storage. This case study analyzed real-time operational effects of using temperature management AI on energy use and consumption within cold storage facilities.

3.2 Materials and Methods

3.2.1 Vapor Compression Refrigeration System Overview

The Vapor-Compression Refrigeration (VCR) system accounts for 80% of the market share in commercial, domestic, and refrigeration transportation sectors in the cold storage industry (Fu et al., 2024b). Utilization of VCR systems accounts for 15% of the world's electricity consumption, and contributes to 10% of global greenhouse gas emissions (Fu et al., 2024b).

Performance and efficiency of the refrigeration systems is measured using value, coefficient of performance (COP). The COP is quantified by the amount of heat expelled from the refrigerator (Q_c) to the work input into the system (W_{in}), together determining the overall cooling efficiency ($K_{cooling}$) (Donev et al., 2024; Redko et al., 2020; Zajac, 2016).

$$K_{cooling} = \frac{Q_c}{W_{in}}$$

The higher a COP value is, the greater the system is at conserving energy resources.

Commercial VCR systems are comprised of the components: evaporators, compressors, expansion valves, and condensers (Fenton et al., 2019; Rodriguez and Rasmussen, 2017). The refrigeration systems use energy to generate low-grade thermal energy, resulting in higher COP values. (Fu et al., 2024b).

3.2.2 Power Utilization

Power utilization data, measured in kilowatts (kW), were collected from two industrial cold storage facilities located in distinct regions of the United States. One facility was located in Colorado, a region known for its industrial activity, while the other was situated in Illinois, noted for its central location. Despite their differing geographical locations, both facilities experience similar seasonal environmental temporal fluctuations. Furthermore, these facilities share similar operational characteristics, facilitating a comprehensive analysis of energy consumption trends before and following the implementation of AI driven temperature management optimization.

Before the implementation of temperature optimization, baseline energy consumption data points were collected through traditional electricity meter readings. Subsequently, following the application of energy AI driven optimization, energy demand measurements were acquired utilizing the CrossnoKaye ATLAS platform (“Home - CrossnoKaye Inc.,” 2023). This application provides a comprehensive, cloud-based platform designed to enhance the management of ambient temperature across a wide range of facilities. The system employs advanced predictive algorithms and site-specific applications to optimize environmental control, thereby ensuring operational efficiency and energy conservation. The predictive algorithms will monitor temporal variations in the facility to maintain the necessary temperature set point established by the facility management team, along with the specified standard deviation. These algorithms will monitor the temperature setpoint and adjust accordingly, considering the dynamic variables of price and environmental conditions, to ensure the system operates at maximum efficiency during off peak energy pricing, avoiding any unnecessary energy consumption during peak rates.

The process begins with ATLAS driven AI calibration, which thoroughly examines and monitors current system operational schedules. This preliminary phase enables precise control of temperature set points and variations, which aids in achieving energy usage goals; and creates a basis for autonomous system control to manage operations based on real-time information. As the system is fully deployed, it continuously learns and adapts to evolving conditions, providing more intelligent and efficient temperature regulations tailored to each facility's distinctive requirements.

Energy consumption data readings related to temperature/cooling management were collected from the ATLAS platform every 15 min over a period of 94 d. Before conducting the statistical analysis, the power consumption metrics, originally recorded at 15 min intervals, were combined into hourly data, with hourly averages calculated accordingly (N = 73,173).

3.2.3 Temperature Fluctuation

Energy usage fluctuations over time, influenced by operational cycles and load variations, remained stable during both treatment phases, indicating a consistent energy consumption pattern throughout the study.

3.2.4 Statistical Analysis

A statistical ANOVA model was utilized to evaluate the influence of facility location (Colorado and Illinois) and the status of energy AI implementation (before and after) on overall energy consumption recorded in power usage. The two-way fixed effects factorial design facilitated the evaluation of both independent and interaction effects, thereby establishing whether AI driven optimization variably influenced energy consumption across different facilities. Integration of both independent and interaction effects within a single model enhances statistical power and ensures that observed differences in energy consumption are not confounded by facility-specific characteristics or environmental variables. To account for potential environmental and seasonal variation in energy consumption, hour of measurement during the day was incorporated as a random discrete covariate. Furthermore, external temperature conditions remained comparable both before and following implementation of AI, as both facilities were subject to all four seasons. This approach minimizes risk of seasonal fluctuations affecting cooling costs. Estimated marginal means were calculated for fixed and interaction effects within each facility and were compared using Tukey-adjusted pairwise comparisons. All statistical analyses were performed utilizing RStudio (version 4.4.1) (R Core Team, 2024), employing the 'broom,' 'car,' and 'emmeans' packages (Fox and Weisberg, 2019; Lenth, 2025; Robinson et al., 2024), with Type I Error established at $\alpha = 0.05$.

3.3 Results

3.3.1 Power Demand Utilization

The case study showed that fixed effects for AI treatment, facility, measurement hour, and the AI-facility interaction were highly significant ($P < 0.0001$). The mean square for facility was a major source of variation in our results, at 2,277,726,278. Another significant source of differences was the variable treatment, at 1,510,918,688. Lastly, the interaction between treatment and facility had a mean square value of 68,969,130, contributing to the overall variation in our final results. The study also presented least square means for overall power utilization in kilowatt hours (kWh), adjusted for the measurement hour at each site (Colorado or Illinois) and whether the AI treatment had been applied before or after implementation (Table 3.1).

After deploying the temperature management AI treatment within the Colorado facility, from the pre-implementation average power of 671.5 kWh decreased to 443.9 kWh. Indicative of a reduction in power utilization by approximately 227.6 kWh, which was statistically significant ($P < 0.0001$), as shown in Figure 3.2. At the Illinois facility, before the implementation of the temperature AI treatment, average power consumption was approximately 1,088.6 kWh the average power consumption declined to 737.3 kWh following implementation. This resulted in an overall reduction of approximately 351.3 kWh, which was statistically significant ($P < 0.0001$), as illustrated in Figure 3.3.

Furthermore, descriptive statistics of power usage across each facility, shown in Table 3.2, indicated that power savings aligned with the results from the previous case study using CrossnoKaye's ATLAS system (Crossnokaye, 2022). With adequate infrastructure, the deployment of temperature AI management systems diminished overall power consumption (Nozari et al., 2022; Salgado et al., 2024). Reductions were substantial and consistent across different facilities, emphasizing the potential impact of these systems in real-world operational environments.

3.4 Discussion

3.4.1 Cost Savings

To contextualize the operational cost advantages of implementing energy AI management, it is imperative to quantify the financial savings derived from reduced power consumption as an consequence of improved temperature control efficiency. An estimation of the economic impact resulting from the observed reductions in energy utilization was calculated using the following formula:

$$Cost\ Savings = \ Reduction\ of\ Power\ (kW) \times Total\ Time\ (h) \times Electricity\ Rate\ (\frac{USD}{kWh})$$

Based on an average peak and non-peak industrial electricity rate of \$0.0823 per kilowatt-hour (U.S. EIA, 2024), the Colorado facility's reduction of 227.6 kWh over a duration of 94 d resulted in an estimated cost saving of approximately \$42,258 (USD). If this reduction were sustained throughout a full year of 365 d, it could potentially yield savings of approximately $164,087 (USD). An average reduction of 351.3 kWh over the same 94 d period in the Illinois facility was associated with an estimated cost saving of $65,225.45 (USD), which could be scaled to approximately $253,269.03 (USD) annually, provided the energy reduction remained consistent. Economic benefits linked to implementing AI driven temperature management optimization in industrial cold storage supply chains seemed apparent.

3.4.2 Operational Efficiency

Although facilities included in the present study exhibited similar operational characteristics and experienced reasonably similar seasonal patterns, the Illinois site initially consumed more energy before implementation of AI and therefore experienced a larger reduction in energy use and cost than the Colorado facility. This discrepancy was attributed to regional energy demand profiles plus differences in operational dynamics among the two facilities, which were more effectively managed following the integration of AI algorithms rather than through manual oversight. The finding of energy and cost reduction within our study aligns with the findings of Salgado et al. (2024), who concluded that AI driven management of facilities ranks among the most effective strategies for reducing operational expenses and enhancing energy efficiency. A comparable study by Nozari et al. (2025) demonstrated that the integration of AI with Internet of Things (IoT) technology successfully reduced operational costs of refrigeration distribution centers by 26% (Nozari et al., 2025).

The ATLAS platform provided real time management capabilities to the refrigeration systems, given the response factor of peak electricity pricing, while ensuring products were held in a temperature safe environment, further highlighting the economic value that this would add to the cold storage supply chain (Wang et al., 2024). The proficiency and adaptability of ATLAS to operational conditions improved energy efficiency beyond the initial manual temperature control capabilities. Automated temperature control promotion not only lowered energy expenses by ensuring more consistent temperature regulation, vital for preserving product quality, safety, and shelf life, but also highlighted progress in energy management technology.

Implementation of temperature control automation for management within facilities not only influences cost savings but also impacts temporal temperature variations, the COP of VCR systems, and the overall greenhouse gas (GHG) emission potential within these facilities (Dong and Miller, 2021; Onyeaka et al., 2025; Salgado et al., 2024). Deployment of artificial intelligence (AI) in temperature management effectively reduced variance, thereby decreasing variability in surface temperature (Onyeaka et al., 2025). This reduction in temperature variability has consequently led to an increased overall product stability (Archer, 2004; Banerjee and Maheswarappa, 2019; Campañone et al., 2006; James and James, 2003; Kitinoja, 2013). In addition to reducing temporal fluctuations, this improvement further supports raising the static frozen storage temperature by at minimum of 3°C (Allouche et al., 2023).

Raising ambient temperature inside cold storage facilities reduces total energy required for cooling, supporting Marchi and Zanoni's findings (2022). By enabling AI to manage the frozen storage temperatures more consistently, there is a greater likelihood for the system to operate at peak COP efficiency. Allowing the system to operate at peak COP not only reduced external energy demands but also kept the compressor and other equipment running at their highest efficiency levels (Belman-Flores et al., 2017). As a result, the amount of refrigerant used would be reduced, and the lifespan of the equipment would be prolonged (Cascini et al., 2016; Nandanwar et al., 2023).

In addition to operational and economic benefits, the implementation of AI driven energy optimization presents opportunities for substantial contributions toward environmental sustainability. By managing refrigeration technologies and by reducing energy usage, this initiative supports the organization's efforts to decrease Scope-II emissions. Scope-II emissions refer to the indirect greenhouse gas emissions resulting from the purchase of electricity, steam, heat, and cooling required for organizational operations (US EPA, 2020). The decrease in electrical consumption at the Colorado and Illinois facilities, achieved through the deployment of AI systems, subsequently resulted in a reduction of classified Scope-II emissions (US EPA, 2020). This observation corroborates existing scientific literature, such as the study conducted by Dong and Miller (2021), which estimated that lower temperature meat products typically produce elevated levels of total greenhouse gas emissions (Dong and Miller, 2021). Furthermore, this research highlighted that, within the post-agricultural stages, refrigerated warehouses rank among the three principal stages in terms of emissions for meat products (Dong and Miller, 2021).

Although AI offers benefits in temperature management, some challenges remain. A key issue is limited financial resources, which prevent adoption of energy-efficient solutions. Without necessary funding, companies may continue using outdated, inefficient technologies, further contributing to increased supply chain energy consumption (Marchi and Zanoni, 2022). However, as consumer demand for sustainability increases, it may become necessary to go beyond this willingness-to-pay threshold to adopt alternative operational approaches (Grunert et al., 2014; Neusel and Hirzel, 2022). Additionally, engaging commercial refrigeration companies in a shift towards technologies that prioritize long-term energy savings over immediate short-term gains is crucial (Garnett and Jackson, 2007). Limitations of this current case study include the fact that the 94 d period only captured the third and fourth quarters of the year. To gain a more holistic view, a subsequent study should be performed over a longer timeframe, ideally a full 365 d, to capture the different temporal variations over the span of a year. Another limitation of the study was the selection of the facilities. To gain a better understanding of site-specific demand, the inclusion of more facilities in the subsequent studies would better illuminate the actual reduction of energy usage. Additionally, being able to monitor the facilities by temperature within each room and the corresponding energy usage to maintain said temperature would be beneficial in drawing future conclusions.

3.5 Conclusions and Industry Implications

Refrigeration of temperature-sensitive products offers extensive benefits, including the preservation of food safety and quality, as well as reduction of overall food waste (Garnett and Jackson, 2007). This case study aligns with existing literature indicating that the application of Industry 4.0 technologies, particularly artificial intelligence (AI), has potential to improve overall efficiency of cold storage (Ivanov et al., 2019). Such improvements are realized through decreased energy consumption, increased cost savings, enhanced temporal management in response to operational challenges, and the potential to support sustainability initiatives. Our findings demonstrated that through temperature management using AI effectively reduced overall power usage in a commercial cold storage facility, regardless of geographic location. Results confirm the transformative potential of integrating temperature management AI into cold storage facilities, with the capacity to revolutionize energy consumption and operational efficiency. Optimization efforts over the 94 d period led to an average energy reduction of 289.5 kWh per facility, representing approximately a 32% decrease from the pre-AI implementations' average consumption of 880 kWh.

This reduction not only decreased the total energy consumption within the facilities but also resulted in sizable economic benefits. Specifically, the average decrease of 289.5 kWh across both facilities during the 94 d period corresponds to an estimated savings of $53,751 (USD) per facility, based on an average electricity cost of $0.0823 (USD) per kWh, as reported by the U.S. Energy Information Administration for February 2025. Such cost savings could hypothetically lower the Scope-II greenhouse gas emissions from the facilities, aligning with the broader environmental objectives of the supply chain and its stakeholders.

The case study showed that technology has contributed to economically viable and environmentally sustainable practices. It provides a foundational framework for the integration of advanced computational solutions aimed to optimize facility operations. The scalability of AI driven optimization has the potential to generate even greater cumulative energy savings, especially if extended to other sectors within the cold storage supply chain, such as distribution centers and retail cold storage facilities that face similar inefficiencies.

Continued empirical research is essential to validate these findings across diverse operational environments and facilities, thereby strengthening industry wide adoption and potentially informing policy incentives and formulations based on AI optimized energy management solutions.

Table 3.1. Means ± SE[1] for energy utilization (kWh) by facilities in Colorado, USA and Illinois, USA before and after energy AI implementation over the 94 d period.

Facility	Energy AI Status		
	Before	After	Tukey's *P*
Colorado, USA	671.5±0.815	443.9±0.822	< 0.0001***
Illinois, USA	1088.6±0.816	737.3±0.816	< 0.0001***

[1] SE = Standard Error

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$;

Table 3.2. Descriptive summary statistics for hourly power usage readings from cold storage facilities in Colorado and Illinois (N = 72,173) before and after AI-driven optimization.

Facility	Hour	AI status	N	Mean (±SD[1])	Min	Max
Colorado	0	before	754	692.99±121.11	52.49	935.98
Colorado	0	after	752	439.7±53.79	139.91	630.24
Colorado	1	before	752	681.32±109.36	49.45	855.07
Colorado	1	after	752	437.77±51.15	148.84	577.6
Colorado	2	before	752	678.56±97.61	48.02	869.07
Colorado	2	after	752	433.38±52.27	150.09	556.79
Colorado	3	before	752	670.47±100.71	47.12	866.5
Colorado	3	after	752	432.07±54.35	146.81	609.72
Colorado	4	before	752	666.17±94.78	58.91	878.14
Colorado	4	after	752	432.71±55.38	138.88	577.34
Colorado	5	before	752	656.86±102.11	54.15	863.61
Colorado	5	after	752	435.23±56.65	65.63	616.14
Colorado	6	before	752	650.1±98.6	52.46	828.76
Colorado	6	after	752	434.93±56.76	108.05	578.44
Colorado	7	before	752	646.75±100.65	53.09	840.02
Colorado	7	after	752	438.95±55.61	117.94	624.55
Colorado	8	before	766	643.43±106.61	51.2	834.67
Colorado	8	after	752	439.82±55.5	104.93	616.58
Colorado	9	before	752	636.62±103.37	49.26	846.75
Colorado	9	after	752	434.21±69.09	52.4	627.64
Colorado	10	before	752	633.85±98.3	51.52	836.68
Colorado	10	after	750	432.71±82.94	0.3	698.68
Colorado	11	before	752	634.46±94.88	210.33	829.01
Colorado	11	after	750	438.61±87.59	0	682.59
Colorado	12	before	752	630.82±90.4	195.35	834.03
Colorado	12	after	750	445.78±84.63	0	679.67
Colorado	13	before	752	631.4±102.1	267.41	839.39
Colorado	13	after	752	453.16±76.99	103.01	673.72
Colorado	14	before	752	655.11±96.4	117.24	855.07
Colorado	14	after	752	462.85±70.29	265.55	689.32
Colorado	15	before	752	669.3±94.27	82.92	858.77
Colorado	15	after	752	464.76±67.64	157.99	732.23
Colorado	16	before	752	681.16±98.62	163.38	869.75
Colorado	16	after	752	461.02±64.99	113.45	712.62
Colorado	17	before	752	700.07±91.97	227.91	890.53
Colorado	17	after	752	459.83±60.28	252.03	670.23
Colorado	18	before	752	703.41±92.51	350.06	907.01
Colorado	18	after	752	454.53±58.68	230.88	673.9
Colorado	19	before	752	711.14±90.65	189.53	890.18
Colorado	19	after	752	450.79±53.55	234.99	637.61
Colorado	20	before	752	716.78±90.95	172.22	894.37
Colorado	20	after	752	440.23±52.23	271.38	604.09
Colorado	21	before	752	716.85±94.49	180.97	899.09
Colorado	21	after	752	442.2±53.55	241.38	613.22
Colorado	22	before	752	708.71±110.21	56.18	904.65
Colorado	22	after	752	444.94±51.61	266.85	627.11

Colorado	23	before	752	698.99±124.82	53.23	914.09
Colorado	23	after	752	443.11±51.92	144.91	644.55
Illinois	0	before	754	1123.26±136.51	424.53	1509.32
Illinois	0	after	752	726.15±109.03	467.7	1136.23
Illinois	1	before	752	1132.8±133.62	425.75	1504.73
Illinois	1	after	752	715.31±106.89	369.34	1071.35
Illinois	2	before	752	1111.95±127.42	437.22	1570.07
Illinois	2	after	752	709.58±105.73	434.14	1088.68
Illinois	3	before	752	1090.59±123	394.35	1512.16
Illinois	3	after	752	707.54±105.36	338.76	1028.45
Illinois	4	before	752	1105.91±119.64	607.3	1556.51
Illinois	4	after	752	713.75±113.28	375.63	1084.53
Illinois	5	before	752	1087.97±126.44	810.09	1813.17
Illinois	5	after	752	723.64±103.96	497.14	1050.82
Illinois	6	before	752	1087.83±126.79	768.69	1780.6
Illinois	6	after	752	738.48±105.46	338.09	1102.22
Illinois	7	before	754	1076.62±132.47	435.95	1716.15
Illinois	7	after	752	760.23±103.33	477.64	1162.91
Illinois	8	before	748	1028.59±127.04	744.37	1666.2
Illinois	8	after	752	751.5±106.51	458.61	1178.29
Illinois	9	before	753	991.69±124.81	718.62	1552.42
Illinois	9	after	749	754.18±106.9	470.77	1246.73
Illinois	10	before	752	1018.91±119.46	750.16	1689.42
Illinois	10	after	748	763.33±112.1	501.8	1170.95
Illinois	11	before	752	1064.21±125.29	382	1647.66
Illinois	11	after	748	754.32±106.07	408.6	1079.77
Illinois	12	before	752	1077.09±127.07	464.89	1651.5
Illinois	12	after	747	738.89±117.37	111.78	1109.29
Illinois	13	before	752	1083.75±127.33	586.23	1635.79
Illinois	13	after	748	755.39±123.61	169.79	1231.37
Illinois	14	before	752	1102.54±119.82	781	1667.77
Illinois	14	after	750	765.39±107.25	301.71	1152.82
Illinois	15	before	752	1122.36±130.3	796.19	1688.21
Illinois	15	after	752	757.14±102.58	470.98	1198.32
Illinois	16	before	752	1097.4±120.85	333.1	1668.34
Illinois	16	after	752	758.59±109.93	419.43	1288.69
Illinois	17	before	752	1105.79±140.29	181.44	1698.6
Illinois	17	after	752	755.63±112.2	422.62	1254.77
Illinois	18	before	752	1116.31±147.55	166.32	1684.08
Illinois	18	after	752	739.7±106.93	503.64	1095.53
Illinois	19	before	751	1114.41±153.82	267.2	1649.35
Illinois	19	after	751	713.26±103.8	132.1	1056.47
Illinois	20	before	749	1089.95±225.4	162.8	1505.12
Illinois	20	after	752	720.08±100.9	357.42	1080.73
Illinois	21	before	748	1060.11±226.85	139.06	1497.37
Illinois	21	after	752	722.26±102	342.93	1061.17
Illinois	22	before	752	1112.85±180.83	150.49	1721.18
Illinois	22	after	752	726.12±114.26	397.39	1156.18
Illinois	23	before	752	1122.22±147.44	441.13	1739.62

Illinois	23	after	753	726.18±110.93	375.31	1080.27

[1] SD = Standard deviation from the mean

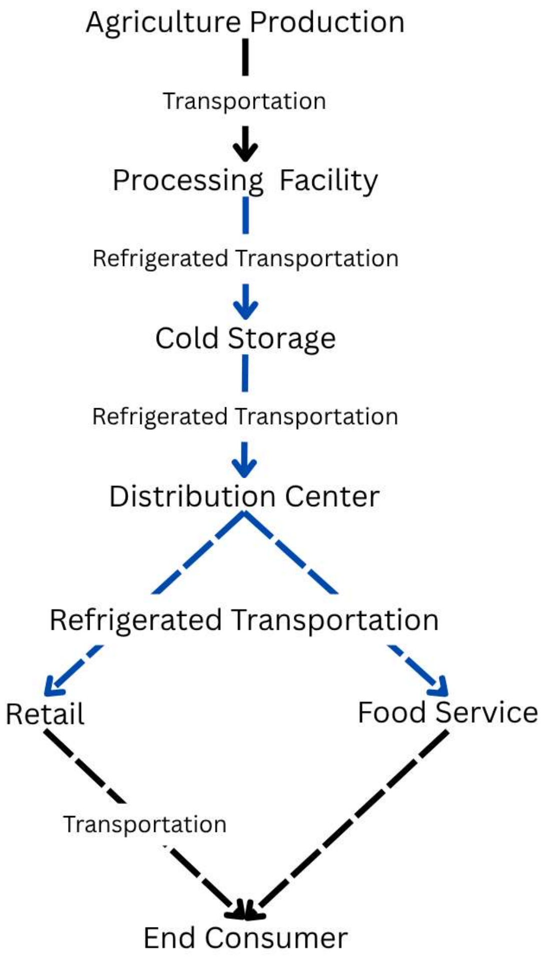

Figure 3.1 – An overview of the cold chain

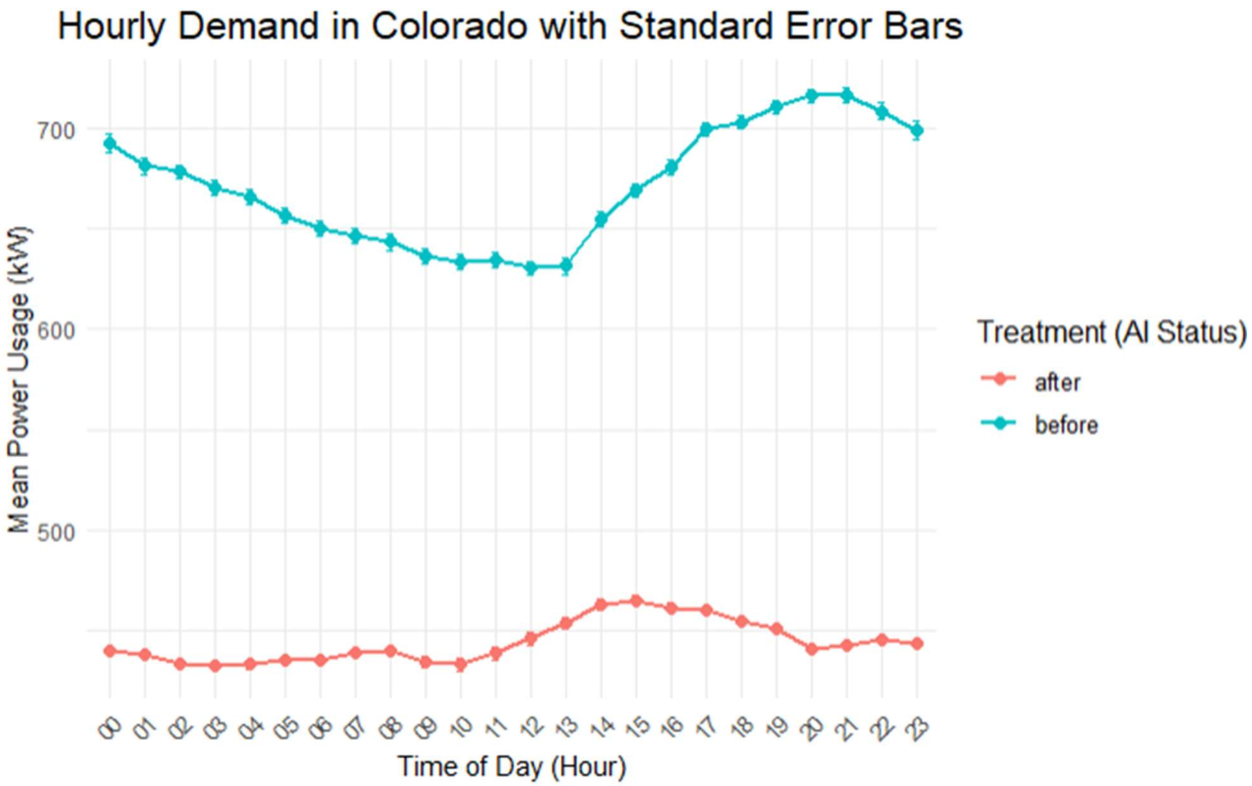

Figure 3.2 – Power Usage (kWh) for Colorado, USA Facility.

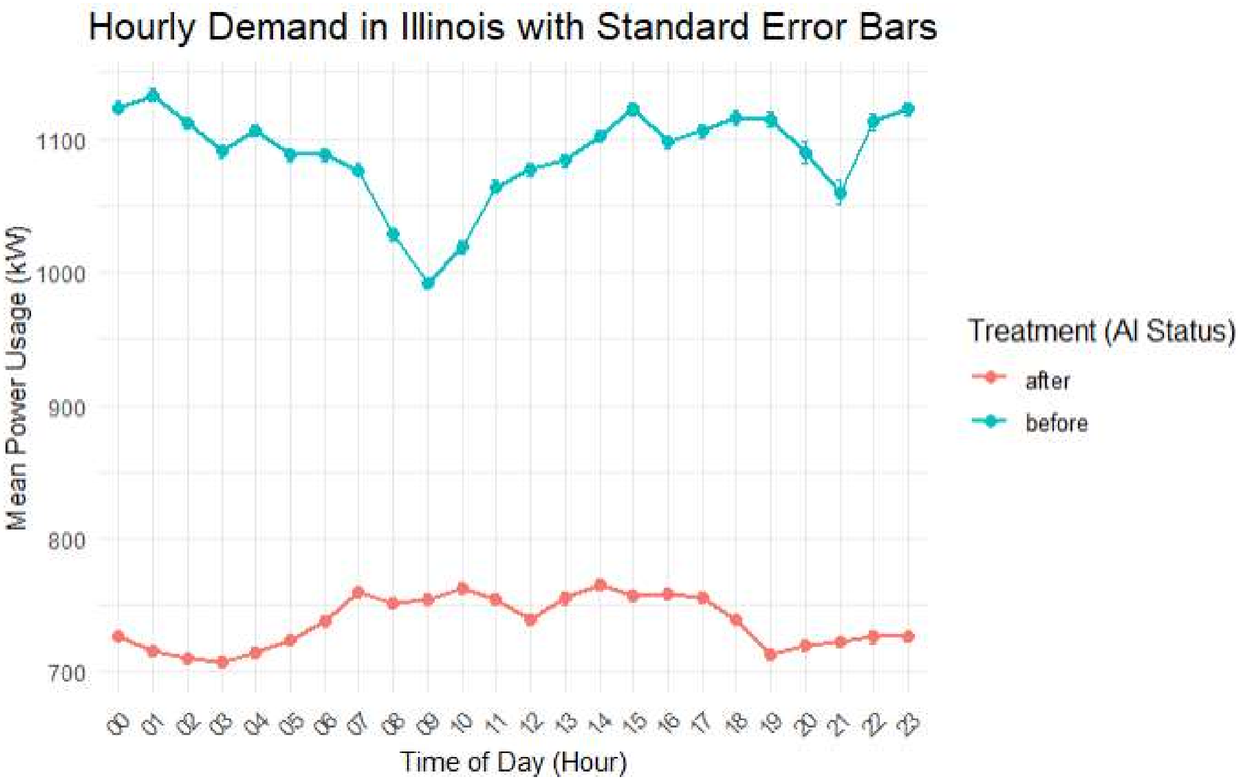

Figure 3.3 – Power Usage (kWh) for Illinois, USA Facility.

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