Microbial Degradation of Food Products

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Abstract

Contamination and degradation of food products by microorganisms are a major problem for the food industry, with serious damage to economic circulation and public health worldwide. Therefore, the objective of this chapter is to clarify how the contamination of microorganisms occurs, the main damage to food, production of toxic substances, economic losses, and new food preservation technologies, with a focus on developing a sustainable and safe food industry. Most foods contain structural ingredients, such as a high concentration of carbohydrates, perfect conditions for an outbreak of microbial infection. The microorganisms that grow in food alter their chemical composition, appearance, taste, texture, and characteristics such as color, shape, and aroma. In addition, some fungi produce mycotoxins that are highly toxic. Finally, contaminated food undergoes degradation and putrefaction, causing irreparable losses to producers.

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Thus, we address new technologies that may be the key to increasing the shelf life of food and reducing the rate of contamination. In conclusion, microorganisms are a persistent problem and difficult to eliminate, especially considering the possibilities of artificial selection. However, emerging technologies can help preserve food for longer, without increasing the risk of artificial selection of the microorganism or even causing changes in food.

Keywords

Pathogens · Food security · Shelf life · Artificial selection · Mycotoxins

6.1 Introduction

Microbial contamination and subsequent degradation of food products due to the biological activities of pathogens are a current problem, faced from the beginning of the food production chain to the consumer base. Since ancient times, human beings have faced problems in food preservation and we can say that human survival itself has been through understanding how to preserve food for long periods. Human civilization has managed to survive using various food preservation strategies, for example, man in the ice age period has adapted to preserve food on ice. In the Bronze Age, we have food preservation by cooking and drying. Although food preservation techniques have evolved, contamination problems still persist, now with new aggravating factors (Ganivet 2020).

The idea of eradicating all microorganisms from the food production chain is almost impossible. And we can say that although all modern techniques help to eliminate pathogens, contamination problems still persist. So, how can we adapt to live with microbes? In fact, microorganisms have been on planet Earth longer than any other living organism, and adapt to survive in the most variable conditions, whether adverse or not. During the entire period of evolution of microorganisms, there was no major change in their primordial states, except for several molecular adaptations. Molecular adaptations are the great advance of microorganisms and perhaps their greatest triumph in the evolutionary chain (Chatterjee and Abraham 2018).

Thus, microorganisms can survive, reproduce, and continue to evolve, adapting their molecular arsenal and conquering new territories. Therefore, it is not easy to eliminate pathogens from the entire food production chain, considering all the evolutionary force (molecular evolution) present in microorganisms. Therefore, we have to think about new food preservation strategies, considering deeper aspects, such as evolution, adaptation, and artificial selection (Newell et al. 2010).

Given this scenario, we realize that microbial contamination has been relentless, especially in modern times. Today we realize that although the academic community has made great efforts to develop technologies to eliminate pathogens, we are losing the war with our weapons. Deeper aspects of microorganisms were not considered, that is, their ability to adapt and evolve. Since the past 50 years, our modern society has selected microorganisms in almost all industrial sectors through artificial selection. Thus, several resistant and better-adapted microorganisms have been the focus of several problems for the food industry, with profound impacts on the health of the population and the financial market (Pittia and Antonello 2016). In this context, this chapter brings to light the reader relevant scientific information on the degradation of food products by microorganisms, the negative effects and forms of preservation, especially new technologies, with a focus on sustainable industry and with high standards of food safety.

6.2 Contamination and Microbial Degradation of Food Products

Food contamination and degradation refer to the presence of pathogenic and non-pathogenic microorganisms that can cause changes in the product, such as odor, taste, texture, and appearance during the proliferation of microorganisms due to their natural metabolic activity. In addition to sensory changes, some microbial agents can produce harmful toxins and chemicals that shorten the shelf life of food and can cause various diseases in humans (Lianou et al. 2016).

Contamination can occur at any production stage, from harvesting or slaughtering to processing, transportation, and storage, due to surfaces that come into contact with food. in addition to factors such as the quality of raw materials and inputs; incorrect cleaning of equipment, utensils, and installations, inadequate packaging of waste and by-products, inadequate handling practices, temperature variations, among others (Singh et al. 2018).

The level of quality and safety of food is based on microbiological assessment since most foods represent a rich source of nutrients for microbial development. The increase in population and, consequently, the increase in food consumption, and changes in the eating habits of the population, such as:

1. Consumption of raw, lightly cooked, or exotic foods
2. Higher consumer demand for food products of superior sensory quality
3. Increase in functional, nutritional properties, and demand for organic products with less pesticides and additives, helped a modern revolution in the food industry.

These examples make the industry seek to develop different technologies and treatments applied in all stages of food processing. How to limit the growth rate in the exponential phase, reducing the maximum microbial population density, and thus maintaining the quality of the final product, the nutritional value, and providing consumers with a healthy and safe product (Havelaar et al. 2010; Jacxsens et al. 2010; Chatterjee and Abraham 2018).

6.2.1 Major Food Pathogens

Each food has a microbial profile that depends on the flora, the source of the contamination, the physical and chemical factors intrinsic to the food (composition, pH, water activity, presence of natural antimicrobial compounds, etc.), and environmental factors such as temperature (Baron and Gautier 2016).

Microorganisms can be classified as deteriorating and pathogenic. The so-called deteriorants cause changes in the food product, which makes it unacceptable for consumption from a sensory point of view. While pathogens are those that can cause a range of diseases in the consumer, such as botulism, food poisoning, and other enteric infections by the production of harmful toxic metabolites. These microorganisms can be bacteria, fungi, yeasts, and viruses (Wirtanen and Salo 2007). Table 6.1 shows some examples of bacteria and fungi responsible for food contamination, changes suffered by food, and methods of inhibition.

Table 6.1 Examples of bacteria and fungi responsible for food contamination, their possible changes, and methods of inhibition. Summary based on studies of Newell et al. (2010) and Faille et al. (2018)

Name	Affected foods	Changes in food	Inhibition methods
Bacteria			
Clostridium botulinum	Meat	Acidification, flavor change, protein breakdown, oxidative rancidity of fats	Storage under refrigeration; thermal treatments
Staphylococcus aureus	Raw milk and dairy products in general		Storage under refrigeration; thermal treatments
Salmonella spp.	Eggs	Yolk blackening; unpleasant odor, changes in skin pigmentation	Eggshell protection (cuticle, internal membranes, and pores); inner protection (lysozymes, conalbumin, and avidin)
Escherischia coli	Meat and animal products	PH increase; production of volatile compost with a strange taste; pigmentation change	Storage under refrigeration; thermal treatments
Fungi			
Aspergillus spp.	Raw milk and dairy in general	Proteolysis and lipolysis; increase in pH; color change; production of volatile compounds with an unpleasant odor, production of toxin	Thermal treatments
Penicillium spp.	Cereals	Toxin production and formation of dark spots	Heat treatments and drying

6.2.2 Contamination and Degradation of Food Products by Bacteria

Microorganisms that degrade food are responsible for most foodborne diseases and have been the most investigated cause of diseases related to intestinal infection, reinforcing the resistance of these pathogens, despite the prevention and control techniques applied in the industry (Newell et al. 2010). In general, bacteria need water to thrive, survive in the presence or absence of oxygen, classified as aerobic and anaerobic, respectively, or both (optional anaerobic). Their shapes are bacilli, coconuts and others, like spirochetes. Prefer protein-rich environments and can produce toxins (Lopez et al. 2018).

Bacteria can grow and multiply from the nutrients present in food, causing undesirable changes that vary depending on the type of bacteria, in addition to factors such as pH, osmolarity, temperature, and oxidation, as well as processing methods, which also influence bacterial development (Chatterjee and Abraham 2018). For example, stainless steel surfaces, floors, conveyor belts, or equipment, have been reported with the presence of bacteria (Faille et al. 2018).

Bacteria can evolve, adapt, and become resistant. In addition, a microbial community formed by bacteria can adhere to a surface, distributing its growth to form a solid matrix (biofilm). Microbial biofilms are a major problem for the food industry, as they are the focus of widespread contamination. Therefore, techniques and treatments to prevent biofilm formation must be effective in inhibiting them and, thus, preventing the spread of more bacteria to other environments (Van Houdt and Michiels 2010; Chandki et al. 2011).

Modern and advanced techniques have been developed and applied in the industry for the preservation and safety of food (Alvarez-Ordóñez et al. 2015). However, not every product can be subjected to heat treatments, as they can degrade nutrients or cause unwanted changes, requiring more sophisticated technologies to overcome bacterial resistance in the food industry, such as microwave pasteurization, ultrasound, plasma gas, ultraviolet light technology, and others (Galis et al. 2013; Motarjemi et al. 2014).

6.2.3 Contamination and Degradation of Food Products by Fungi

Fungi grow in a wide range of pH, temperatures, and water activity and use various types of substrates such as lipids, proteins, and carbohydrates. They are divided into molds and yeasts and are capable of deteriorating foods with intermediate humidity, bakery products, cherries, drinks, and fermented products causing economic losses (Chalupová et al. 2014).

The presence of fungi is related to the production of harmful toxins and microtoxins in foods, making them of low quality (Njobeh et al. 2009). Toxins are secondary metabolites produced by some types of fungi (Terzi et al. 2014) detected in several products (Njobeh et al. 2010), aflatoxins being considered the most important due to their significant negative impact on health and trade. These

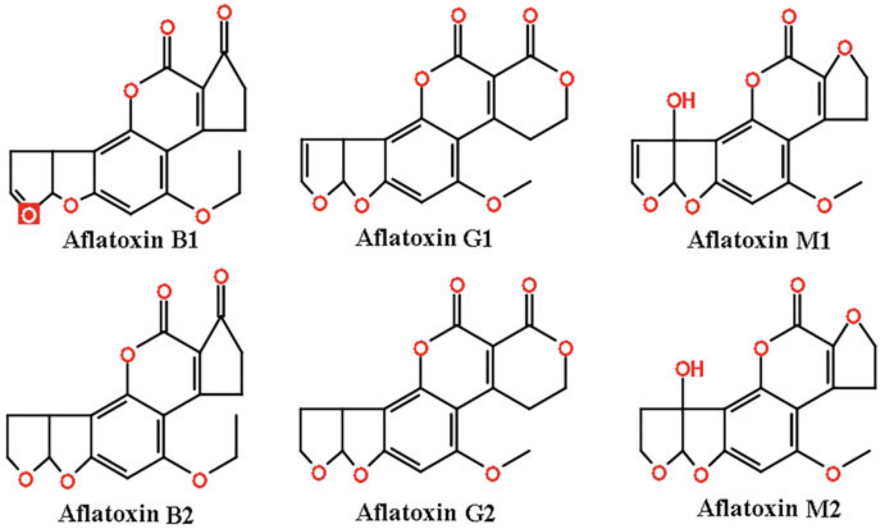

Fig. 6.1 Main aflatoxins produced by fungi of the genus *Aspergillus* spp. Source: the authors

products resulting from the secondary metabolism of fungi may vary according to the substrate and environmental conditions (pH, temperature, humidity, and the presence or absence of oxygen) (Rocha et al. 2014; Yehia 2014).

The identification of fungi that spoil food is an important step in managing food safety and quality. Prevention or decontamination methods can involve removing contaminated products, inactivating or reducing the level of toxins in food, and can be done by physical, chemical, and biological means, depending on the type of product and the toxin (Halasz et al. 2009).

The toxins produced by fungi (also known as mycotoxins) are a mixture of highly toxic secondary metabolites. There are numerous known mycotoxins and they are all easily produced in natural environments. Fungi like *Aspergillus* spp., *Fusarium* spp., and *Penicillium* spp., are known to easily contaminate fruits, processed foods, and vegetables, producing highly toxic toxins. Three groups of mycotoxins are currently recognized, namely aflatoxins, ochratoxin, and fumonisins, both of which will be addressed below.

Aflatoxin is produced by fungi of the genus Aspergillus spp., mainly in foods rich in carbohydrates such as bakery products, peanuts, and industrialized products in general, such as almonds, cheese, fluid beds, among others. Aflatoxin is highly toxic and has mutagenic and carcinogenic properties.

Fungi of the genus *Aspergillus* spp. produce four types of aflatoxin, known as B1 (AFB1), aflatoxin B2 (AFB2), aflatoxin G1 (AFG1), and aflatoxin G2 (AFG2). In addition to these, there is a group of aflatoxins known as M (AFM1 and AFM2), which are highly toxic and have direct effects on cell dysregulation, resulting in programmed cell death (Terzi et al. 2014). The main aflotoxins produced by fungi of the genus *Aspergillus spp* can be seen in (Fig. 6.1).

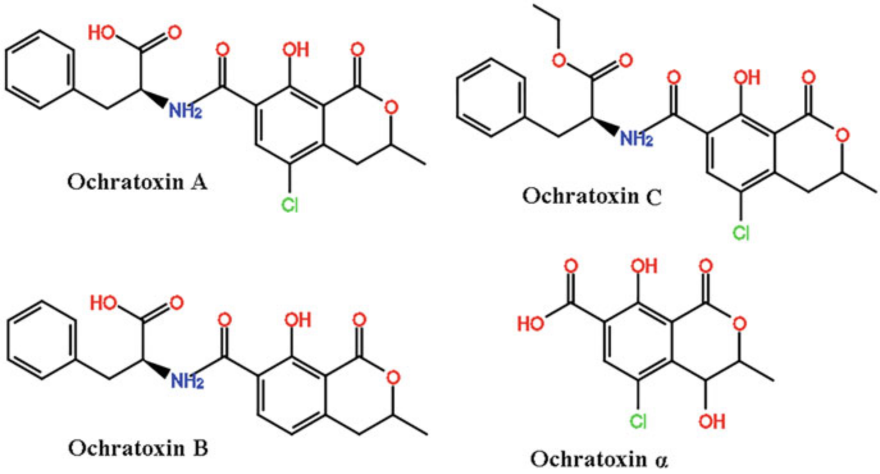

Fig. 6.2 Examples of Ochratoxin produced by fungi. Source: the authors

Ochratoxin is a secondary metabolite produced by a variety of species of fungi; especially those of the genus *Penicillium* spp. and *Aspergillus spp*. *Aspergillus* are the most common, among them *A. carbonarius*, *A. melleus*, *A. ochraceus, A. sclerotiorum*, and *A. sulphureus*. Among *Penicillium* spp., we have two relevant species, namely, *P. verrucosum* and *P. nordicum*. Ochratoxin is found mainly in cereals, dry beans, wine, coffee, and other food products. Even in small amounts, ochratoxin is highly toxic and is associated with endemic nephropathy in Balças, a degenerative disease of kidney and liver functions, leading to organ failure and death (Ayofemi Olalekan Adeyeye 2020). The main ochratoxins produced by fungi can be seen in (Fig. 6.2).

Finally, fumonisins are mycotoxins produced by fungi of the genus *Fusarium spp*, and like the other mycotoxins already reported, they also have harmful effects if ingested. The main fungi that synthesize fumonisins are *F. oxysporium* and *F. verticillioides*. These mycotoxins are found mainly in foods such as corn and are a major cause of production losses if fungi are not controlled. The concentration of these mycotoxins depends on several factors, however, studies with corn contaminated by the fungus *F. oxysporium* showed that fumonisin B1 is found in a higher concentration than fumonisin B2. The consumption of these toxins can lead to serious effects, especially on organs such as the lung and liver, and even more severe effects such as cerebral necrosis (Ayofemi Olalekan Adeyeye 2020). The main fumonisin produced by fungi can be seen in (Fig. 6.3).

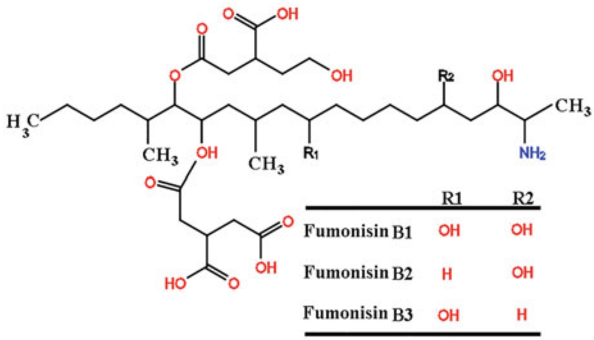

Fig. 6.3 Examples of Fumonisins produced by fungi. Source: the authors

6.3 Major Problems Associated with Contamination and Spoilage of Food

Contamination and spoilage of food are problems that can cause very serious damage to human health, and in some situations it can be irreversible, leading to death. For that reason, one of the main purposes of the food industry is to produce foods free of any contamination, ensuring the food safety of the product and its consumers. Problems in the supply and processing chain usually change the environmental conditions of food and promote the growth of microbial agents, instigating contamination and spoilage. The contamination and spoilage of food can be caused by three main agents physical, chemical, and biological. In this chapter, we will only deal with the main problems related to the contamination and spoilage caused by biological agents, which cover most of the reported problems of food contamination.

6.3.1 Problems Related to Meats

Meat is known as one of the most perishable foods due to its chemical composition that allows microbial growth at high levels, contributing significantly to the deterioration or deterioration of meat (Doulgeraki et al. 2012). Various problems related to contamination and deterioration of meat is reported by the scientific food community. Most of these problems are linked to the observed effects, which are quite variable, including discoloration, slime formation (visible growth), development of strange odors and flavors, and changes in texture (degradation), among others (Nychas et al. 2008).

The color of the meat is one of the main factors that consumers take into account when purchasing the product and the change in the natural color of the meat is always associated with problems about its quality. Meat discoloration occurs because the microbial agents are capable to destroy meat myoglobin causing the greening/graying of meat by combining with hydrogen sulfide of microbial origin. In addition, these agents can be broken down to create green or yellow bile pigments by microbial hydrogen peroxide (Pellissery et al. 2020). *L. sakei, Hafnia alvei, S. putrefaciens,* and lactic acid bacterias (LAB) are the main responsible for the mentioned types of coloring (Dušková et al. 2013). The production of microbial pigment is also visually perceptible on meats where Pseudomonads, molds of the genera *Cladosporium, Sporotrichium, and Penicillium* produce a range of black, white, blue, green, and yellow pigments (Pellissery et al. 2020). In some situations, the yellow fluorescent pigment is associated with the action of *P. fluorescens* (Cornelis 2010). In addition to the changes in the food visual aspect, the consumption of contaminated and spoiled meats can cause the development of several foodborne diseases.

Gas production is another problem caused by microorganisms that can affect meat quality. Unwanted gas formation generally occurs in vacuum-packed meat. Hydrogen and carbon dioxide are produced and released by the action of anaerobic bacteria, causing problems such as changes in pH, putrid odors, exudate formation, and color changes (Iulietto et al. 2015). Similarly, ropy slime appearance on the surface of meat due to the presence of *Lactobacillus spp*. and *Leuconostoc spp*. is another visual problem that has a negative impact on consumer purchase decision (Iulietto et al. 2015).

In the same way, off-odors and off-flavors are generally noticeable in meats before any other signs of deterioration. Volatile acids such as formic, butyric, and propionic are responsible for the acidic odor of microbially spoiled meat (Pellissery et al. 2020). Usually, off-odors are perceived when the bacterial population reaches 10^7 CFU/g (Casaburi et al. 2015). In addition, when the meat is stored at temperatures close to freezing, occurs the formation of a cotton bud mycelium, without sporulation, of white color, called whiskers. The main agents of this deterioration are *Pseudomonas, Moraxella, Alcaligens, Aeromonas, Serratia, Pantoea Thamnidium, Nucor*, and *Rhizopu*s species (Nychas et al. 2008).

6.3.2 Problems Related to Milk and Dairy Products

Milk and dairy products can be contaminated by microorganisms at any stage of the processing chain, causing problems to these products. The main forms of contamination that cause problems to milk and dairy products are the inadequate cleaning of milking equipment, cross-contamination by handling, animal feces, oscillation in temperature of refrigeration tank, inadequate pasteurization, and storage (Lopez et al. 2018; Beletsiotis et al. 2011). There are a large number of infectious diseases that can be transmitted to humans through milk. The most important pathogens are *Salmonella sp., Pathogenic Escherichia coli, Listeria monocytogenes, Campylobacter jejuni, Yersinia enterocolitica*, and *Staphylococcus aureus*, which can cause several fatal diseases (Iqbal et al. 2016). However, most of these pathogenic microorganisms are destroyed by pasteurization. That is why the thermal process should be well executed.

Furthermore, under refrigeration temperatures, some psychrotrophic bacteria present a higher growth rate, producing lipolytic and proteolytic enzymes, which cause problems on the quality of milk and dairy products due to their ability to resist thermal treatments (Nsofor and Frank 2013). Milk and dairy products are also susceptible to contamination by yeasts and molds that can cause changes in the color, texture, and odor of these products. The main microorganisms responsible for these contaminations are *Candida, Galactomyces*, and *Yarrowia* for yeasts and *Penicillium, Mucor*, and *Cladosporium* for molds (Garnier et al. 2017)

6.3.3 Problems Related to Eggs

Eggs are widely consumed worldwide because it is considered a low-cost food and partially meets the nutritional requirements of a person. However, some problems with the contamination and spoilage of this food can cause damage to consumer. The main form of contamination occurs through the eggshell when the microorganisms penetrate through its membranes and access the egg content, resulting in deterioration (Whiley and Ross 2015). Bacteria, yeasts, and molds can be the cause of undesirable alterations such as the release of gas, formation of visible colonies, changes in the coloration, texture, and odor of the eggs (Techer et al. 2014). *Salmonella Enteritidis*, which do not changes the color, smell, and consistency of the egg content, is a pathogenic bacterium that is considered the main factor for foodborne illnesses in developed countries, and it is important to highlight that eggs are the most common food that causes these infections (Jan et al. 2018).

6.3.4 Problems Related to Vegetables and Fruits

Fruits and vegetables are perishable foods due to their high water content (approximately 95%), which can facilitate the growth of both pathogenic and spoilage microorganisms, therefore these foods are more susceptible to contamination (Yousuf et al. 2020). The contamination and spoilage of fruits and vegetables can occur during the growing, harvest, and processing steps. In fruits with low pH, especially citrus fruits, the bacteria do not grow and fail to develop, since the pH changes the metabolism. However, molds and yeast are able to grow on these foods (Carlin 2013). For instance, Basidiomycetous can cause problems such as dark spots, dark lesions, gangrene, sour and soft roots, as well as it can produce cutinase that is related to the cuticle degradation of various fresh vegetal products (Carlin 2013). Another problem that affects some types of fruits and vegetables is the contamination by pathogenic microorganisms, mainly related to ready-to-eat products, which are responsible for several outbreaks caused by *Salmonella* ssp. and *E. Coli*. O157: H7. Infection by these microorganisms can cause serious damage to human health such as gastroenteritis (Amrutha et al. 2017).

6.3.5 Problems Related to Canned Foods

The main problem caused by the consumption of canned foods is botulism. Botulism is a rare and serious bacterial disease caused by the ingestion of toxins from *Clostridium botulinum*, which is considered the most potent among the known toxins (Momose et al. 2014). Botulism is characterized as an extremely serious disease, with acute evolution, causes digestive and neurological disorders, as a result of the ingestion of several types of food, including canned foods (Rhodehamel et al. 1992) As *Clostridium botulinum* is a sporogenic bacterium, the spores can remain in the food in case of inadequate sterilization in the heat treatment stage, and start to multiply and produce toxins (Featherstone 2015). Therefore, all low-acid canned foods (pH $>$ 4.5) that are contained in packages completely free of oxygen, such as canned products, are potentially botulogenic and attention needs to be doubled in order to avoid serious health problems (Peck et al. 2020)

6.4 New Technologies to Reduce the Rate of Contamination and Degradation of Food Products

The quest for food preservation dates back to prehistoric times through the empirical use of salt, sugar, drying, and smoking. In some places where there were caves, snow and ice were used as a cooling resource. Later, ice was first transported in 1799, and in 1877 to Linde, the first industrial compact refrigerator process. Nicholas Appert, in 1810, took a big step in preserving food by adding heat, developing the tightening process that gave rise to canning and heat sterilization. Over the years, new technologies have been and are being developed in response to historical events such as the Second World War and space exploration, and to the intense population growth that went from 2.5 to 8 billion between 1950 and 2020. In this way, the objective of modern studies is to increase the shelf life of food, safety in consumption without causing damage to health, and the maintenance of sensory qualities and the chemical composition of macro and micronutrients. Next, we will discuss some of these emerging technologies (Ganivet 2020; Jiang et al. 2019; Misra et al. 2017; Pittia and Antonello 2016).

Cold plasma (CP) is a nonthermal technique that uses as a principle the active species contained in ionized gas such as electrons, free radicals, ions, and so on, leading to excitation, de-excitation, and ionization reactions that result in the decontamination and sterilization of food products affected by spores and products of deterioration of pathogenic organisms. It is also used in packaging processing to improve its barrier properties and add antimicrobial activity. The technique can be used by direct CP contact; indirect CP applying plasma-activated water during washes, sprays, or mists; and package CP discharge (Bourke et al. 2018; Ekezie et al. 2017; Fang et al. 2017).

Bosch et al. (2017) studied the effectiveness of using cold plasma (CP) at atmospheric pressure with ambient air in the degradation of mycotoxins produced by species of *Fusarium* spp., *Aspergillus* spp., and *Alternaria alternata*. The study showed the effectiveness of the method in the degradation of pure mycotoxins, being considered a promising method in the decontamination of food products affected internally or externally as on the surface of cereal grain. Connolly et al. (2013) performed the inactivation of *Escherichia coli* in plastic packaging with helium/air plasma at atmospheric pressure. The authors concluded that the technique was effective in reducing 1.5 logarithmic cycles of specific cell regulatory systems, generating the decontamination of the packaging of fresh products.

The inactivation of microorganisms through sterilization by supercritical carbon dioxide (SC-CO_2) is also considered an emerging and promising technique in the treatment of food, as it is a green and environmentally safe technique, preserving the integrity of foods, especially thermosensitive and hydrolytically sensitive. The technique is dependent on variables such as pressure, temperature, CO_2 density, treatment time, CO_2 flow rate, depressurization rate, and use of additives. The mechanism involved in inactivation has not yet been fully elucidated; however, it is believed that cell rupture may occur due to volume expansion when high-pressure CO_2 is suddenly depressurized and/or the physiological deactivation that consists of the simultaneous or isolated occurrence of seven steps:

1. It is derived from acidification of the medium due to the dissociation and formation of carbonic acid from the contact of CO_2 with the water present in the medium
2. It results from the change in the cell membrane
3. There is a decrease in intracellular pH
4. The low pH inactivates key enzymes and inhibits cell metabolism
5. There is an inhibitory effect on microbial metabolism through molecular CO_2 and HCO_3 affecting the reactions of carboxylation and decarboxylation
6. Intracellular electrolyte balance disorder occurs
7. There is the removal of the vital constituents of cells and their membranes (Ribeiro et al. 2020; Soares et al. 2019).

Checinska et al. (2011) validated the sterilization method of biological pathogens using SC-CO_2, water (3.3%), and hydrogen peroxide (0.1%), at 80 atm, 50 °C and 30 min. The authors described that the method was effective in sterilizing pathogens present in biofilm structures, fungal spores associated with nosocomial infections, and SAFR-032 endospores of *Bacillus pumilus*. Silva et al. (2013) studied the inactivation of pathogenic *E. coli* through sterilization with SC-CO_2, evaluating the effects of pressure, rate of depressurization, and pressure cycling, the authors concluded that the increase in the number of cycles and pressure of the system increased the efficiency of inactivation inferring that the process is useful in non-thermal sterilization of food. Sterilization of palm fruits with SC-CO_2 was performed by Omar et al. (2017) to inactivate microorganisms and enzymes responsible for the degradation of palm oil. The authors pointed out that the process was able to completely inactivate lipase-producing microorganisms at 10 MPa, 80 °C and 60 min, avoiding the formation of free fatty acids, moreover, with the process they managed to drastically reduce the amount of oil from the palm oil plant (POME) generated during steam sterilization.

Active packaging has been used in food preservation in order to isolate them from the external environment and protect them from the action of microbiological, chemical, and physical hazards, maintaining quality and prolonging shelf life. Active packaging interacts with food and is considered more effective compared to the simple addition of active agents on food surfaces by sprays or drips, because in these processes there is a rapid diffusion and the denaturation of the agents by the food may occur, reducing its effectiveness. Agents added to packaging must be safe and have been regulated by competent bodies such as the United States-Food and Drug Administration (U.S.FDA), Brazil National Health Surveillance Agency (ANVISA), and European Commission (Ribeiro-Santos et al. 2017; Sung et al. 2013). Studies have been developed to evaluate, mainly, the antimicrobial and antioxidant effect of active agents from plant extracts as an alternative to the synthetic ones, commonly used.

Ramos et al. (2012) evaluated the antimicrobial activity of polypropylene films incorporated with thymol and carvacrol. The authors pointed out that the thymol impregnated film showed greater inhibition against bacterial strains of *S. aureus* and *E. coli* compared to carvacrol, the compounds also had the potential to be used as agents antioxidants. Peng et al. (2013) investigated the addition of green tea and black tea extracts to chitosan films in the preparation of active packaging film. According to the authors, the addition of the extracts resulted in a decrease in water vapor permeability and increased the antioxidant capacity of the films due to the significant improvement in DPPH radical scavenging activity. Albuquerque et al. (2020) impregnated *Piper divaricatum* essential oil with supercritical CO_2 in fish (*Cynoscion acoupa*) skin gelatin films. The authors concluded that the addition of the oil resulted in 41.63% of antioxidant activity index and the film presented greater flexibility and opacity when compared to the control film.

Nanotechnology is a resource that has been used in food science and technology to improve production and application in the areas of processing, packaging, storage, transportation, functionality, and safety. Nanomaterials vary from 1 to 100 nm and are appreciated due to their properties that allow to increase solubility, bioavailability, and protection during the processing and storage of active compounds that are generally chemically unstable, thermosensitive, and photosensitive. Several antimicrobial agents are used in nanoencapsulation such as alkaloids, antioxidants, phytochemicals, essential oils, plant extracts, and so on, reducing microbial contamination and adding functional and sensory properties to foods (Bajpai et al. 2018; Prakash et al. 2018).

Mohammadi et al. (2016) evaluated the antibacterial activity of microparticles prepared with three different molecular weights and nanoparticles of chitosan crab shells against *P. fluorescens*, *E. carotovora*, and *E. coli*. The authors found from the results that there was a positive correlation between the particle size and the molecular weight of chitosan, the maximum antibacterial activity was verified in the nanoparticles and the microparticles, the antibacterial activity was dependent on the application. In the study by Mohanta et al. (2017), the biosynthesis of silver nanoparticles with the extract of leaves of the *Protium serratum* was performed. The nanoparticles showed antibacterial activity against *P. aeruginosa*, Escherichia coli, and *B. subtilis*. As well as antioxidant activity to DPPH and hydroxyl radicals, and biocompatibility to the L-929 fibroblast cell line, not being harmful to the human body. In addition to oral administration being effective against gastrointestinal diseases and stomach ulcers, the study highlights the potential application in the food industry.

6.5 Conclusions

The degradation of food products by microorganisms is almost inevitable, because, like everything in nature, the cycle of the food chain is relentless. However, although the existence of microorganisms predates any other living thing on earth, the evolutionary process has improved these living organisms, especially at the molecular level, with incredible adaptations. The survival of microorganisms in specific conditions and their evolution are the key to a deep understanding of how to intervene in their development, growth, and reproduction and, thus, increase food preservation.

Our modern civilization has had great economic losses due to contamination and subsequent degradation of food. However, modern studies have helped to understand how new technologies can be applied to increase the shelf life of food, or even to reduce contamination rates in products still in the field.

One of the main problems of food contamination by microorganisms is directly related to public health. Foods contaminated with fungi produce mycotoxins, which even in small amounts can be fatal. Therefore, the issue of food security persists as a current issue and one that deserves constant efforts to reduce impacts on human health. Thus, we conclude that in-depth studies are still needed to understand relevant aspects about the evolutionary biology of the main food pathogens and more efforts in the development of green technologies to reduce the contamination rate and increase the shelf life of food.

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