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Risk Factors Influencing Microbial Contamination in Food Service Centers

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Abstract

An improvement of food service centers in recent years has been made based on the implementation of the principles of the Hazard Analysis and Critical Control Points (HACCP) system. Food safety preventive measures have been focused on training of handlers in hygiene practices and on improving the sanitary quality of meals. In Europe, an increasing trend in foodborne outbreaks has been attributed to catering businesses. This fact highlights that the impact of preventive measures in the past few years has not been sufficiently effective as expected. Special attention should be paid to food services destined to susceptible population, such as hospitals, long-term care facilities, or school canteens, because people could be more susceptible to become ill when exposed to foodborne agents. There are numerous relevant factors influencing microbial contamination of foods, according to the preparation method, hygienic sanitary conditions of catering facilities, or food handling, storage, and distribution. In the present chapter, a review of the most significant risk factors influencing microbial contamination of foods in food service centers are described with special focus on those establishments where susceptible population (i.e., children, elderly, immunocompromised people) is present. Besides, potential preventive measures to be considered in that establishments and correct implementation of food safety actions are given to provide useful recommendations to food handlers, food operators, and risk managers.

Keywords: Food safety, catering establishments, risk factors, handlers, microbial indicators

1. Introduction

Some individuals tend to eat out of the home, often at food service operations, such as cafeterias, canteens, fast food outlets, bars, and restaurants [1]. In recent years, the catering



sector has been experiencing an increase in technological innovation in correspondence with changes in consumer habits of the population, transformed by numerous factors and changing lifestyles, demographic trends, and so on, which have increased consumer preferences for healthy, safe, and convenient foods.

Food service or catering industry defines those businesses, institutions, and companies responsible for any meal prepared outside home. These industries include restaurants, school and hospital cafeterias, catering operations, and some other small- and big-scale establishments. A catering food establishment also means an approved food establishment that is serving or preparing food at a location other than its permitted location for a contracted food service event. During these operations, foods are often transported, distributed, handled, and consumed in a short-time framework (often less than 1 week).

The catering sector can be divided into three groups based on the population they serve, the way of working, the technology used, or the food types served (Table 1): commercial (residential and non-residential) catering sector, and non-commercial residential establishments.

Residential	Hotels, guest houses, holiday parks,		
	farmhouses, public houses, bed, and		
	breakfasts		
Non-residential	Restaurants, cafes, fast-food outlets, wine		
	bars, delicatessen and salads, bars, take-		
	away outlets, schools catering, and burger		
	vans		
Non-commercial residential establishments	Hospitals, residential homes, prisons, and		
	armed services		

Table 1. Types of catering establishments

The Hazard Analysis and Critical Control Points (HACCPs) system has been recognized as a useful tool to prevent food contamination in food service establishments. Control measures are mainly focused on training of food handlers in hygiene practices to improve safety of served meals.

Unquestionably, consumption outside home is linked to a fact of modern life that causes some concern among health professionals and food authorities because of the potential adverse effects that may entail for human health. Food poisoning poses a serious problem for public health worldwide, and the most vulnerable population (children, elderly, pregnant, and sick) counts among the most affected.

Data from the World Health Organization assert that about 2.2 million people become daily ill in the world for more than 200 foodborne diseases and about two-thirds of the outbreaks that occur originate in their homes and in catering establishments. Given the higher volume of meals prepared in the later ones together with meals exposure to handling and environmental factor, the risk of becoming ill is expected to be considerably higher [2]. In Europe, an

increasing trend in foodborne diseases outbreaks has been attributed to catering businesses [3]. This fact highlights that the impact of prevention measures in the past few years has not been sufficiently effective as expected. Important aspects such as the size of establishments and heterogeneity of foods served justify the creation of specific regulations on food safety management, to reduce the risks of foodborne illness.

Moreover, it seems to be necessary to strengthen food hygiene and compliance with HACCP system to prevent food outbreaks. These outbreaks are mainly caused by foodborne pathogens and norovirus. Special attention should be paid to food services destined to susceptible population, such as hospitals, long-term care facilities, or school canteens, because people could be more susceptible to become ill when exposed to foodborne agents. Besides this, the large number of meals served per day could have an impact on public health if prepared foods are contaminated.

In food service environments, various factors may be related to foodborne diseases. According to the Food and Drug Administration (FDA), these factors are the food served coming from unsafe sources, poor personal hygiene, inadequate cooking, improper holding temperatures, or utilization of contaminated equipment. On the other hand, the World Health Organization (WHO) [4] suggested that drinking water could be an important factor related to foodborne diseases. It was already demonstrated that training of food handlers and knowledge acquisition in hygienic food preparation, processing, and distribution of meals is crucial in the prevention of most types of foodborne diseases [5]. The use of normalized questionnaires on good manufacturing practices (GMPs) was achieved to evaluate training of food handlers. It was found that, after implemented, there is a substantial improvement in GMP [6–8]. However, it is recognized that these training activities should be repeated over time to minimize the reluctance of food handlers to apply the acquired knowledge [9].

Legislation in food hygiene at EU level prioritizes control measures to protect public health, making food operators responsible to assure product safety [10]. Implementation of HACCP system in food service operations may increase food safety management. However, given the complexity of the food chain and variety of menus and meals prepared, simplified and flexible self-control measures must be required in most cases to increase efficiency and homogeneity of implemented food safety management systems [7]. Evaluation of microbial risks is crucial to determine food safety of prepared meals [11].

In the present chapter, a review of the most significant risk factors influencing microbial contamination of foods in food service centers will be described with special focus on those establishments where susceptible population (i.e., children, elderly, immunocompromised people) is present. Besides, potential preventive measures to be considered in that establishments and correct implementation of food safety actions will be given to provide useful recommendations to food handlers, food operators, and risk managers.

2. Factors enhancing microbial foodborne outbreaks worldwide

The increase in the global burden of foodborne diseases constitutes a concern to governments and food operators today than a few years ago. Identification of emerging pathogens (or

environmentally adapted) causing life-threatening conditions, introduction of ethnic foods, environmental changes, food security aspects, migrant populations, the ease of worldwide shipment of fresh and frozen food, and the development of new food industries including aquaculture are some of the reasons for this concern. Over the past years, foodborne diseases caused by bacteria, viruses, parasites, and prions have been prioritized by governments of industrialized countries generating substantial media attention.

However, to monitor foodborne diseases, an effective surveillance system at the local, national, and international levels should be implemented. The appearance of multistate outbreaks (i.e., contamination in a commercial product occurring in one country and affecting persons in several other countries, or tourists being infected abroad and possibly transmitting the pathogen to others at home) contributes to the increase in the number of illnesses at international level. Currently, funding sources are not easily available for non-developed countries and regions where most of the information comes from passive reporting mechanisms [12]. In the industrialized countries, a need for funding sources allocated to the improvement of foodborne disease surveillance, and control is required.

Several factors (such as environmental, socioeconomic, chemical, physical, and biological) are influencing on such public concern.

Environmental factors such as weather influence the transport and dissemination of microbial hazards via rainfall and runoff and their survival and/or growth through according to temperature or humidity conditions [13]. It is shown that the increase in climate variability influences on current and future deficiencies in areas, such as watershed protection, infrastructure, and storm drainage systems, thus enhancing the risk of food contamination events. More knowledge is needed about transport processes and fate of microbial hazards to predict risks associated to weather variability. In this sense, application of existing technologies such as molecular fingerprinting to track contaminant sources could be expanded.

On the other hand, despite of the development of novel traceability systems and inspection controls, infectious diseases still remain a leading cause of global disease burden with high morbidity and mortality in non-developed countries. The emerging and re-emerging diseases have been a big impact at socioeconomic and public health levels. Their control requires continuing surveillance, research and training, better diagnostic facilities, and improved public health system. Food safety is of particular concern in a developing country context given the higher incidence of foodborne illnesses and their associated economic and social cost [14]. Furthermore, the economic and social changes associated with development (i.e., urbanization, changes in food production systems, and consumption patterns) could increase the risk of emerging foodborne illnesses. This recognizes the connections between disease and socioeconomic factors such as poverty and malnutrition and the wider economic, social, physical, and cultural environment in which people live [15]. Promotion of trade together with improvement of agricultural and animal practices at primary production will definitely help developing countries to better manage food safety. Improving food safety in international trade would also require numerous policy and technical interventions that include an effective market access; a better analysis of the costs and benefits of global trade rules for developing countries or integration of developing countries into the global economy [16].

Besides environmental and socioeconomic factors, according to the Center for Disease Control and Prevention (CDC), the absence of control, a key elementary factor along the production chain, contributes to the extension of foodborne outbreaks, some of them affecting to food service centers:

- Producing and harvesting food: lack of quality assurance programs and unsafe agricultural practices.
- · Processing foods: absence or undefined inspection systems at industrial facilities; poor processing preservation technologies (i.e., thermal treatments, addition of preservatives, and unsafe food formulations).
- Distributing and preparing foods: absence of food purchasing specifications, untrained food handlers, improper hand-washing procedures and facilities, and lack of food safety education programs for consumers.

3. Residential catering

Institutional food establishments have a key role to public health. Consumers that are generally more likely to suffer from foodborne diseases occupy them, such as children, elderly, sick, or immunocompromised individuals. Their physiological characteristics often require high degree of food safety in the meals served.

According to the European Federation of Contract Catering Organizations (FERCO), approximately 33% of firms or collective organizations currently have a contract with a Contract Catering company. Indeed, this is a sector that represents an alternative to meet the basic food needs of a group of very important people. Among institutional catering companies stand Eurest SA authorities; Serunión S.A; Sodexho S.A. Spain; and Aramark catering services, SL, among others, that allocate their production mainly to hospitals, nursing geriatric, dining, and study centers.

The fact of preparing and serving large volumes of food in a relatively short-time framework involves the use of new technologies for conservation and/or optimization of existing technologies, to ensure the hygienic and sanitary quality and shelf life of food.

Studies related to systems development, maintenance, and transportation of prepared meals cite the refrigerated cold chain, frozen cold chain, hot chain, or vacuum cooking [17]:

- In the refrigerated cold chain, food is abated once drawn up, from 65°C to 10°C in a time not exceeding 90 min, before being stored at 0-3°C. Afterward, food is regenerated until reaching the appropriate temperature before serving.
- In the frozen cold chain, food is placed in a blast chiller given the same procedures above and subsequent cold storage at -18° C until consumption, at which time must be regenerated to its proper serving temperature (65°C).

- In the hot chain, foods are maintained at temperatures above 65°C and should be consumed within a maximum of 12 h.
- In the vacuum cooking, the food is prepared at low temperatures and for a long time. The food is then packaged in a waterproof material, which is not affected by high temperatures. Before cooking, air is extracted and sealed. After cooking, the product is cooled quickly and is cooled, and then regenerated (65°C) before consumption.

Utilization of more traditional food preservation techniques is also common [17] such as industrial pasteurization or chemical preservation (salting, smoking, marinating, and pickling).

Lastly, other preservation technologies are used to reduce the amount of food handling in the kitchen, such as high hydrostatic pressure, the light pulses, dehydration, irradiation, and modified atmosphere preservation, among others [17–19].

4. Health and socioeconomic implications of microbial contamination and its effects on international trade

Provision of safe foods supports national economies, international trade, and consumer confidence, thus underpinning sustainable development. However, globalization and changes in consumers' habits to a more convenient and healthy foods led to increase the awareness of potential and/or emerging hazards for public health. This also triggered a growing consumer demand for a wider variety of foods, thus leading to a more complex food chain.

The consequence of the population growth is an intensification of agriculture and animal production to meet consumer demands. Food safety challenges should consider the potential effect of climate change because temperature changes can modify the risk profile of a given food commodity during the whole production chain [20]. These effects produce a greater degree of responsibility to food producers and handlers to ensure food safety. It should be highlighted that the spread of a localized outbreak can increase largely due to the globalized food chain and international trade. Examples include the contamination of infant formula with melamine in 2008, and the 2011 Enterohemorrhagic *Escherichia coli* outbreak in Germany linked to contaminated sprouts. This multistate outbreak was reported in eight countries in Europe and North America, leading to 53 deaths. Losses caused to farmers and food industries were quantified in US\$ 1.3 billion, while emergency aid payments to 22 EU member states were around US\$ 236 million [21].

According to data reported by USDA, foodborne illnesses are annually costing the economy more than \$15.6 billion. Each year, more than 8.9 million Americans will be sickened by one of the 15 pathogens, with more than 5.4 million of those illnesses due to the stomach churning, but usually short-lived, Norovirus. In EU countries, 5196 foodborne outbreaks, such as waterborne outbreaks, were reported [22]. Governmental authorities should make food safety a public health priority, as they play key role in developing policies and regulatory frameworks. They also are in charge of establishing and implementing effective food safety systems

that ensure that food producers and suppliers along the whole food chain operate responsibly and supply safe food to consumers.

Among the food commodities causing outbreaks, meat products are the most frequently reported, given the high consumption associated to these products. Vegetable salads are recognized as potential vehicles for enteric pathogens, as they are not subjected to any heat treatment before consumption. In Table 2, notified outbreaks in EU and the United States for produce commodities are reported. Scientific studies highlight the importance of an adequate training of food handlers and implementation of GMPs when elaborating vegetable salads in catering establishments.

Regarding cooked meat products, they are susceptible to be contaminated after heat treatment, during storage, and distribution. This is because they can be subjected to poor handling practices (i.e., slicing, packaging) during preparation in catering establishments. As an example, in Table 3, notified outbreaks in EU countries and the United States associated to consumption of meat products are reported. It is concluded that problems in kitchen design, inadequate handling and disinfection practices, and lack of knowledge on food safety by handlers are the main risk factors influencing microbial contamination.

Year	Vehicle	Microbial hazard	Place	Country	Number of	Reference
		involved			cases	
2012	Romaine lettuce	E. coli O157:H7	Retail outlets	The United States	58	[23]
2011	Bean sprouts	E. coli O104:H4	Multiple places	Multistate EU outbreak	3910	[24]
2011	Romaine lettuce	E. coli O157:H7	Retail outlets	The United States	60	[25]
2011	Fresh basil	Shigella sonnei	Not available	Norway	46	[26]
2010	Lettuce	E. coli enterotoxigénica and norovirus	Catering establishments	Denmark	260	[27]
2007	Shredded lettuce	E. coli O157	Processing industries	The Netherlands, Iceland	50	[28]
2007	Alfalfa sprouts	Salmonella stanley	Domestic homes	Sweden	51	[29]
2006	Shredded romaine lettuce	E. coli O145	Processing industries	The United States	26	[30]
2004	Salad lettuce	Salmonella newport	-	The United Kingdom	375	[31]
1994	Iceberg lettuce	S. sonnei	Domestic homes	The United Kingdom, Norway	-	[32]

Table 2. Notified foodborne outbreaks in Europe and the United States by the consumption of produce commodities

Year	Vehicle	Microbial hazard involved	Place	Country	Number of cases	Reference
2011	Turkey meat	Salmonella Heidelberg	_	The United States	77	[33]
2009	Cured RTE meat	Salmonella montevideo	- 	The United States	272	[34]
2009	Cooked meat	E. coli O157	School canteens	Wales	150	[35]
2008	Not available	Listeria monocytogenes	Processing meat industry	Canada	22	[36]
2006	RTE pork meat	Yersinia enterocolítica O:9	ı –	Norway	11	[37]
2006	Fermented sausages	E. coli O103:H25	_	Norway	17	[38]
2005	Minced beef	E. coli O157:H7	_	France	69	[39]
2002	Turkey meat	L. monocytogenes	Processing industry	The United States	54	[40]
2001	Cooked meat	E. coli	Butchery	The United Kingdom	30	[41]
_	Minced beef	E. coli O157	_	-	732	[42]
2000	Ham	L. monocytogenes	_	New Zealand	28	[43]
2000	Turkey meat slices	L. monocytogenes	Processing industry	The United States	11	[44]

Table 3. Notified foodborne outbreaks in Europe and the United States by consumption of meat commodities

5. Microbial contaminants of prepared meals in catering establishments

During the whole production chain, there is constant exposure of food to microbial contamination. Therefore, a strict quality and safety food control should be promoted with a view to minimize the incidence of food poisoning.

Undoubtedly, for catering establishments, the HACCP system assesses the condition under which the product was elaborated, determines the main risk factors of food contamination, and manages effective measures to reduce contamination by pathogenic and spoilage microorganisms.

Microbial indicators are able to highlight deficiencies in the hygienic and sanitary food quality. Indeed, their presence at high levels leads to a reduction of shelf life and is probably related to the presence of pathogenic microorganisms.

According to the International Commission on Microbiological Specifications for Foods (ICMSF), microbial indicators do not offer a direct risk to human health. These groups are

mainly aerobic mesophilic bacteria, lactic acid bacteria, total coliforms, fecal coliforms, enterococci, enterobacteriaceae, Staphylococcus aureus and E. coli [45]. In the next subsections, the most representative microbial indicators will be described. Besides, Listeria monocytogenes is included due to its relevance and presence in a wide range of food commodities as well as for the current EU regulation (No. 1441/2007) [46] where it is included as safety criteria for ready to eat foods.

5.1. Aerobic mesophilic microorganisms

Microbial species belonging to this group are quite heterogeneous and include all bacteria, fungi, and yeasts growing at aerobic conditions. The presence of aerobic mesophilic microorganisms in fresh foods demonstrates the effectiveness of sanitary procedures during processing, handling, and storage before [47].

Ready to eat foods (apart from fermented foods, cheeses, and dairy products) with significant concentration levels of aerobic mesophilic microorganisms should not be considered suitable for human consumption, even if microbial species are not pathogenic.

Generally, contamination occurs because of the use of contaminated raw materials or inefficient health treatments as well as inadequate conditions of storage time and temperature [48].

In general, high levels pose a greater risk of pathogen contamination. Several authors agree that the recommended concentrations for ready to eat foods should be less than 5.0 log cfu/g [49]. However, other guidelines for ready to eat foods such as those proposed by the Health Protection Agency (UK) [50] establish acceptable limits between 6 and 8 log cfu/g, depending on the food type.

5.2. Lactic acid bacteria

Lactic acid bacteria comprise a wide range of microorganisms with common morphological, metabolic, and physiological characteristics. Some of the most representative species are Streptococcus spp., Pediococcus spp., Leuconostoc spp., Lactobacillus spp., and Lactococcus spp. [51].

In the food industry, they have multiple uses as starter cultures in the manufacture of cheese, yogurt, and fermented meats. They are also recognized as natural antimicrobial agents against foodborne pathogens in biopreservation processes [52]. They represent the predominant group in fermented meat products reaching levels between 8 and 9 log cfu/g during the maturation processes. The most common species are Lactobacillus sakei, Lactobacillus curvatus, Lactobacillus plantarum, Lactobacillus pentosus, Pediococcus acidilactici, and Pediococcus pentasaceus [53].

Despite its protective function, they are able to produce end metabolites that lead to food spoilage and thus shortening its shelf life. Their final levels depend largely on the storage temperature and packaging methods [54]. Deterioration caused by the growth of lactic acid bacteria is shown by undesirable changes in smell, taste, color, and gas production. Some studies have found these changes in vacuum-packed meat products or modified atmosphere products [55].

5.3. Enterobacteriaceae

Enterobacteriaceae are considered as food quality indicators including *E. coli* being mainly related to fecal contamination. Generally, the presence of these microorganisms in foods is closely linked with the implementation of inadequate handling practices, inefficient cooking processes, cross-contamination, inadequate personal hygiene of food handlers, equipment and food-contact surfaces as well as inadequate holding time and temperature conditions [56].

Enterobacteriaceae species are Gram-negative bacteria, aerobic or facultative anaerobic, non-sporulated, mobile or immobile, and being able to ferment glucose and to reduce nitrate to nitrite. Some of the most representative species include *Salmonella enterica*, *Shigella* spp., *Yersinia* spp. (intestinal pathogens in humans), *Edwarsiella* spp., *Hafnia* spp., *Proteus* spp., *Morganella* spp., *Erwinia* spp., *E. coli*, *Enterobacter* spp., *Citrobacter* spp., *Serratia* spp., or *Klebsiella* spp. Most of them produce endotoxins and thermolabile and/or thermostable exotoxins. Some *E. coli* serotypes are producing verotoxins and shigatoxins, which are linked to a high rate of morbidity and mortality in humans [57].

Food commodities where Enterobacteriaceae can be found are processed meat products [58], nutritional formulas for infants [59], mixed salads, raw vegetables, and milk/dairy products, among others [60].

5.4. Total and fecal coliforms

Total and fecal coliforms are specific groups within the Enterobacteriaceae family, including species, such as *E. coli*, *Klebsiella* spp., *Enterobacter* spp., or *Citrobacter* spp. These are Gramnegative bacteria, aerobic or facultative anaerobes, non-sporulated, whose optimal growth temperature is around 35–40°C. These food quality indicators are taking part of the intestines of humans and warm-blooded animals and other organisms often located on the ground or plant.

The main difference between total and fecal coliforms is that the latter group ferments lactose at temperatures between 44 and 45°C. The group includes primarily *E. coli* (~90%) with certain *Klebsiella* and *Citrobacter* species. Coliforms are considered a reliable indicator of fecal contamination and are sometimes found in contaminated equipment and utensils, as well as in a wide variety of foods.

Contamination of ready to eat foods by coliforms is commonly attributed to environmental contamination, the use of inadequate hygiene practices, and/or insufficient control of the storage temperature. In the case of thermally treated food, the presence of coliforms is indicative of inadequate treatment or post-processing contamination as they are thermolabile microorganisms [47].

5.5. Escherichia coli

Enteropathogenic *E. coli* comprise different serotypes that can be present in contaminated foods. Most of them are able to produce Shiga-like toxins and/or other heat-labile or heat-stable

toxins that can potentially cause diarrheagenic diseases in humans [61]. Besides, some serotypes of enterotoxigenic *E. coli* can also produce a cytotoxin to Vero cells (VTEC *E. coli*).

Normally, outbreaks caused by VTEC serotypes are of low prevalence (1.2 cases per 100,000 people in the EU) [39]; however, the high infectivity and severity of the disease increase the importance of performing novel research on this pathogen. It is reported that human outbreaks attributed to *E. coli* serotypes were mostly originated from catering services or restaurants [62].

Generally, E. coli can be present in animal origin foods (pork, beef, and poultry), water sources, or produce such as cabbage, lettuce, or spinach. They can enter the food chain through cross-contamination or recontamination phenomena [63] or through the irrigation with contaminated water, which may result in the internalization of certain E. coli serotypes in vegetables [64].

E. coli O157:H7 was the most studied serotype due to the severity of the illnesses caused and its low infective dose, around 100 cells [65]. However, other non-O157 serotypes have been associated to human infections through the ingestion of risk food products, such as fermented and minced meats or raw milk [66].

5.6. Staphylococcus aureus

S. aureus has been reported as a microbial indicator most likely associated to reduced water activity (a_w) foods, such as ready to eat cooked or cured meats [67, 68]. The presence of S. aureus is often associated to contamination of raw material, such as poultry carcasses or raw chicken samples [69] or cross-contamination events occurring because of mishandling during processing and storage [63].

Food poisoning is attributed to the ingestion of foods that contain thermotolerant Staphylococcal Enterotoxins (SEs) in doses around 20-100 ng [70]. The staphylococcal enterotoxin A (SEA) is the one most frequently reported. A wide range of environmental factors, such as pH, $a_{\rm w}$, temperature, food type, and processing conditions, have been suggested to play an important role on SEs production. Generally, growth of S. aureus is necessary for SE production, although this phenomenon does not always accompany growth [67]. Indeed, some published studies consider hazardous S. aureus levels from 6 log cfu/g in contaminated foods for SE production [71].

5.7. Listeria monocytogenes

L. monocytogenes is a foodborne pathogen causing listeriosis, with high mortality rates between 20% and 30% [72]. It is mainly distributed in the field, soil, contaminated water sources, and decaying vegetation. It is also categorized as a psychrotrophic microorganism, being ubiquitous in food-processing environments. Consequently, *L. monocytogenes* is often found as a postcontamination pathogen in food products like sliced cooked meat products, smoked fish, cut vegetables, or ready-to-eat (RTE) products. Raw chicken, milk, and raw meat are frequently implicated in foodborne outbreaks [73]. The associated high mortality rates to pregnant women and their unborn child, neonates, elderly people, and immunocompromised people makes that its level in food products should remain low. The Commission Regulation No. 1441/2007 on microbiological criteria for foodstuffs states that, for *L. monocytogenes*, in the food category *RTE foods able to support the growth of L. monocytogenes other than those intended for infants and for special medical purposes, two different microbiological criteria are proposed: (i) <i>L. monocytogenes levels should not exceed 100 cfu/g throughout the shelf life of the product and (ii) absence in 25 g of the product before the food has left the immediate control of the food business operator*, who has produced it. Their application depends on the ability of the food operator to demonstrate that the targeted food is able or not to support the growth of *L. monocytogenes* up to the end of the shelf life. Also in the United States, the limit of 100 cfu/g for *L. monocytogenes* that does not support growth of the microorganism in foods is being considered [74].

6. Risk factors associated to microbiological contamination and foodborne outbreaks

Foods can become contaminated during growth and harvesting of raw materials, storage and transport to the industry, and processing into finished products. Recontamination can also occur during transport to retail outlets and before consumption at domestic homes and/or in catering establishments. Contamination vectors are mainly animals, surfaces, environment (air, water), and people in contact with foods (food handlers). Processing conditions, packaging materials, and equipment used can also be contamination sources. Survival of microorganisms on contaminated surfaces could lead to their multiplication at high levels, thus compromising food quality and safety [75].

Animals are important reservoirs of microorganisms, and slaughter of animals could introduce high concentration of microorganisms in food industries. Zoonotic pathogens are normally present on the skin and in the gastrointestinal and respiratory tracts. Pathogens carried on the hands are also a major source of contamination [76].

Airborne contamination represents a significant medium for the microbial transfer to food products. [77] Installation of proper air filters is recommended; otherwise, microorganisms can be present together with dust, debris, or insects.

Water sources are used in the food industry as an ingredient, a processing aid, and for cleaning. Therefore, not to increase both microbial and chemical contaminations, it is important to use decontaminated water (i.e., chlorinated and electrolyzed). Water used in hand-washing facilities can pose a potential risk because of the presence of condensations, leaking pipes, or aerosols. Microorganisms colonizing these surfaces can multiply rapidly if conditions are favorable. Thus, checking the microbiological quality of water is essential to guarantee food safety.

Food handlers can act as vectors for food contamination leading to the transmission of enteric and respiratory pathogens to food, e.g., through aerosol droplets from coughing near the processing line [78]. They can also favor cross-contamination through the skin if hand-washing is not properly done.

Finally, pests, such as birds, insects, and rodents, are potentially a major contamination problem. Therefore, care must be taken to avoid their entrance into food factories. Facilities should be designed in such a way they cannot live and breed. To do so, appropriate disinsectization fumigation methods must be achieved.

The above factors when combined together may increase the risk of food contamination. According to CDC data, 1527 foodborne disease outbreaks, resulting in 29,444 cases of illness, 1184 hospitalizations, and 23 deaths were reported within 2009-2010. [79] Among the 790 outbreaks with a laboratory-confirmed illness, norovirus was the most commonly reported infection, accounting for 42% of outbreaks, followed by Salmonella, with 30% of outbreaks. Outbreaks caused by some pathogens were particularly severe. For example, Listeria outbreaks resulted in the highest proportion of persons hospitalized (82%), followed by Clostridium botulinum (67%). Among the 23 deaths, 22 were linked to bacteria (9 Listeria, 5 Salmonella, E. coli O157, 3 Clostridium perfringens, and 1 Shigella), and 1 was linked to norovirus. Regarding European data [22], in 2013, Campylobacter continued to be the most commonly reported gastrointestinal bacterial pathogen in humans in the European Union (EU) and has been so since 2005 (214,779 cases) with an EU notification rate of 64.8 per 100,000 population. However, the high mortality rate associated to L. monocytogenes was confirmed since 191 deaths were reported in 2013, much higher than deaths associated to Campylobacter or Salmonella (59 or 56, respectively).

6.1. Risk factors affecting microbial safety of foods in catering establishments

6.1.1. Hygienic food handlers` practices

In production processes, storage, and distribution of prepared foods, the role of food handlers seems essential to ensure food safety, supported mainly on good hygienic practices and implementation of improved self-control measures. The food handler is defined as "anyone who by their work have direct contact with food during preparation, manufacture, processing, manufacturing, packaging, storage, transport, distribution, sale, supply and service."

Then existing laws applied to food handlers are cited as follows:

- Regulation (EC) 852/2004 of the European Parliament and of the Council of 29 April 2004 on the hygiene of foodstuffs [10].
- Regulation (EC) 882/2004 of the European Parliament and of the Council of 29 April 2004 on official controls performed to ensure the verification of compliance with the legislation on feed and food law, animal health, and animal welfare [80].

In a food catering environment, hygiene procedures may be improved as part of food poisoning occurs as a result of the risk factors associated with food handling, related to poor hygiene, improper cooking procedures, cross-contamination, or improper storage of food [81].

Handlers sometimes act as vehicles for the spread of indicators or pathogens directly and indirectly through the hands to other food-contact surfaces and handlers. In the United States, the hygienic practices of food handlers are one of the five most important risk factors of food poisoning and about 89% of the outbreaks occur mainly by inadequate hygiene [78].

In certain circumstances, the hands may represent the most important vehicle of fecal and respiratory microorganisms [82]. It has been shown that microorganisms, such as *S. aureus*, *E. coli*, and *S. enterica.*, can survive on the hands if hygiene measures are not sufficiently appropriate. Several studies describe outbreaks of food poisoning associated with catering establishments [83]; these studies indicate that sanitary measures may be insufficient when dealing with consumer food safety.

Ayçiçek et al. [84] evaluated the sanitary measures of food handlers in a hospital central kitchen. They concluded that these measures were insufficient as significant counts of S. aureus and E. coli in both gloved hands and bare hands were presented when handling food. Specifically, S. aureus positive samples were obtained in 70% of the isolates. Other microorganisms, such as Bacillus spp. or Staphylococcus coagulase negative, were also isolated. Lues and Van Torden [85] attempted to relate the microbiological contamination found in the hands of food handlers and that presented in cloths and aprons. To do this, they visited several retail points in South Africa dedicated to the selling of RTE meat. In the study, it was found aerobic mesophilic counts (hands and aprons, respectively) in 98% and 8% of analyzed samples, total coliforms (40% and 26%), Enterobacteriaceae (44% and 16%), and S. aureus (88% and 40%). However, they did not find any significant correlation between the microbial counts, so potential cross-contamination could not be concluded. Besides, they considered that inadequate hygiene could be a potential risk factor in the microbiological contamination of food, as 32% of the hands analyzed presented high counts for total coliforms. Fecal coliforms also prevailed in more than half of the samples from the hands (55.6%) of food handlers of several school canteens in Brazil [8]. The lack of the annual medical examination (51.9%), lack of regular training for handlers (74.1%), and poor hygiene practices (100%) could have an influence of these results, as stated by other authors [86]. Other microbial agents, such as norovirus and hepatitis A virus in humans, can survive in the hands of food handlers when they do not follow good hygienic practices [2]. In this case, contamination is often associated with asymptomatic carriers and direct hand contact with contaminated food. At the same time, contamination of food-contact surfaces is also promoted.

While hand-washing is a quick and simple method, it is also considered by many authors as the most convenient and effective way to reduce foodborne pathogens [82]. If done correctly, it prevents the risk of cross-contamination and the presence of high microbial loads in foods not submitted to intense inactivation treatments.

The principles for hand-washing are universal, though effective reduction of microorganisms depends on the following considerations [82]:

- Origin and level of organic and microbial contamination.
- The use of water power.
- Washing time (15–30 s).
- Type of soap and amount used.
- Degree of exposure to the washing process of the fingers, palms, back, wrists, nails, and subungual region.

• Frequency and intensity of rubbing fingers and palms during rinsing.

In the light of the importance of maintaining adequate hygiene habits and attitudes of food handlers to ensure food safety, various methods have been used according to legal regulations for guidance handlers' hygiene. The Theory of Planned Behavior (TPB) has been advocated by many researchers to predict determinants of food handler's behavior [87].

However, some studies warn of possible deficiencies in hygiene by food handlers. For example, Green et al. [88] conducted a study to identify factors related to the hygienic practices of food handlers. To do this, they performed an observational study that evaluated the hand-washing and use of gloves of 321 food handlers. The results showed that washing hands and wearing gloves were most frequent in food preparation activities than when handling soiled equipment and direct contact with body parts. They also found that the use of gloves resulted in a decrease in the frequency of washes hands. In Clayton and Griffith [89], the habits of 115 handlers in 29 establishments during food preparation together with their corresponding hygiene measures were analyzed. The results indicated that only 9% of handlers washed their hands after touching their hair or face and 25% washed their hands after handling of contaminated equipment. Lubran et al. [90] examined the frequency of contact of the hands of food handlers with objects and food ready for consumption before the sale, washing hands and changing gloves during food preparation, as well as hygienic measures used for the equipment, utensils, and food-contact surfaces. The study was conducted in nine retail stores of RTE food and found a high percentage (60-80%) of handlers that did not wash their hands when handling food while maintaining contact with other surfaces. Likewise, the hygiene of the hands of food handlers in 15 retail shops was evaluated in another study [91]. Out of the 29 food handlers examined, only 48.3% used soap to wash their hands properly and rubbed hands including washing between fingers, fingertips, and wrists. They also found that most handlers who washed their hands in less than 10 s (41.4%) had slightly higher levels of contamination of aerobic mesophilic bacteria and *S. aureus* than those food handlers who took a longer time for hand-washing. To this end, some studies [82] highlight the importance of rubbing hands and increase in the frequency of hand-washing to have higher efficiency to remove microbial load.

On the other hand, hand-drying is also a very important in the hand-washing stage. Some authors consider most critical is the last washing stage and needs to be implemented correctly to ensure proper hygiene, thus reducing the risk of cross-contamination [92]. According to the study conducted by Michaels et al. [93], effective hand-drying may reduce microbial population up to 90%. As result of inappropriate drying practices, residual moisture of hands, drying hands with sheets of cloth, or inefficient air dryers that lengthen the drying time are encountered [94]. It should be pointed out that rubbing hands during drying could promote skin contamination after washing. In an observational study by Clayton and Griffith [89], it was found that the effectiveness of hand-washing was affected by a high percentage of inappropriate drying practices (61%).

6.1.2. Availability of health resources

Obviously, the availability of resources and the functioning of health facilities (sink, hot water, soap, etc.) in the catering establishments constitute also another factor that may adversely affect the personal hygiene practices of food handlers [94]. Another study aimed at evaluating the implementation of the HACCP system and knowledge of hygiene and food safety as well as food handlers' attitudes and practices in 20 establishments in Spain. [95] revealed that the difficulties encountered were improper maintenance of sinks and showers in 95% of the establishments visited, as well as the availability of hot water in bathrooms and changing rooms (50%). Likewise, another study evaluated 123 food retail outlets in the United States to investigate the knowledge and hygienic practices of food handlers, as well as the availability of resources for hand-washing [96]. The results indicated that the main constraints were linked to the absence of brushes to wash the nails (38%) and insufficient sinks (24%) concluding that only the fully equipped establishments (55%) had a properly trained supervisory staff.

6.1.3. Gloves

It is already demonstrated that hand-washing does not always guarantee complete removal of microorganisms. Thus, the use of gloves is necessary to prevent direct hand contact with food and food-contact surfaces as a measure of increasing food safety and minimizing risks of cross-contamination in the food industry. The most important issue is that sometimes the use of gloves can create a false sense of hygiene among handlers [97] and jeopardize the food safety.

It should be noted that the irregular change of gloves as well as their improper use could enhance cross-contamination. In the United States, this is attributed as one of the main reasons that favor the occurrence of food poisoning [98]. In another study [99], it was found that the use of the same pair of gloves for handling different foods increases the risk of transfer of pathogens. In this case, they found an increased risk of transfer of *L. monocytogenes* from contaminated raw chicken to cooked meat slices. In turn, if gloves are changed, this risk was minimized. Besides, not only the frequency of gloves changing but also their integrity is highly important to avoid cross-contamination. Some authors state that washing hands before and after the change of gloves is highly desirable to reduce this microbial contamination [81]. Even the process of changing gloves is fraught with hazards, because many glove materials cause excess moisture build-up, causing difficulty in disinfecting contamination from the nail region to spread all over the hand. However, gloves should be changed regularly because the risk of transmission of foodborne pathogens could be higher from dirty, unchanged gloves than from bare hands.

The pros and cons of using gloves are extensively explained [97]. However, it should be highlighted that a proper glove change must be done because their efficiency as physical barrier for microorganisms is limited over working time.

Besides this, physical properties of gloves (tensile strength, flexibility, resistance to puncture, and tears), material used (natural rubber gloves, vinyl, nitrile, polyurethane), and glove features (single use vs. multiuse, powder-free, allergenic reactions, etc.) should be primarily considered in the quality control system as they have a great impact on pathogens transmission from food handlers to prepared meals.

Gloves can be made of different types of material, according to their use and physical properties. Polyethylene copolymer gloves could be the least expensive of all glove types. Density materials are quite variable, and they usually have a loose fit. They are built for using in short-

time periods, and some glove types contain antibacterial compounds. However, their use is quite limited in comparison to other gloves.

Vinyl gloves (PVC) can be used as an alternative to latex gloves being more resistant to heat damage. However, they are susceptible to being torn with snag on nail edges. They also have a limited use in food industries because of their short shelf life.

Nitrile gloves were also developed to replace latex gloves. They are less elastic but have a longer shelf life. They are more resistant to chemicals but sensitive to alcohol degradation. Although they are sensible to be torn, their garish color helps them to be distinguished within a food lot.

Natural rubber latex gloves are most commonly used because they are most cost-efficient and comfortable. They provide good tactile sensitivity and good dexterity. However, latex and chemical compounds added to the gloves can produce allergies and migration of particles to food, especially in the presence of bleach.

In summary, according to the intended use, convenience, and cost-effectiveness, glove material should be carefully chosen. However, it has been demonstrated that regardless of the gloves used, handling practices and gloves changing are critical steps that influence microbial transfer to foods.

6.1.4. Cross-contamination

As mentioned above, the cross-contamination is a major cause of food poisoning worldwide. Cross-contamination phenomena arise as a consequence of the application of inadequate hygiene practices, contact with contaminated equipment and utensils, by direct hand contact with foods prepared by the improper storage of food, bad processing food, by direct food contact with air or contaminated environments, and so on [94]. Recontamination routes and sources (e.g., raw materials, food contact surfaces, food handlers) were revised [100] demonstrating their relevance to foodborne disease outbreaks. These information sources should also be incorporated in Quantitative Microbiological Risk Assessments (QMRAs) to perform mitigation strategies and reduce foodborne disease [101].

Particularly, RTE foods are highly susceptible to be contaminated during handling. Some studies refer to the direct contact of the RTE food (e.g., meat) with food-contact surfaces and contaminated cutting utensils [58]. It is also emphasized its importance as a source of transmission of enteric pathogens, *L. monocytogenes*, or *S. aureus* [102].

6.1.5. Food storage

Temperature is the most important factor that governs microbial growth in food. Most microorganisms grow at temperatures between 5°C and 60°C (called danger zone), being the optimum growth temperature at 37°C. Thus, maintaining the cold chain and a correct heat treatment for hot foods are essential measures to maintain food safety, where food handlers play a key role throughout the production chain.

Table 4 presents the recommended temperature storage/transport and freezing of raw materials and finished products in catering establishments.

Maintaining food to inadequate time intervals/storage temperature often constitutes one of the most common risk factors for food poisoning. Previous studies indicate that in school canteens and other related catering establishments exposure to abuse refrigerated temperature for extended periods of time could lead to an increase of pathogens at hazardous levels for human health from preparation to the distribution thereof [103]. Other possible deficiencies are related to temperature control storage of raw and processed ready for consumption, lack of knowledge of food handlers about cooking and refrigeration temperatures suitable to prevent the growth and survival of microorganisms, inadequate cooling and warming food and preparing several hours before consumption [104], or joint cooling of raw and cooked foods [105].

Food product	Tem	Temperatures		
	Storage/	Freezing		
	Transport			
Meat	≤7°C	≤-18°C		
Meat wastes	≤3°C	≤-18°C		
Broiler meat	≤4°C	≤-18°C		
Minced and mechanically separated meat	≤2°C	≤-18°C		
Meat preparations	≤4°C	≤-18°C		
Ham, cooked meat, deli meats	0–5°C	≤-18°C		
Prepared meals to be consumed within 24 h from preparation	≤8°C			
Prepared meals to be consumed after 24 h from preparation	≤4°C			
Prepared meals (frozen)	≤-18°C			
Hot meals	≥65°C			
Frozen fruits and vegetables	≤-18°C			

Table 4. Recommended storage temperatures of raw ingredients and prepared meals

6.1.6. Training of food handlers

Training of food handlers has been considered an important measure as a part of the HACCP systems, given that it helps to prevent most foodborne diseases. Although knowledge alone is not enough to change practices, food handlers with adequate knowledge can change their practices easier if they are closely supervised and supported by their onsite managers. In addition, guidance and supervision by their managers during work improve attitudes and practices [106].

For some time ago, questionnaires or "checklists" have represented an effective tool to evaluate the level of knowledge and skills on food hygiene and safety of food handlers [98]. Neverthe-

less, further studies detailing more sophisticated methods to obtain a greater quantity and reliability of information to improve the training of food handlers are needed.

Additionally, novel strategies leading to more effective training methods have been performed. For example, some studies proposed to strengthen the training of less experienced food handlers and validate the knowledge of those more experienced in a period not exceeding 10 years [107]. In this sense, a better knowledge on food safety by food handlers ensures better performance and motivation [108]. In this sense, it is highly important to food handlers in the HACCP systems companies to correct their attitudes and behavior at work. Also, a periodic training is found as an effective way to raise awareness of food handlers [9].

Training of food handlers in food hygiene is a mandatory requirement for the food industry. At EU level, requirements on food safety and hygiene procedures are stated in Regulation (EC) 852/2004 [10].

Specifically, in its Annex II (Chapter XII), the food business operators must ensure the following:

- "Supervision and instruction or training of food handlers in food hygiene matters, according to their work."
- "That those who are in charge of the development and maintenance of the procedure referred to in paragraph 1 of Article 5 of this Regulation or the application of relevant guides have received adequate training in regard to the application of the HACCP principles."
- "Compliance with all requirements of national legislation concerning training programs for persons working in certain food sectors."

Recently, the European Federation of Food, Agriculture and Tourism (EFFAT) and the FERCO launched the project "Food hygiene training for all" [109]. The project is based on the development of a software tool for basic training of food handlers of the contract-catering sector across Europe. This tool is available online free of charge (www.contract-catering-guide.org/ food-hygiene-training-for-all) and aims at obtaining a better qualification of workers in catering establishments and also offers the opportunity for training staff in those companies that do not have sufficient resources to invest in training.

6.1.7. Intervention strategies against microbial foodborne outbreaks

The burden reduction of foodborne diseases is a major goal of societies. The strategies developed by countries to achieve this goal are numerous and very different depending on issues, such as political and socioeconomic status, actual or emerging pathogens, resources, trade (import/exports), temporal limitations, and inter-regional cooperation.

Woteki and Kineman [110] described different approaches to reducing foodborne illness and grouped them into four categories: (i) population surveillance and better outbreak detection, (ii) prevention-based regulatory approaches, (iii) information and education, and (iv) riskbased system.

Early identification of foodborne outbreaks and the implicated organism should be directed to controlling the outbreak, stopping exposure, and perhaps more importantly, preventing future outbreaks [111]. Also, a rapid and coordinated response is needed among state officials and federal agencies. Some authors [111, 112] pointed out that surveillance based on molecular analysis of foodborne pathogens involved in outbreaks and sporadic cases together with the creation of a platform to share this information would allow for anticipation of potential future episodes. In this sense, Fisher et al. [113] reported the creation in the EU of platforms where data and information on potential outbreaks of foodborne pathogens are available and can be disseminated rapidly to those who need to know; the Enter-net is a surveillance network database of bacterial enteric pathogens, while Salm-gene is a molecular typing network. In the United States, similar platforms are available [110], i.e., FoodNet, a system of disease surveillance that provides information on the incidence of foodborne illness, and Pulse-net, a common name for National Molecular Subtyping Network for Foodborne Disease Surveillance.

Regulatory agencies allow food business operators to set up performance standards in the industry through the well-known HAZARD plan. The HAZARD plan should be designed upon the analysis of the likely hazards in the food and the strategies put in place to eliminate them or to reduce them to acceptable levels [114]. This system has shown to have a very positive and crucial impact on food safety, thus on public health, as has been recognized by key organizations like the World Health Organization [115]. Special attention deserves establishments, which deliver meals to a large number of people, and even more, in those centers where there is an important proportion of consumers with a weak or impaired immune system like hospitals or nurseries. Unfortunately, in the past years it has been reported some cases where the HACCP plan was not fully implemented. This is the case of the study by Kokkinakis et al. [116], who reported that only two out of the seven major hospitals interviewed in Crete (Greece) had implemented the HACCP plan during the period of 2004–2009. These authors identified 14 crucial elements for HACCP implementation in hospitals. Shih and Wang [117], in their study on factors influencing HACCP implementation in 23 public hospitals in Taiwan, revealed that the most important concern perceived by managers was related to economic issues, i.e., "getting funds from the hospital" and "difficulty of allocation funds for facility improvement." In addition, it was shown that more support, HACCP training, and coordination with other hospitals were necessary to avoid staff reluctance to implement the HACCP plan. Shih and Wang [117] also pointed out that kitchen design and flow charts of food production are the first two issues to consider before the HACCP implementation. The lack of financial support and poor HACCP training were also reported by Garayoa et al. [95] in their survey directed to staff from 20 contract catering companies throughout Spain.

In the food industry, emerging and existing technologies should be assessed in terms of food safety [118]. However, a new concept of food safety arisen in the early 2000s, with Regulation 178/2002, laying down the general principles and requirements of food law, establishing the European Food Safety Authority and laying down procedures in matters of food safety. In this document, risk analysis was introduced as the pillar on which food law should be based on the aim to achieve the general objective of a high level of protection of human health and life. This risk-based approach would enable the setting of national and international targets for disease reduction as well as provide the basis for such reduction efforts [110, 119].

Education and information of consumers are highly relevant to prevent outbreaks, and above all, sporadic cases. The World Health Organization launched in 2001 an educational campaign called "Five keys to Safer Foods," where five brief and clear messages were given to food handlers: (i) keep clean, (2) separate raw and cooked, (3) cook thoroughly, (4) keep food at safe temperatures, and (5) use safe water and raw materials (WHO, 2006). Other campaigns like Fight bac™ [120] or Thermy™ [121] aimed at getting consumers informed about hygienic food handling practices the former, and the use of thermometers in the cooking of food products the latter. Other most recent campaigns like "The chicken challenge" clearly show short messages with the objective of cutting *Campylobacter* food poisoning in half by the end of 2015 [122].

The reduction of foodborne illness incidence is a challenge for governments, which should manage the different strategies to lower the risk posed by food hazards up to acceptable levels. Current knowledge and tools on risk assessment allow for science-based decision-making.

7. Conclusions and recommendations

By following a systematic approach in assessing risks from production to serving food safety managers will better define the control measures to be adopted in catering settings to prevent foodborne infections. GMPs and HACCP principles should be followed together with special training of food handlers. Although microbiological quality of prepared meals is often satisfactory, special care should be taken regarding indicator microorganisms or prevalence of pathogens such as L. monocytogenes. Control of time and temperature along the food chain might prevent microbial growth until risk levels. Other measures such us excluding key highrisk foods to the most susceptible population (i.e., children, elderly, immunocompromised people) would also be advisable.

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