Chapter 3
Chemistry and Technology of Ready-to-Eat Vegetable Foods

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Abstract Ready-to-eat vegetable food refers to minimally processed fruits and vegetables, which have undergone treatments of mild intensity, without the alteration sensorial characteristics such as freshness. This type of product is ready for consumption: in recent years, it has emerged as a growing reality as it responds to consumers' needs by offering new services (convenience food). Given the direct consumption, the producer must associate a high quality of the product. This food, normally fresh and without added preservatives, is exposed to chemical and microbiological alterations; as a result, it is surely associated with a reduced shelf life. Even if these products receive some degree of minimal technological processing before market distribution, the used processing technology may be not sufficient, in most cases, with reference to microbiological stability and the complete removal of pathogens. Numerous techniques are currently been used in order to reduce microbiological and chemical spoilage, including chlorine washing, irradiation and modified atmosphere packaging. This chapter concerns recent updates about correlated technologies, including new recyclable trays, and correlated chemical and physical modifications of ready-to-eat packed products: the 'respiration' of vegetables, colorimetric modifications and other sensorial alterations.

Keywords Enzymatic browning · Ethylene · Lactic acid bacteria · Microbial spoilage · Modified atmosphere packaging · pH · Ready-to-eat vegetable food · Respiration · Shelf life · Water activity

Abbreviations

AEW	Acidic electrolysed water
CO_2	Carbon dioxide
ClO_2	Chlorine dioxide
DNA	Deoxyribonucleic acid
EHEC	Enterohaemorrhagic
HPP	High pressure processing

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H_2O_2	Hydrogen peroxide
LAB	Lactic acid bacteria
MAP	Modified atmosphere packaging
N_2	Nitrogen
O_2	Oxygen
ppm	Parts per million
POD	Peroxidase
PAL	Phenylalanine ammonia lyase
PPO	Polyphenoloxidase
RTE	Ready-to-eat
RNA	Ribonucleic acid
UV	Ultraviolet light
A_w	Water activity

3.1 Introduction

In recent years, the demand for minimally processed and ready-to-eat (RTE) fresh food products has increased dramatically in developed countries. The main reason is substantially correlated with the offer of a suitable choice for contemporary lifestyles: RTE products provide incorporated services (convenience food) to consumers. Moreover, the awareness of benefits of a diet rich in fruits and vegetables has simultaneously risen with clinical investigations and the epidemiological research. In particular, recent studies have associated the consumption of vegetable foods to a reduced risk of cardiovascular, chronic and neurological diseases, as well as some kinds of cancer (Ragaert et al. 2004; Su and Arab 2006). As a matter of fact, RTE foods contain high levels of micronutrients, fibres and antioxidants, including carotenoids and flavonoids.

Minimally processed vegetable foods are fruits and vegetables which have undergone treatments of mild intensity with the aim of increasing their functionality. On the other hand, these processing techniques do not alter sensorial features, such as freshness, and the expected nutritional quality (Allende et al. 2006). The initial quality of produce before processing has high relevance when speaking of the final RTE product. In fact, vegetables are in a raw state and ready for consumption. Consequently, these foods require very special attention because of their peculiar physiological, enzymatic and respiratory features. In addition, the problem of microbiological risks for consumers' health has to be considered.

3.2 Shelf Life and Processing

Generally, vegetable foods are known to be among the most perishable edible products. In fact, they display a high water activity (A_w) together with a neutral to slightly acidic pH value and higher carbohydrate contents with respect to proteins (Ramos et al. 2013).

In addition, minimally processed vegetable foods differ from traditional and intact products both for their physiology and their handling and storage requirements. As a matter of fact, processing procedures include very often cutting, slicing, shredding, dicing, peeling, washing and other procedures; these steps can affect the final storage life (Siddiqui et al. 2011). Washing water serves to reduce microbial contaminations because of the presence of sanitising agents such as chlorine (Sect. 3.5.1) and other chemicals (Gurnari 2015a, b).

First of all, some fruits and vegetables require the peeling step because of the necessity of removing inedible parts. Subsequently, chopping operations are required with the aim of facilitating prompt consumption. The disruption of tissues and cells integrity caused by processing can decrease shelf life.

In fact, wounded tissues undergo enhanced deterioration; as a result, derived products have a very short shelf life: 4–7 days, depending on the initial quality, the initial microbial load and the used processing technology (Watada and Qi 1999). However, various factors can influence the extent of disruption and senescence during cutting process: in particular, the size of vegetable pieces, the sharpness of cutting blades and mechanical properties of the product have to be carefully studied (Siddiqui et al. 2011).

3.3 Chemical and Biochemical Mechanisms of Spoilage

Minimally processed fruits and vegetables have different physiological rates if compared with intact products: their metabolism is accelerated similarly to the observed situation of stressed plant tissues. Even minimal processing can lead to an increase in respiration, ethylene production, water loss, microbiological replication, as well as enzymatic browning, formation of volatiles, loss of chlorophyll and lipid oxidation (Toivonen and DeEll 2002). These modifications influence directly the appearance of the final product; unfortunately, the consumer' approach is first focused on the estimation of appearance, colour and texture (Wismer 2009).

Ethylene has been reported to increase in minimally processed vegetable foods even if this phenomenon is dependent on intrinsic factors (i.e. climacteric vs. non-climacteric produce). Temperature has also an effect on the induction of ethylene production: for instance, it has been found in cantaloupes stored at very low temperatures. In this situation, the suppression wound-induced ethylene has been recognised (Madrid and Cantwell 1993). Generally, ethylene increases ripening, senescence and textural modifications by means of the stimulation of enzymatic activity; enzymes can be peroxidase (POD) and polyphenoloxidase (PPO) as well as phenolic compounds (Saltveit 1999). The initiation of wound ethylene response starts usually within 1 hour; the maximum rate is achieved between 6 to 12 h (Abeles et al. 1992).

In turn, ethylene stimulates the respiration rate: consequently, a notable enhancement of the tricarboxylic acid cycle, the electron transport chain and starch breakdown can be observed. In fact, post-harvest vegetables are living tissues similar to normal vegetables; therefore, these tissues utilise reserve energy during ageing. For instance, respiration rates have been reported to increase in baby carrots by two–threefolds after peeling and slicing (Simõ et al. 2011). In agreement, tissues with high respiration rates and low energy reserve have a shorter shelf life (Eskin 1990). However, the augmented respiration is not only due to the enhancement of aerobic respiration: the role of α-oxidation of long-chain fatty acids with the production of carbon dioxide (CO_2) has been also proposed as synergic cause (Rolle and Chism 1987).

Moreover, minimally processed products are more susceptible to water loss because peeling and cutting operations expose interior tissues. As a consequence, the peridermal tissue—which acts as a protection against excessive transpiration—is removed and surface-to-volume ratios are forced to increase (Toivonen and DeEll 2002). The decrease in water leads to a loss of turgor, reducing the firmness of the products and hence the consumer's acceptance.

Another factor correlated with the consumer's evaluation of vegetable foods is enzymatic browning. This phenomenon is primarily caused by

(a) Cell disruption, which activates metabolic pathways, ultimately leading to the synthesis of enzymes and substrates, and by
(b) Loss of cellular compartmentation, which brings cell units together.

Phenylalanine ammonia lyase (PAL) is one of the key enzymes in phenylpropanoid metabolism and is wound induced. As a matter of fact, PAL produces various phenolic compounds, which are then oxidised in reactions involving POD and PPO (Barry-Ryan and O'Beirne 1998). POD, widespread in plant cells, is iron-porphyrin organic catalyst with a notable role in browning through two possible routes. The first of these mechanisms involves the formation of hydrogen peroxide (H_2O_2) during the oxidation of phenolic compounds, whereas the second reaction route utilises quinonic forms as substrates (Richard-Forget and Gauillard 1997).

PPO is a tetramer that contains four atoms of copper per molecule and catalyses the hydroxylation of monophenols to *o*-diphenols. PPO can also further catalyse the oxidation of *o*-diphenols with the consequent production of *o*-quinones. As a result, quinones can react with non-enzymatic reactions with other quinones, amino acids or proteins. The result is a melanin pigment, responsible for the well-known black to brown colour. Another enzyme involved in senescence is lipoxygenase, an iron-containing enzyme that catalyses the oxidation of polyunsaturated fatty acids in lipids containing a *cis*-*cis*-1,4-pentadiene structure (Lamikanra 2002). Therefore, lipoxygenase generates free radicals with the ability of provoking further membrane rupture; the structural lipidic membrane is degraded. In addition, lipoxygenase is responsible for production of certain volatiles: involved biochemical pathways are usually triggered by cell damage.

As a matter of fact, plants produce secondary metabolites in response to wounding: these secondary compounds may affect dramatically the perceived odour. Each vegetable species is believed to synthesise its own characteristic volatile pattern (Pichersky et al. 2006), even if phenylpropanoid and polyketide phenolics, aldehydes, alcohols and terpenoids are the main compounds.

Sulphur-containing compounds may also accumulate during time as a result of the loss of cellular compartmentation. Enzymes such as cysteine sulfoxide lyase can oxidise various substrates and convert these compounds into sulphur-containing molecules which may be responsible for off-odours. Peculiar examples can be methanethiol, dimethyl disulfide and allyl isothiocyanate in cut cabbage tissues (Chin and Lindsay 1993; Dan et al. 1997).

Furthermore, discoloration can also occur with a general loss of green colour, due to chlorophyll degradation. Two enzymes are considered responsible for chlorophyll breakdown: chlorophyllase and magnesium dechelatase. Two alternative alternative pathways have been reported at present, both resulting in the formation of a common product: pheophorbide *a*, an olive-brown compound, which is the precursor of the colourless product in a reaction mediated by pheophorbide *a* oxygenase (Toivonen and Brummell 2008).

Finally, the residential microbial flora also affects the quality of vegetable products through spoilage and/or with possible risks for consumers' health. Processing operations can provide further opportunities for microbial contaminations; in addition, they can also cause leaking of small molecular weight compounds and cellular fluids from damaged tissues. In fact, microbial growth is usually higher in fresh-cut products with respect to the whole product. As a result, spoilage may occur: peculiar signs are loss of texture, brown colours, production of off-odours and soft rot.

3.4 Microbiological Quality

Vegetable food possess a natural saprophyte microflora deriving from soil, water, insects and consisting of bacteria, yeasts, moulds that find favourable pH and A_W conditions. As a consequence, microbial flora tends to increase during all post-harvesting stages.

The number and species of microorganisms can vary depending on the type of produce and growing conditions; however, normal counts usually range from 10^3 to 10^9 colony forming units/g, with a general predominance of Gram-negative bacteria in vegetables, and of yeasts and moulds in fruits (Oliveira et al. 2010). Even biofilms may occur in vegetable leaves, mainly composed of environmental species which may act either preventing adhesion to plant surfaces by other bacteria. Alternatively, pathogens may be embedded in their matrix, hence decreasing the efficacy of sanitising treatments.

The dominant microflora in vegetables is composed of *Pseudomonas*, generally up to 50–90 % (Arvanitoyannis and Stratakos 2010). The most abundant species appear to be *Pseudomonas fluorescens*, *P. putida* and *P. cepacia*, whose role as spoilage microorganisms is notable. As a matter of fact, they can synthetise enzymes—also under refrigeration conditions—such as pectinases, cellulases, glycoside hydrolases and lipoxygenase, in addition to well-recognised proteolytic and lipolytic activities (Heard 2002). Pectic substances are very abundant in vegetable cell walls. Chemically, these compounds are linear chains of α-(1–4)-linked D-galacturonic acid, with carboxyl groups either esterified (pectin) or non-esterified (pectic acid) with methanol.

Pectic substances are used by many microorganisms as energy source, resulting in enzymatic liquefaction of these compounds and consequently in tissue softening (Chen 2002). Involved enzymes are pectinases: these compounds exist in a wide variety of forms and are classified according to the reaction. In detail, should the mechanism of action involve β-elimination or hydrolysis, two categories would be considered: pectinesterases and depolymerising enzymes. Pectinesterases catalyse a de-esterification reaction of pectin resulting in pectate and methanol, whereas the second type of enzymes is able to cleave the pectinic chain, thereby releasing shorter portions (Sakai et al. 1993).

A peculiar enzyme, cellulose, catalyses the decomposition of cellulose, specifically by hydrolysis of the 1, 4-β-D-glucosidic bond. Basically, cellulases break down the cellulose molecule into monosaccharides such as glucose, or shorter chain of oligosaccharides. These enzymes are used by bacteria with the aim of obtaining short soluble sugars as food resources: they are divided into three general major types, based on the type of catalysed reaction:

- Endocellulases, which cleave internal bonds at random sites, thus creating new chain ends
- Exocellulases or cellobiohydrolases, which cleave two to four monomers from one end of the chain, producing cellobiose and/or glucose
- Cellobiases or β–glucosidases, which can hydrolyse exocellulase products into single monosaccharides (Singh and Hayashi 1995).

Enterobacteriaceae are well represented: generally, the most reported life forms are *Enterobacter*, *Pantoea* and *Serratia*. With the notable exception of *Erwinia carotovora*, a well-known plant pathogen, these bacteria are environmental microbes, encompassing a wide variety of ecological niches (Caponigro et al. 2010). Their role in the spoilage process has not been so well examined until now: consequently, more research would be needed at present.

Lactic acid bacteria (LAB) such as *Lactobacillus*, *Leuconostoc* and *Pediococcus* are also commonly found. LAB may affect the observed shelf life of fresh-cut products during storage (Stiles and Holzapfel 1997) through their fermentative metabolism (souring of products and gas production in anaerobic conditions). Finally, fermentative yeasts like *Kloeckera*, *Saccharomyces* and *Hanseniaspora* may cause spoilage in damaged fruits and salads, growing at low temperatures (Barnett et al. 2000).

Beside environmental microflora, human pathogens may also be conveyed by fresh produce. In fact, these products have been increasingly involved in foodborne outbreaks by bacterial, viral and parasitic pathogens. Among most common bacterial infectious agents, *Salmonella* spp. is a main concern with respect to the number of reported situations; on the other side, other species can be a major concern with concern to the severity of caused diseases.

For instance, the Gram-positive psychrophilic bacterium *Listeria monocytogenes* can determine listeriosis in pregnant women, elderly and immunosuppressed subjects. Consequences include gastroenteritis, meningitis, septicemia, abortion and death also.

Another dangerous bacterium with food safety and public health implications, *Escherichia coli*, has to be considered. In fact, aside from commensal strains, many different enteropathogenic strains are reported: enterotoxigenic, enteropathogenic and enterohaemorrhagic *E. coli* are the ones involved in foodborne outbreaks (Caruso and Parisi 2015). In particular, enterohaemorrhagic (EHEC) *E. coli* have been increasingly linked to the consumption of fresh vegetable foods. The main symptom of EHEC infections is hemorrhagic colitis; hemolytic uremic syndrome and other potentially lethal complications may also arise.

Viruses such as Norovirus and Hepatitis A virus and parasites, as *Cyclospora*, *Cryptosporidium* and *Toxoplasma*, can be a notable concern (Heard 2002) because of their involvement in foodborne outbreaks (contamination of foods from water and sewage). Moreover, RTE salads may be also a vehicle for the dissemination of antibiotic-resistant bacteria with clinical interest and genes that can be acquired by other opportunistic pathogens (Campos et al. 2013).

The multiplicity of bacteria and pathogens found in these products suggests that washing and disinfection procedures may be not sufficient to ensure a good microbiological quality, highlighting the necessity of implementing more efficient post-harvesting decontamination methods.

3.5 Methodologies to Improve Quality

Physiological and microbial-induced modifications in appearance and quality of minimally processed vegetable foods can be slowed down and minimised through a multi-phase approach, combining pre-harvest, pre- and post-processing treatments and management procedures. Obviously, the primary objective is to prevent microbial contamination and extend shelf life of food products; because of the intrinsic difficulty, various techniques, above all chemical and physical ones, are available at present.

3.5.1 Chemical Methods

Among sanitising agents, chloride-based rinses are the most widely used in the produce industry. Chlorine compounds are usually utilised in a concentration range between 50 and 200 parts per million (ppm) for less than 5 min (Rico et al. 2007). Theoretically, chlorine is more efficient at acidic pH levels, but usually it is used at pH between 6.0 and 7.5; the reason is the necessity of minimising machinery corrosion (Beuchat 2000). Although observed the advantages (reduction of microbial counts), chlorine can lead to the formation of chlorine vapours or chlorinated by-products, that may have potential harmful health effects (Parish et al. 2003). Therefore, chlorine dioxide (ClO_2) has been introduced as an alternative to chlorine, as it does not form noxious chloramine compounds. Moreover, this chemical has a higher oxidation capacity: about 2.5 times greater than normal chlorine. ClO_2 has also shown (Ramos et al. 2013):

(a) A higher level of penetration with respect to the liquid agent
(b) A high efficacy against pathogens, acting on cellular aminoacids and ribonucleic acid (RNA).

H_2O_2 has a strong oxidising power leading to the generation of cytotoxic reactive oxygen species, hydroxyl radicals above all. H_2O_2 has hence a notable bactericidal activity: it is used up to 80 ppm in washing water (Alexandre et al. 2012).

Organic acids (e.g. ascorbic, lactic, citric and tartaric acid) are also frequently used as antimicrobial agents, as they have a role in environmental and intracellular pH reduction, anion accumulation and damage of membrane permeability and transport (Beuchat 2000). Ascorbic acid is frequently used as antioxidant in fruits and vegetables because of its antioxidant activity which prevents browning and inhibits polyphenol oxidase reactions.

Ozone is a potential method for extending shelf life of fresh commodities, due to its high reactivity and penetrability. It can be used both in water and in gas form where higher concentrations—around 20,000 ppm—can be reached even if gaseous ozone is considered to be more effective (Klockow and Keener 2009). Ozone has shown various advantages, including decomposition in non-toxic products, reduction in enzyme activity, decomposition of some pesticides and reduction in the oxygen demand. On the other hand, this agent has also some side effects, as it rapidly disappears; moreover, ozone is reported to be associated with lower crispiness and colour degradation (Guzel-Seydim et al. 2004; Rico et al. 2006).

Calcium-based additives (e.g. calcium lactate) are also used for products with a high senescence index. In fact, calcium helps in maintaining firmness by interacting with cell walls and middle lamella pectins to form calcium pectate. Furthermore, calcium-based solutions have been shown to reduce chlorophyll and protein loss, as well as inhibit tissue senescence (Smout et al. 2005).

Lastly, electrolysed water is utilised for its bactericidal effect. Generally, it is generated by the electrolysis of water containing dissolved sodium chloride. This process leads essentially to the production of gaseous hydrogen and hydroxide ions at the cathode, hence forming an alkaline solution consisting of sodium hydroxide. At the anode, chloride and hydroxide ions are oxidised to gaseous chlorine, hypochlorous acid, hydrochloric acid and hypochlorite ions. Should the formation of these compounds be allowed, acidic electrolysed water (AEW) would be obtained with a pH value between 2.1 and 4.5. Despite its strong bactericidal activity, AEW has shown adverse effects on produce quality because of pH values and high oxidation-reduction potentials (Rico et al. 2007; Wang et al. 2004). On the contrary, pH can be raised to neutral values (i.e. neutral electrolysed water): this solution does not affect colours and the general appearance of products (Izumi 1999).

3.5.2 Physical Methods

In recent years, different physical technologies are emerging as processing applications in the food industry.

Modified atmosphere packaging (MAP) is still commonly used in the food industry as preservation technique and consists in the alteration of the normal air composition, usually by lowering oxygen (O_2) percentage and replacing it with CO_2 or nitrogen (N_2). The gas modification is reached either actively, by flushing a gas mixture before sealing, or passively. However, gas composition will inevitably be modified in both cases during the commercial life of MAP products, due to respiration and film permeability to gases (Sivertsvik et al. 2002).

CO_2 and N_2 concentrations vary depending on the type of product and on processing methods. MAP extends storage life of both whole and processed commodities of about 50–400 % by reducing ethylene production, respiration rates and other metabolic activities (Ramos et al. 2013). Moreover, MAP delays enzymatic browning and growth of aerobic bacteria, even if excessively reduced O_2 concentrations may lead to the overgrowth of anaerobes with fermentative metabolism and consequent off-odours. The increase of microbial counts ascribed to potential pathogens has to be also considered.

Irradiation is an innovative and very effective method of decontamination: the application of this technology is gradually increasing at a global level. Irradiation is a physical treatment that consists in exposing foods to an energy source such as gamma rays and X-rays. It is effective against microorganisms because it ionises atoms, removing electrons from their orbits with the generation of free radicals. This process destabilises essential cellular macromolecules such as proteins, deoxyribonucleic acid (DNA) and RNA (Kundu et al. 2014), hence delaying also senescence. On the other side, irradiation treatments are essentially safe from the toxicological point of view when speaking of consumers' health. Moreover, adequate doses do not compromise organoleptic and nutritional quality of irradiated foods (Ahn et al. 2004).

The irradiation of foodstuffs can be performed by means of gamma rays, emitted by sources of caesium-137, cobalt-60 or, alternatively, by electron beams. The formation of radicals and their spread depend on A_w values of the irradiated food: the treatment is reported to be less effective in anhydrous and frozen products. On the other hand, irradiation is not widely accepted by consumers; in addition, it may produce some textural alteration.

Ultraviolet light (UV) is used as antimicrobial agent because of its direct damage to DNA: in fact, UV rays cause the production of pyrimidine dimers, a disruption in the genetic sequence. UV light is subdivided into three different types according to wavelengths:

- UV-A rays (range: from 315 to 400 nm)
- UV-B light (range from 280 to 315 nm)
- UV-C, also named 'far UV' rays (range from 100 to 280 nm).

UV light, especially UV-C rays, is commonly used because of the inexpensiveness of equipment and the induction of the synthesis of health-promoting molecules such as anthocyanins and stillbenoids (Cantos et al. 2001). However, it has to be noted that the application of UV light has various limitations. For instance, this technology can increase respiration rate of the produce and induce lignification-like processes (Ramos et al. 2013).

High pressure processing (HPP) is another method for the inactivation of microorganisms and enzymes: the technology is based essentially on the application of elevated pressures (100–1000 MPa) on foods. Although a high pressure is achieved, flavours and the general nutritional quality appear to remain unchanged, even if this method shows some adverse effects on vegetables. In fact, high pressures can damage the integrity of porous products due to the intrinsic compression and expansion cycle of the process (Palou et al. 2000). In addition, another limitation that can become a notable concern for traders is the expensiveness of the technological system.

Ultrasound technology has also been studied because of its application in food science: it is environmentally sustainable and considered one of the new ‘green’ technologies (Chemat et al. 2011). In detail, high-intensity ultrasound (low frequencies from 20 to 100 kHz) is used in order to inactivate bacteria and enzymes. Basically, ultrasounds in a liquid medium can determine the production of high energy amounts through the compression and expansion of particles of the treated medium (Butz and Tauscher 2002). Its bactericidal activity depends on the cavitation phenomenon: in other words, the formation, growth and subsequent collapse of bubbles are observed during the treatment. The result is the creation of a localised mechanical energy that causes disruption of cellular walls and membranes. In addition, free radicals and highly reactive molecules such as protons, hydroxide ions and H_2O_2 are generated by means of a peculiar reaction, water sonolysis, thus targeting DNA and lipid membranes (Bermúdez-Aguirre et al. 2011; Rastogi 2011).

Enzyme inactivation could be due to the breakage of hydrogen bonds and van der Waals interactions through polypeptide chains, with the consequent disruption of secondary and tertiary enzyme structures and hence of biological functions (São Jose et al. 2014). Ultrasound technology has proved to be more effective in disaggregating microbial biofilms and accessing surfaces that are difficult to reach with respect to other disinfection methods. On the other hand, a large-scale usage is still being discussed: at present, best results for disinfection are provided by utilising ultrasound in combination with other technologies and agents such as peracetic acid (Gao et al. 2014; São José and Vanetti 2012).

Lastly, another innovative option is the realisation of innovative packaging materials (active or ‘smart’ packaging) by means of the use of active agents of various types: ions, enzymes, fungicides, organic acids, ethanol, etc. The final aim is to improve food safety and quality.

As a matter of fact, active packaging strategies include principally

- Addition of volatile antimicrobial agents as ethanol generators, oxygen and moisture absorbers into packages
- Incorporation of bioactive agents into packaging polymers. Examples: silver ions into polyethylene, polypropylene and butadiene styrene
- Use of antimicrobial polymers such as chitosan and polylysine (cationic polymers) that interact directly with cell membranes (Appendini and Hotchkiss 2002).

3.5.3 Biological and 'Generally Recognized as Safe' Methods

Because of the increased consumer concern about the toxicological safety of chemicals and synthetic additives, the request for 'Generally Recognized As Safe' (GRAS) substances or natural food preservative is rising.

Research has been carried out on biocontrol agents, specific species of bacteria which are known for their antagonistic potential on pathogens: LAB. Biocontrol bacteria are strong competitors for physical space and nutrients; they may generate diverse antimicrobial metabolites such as bacteriocins (Sagong et al. 2011).

Bacteriocins are proteinaceous toxic compounds with either a broad or a narrow spectrum of inhibition. Numerous bacteriocins have been tested for their application as food preservatives: some of these compounds are already commercially available such as nisin. They can be added in concentrated preparations or produced in situ by LAB starter cultures. In addition, bacteriocins have been used in polymers (bioactive food packaging materials).

Another promising strategy concerns the use of essential oils because of their own antioxidant properties. These organic substances, derived from spices and other plants, are attracting interest for their potential in enhancing storage life as antimicrobial agents. For instance, oregano (*Origanum vulgare*) and thyme (*Thymus vulgaris*) oils contain two strong antibacterial compounds, carvacrol and thymol, respectively. Many essential oils have been recognised as GRAS at present; however, their practical application is still limited because of the altering effect on food organoleptic properties (Oussalah et al. 2006).

Edible coating films are now being recognised for their potential applications. First of all, these coatings can remarkably delay sensorial modifications (appearance and aroma) during storage. Moreover, these materials can act as carriers of active compounds as antimicrobials, nutrients and anti-browning agents. A wide variety of substances can be used in edible films including lipids, resins, polysaccharides and proteins, either individually or combined. Some of the most used compounds are vegetable starch, fruit wax, gum, pectin, carboxymethyl cellulose, chitosan, alginates and carrageenan.

References

Abeles FB, Morgan PW, Saltveit ME Jr (1992) Ethylene in plant biology, 2nd edn. Academic Press Inc, San Diego

Ahn HJ, Kima JH, Jo C, Lee J, Yook H, Kim HY, Byun MW (2004) Combined effects of gamma irradiation and a modified atmosphere packaging on the physico-chemical characteristics of sausage. Radiat Phys Chem 71(1–2):51–54. doi:10.1016/j.radphyschem.2004.04.006

Alexandre EMC, Brandão TRS, Silva CLM (2012) Assessment of the impact of hydrogen peroxide solutions on microbial loads and quality factors of red bell peppers, strawberries and watercress. Food Control 27(2):362–368. doi:10.1016/j.foodcont.2012.04.012

Allende A, Tomas-Barberan FA, Gil MI (2006) Minimal processing for healthy traditional foods. Trends Food Sci Tech 17(9):513–519. doi:10.1016/j.tifs.2006.04.005

Appendini P, Hotchkiss JH (2002) Review of antimicrobial food packaging. Innov Food Sci Emerg Tech 3:113–126. doi:10.1016/S1466-8564(02)00012-7

Arvanitoyannis IS, Stratakos AC (2010) Irradiation of fruits and vegetables. In: Arvanitoyannis IS (ed) Irradiation of food commodities. Academic Press, San Diego, pp 467–535

Barnett JA, Payne RW, Yarrow D (2000) Yeasts: characteristics and identification, 3rd edn. Cambridge University Press, Cambridge, pp 395–401

Barry-Ryan C, O'Beirne D (1998) Quality and shelf-life of fresh-cut carrot slices as affected by slicing method. J Food Sci 63:851–856

Bermúdez-Aguirre D, Mobbs T, Barbosa-Cánovas GV (2011) Ultrasound applications in food processing. In: Feng H, Barbosa-Cánovas GV, Weiss J (eds) Ultrasound technologies for food and bioprocessing. Springer, New York, pp 65–105

Beuchat LF (2000) Use of sanitizers in raw fruit and vegetable processing. In: Alzamora SM, Tapia MS, Lopez-Malo A (eds) Minimally processed fruits and vegetables. Fundamental aspects and applications. Aspen, Gaithersburg, pp 63–78

Butz P, Tauscher B (2002) Emerging technologies: chemical aspects. Food Res Int 35(2–3):279–284. doi:10.1016/S0963-9969(01)00197-1

Campos J, Mourão J, Pestana N, Peixe L, Novais C, Antunes P (2013) Microbiological quality of ready-to-eat salads: an underestimated vehicle of bacteria and clinically relevant antibiotic resistance genes. Int J Food Microbiol 166(3):464–470. doi:10.1016/j.ijfoodmicro.2013.08.005

Cantos E, Espin JC, Tomas-Barberan FA (2001) Effect of wounding on phenolic enzymes in six minimally processed lettuce cultivars upon storage. J Agri Food Chem 49(1):22–30. doi:10.1021/jf000644q

Caponigro V, Ventura M, Chiancone I, Amato L, Parente E, Piro F (2010) Variation of microbial load and visual quality of ready-to-eat salads by vegetable type, season, processor and retailer. Food Microbiol 27(8):1071–1077. doi:10.1016/j.fm.2010.07.011

Caruso G, Parisi S (2015) The importance of Escherichia coli, a versatile enemy. In: Laganà P, Caruso G, Barone C, Caruso G, Parisi S, Melcarne L, Mazzù F, Santi Delia A (eds) Microbial toxins and related contamination in the food industry. Springer Briefs in Chemistry of Foods, Springer. doi:10.1007/978-3-319-20559-5_4

Chemat F, Zill-e-Huma, Khan MK (2011) Applications of ultrasound in food technology: processing, preservation and extraction. Ultrason Sonochem 18(4):813–835. doi:10.1016/j.ultsonch.2010.11.023

Chen J (2002) Microbial enzymes associated with fresh-cut produce. In: Lamikanra O (ed) Fresh-cut fruits and vegetables: science, technology and market. CRC Press, Boca Raton, pp 249–266

Chin HW, Lindsay RC (1993) Volatile sulfur compounds formed in disrupted tissues of different cabbage cultivars. J Food Sci 58(4):835–839. doi:10.1111/j.1365-2621.1993.tb09370.x

Dan K, Todoriki S, Nagata M, Yamashita I (1997) Formation of volatile sulfur compounds in broccoli stored under anaerobic condition. J Jpn Soc Hort Sci 65(4):867–875. doi:10.2503/jjshs.65.867

Eskin MNA (1990) Biochemical changes in raw foods: fruits and vegetables. In: Eskin MNA (ed) Biochemistry of foods, 2nd edn. Academic Press, San Diego, pp 70–78

Gao S, Lewis GD, Ashokkumar M, Hemar Y (2014) Inactivation of microorganisms by low-frequency high-power ultrasound: 1. Effect of growth phase and capsule properties of the bacteria. Ultrason Sonochem 21(1):446–453. doi:10.1016/j.ultsonch.2013.06.006

Gurnari G (2015a) The chemistry of disinfection: ally or enemy? In: Gurnari G (ed) Safety protocols in the food industry and emerging concerns. Springer briefs in Chemistry of Foods. Springer, pp 75–92. doi:10.1007/978-3-319-16492-2_6

Gurnari G (2015b) Reasons for the substitution of chlorine in the disinfection. In: Gurnari G (ed) Safety protocols in the food industry and emerging concerns. Springer briefs in Chemistry of Foods. Springer, pp 75–92. doi:10.1007/978-3-319-16492-2_7

Guzel-Seydim ZB, Greene AK, Seydim AC (2004) Use of ozone in the food industry. LWT Food Sci Tech 37(7):453–460. doi:10.1016/j.lwt.2003.10.014

Heard GM (2002) Microbiology of fresh-cut produce. In: Lamikanra O (ed) Fresh-cut fruits and vegetables: science, technology and market. CRC Press, Boca Raton

Izumi H (1999) Electrolysed water as a disinfectant for fresh-cut vegetables. J Food Sci 64(3):536–539. doi:10.1111/j.1365-2621.1999.tb15079.x

Klockow PA, Keener KM (2009) Safety and quality assessment of packaged spinach treated with a novel ozone-generation system. LWT Food Sci Tech 42(6):1047–1053. doi:10.1016/j.lwt.2009.02.011

Kundu D, Gill A, Lui C, Goswami N, Holley R (2014) Use of low dose e-beam irradiation to reduce E. coli O157:H7, non-O157 (VTEC) E. coli and Salmonella viability on meat surfaces. Meat Sci 96(1):413–418. doi:10.1016/j.meatsci.2013.07.034

Lamikanra O (2002) Enzymatic effects on flavor and texture of fresh-cut fruits and vegetables. In: Lamikanra O (ed) Fresh-cut fruits and vegetables: science, technology and market. CRC Press, Boca Raton

Madrid M, Cantwell M (1993) Use of high CO_2 atmospheres to maintain quality of intact and fresh cut melon. In: Proceedings of the 6th international controlled atmosphere research conference, vol 2, 14–17 June 1993, NRAES-71, Cornell University, Ithaca, p 736–745. Northeast Regional Agricultural Engineering Service Cooperative Extensions, Ithaca

Oliveira M, Usall J, Viñas I, Anguera M, Gatius F, Abadias M (2010) Microbiological quality of fresh lettuce from organic and conventional production. Food Microbiol 27(5):679–684. doi:10.1016/j.fm.2010.03.008

Oussalah M, Caillet S, Saucier L, Lacroix M (2006) Inhibitory effects of selected plant essential oils on the growth of four pathogenic bacteria: E. coli O157:H7, Salmonella Typhimurium, Staphylococcus aureus and Listeria monocytogenes. Food Control 18(5):414–420. doi:10.1016/j.foodcont.2005.11.009

Palou E, Lopez-Malo A, Barbosa-Canovas GV, Welti-Chanes J (2000) High hydrostatic pressure and minimal processing. In: Alzamora SM, Tapia MS, Lopez-Malo A (eds) Minimally processed fruits and vegetables. Fundamental aspects and applications. Aspen, Gaithersburg, pp 205–222

Parish ME, Beuchat LR, Suslow TV, Harris LJ, Garrett EH, Farber JN, Busta FF (2003) Methods to reduce/eliminate pathogens from fresh and fresh-cut produce. Compr Rev Food Sci Food Safety 2(Supplement):161–173. doi:10.1111/j.1541-4337.2003.tb00033.x/pdf

Pichersky E, Noel JP, Dudareva N (2006) Biosynthesis of plant volatiles: nature's diversity and ingenuity. Science 311(5762):808–811. doi:10.1126/science.1118510

Ragaert P, Verbeke W, Devlieghere F, Debevere J (2004) Consumer perception and choice of minimally processed vegetables and packaged fruits. Food Qual Pref 15(3):259–270. doi:10.1016/S0950-3293(03)00066-1

Ramos B, Miller FA, Brandão TRS, Teixeira P, Silva CLM (2013) Fresh fruits and vegetables—an overview on applied methodologies to improve its quality and safety. Innov Food Sci Emerg Tech 20:1–15. doi:10.1016/j.ifset.2013.07.002

Rastogi N (2011) Opportunities and challenges in application of ultrasound in food processing. Crit Rev Food Sci Nutr 51(8):705–722. doi:10.1080/10408391003770583

Richard-Forget FC, Gauillard FA (1997) Oxidation of chlorogenic acid, catechins, and 4-methylcatechol in model solutions by combinations of pear (Pyrus communis cv. Williams) polyphenol oxidase and peroxidase: a possible involvement of peroxidase in enzymatic browning. J Agri Food Chem 45(7):2472–2476. doi:10.1021/jf970042f

Rico D, Martin-Diana AB, Frias JM, Henehan GTM, Barry-Ryan C (2006) Effect of ozone and calcium lactate treatments on browning and texture properties of fresh-cut lettuce. J Sci Food Agri 86(13):2179–2188. doi:10.1002/jsfa.2594

Rico D, Martin-Diana AB, Barat JM, Barry-Ryan C (2007) Extending and measuring the quality of fresh-cut fruit and vegetables: a review. Trends Food Sci Tech 18(7):373–386. doi:10.1016/j.tifs.2007.03.011

Rolle RS, Chism GW III (1987) Physiological consequences of minimally processed fruits and vegetables. J Food Qual 10(3):157–177. doi:10.1111/j.1745-4557.1987.tb00856.x

Sagong HG, Lee SY, Chang PS, Heu S, Ryu S, Choi YJ et al (2011) Combined effect of ultrasound and organic acids to reduce Escherichia coli O157:H7, Salmonella Typhimurium, and Listeria monocytogenes on organic fresh lettuce. Int J Food Microbiol 145(1):287–292. doi:10.1016/j.ijfoodmicro.2011.01.010

Sakai T, Sakamoto T, Hallaert J, Vandamme EJ (1993) Pectin, pectinase, and protopectinase: production, properties, and applications. Adv Appl Microbiol 39:213–294

Saltveit M (1999) Effect of ethylene on quality of fresh fruits and vegetables. Postharvest Biol Technol 15(3):279–292. doi:10.1016/S0925-5214(98)00091-X

São Jose JFB, Andrade NJ, Ramos AM, Vanetti MCD, Stringheta PC, Chaves JBP (2014) Decontamination by ultrasound application in fresh fruits and vegetables. Food Control 45:36–50. doi:10.1016/j.foodcont.2014.04.015

São José JFB, Vanetti MCD (2012) Effect of ultrasound and commercial sanitizers on natural microbiota and Salmonella enterica Typhimurium on cherry tomatoes. Food Control 24(1–2):95–99

Siddiqui MW, Chakraborty I, Ayala-Zavala JF, Dhua RS (2011) Advances in minimal processing of fruits and vegetables: a review. J Sci Ind Res 70(10):823–834

Simõ AND, Allende A, Tudela JA, Puschmann R, Gil MI (2011) Optimum controlled atmosphere minimise respiration rate and quality losses while increase phenolics compounds of baby carrots. LWT Food Sci Technol 44(1):277–283. doi:10.1016/j.lwt.2010.06.002

Singh A, Hayashi K (1995) Microbial cellulases: Protein architecture, molecular properties, and biosynthesis. Adv Appl Microbiol 40:1–44

Sivertsvik M, Rosnes JT, Bergslien H (2002) Modified atmosphere packaging. In: Ohlsson T, Bengtsson N (eds) Minimal processing technologies in the food industry. Woodhead Publishing Ltd, Cambridge, pp 61–86

Smout C, Sila DN, Vu TS, Van Loey AML, Hendrickx MEG (2005) Effect of preheating and calcium pretreatment on pectin structure and thermal texture degradation: a case study on carrots. J Food Eng 67(4):419–425. doi:10.1016/j.jfoodeng.2004.05.010

Stiles ME, Holzapfel WH (1997) Lactic acid bacteria of foods and their current taxonomy. Int J Food Microbiol 36(1):1–29. doi:10.1016/S0168-1605(96)01233-0

Wismer WV (2009) Consumer eating habits and perceptions of fresh produce quality. In: Florkowski WJ, Prussia SE, Shewfelt RL, Brueckner B (eds) Postharvest handling: a systems approach, 2nd edn. Academic Press, Burlington

Su LJ, Arab L (2006) Salad and raw vegetable consumption and nutritional status in the adult US population: Results from the third national health and nutrition examination survey. J Am Diet Assoc 106(9):1394–1404. doi:10.1016/j.jada.2006.06.004

Toivonen PMA, DeEll JR (2002) Physiology of fresh-cut fruits and vegetables. In: Lamikanra O (ed) Fresh-cut fruits and vegetables: science, technology and market. CRC Press, Boca Raton

Toivonen PMA, Brummell DA (2008) Biochemical bases of appearance and texture changes in fresh-cut fruit and vegetables. Postharvest Biol Technol 48(1):1–14. doi:10.1016/j.postharvbio.2007.09.004

Wang H, Feng H, Luo Y (2004) Microbial reduction and storage quality of fresh-cut cilantro washed with acidic electrolyzed water and aqueous ozone. Food Res Int 37(10):949–956. doi:10.1016/j.foodres.2004.06.004

Watada AE, Qi L (1999) Quality of fresh-cut produce. Postharvest Biol Technol 15(3):201–205. doi:10.1016/S0925-5214(98)00085-4