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Polysaccharides as Carriers and Protectors of Additives and Bioactive Compounds in Foods

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Additional information is available at the end of the chapter

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1. Introduction

Overall quality and shelf-life of fresh foods post-harvest or -slaughter is reduced by several factors including microbial growth, water loss, enzymatic browning, lipid oxidation, off-flavor, texture deterioration, rise in respiration rate and senescence processes, among others. In fresh-cut fruits and vegetables, these events are accelerated due to lesions of tissues during peeling, slicing and cutting [1]; whereas, in meat products these events are accelerated due to lesions of tissues during cutting and grinding.

Fresh-cut fruits and vegetables during mechanical operations are exposed to spoil because the natural protection of fruit (the peel or skin) is generally removed and hence, they become highly susceptible to microbial growth due to the leakage of juices and sugar from damaged tissues which allow the growth and fermentation of some microorganism. Likewise, during processing, enzymes such as polyphenol oxidase, polygalacturonase and pectin methylesterase are released, thus causing, browning and softening from the tissues, respectively. These enzymes come in contact with phenolic compounds for forming brown pigments, and hydrolyze the α -1,4-glucosidic bonds to degrade the tissues [2].

Appearance/color, texture, and flavor are the main quality attributes that affect consumer acceptance of meat, and lipid oxidation is one of the primary causes by quality deterioration in meat and meat products [3]. The meat once cut or sliced is exposed to the surrounding environment, and cell compounds released during mechanical operations react with the environment and cause quality deterioration of tissues. Lipid oxidation occur when oxygen come in contact with lipid present in pieces of meats, being the iron the major catalyst in lipid oxidation processes. This process is associated with the presence of free radicals that lead to the production of aldehydes, which are responsible for the development of rancid flavors and changes in the color of meat [4].

On the other hand, dairy products such as fresh and semi-hard cheeses are complex food products consisting mainly of casein, fat, and water. Such products are highly perishable due to the high content of moisture (only in fresh cheeses) and microorganisms, and some cases high fat-content [5]; therefore, off-flavor, lipid oxidation and microbial spoilage are the major quality deteriorations.

Because of happened issues in the past with fresh or fresh-cut products, new technologies have been applied to counteract these negatives effects. Among them, polysaccharide-based edible films and coatings have emerged like good alternative for enhancing the quality and safety of such foods. Edible films and coatings have been used to reduce the deleterious effect caused by minimal processing. The semipermeable barrier provided by edible coatings is focused to extend shelf-life by reducing moisture and solutes migration, gas exchange, respiration and oxidative reaction rates, as well as suppress physiological disorders on fresh and/or fresh-cut foods [6]. However, the use of edible films and coatings for a wide range of food products, including fresh and minimally processed vegetables and fruits, has received an increasing interest because films and coatings can serve as carriers for a wide range of food additives including: antimicrobials, antioxidants and antisoftening compounds into edible films or coatings to provide a novel way for enhancing the safety and shelf-life of fresh, fresh-cut or ready-to-eat foods [7-10].

The new generation of edible films and coatings is being especially designed to increase their functionalities by incorporating natural or nutraceutical/functional ingredients such as probiotics, minerals and vitamins [10-11]. In addition, the sensory quality of coated products with edible materials can be also improved [2,7-9]. On the other hand, encapsulation (microencapsulation or nanoencapsulation) are being currently applied to foods to preserve and protect the additive or bioactive compounds from the surrounding environment [12-14].

In this chapter will discuss the use of polysaccharide-based edible films and coatings as polymeric matrix to carrier additive or bioactive compounds such as antimicrobial, antioxidant, antisoftening and nutraceutical for enhancing the shelf-life, safety and sensory attributes of fresh food products, as well as methodologies of forming and application of edible films and coatings and futures trends using microencapsulation or nanotechnology.

2. Use of polysaccharide-based edible films and coatings as carriers of additives and bioactive compounds on foods

The overall quality of food products decreases from harvest or slaughter until they are consumed. Quality loss may be due to microbiological, enzymatic, chemical, or physical changes. Therefore, food additives should be added to prevent the quality loss and extend the shelf-life of foods. The use of films and coatings have been a good alternative for carrier different additives and bioactive compounds to the food, as well as, to protect them of the water loss, volatile compounds loss, discoloration, gas permeability and microbial spoilage; since, these can to guarantee the controlled supply of antimicrobial, antioxidant, antisoftening and nutraceutical compounds. Tables 1, 2, 3 and 4 show the additives

(antimicrobial, antioxidant, antisoftening and nutraceutical compounds) added on foods through polysaccharide-based edible films and coatings for improving the food quality and safety. Each of these additives is studied in the following sections of this chapter.

2.1. Carriers of antimicrobial compounds

Foods may be contaminated with pathogenic and spoilage microorganisms if bad manufacture practices are carried out during handling, processing, distribution and commercialization [15]. Therefore, antimicrobial compounds should be used during processing and packaging for controlling the microbiological safety and quality, and prolonging the shelf-life of foods. Food antimicrobials are chemical compounds added or naturally occurring in foods to inhibit or inactivate populations of pathogenic and spoilage microorganisms.

Several studies have demonstrated that antimicrobials such as organic acids, enzymes, essential oils, spices and bacteriocin incorporated into polysaccharide-based edible films and coatings have been effective for controlling pathogenic and spoilage microorganisms in different foods (Table 1). In this context, different researchers have demonstrated that the incorporation of bacteriocins into alginate-based film have been effective to inactivate or delay the growth of some pathogenic microorganisms. The alginates are anionic polysaccharides from the cell walls of brown algae that can serve to prepare carriers solution of antimicrobial substances. Hence, Cutter and Siragusa [16], Natrajan and Sheldon [17] and Milette et al. [18] achieved to reduce populations of *Brochothrix thermosphacta* ($> 3.0 \log \text{CFU/g}$), *Salmonella enterica* ser. Typhimurium ($> 4.0 \log \text{CFU/g}$) and *Staphylococcus aureus* ($> 2.5 \log \text{CFU/g}$) on ground beef, poultry skin and beef fillets, respectively, using calcium alginate-based film and coating, and palmitoylated alginate incorporated with nisin (from 5 to 100,000 mg/mL) during storage refrigerated. Likewise, Datta et al. [19] indicated that the growth of *Listeria monocytogenes* and *Salmonella enterica* ser. Anatum was suppressed in the range of 2.2 to 2.8 log CFU/g in smoked salmon coated with an alginate coating containing oyster lysozyme at 160,000 mg/g plus nisin at 10 mg/g during storage at 4°C by 35 days. Neetoo et al. [20] delayed the growth of *L. monocytogenes* on cold-smoked salmon slices and fillets during the 30 days storage at 4°C using alginate-based edible coating with sodium lactate (2.4%) and diacetate (0.25%). Marcos et al. [21] reported a bacteriostatic effect against *L. monocytogenes* inoculated in sliced cooked ham during 60 days of storage at 1°C, when enterocins A and B (2,000 AU/cm²) were incorporated into an alginate film.

On the other hand, essential oils and their active compounds have been also incorporated into the alginate-based films and coatings to control the growth of pathogenic and spoilage microorganisms in several foods [22-26]. Hence, Oussalah et al. [22] evaluated the effect of an alginate-based film containing essential oils of Spanish oregano, Chinese cinnamon or winter savory at 1% w/v against populations of *S. enterica* ser. Typhimurium or *E. coli* O157:H7 inoculated in beef muscle slices stored at 4 °C by 5 days. These authors reported that films including essential oils of oregano or cinnamon were more effectives against *S. enterica* ser. Typhimurium ($>1 \log \text{cycle}$); whereas, films including essential oils of oregano

Type of polysaccharide matrix		Food	Antimicrobial compounds	Reference
Film	Coating			
Alginate	-	Ground beef	Nisin	[16]
Alginate	-	Poultry skin	Nisin	[17]
Alginate	-	Beef fillets	Nisin	[18]
Alginate	-	Smoked salmon	lysozyme / nisin	[19]
Alginate	-	Cooked ham sliced	Enterocin A and B	[21]
Alginate	-	Beef fillets	EOs of oregano, cinnamon, savory	[22]
Alginate	-	Cooked ham and bologna sliced	EOs of oregano, cinnamon, savory	[23]
-	Alginate	Cold-smoked salmon slices and fillets	Sodium lactate and diacetate	[20]
-	Alginate/ apple puree	Fresh-cut apple	Vanillin and EOs of lemongrass, oregano	[24]
-	Alginate	Fresh-cut melon	Malic acid and EOs of lemongrass, cinnamon, plamarose, eugenol, citral, geraniol	[25]
-	Alginate	Fresh-cut apple	Malic acid and EOs of lemongrass, cinnamon, clove, cinnamaldehyde, eugenol, citral	[26]
-	Alginate	Fresh-cut apple	Potassium sorbate	[27]
-	Alginate	Roasted turkey	Sodium lactate and diacetate	[29]
Carrageenan	-	Fresh chicken breast	Ovotransferrin	[28]
Chitosan	-	Cooked ham, bologna and pastrami	Cinnamaldehyde, acetic, propionic and lauric acids	[32]
Chitosan	Chitosan	Mozzarella cheese	Lysozyme	[74]
Chitosan	-	Cheese	Natamycin	[38]
-	Chitosan	Whole strawberry	Potassium sorbate	[33]
-	Chitosan	Rainbow trout	Cinnamon oil	[37]
-	Chitosan	Roasted turkey	Sodium lactate and diacetate	[29]
Chitosan	Chitosan	Cold-smoked salmon	Potassium sorbate, sodium lactate and diacetate	[30]
-	Chitosan / Plastic	Ham steaks	Sodium lactate, diacetate and benzoate, potassium sorbate, or nisin	[36]
Chitosan	-	Pork sausages	Green tea extract	[39]
-	Chitosan / MC	Fresh-cut Pineapple and melon	Vanillin	[35]
CMC	-	Fresh pistachios	Potassium sorbate	[43]
Cellulose	-	Cooked ham sliced	Pediocin	[42]
Cellulose	-	Frankfurter sausages	Nisin	[41]
MC / HPMC	-	Hot Dog Sausage	Nisin	[40]
-	HPMC	Whole oranges	Potassium sorbate, sodium benzoate, sodium propionate	[44]
-	HPMC	Whole strawberry	Potassium sorbate	[33]
-	Pectin	Pork patties	Powder of green tea	[47]
-	Pectin	Roasted turkey	Sodium lactate and diacetate	[29]
-	Starch	Minimally processed carrots	Chitosan	[45]
-	Starch	Roasted turkey	Sodium lactate and diacetate	[29]
-	Starch / Gum	Fruit-based salad, romaine hearts and pork slices	Green tea extract	[46]

MC: methyl cellulose; CMC: carboxy methyl cellulose; HPMC: hydroxyl propyl methyl cellulose; EOs: essential oils

Table 1. Major antimicrobial compounds applied on foods through polysaccharide-based edible films and coatings

Type of polysaccharide matrix		Food	Antioxidant compounds	Reference
Film	Coating			
-	Alginate	Fresh-cut apple	Glutathione and N-Acetyl-cysteine	[61-62]
-	Alginate	Fresh-cut pears	Glutathione and N-Acetyl-cysteine	[63]
-	Alginate	Fresh-cut apples	Calcium chloride	[27]
-	Alginate	Bream (freshwater fish)	Vitamin C and tea polyphenols	[53]
-	Carrageenan	Fresh-cut apples	Ascorbic, citric and oxalic acids	[59]
-	Carrageenan	Fresh-cut banana	Ascorbic acid and cysteine	[60]
-	CMC	Fresh-cut apples and potatoes	Ascorbic acid and TBHQ	[57]
-	Gellan	Fresh-cut apple	Glutathione and N-Acetyl-cysteine	[61-62]
-	Gellan	Fresh-cut pears	Glutathione and N-Acetyl-cysteine	[63]
-	HPMC	Toasted almonds	Ascorbic, citric and EO ginger	[52]
-	MC	Fresh-cut apples	Ascorbic acid	[58]
-	Maltodextrin	Fresh-cut apples	Ascorbic acid	[58]
-	Pectin	Fresh-cut pears	Glutathione and N-Acetyl-cysteine	[63]
-	Pectin	Pork patties	Powder of green tea	[47]

MC: methyl cellulose; CMC: carboxy methyl cellulose; HPMC: hydroxyl propyl methyl cellulose; EOs: essential oils

Table 2. Major antioxidant compounds applied on foods through polysaccharide-based edible films and coatings

Type of polysaccharide matrix		Food	Antisoftening compounds	Reference
Film	Coating			
-	Alginate	Fresh-cut apples	Calcium lactate	[26]
-	Alginate	Fresh-cut melons	Calcium lactate	[27]
-	Alginate	Fresh-cut apples	Calcium chloride	[61-62]
-	Alginate	Fresh-cut melons	Calcium chloride	[67]
-	Alginate	Fresh-cut pears	Calcium chloride	[63]
-	Alginate	Fresh-cut apples	Calcium chloride	[27]
-	Alginate	Fresh-cut papayas	Calcium chloride	[68]
-	Alginate / Apple puree	Fresh-cut apples	Calcium chloride	[24]
-	Carrageenan	Fresh-cut apples	Calcium chloride	[59]
-	Carrageenan	Fresh-cut banana	Calcium chloride	[60]
-	Gellan	Fresh-cut papayas	Calcium chloride	[68]
-	Gellan	Fresh-cut apples	Calcium chloride	[61-62]
-	Gellan	Fresh-cut melons	Calcium chloride	[67]
-	Gellan	Fresh-cut pears	Calcium chloride	[63]
-	Pectin	Fresh-cut melons	Calcium chloride	[67]
-	Pectin	Fresh-cut pears	Calcium chloride	[63]

Table 3. Major antisoftening compounds applied on foods through polysaccharide-based edible films and coatings

Type of polysaccharide matrix		Food	Nutraceutical compounds	Reference
Film	Coating			
-	Alginate	Fresh-cut papayas	Probiotics	[11]
-	Gellan	Fresh-cut papayas	Probiotics	[11]
-	Chitosan	Lingcod fillets	Omega 3 and Vitamin E	[34]
-	Chitosan	Whole strawberry	Calcium	[72]
-	Chitosan	Whole strawberry and red raspberry	Calcium and Vitamin E	[73]
-	Xanthan gum	Peeled baby carrots	Calcium and Vitamin E	[71]

Table 4. Major nutraceutical compounds applied on foods through polysaccharide-based edible films and coatings

was more effective against *E. coli* O157:H7 (> 2 log cycles). Similarly, Oussalah et al. [23] studied the effect of alginate-based edible film containing essential oils of Spanish oregano, Chinese cinnamon, or winter savory at 1% (w/v) against *S. enterica* ser. Typhimurium or *L. monocytogenes* inoculated onto bologna and ham slices. These authors concluded that alginate-based films containing essential oil of cinnamon was the most effective in reducing the populations of both pathogenic microorganisms by more than 2 logs CFU/g on bologna and ham sliced. In the same way, Rojas-Grau et al. [24] studied the antimicrobial effect of essential oils of lemongrass (1 and 1.5%) and oregano (0.1 and 0.5%), and vanillin (0.3 and 0.6%) incorporated into coating forming solutions based on alginate and apple puree against the naturally occurring microorganisms and *Listeria innocua* inoculated on fresh-cut apples. These authors found that all the essential oils used significantly inhibited the native flora during 21 days of storage at 4°C, being lemongrass and oregano oils more effective against *L. innocua* than vanillin. Likewise, Raybaudi-Massilia et al. [25] reported significant reduction (3–5 log cycles) of the inoculated *Salmonella enterica* var. Enteritidis population in pieces of melon when an alginate-based edible coating containing malic acid (2.5%), alone or in combination with essential oils of cinnamon, palmarose or lemongrass at 0.3 and 0.7% or their actives compounds eugenol, geraniol and citral at 0.5%, were applied. In addition, inhibition of the native flora by more than 21 days of storage was also observed at 5°C. Similar results were found by Raybaudi-Massilia et al. [26], who evaluated the antimicrobial effect of an alginate-based edible coating with malic acid (2.5%) incorporated, alone or in combination with essential oils of cinnamon bark, clove or lemongrass at 0.3 and 0.7% or their actives compounds cinnamaldehyde, eugenol or citral at 0.5% on fresh-cut apples. They reached to reduce population of *Escherichia coli* O157:H7 (4 log cycles) after 30 days of refrigerated storage (5°C), as well as to inhibit the native flora by more than 30 days.

Other antimicrobial compounds such as potassium sorbate, ovotransferrin, sodium lactate and sodium acetate have been also applied to fresh-cut apples, fresh chicken breast and ready-to-eat roasted turkey through an alginate-based edible coating to inhibit the native flora growth. In such sense, Olivas et al. [27] inhibited the microbial growth of mesophilic and psychrotropic bacteria, moulds and yeasts in apple slices coated with an edible alginate

coating containing 0.05% potassium sorbate during 8 days of storage at 5°C. Likewise, Seol et al. [28] reduced populations of total microorganisms (about 2 log cycles) and *E. coli* (about 3 log cycles) on fresh chicken breast stored at 5°C after 7 days, when a κ -carrageenan-based edible film containing ovotransferrin (25 mg) and EDTA (5 mM) was applied on its surface. Jiang et al. [29-30] showed that potassium sorbate (0.15%), sodium lactate (1.2-2.4%) and sodium diacetate (0.25-0.50%) incorporated into chitosan- or alginate-based edible coating and film were able to inactivate *L. monocytogenes* (about 1-3 log CFU/g) on ready-to-eat cold-smoked salmon and roasted turkey stored at 4°C. All these results have demonstrated that alginate- κ -carrageenan-based film and coating are excellent carriers of antimicrobial substances on meat, poultry and fruits and vegetables products for reducing populations of pathogenic microorganisms.

In the same way, chitosan which is a linear polysaccharide consisting of (1,4)-linked 2-amino-deoxy-b-D-glucan, and a deacetylated derivative of chitin, and the second most abundant polysaccharide found in nature after cellulose [31] has been used as carrier of antimicrobial compounds in other foods. In such sense, Ouattara et al. [32] evaluated the effectiveness of chitosan films incorporated with acetic or propionic acid, with or without addition of lauric acid or cinnamaldehyde to preserve vacuum-packaged bologna, cooked ham and pastrami during refrigerated storage. The efficacies of the films to inhibit the microbial growth were tested against native lactic acid bacteria, Enterobacteriaceae, and against *Lactobacillus sakei* or *Serratia liquefaciens* inoculated on the surface of products. The authors indicated that the growth of lactic acid bacteria were not affected by the antimicrobial films, but the growth of Enterobacteriaceae and *S. liquefaciens* was delayed or completely inhibited after application. Park et al. [33] showed the antifungal effect of a chitosan-based edible coating containing potassium sorbate (0.3%) to inhibit the *Cladosporium* sp. and *Rhizopus* sp, total aerobic count and coliforms growth, and in fresh strawberries stored at 5°C and 50% RH by 23 days. Coating treatment also reduced total aerobic count, coliforms, and weight loss of strawberries during storage. Duan et al. [34] reduced about 1 log cycle the populations of *L. monocytogenes*, *E. coli*, or *Pseudomonas fluorescens* inoculated on the surface of Mozzarella cheese using chitosan composite films and coatings incorporated with lysozyme and storage at 10 °C. Sangsuwan et al. [35] studied the antimicrobial effect of a chitosan/MC film incorporated with vanillin against *E. coli* and *Saccharomyces cerevisiae* inoculated on fresh-cut cantaloupe and pineapple. They found that antimicrobial film inactivated populations of *E. coli* and *S. cerevisiae* on fresh-cut cantaloupe by more than 5 and 0.6 log CFU/g during 8 and 20 days of storage, respectively, at 10°C. Whereas, this antimicrobial film inactivated *S. cerevisiae* on fresh-cut pineapple by more than 4 log CFU/g during 12 days of storage at 10°C, but against *E. coli* there was not significant reductions. Ye et al. [36] used a plastic film coated with chitosan and Sodium lactate (1%), diacetate (0.25%), and benzoate (0.1%), potassium sorbate (0.3%) or nisin (5 mg/cm²) for inhibiting the growth of *L. monocytogenes* on strawberries during 10 days of storage at room temperature (20°C). Ojagh et al. [37] extended the shelf-life of Rainbow trout (a fish native of North America) during 16 days at 4°C incorporating cinnamon oil (at 1.5%) into a matrix of chitosan-based edible coating. Fajardo et al. [38] evaluated the antifungal activity of

chitosan-based edible coating containing 0.5mg/mL natamycin on semi-hard "Salvio" cheese; and demonstrated that populations of moulds and yeasts were reduced by about 1.1 log CFU/g compared to control samples after 27 days of refrigerated storage. Jiang et al. [29] showed that a combination of sodium lactate and sodium diacetate incorporated into chitosan edible coating was able to inactivate *L. monocytogenes* on ready-to-eat roasted turkey stored at 4°C. Siripatrawan and Noipha [39] used a chitosan film containing green tea extract as active packaging for extending shelf-life of pork sausages. These authors completely inhibited the microbial growth in pork sausages refrigerated (4 °C). Hence, chitosan can be used as a natural antimicrobial coating on fresh strawberries to control the growth of microorganisms, thus extending shelf-life of the products

Films and coatings based on cellulose or derivatives such as methyl cellulose (MC), carboxy methyl cellulose (CMC) or hydroxy propyl methyl cellulose (HPMC) containing antimicrobial compounds have been used to control microbial growth and extend the shelf-life of several foods. In such sense, Franklin et al. [40] determined the effectiveness of packaging films coated with a MC/HPMC-based solution containing 100, 75, 25 or 1.563 mg/ml nisin for controlling *L. monocytogenes* on the surfaces of vacuum-packaged hot dogs. They found that packaging films coated with a cellulose-based solution containing 100 and 75 mg/ml nisin significantly decreased ($P \leq 0.05$) *L. monocytogenes* populations on the surface of hot dogs by greater than 2 logs CFU/g throughout the 60 days of storage. Nguyen et al. [41] developed and used cellulose films produced by bacteria containing nisin to control *L. monocytogenes* and total aerobic bacteria on the surface of vacuum-packaged frankfurters. Bacterial cellulose films were produced by *Gluconacetobacter xylinus* K3 in corn steep liquor-mannitol medium and were subsequently purified before nisin was incorporated into them. Cellulose films with nisin at 25 mg/ml significantly reduced ($P < 0.05$) *L. monocytogenes* (approximately 2 log CFU/g) and total aerobic bacteria (approximately 3.3 log CFU/g) counts on frankfurters after 14 days of storage as compared to the control samples. Whereas, Santiago-Silva et al. [42] developed and evaluated the antimicrobial efficiency of cellulose films with pediocin (antimicrobial peptide produced by *Pediococcus* sp.) incorporated at 25% and 50% of cellulose weight on sliced ham. They found that antimicrobial films were more effective against *L. innocua* than *Salmonella* sp., since the 50% pediocin-film showed a reduction of 2 log CFU/g in relation to control treatment after 15 days of storage; whereas, the 25% and 50% pediocin-films had similar performance on *Salmonella* sp. about 0.5 log CFU/g reductions in relation to control, after 12 days of storage at 12°C. On the other hand, Park et al. [33] achieved to inhibit the growth of *Cladosporium* sp., *Rhizopus* sp, total aerobic count and coliforms on fresh strawberry through a HPMC-based edible coating containing potassium sorbate (0.3%) stored at 5°C and 50% RH by 23 days. Sayanjali et al. [43] evaluated the antimicrobial properties of edible films based on CMC containing potassium sorbate (at 0.25, 0.5 and 1.0%) applied on fresh pistachios, and reported that all concentrations of potassium sorbate used inhibited the growth of molds. Valencia-Chamorro et al. [44] studied the antifungal effect of HPMC based coatings with potassium sorbate (2%), sodium benzoate (2.5%), sodium propionate (0.5%) and their combinations on the postharvest conservation of "Valencia" oranges. These authors reported that the

application of HPMC coatings reduce significantly the effects caused by *Penicillium digitatum* and *Penicillium italicum* inoculated in the surface of the oranges, resulting more effective those coatings with potassium sorbate and sodium propionate combined.

Others polysaccharides-based films and coatings such as pectins and starches have been used also as carriers of antimicrobials compounds in foods. Durango et al. [45] controlled the growth of mesophilic aerobes, yeasts and moulds and psychrotrophics populations in processed minimally carrots during the first 5 days of storage at 15°C using yam starch-based edible coatings containing chitosan (0.5 and 1.5%). In the same way, Chiu and Lai [46] studied the antimicrobial properties of edible coatings based on a tapioca starch/decolorized hsian-tso leaf gum matrix with incorporated green tea extracts on fruit-based salads, romaine hearts and pork slices. The authors indicated that when green tea extracts at 1% were added into edible coating formulations, the aerobic count successfully decreased and growth of yeasts/molds decreases by 1 to 2 logs CFU/g in fruit-based salads. In addition, they reported that romaine hearts and pork slices coated with these antimicrobial edible coatings reduced populations of Gram positive bacteria from 4 to 6 logs CFU/g during 48 h of refrigerated storage. On the other hand, Kang et al. [47] evaluated the microbiological quality of pork hamburger coated with a pectin-based edible coating with incorporated green tea powder (0.5%), and packed in air or vacuum during 14 days at 10°C. These authors reported that initial population of total aerobic microorganisms (10^4 CFU/mg) decreased until undetectable levels by more than 7 days under vacuum conditions; whereas, in normal conditions of atmosphere (air) a level of 10^5 CFU/mg was reached at the same time. Jiang et al. [29] showed that a combination of sodium lactate and sodium diacetate incorporated into pectin-based edible coating was able to inactivate populations of *L. monocytogenes* on ready-to-eat roasted turkey stored at 4°C.

Previous results have showed that several polysaccharides-based films and coatings (alginate, carrageenan, chitosan, cellulose derivatives, pectin, starch and apple puree) could be used as outstanding carriers of antimicrobial substances for ensuring the quality and safety of foods in the meat, poultry, seafood, dairy, fruits and vegetables industries. In addition, the incorporation of essential oils into films and coatings formulations may contribute to prevent the water vapor permeability and decreases the solubility of films and coatings in foods with high content of humidity.

3. Carriers of antioxidant compounds

Antioxidant compounds can also be incorporated into edible films and coatings to avoid the food oxidation and browning. In such sense, rosemary oleoresin, an extract of spice with antioxidant activity, has been added into starch-alginate coatings to inhibit the lipid oxidation and warmed-over flavor (WOF) development in precooked pork chops [48] and beef patties [49]. In the same way, tocopherols have been incorporated into starch-alginate coatings to retard the formation of WOF in precooked pork chops [50]. Wu et al. [51] studied the effect of starch-alginate (SA), SA-stearic acid (SAS), SA-tocopherol (SAT), SAS-tocopherol (SAST), SAT-coated (SATC), and SAST-coated (SASTC) films on moisture loss

and lipid oxidation in precooked ground-beef patties. These authors reported that tocopherol-treated films were more effective ($P < 0.05$) in inhibiting lipid oxidation than those tocopherol-untreated films on ground-beef patties. However, SAS-based films were more effective ($P < 0.05$) in controlling moisture loss than lipid oxidation. Atarés et al. [52] evaluated the antioxidant efficiency of HPMC coatings with ascorbic acid, citric acid or ginger essential oil incorporated on toasted almonds to avoid the lipid oxidation. They concluded that films with ascorbic and citric acid showed a cross-linking effect, and were the most effective protectors against oxidation of almonds, due to both their antioxidant effect and the tighter structure which leads to lower oxygen permeability. Khang et al. [47] found that lipid oxidation decreased and radical scavenging increased in the pork patties coated with a pectin-based edible coating containing green tea leaf extract (0.5%) during 14 days at 10°C. These authors indicated that coated patties held higher moisture contents than the controls in both air- and vacuum packaging. Song et al. [53] indicated that sodium alginate-based edible coating containing vitamin C (5%) or tea polyphenols (0.3%) were able to delay the chemical spoilage and water loss of bream (*Megalobrama amblycephala*), in addition to enhancing the overall sensory attributes, in comparison with uncoated bream during 21 days storage at $4 \pm 1^\circ\text{C}$.

On the other hand, the color in fresh-cut fruits and vegetables is of great importance, since oxidation and enzymatic browning take place quickly upon contact with oxygen during processing, leading to discoloration [54]. Browning phenomena in fresh-cut products are caused when, after mechanical operations (cutting, slicing, coring, shredding, etc) during processing, enzymes, which are released from wounded tissues, come in contact with phenolic components to give dark colored pigments [55]. Such phenomenon is caused by the action of a group of enzymes called polyphenol oxidases (PPOs), which can oxidize the phenolic substrates to *o*-quinones in presence of oxygen [56]. Therefore, the application of antioxidant agents incorporated into edible coatings would be a good alternative to ensure the inhibition of browning, to prevent ascorbic acid or vitamin C loss, and extend the shelf-life of fresh-cut fruits and vegetables [9]. In such sense, Baldwin et al. [57] reported that ascorbic acid (0.5%) and *ter*-butyl-hydro-quinone (0.2%) had a better effect on the inhibition of browning in fresh-cut apples and potatoes throughout storage when these antioxidants were incorporated into an edible coating based on CMC than when these were used in an aqueous dipping solution after 14 days at 4°C. Both methods were effective during the first day of storage, but samples coated with the edible coating prevented browning for a longer time than those samples dipped in an aqueous solution alone. Brancoli and Barbosa-Cánovas [58] achieved a decreasing browning in surface of apple slices during 21 days of storage at 4°C using maltodextrin and MC-based coatings containing ascorbic acid (1%). Likewise, Lee et al. [59] delayed the browning of fresh-cut apples using antibrowning agents such as ascorbic (1%), citric (1%), oxalic (0.05%) acid or their combinations incorporated into edible coatings based on carrageenan. These authors observed an inhibition of the enzymatic browning in fresh-cut apples during 14 days storage at 3°C. In addition, edible coating with antioxidants obtained higher sensory scores (positive effect) during sensory evaluation than non-coated apples. In the same way, Bico et al. [60] reached to retard the

browning of fresh-cut bananas using ascorbic acid and cysteine at 0.75% incorporated into an edible coating based on carrageenan during 5 days of storage at 5°C. Rojas-Grau et al. [61-62] inhibited the browning in fresh-cut apples using edible coatings based on alginate or gellan with the addition of glutathione (up to 2%) or N-acetyl-cysteine (up to 2%), or their combination. These authors indicated that a concentration of 1% each of the antibrowning agents was needed to maintain the color of cut apples. Similar results were also obtained by Oms-Oliu et al. [63], who achieved browning inhibition of fresh-cut “Flor de invierno” pears for 14 days at 4°C using N-acetyl-cysteine (0.75%) and glutathione (0.75%) incorporated into edible coatings based on alginate, gellan or pectin. Olivás et al. [27] delayed the development of browning in apple slices during 8 days of storage at 5°C after applying alginate coatings containing calcium chloride (10%). Calcium chloride is an anti-browning agent known to inhibit PPO by interaction of the chloride ion with copper at the PPO active site [64].

Based on the different works reported in the bibliography it is possible to indicate that several polysaccharides-based films and coatings (alginate, carrageenan, cellulose derivatives, pectin, gellan and maltodextrin) could be used as excellent carriers of antioxidant substances for avoiding the lipid oxidation and enzymatic browning of meat and fruits products.

4. Carriers of antisoftening compounds

Foods more susceptible to the texture loss are fresh-cut fruits and vegetables. This fact is due to that during mechanical operations (peeling, cutting, sliced, shredded) plant tissues are broken down and enzymes such as pectinolytic and proteolytic are released, thus causing softening [1]. In addition, these enzymes could also affect the morphology, cell wall middle lamella structure, cell turgor, water content, and biochemical components [65]. Pectinase enzymes such as polygalacturonase and pectin methylesterase are responsible for texture losses in plant tissues. Polygalacturonase hydrolyses the α -1,4-glucosidic bond among anhydrogalacturonic acid units, whereas, pectin methylesterase hydrolyses the methyl-ester bonds of pectin to give pectic acid and methanol, thus resulting in texture degradation because of hydrolysis of the pectin polymers [1]. Nonetheless, treatments with calcium can help to counteract this problem improving the firmness of fruit tissues by reacting with pectic acid present in the cell wall to form calcium pectate, which reinforces the molecular bonding among constituents of the cell wall, thus delaying the senescence and controlling physiological disorders in fruits and vegetables [8,66]. Different studies have demonstrated that the use of polysaccharide based films and coatings (alginate, carrageenan, pectin, gellan and apple puree) as carriers of calcium chloride or lactate have resulted to be a good alternative to prevent the firmness or texture loss of the fresh-cut fruits, which could be beneficial to the fresh-cut fruits industry.

In this sense, Oms-Oliu et al. [63,67] and Rojas-Graü et al. [24,61-62] observed that fresh-cut melons, pears, and apples coated with alginate-, gellan-, pectin- or apple-puree edible coatings containing calcium chloride (2%) maintain in excellent conditions their initial firmness during refrigerated storage (4°C) from 14 to 21 days. The authors indicated that

polysaccharide matrices with substances increased the water vapor resistance, thus preventing dehydration, and they had an inhibitory effect on ethylene production, but O₂ and CO₂ production was not affected. Similar effects were achieved by Olivas et al. [27], who preserved the firmness of apple slices stored at 5°C for 10 days by using an alginate edible coating containing calcium chloride (10%). Raybaudi-Massilia et al. [25,26] showed that the incorporation of calcium lactate (2%) into an alginate-edible coating maintained the firmness of fresh-cut apples and melons during 21 days at 5°C. Similarly, Tapia et al. [68] improved the firmness of fresh-cut papaya with the addition of calcium chloride (2%) into alginate- and gellan edible coating during the period studied (8 days at 4°C). Likewise, Lee et al. [59] and Bico et al. [60] kept the firmness of fresh-cut apple and banana slices storage at refrigerated temperature using a carrageenan-based edible coating containing calcium chloride (1%).

5. Carriers of nutraceutical compounds

Nutraceuticals are chemical compounds found as natural components of foods or other ingestible forms that have been determined to be beneficial to the human body in preventing or treating one or more diseases or improving physiological performance [69]. Calcium and vitamin E are the most important nutraceutical compounds and, they can play significant roles in the human body in preventing certain diseases [70]. Nonetheless, probiotics are being used currently as a functional compound in foods, since potential health benefits and biological functions of bifidobacteria in humans like the intestinal production of lactic and acetic acids, pathogens inhibition, reduction of colon cancer risks, cholesterol reduction in serum, improved calcium absorption, and activation of the immune system, among others [11]. Thus, nutraceutical compounds carried into edible coatings and films to strengthen and increase the nutritional value of foods have been researched.

Edible coatings can provide an excellent vehicle to further enhance the health benefit of products like berry fruits where the lack of some important nutraceuticals, such as vitamin E and calcium may be compensated by incorporating them into the coatings [10]. In this way, Mei et al. [71] used xanthan gum coating as a carrier of calcium (as calcium lactate at 5%) and vitamin E (as α -tocopheryl acetate at 0.2%) for covering peeled baby carrots. The authors indicated that calcium and vitamin E contents of the coated samples (85g per serving), increased from 2.6 to 6.6% and from 0 to about 67% of the Dietary Reference Intakes values, respectively. In addition, they found that edible coatings improved the desirable surface color of carrots without significant effects on the taste, texture and fresh aroma. Hernández-Muñoz et al. [72] coated strawberries (*Fragaria x ananassa* Duch.) with a chitosan-based edible coating containing 1% calcium gluconate and stored during 4 days at 20°C. These authors found that strawberries coated with chitosan-based edible coating with incorporated calcium were better retained in coating (3,079 g/kg dry matter) than in strawberries dipped in calcium solutions alone (2,340 g/kg), thus resulting in increased nutritional value. Likewise, Han et al. [73] used chitosan-based edible coatings containing 5% Gluconal® CAL or 0.2% DL- α -tocopheryl acetate to enhance the nutritional value of

strawberries (*Fragaria × ananassa*) and red raspberries (*Rubus ideaus*) stored at 2°C and 88% relative humidity (RH) for 3 weeks or at 23°C up to 6 months. They concluded that chitosan-based coatings containing calcium or Vitamin E significantly increased the content of these nutrients in both fresh and frozen fruits. These researchers also indicated that adding high concentrations of calcium or Vitamin E into chitosan-based coatings did not alter their anti-fungal and moisture barrier functions. Moreover, the coatings significantly decreased decay incidence and weight loss, drip loss and delayed the change in color, pH and titratable acidity of strawberries and red raspberries during cold storage. Duan et al. [74] increased total lipid and omega-3 fatty acid contents of fresh and frozen lingcod by about 3-fold and reduced TBARS (Thio-barbituric acid reactive substances) values in both fresh and frozen samples, incorporating 10% fish oil (containing 91.2% EPA (eicosapentaenoic acid) and DHA (docosahexaenoic acid)) plus 0.8% vitamin E into chitosan-based edible coating.

Developing edible coatings to carry high concentrations of nutraceuticals for nutritionally fortified foods can also be considered as an important way to afford functional characteristics to coated foods. In this context, Tapia et al. [11] managed to incorporate viable *Bifidobacterium lactis* Bb12 strains into alginate and gellan film-forming solutions to coat fresh-cut apples and papayas, and evaluated the effectiveness of such edible coatings to carry and support the probiotic culture. The authors reported that populations > 10⁶ CFU/g of the microorganism were kept during 10 days of refrigerated storage. A viable bifidobacteria population of 5 logs CFU/g in the final product has been pointed out as the therapeutic minimum to attain health benefits [75].

In general, fruits, vegetables and seafood industries could apply different polysaccharides-based coatings (alginate, gellan, chitosan and gum) as excellent carriers of nutraceutical compounds for adding nutritive value and functional properties to the products.

6. Methodology for film and coating formation, incorporation of additives/bioactive compounds and ways of applications

6.1. Film and coating formation, and incorporation of additives/bioactives compounds

An edible film is essentially an interacting polymer network of three-dimensional gel structure. Despite the film-forming process, whether it is wet casting or dry casting, film-forming materials should form a spatially rearranged gel structure with all incorporated film-forming agents, such as biopolymers, plasticizers, other additives, and solvents in the case of wet casting. Biopolymers film-forming materials are generally gelatinized to produce film-forming solutions. Sometimes drying of the hydrogels is necessary to eliminate excess solvents from the gel structure. This does not mean that the film-forming mechanism during the drying process is only the extension of the wet-gelation mechanism. The film-forming mechanism during the drying process may differ from the wet-gelation mechanism, though wet gelation is initial stage of the film-forming process. There could be a critical stage of a transition from a wet gel to a dry film, which relates to a phase

transition from a polymer-in-water (or other solvents) system to a water-in-polymer system [76].

Two processes can be used for film-production: dry and wet. The dry process of edible film production does not use liquid solvent, such as water or alcohol. Molten casting, extrusion, and heat pressing are good examples of dry process. For the dry process, heat is applied to the film-forming materials to increase the temperature to above the melting point of the film-forming materials, to cause them to flow. The wet process uses solvents for the dispersion of film-forming materials, followed by drying to remove the solvent and form a film structure. For the wet process the selection of solvents is the one of the most important factors. Since the film-forming solution should be edible and biodegradable, only water, ethanol and their mixtures are appropriated as solvents. To produce a homogeneous film structure avoiding phase separation, various emulsifiers can be added to the film forming solution. This solvent compatibility of ingredients is very important to develop homogeneous edible film and coating systems carrying active agents. All ingredients, including active agents as well as biopolymers and plasticizers should be homogeneously dissolved in solvent to produce film-forming solutions [76].

7. Ways of application of films and coatings

Different ways for film and coating application have been reported in the literature; being dipping, spraying, brushing, casting and wrapping the more commons methods [7,76-79]:

- *Dipping*: This method lends to food products that require several applications of coating materials or require a uniform coating on an irregular surface. After dipping, excess coating material is allowed to drain from the product and it is then dried or allowed to solidify. This method has been generally used to apply coating of alginate, gellan, chitosan, MC and pectin to fresh-cut fruit.
- *Spraying*: Film applied by spraying can be formed in a more uniform manner and thinner than those applied by dipping. Spraying, unlike dipping, is more suitable for applying a film to only one side of a food to be covered. This is desirable when protection is needed on only one surface, e.g., when a pizza crust is exposed to a moist sauce. Spraying can also be used to apply a thin second coating, such as the cation solution needed to cross-link alginate or pectin coatings.
- *Brushing*: This method consists in the direct application and distribution of the coating material in a liquid form using a hand brush.
- *Casting*: This technique, useful for forming free-standing films, is borrowed from methods developed for not edible films. For formation of a film the film-forming biopolymers are first dissolved in the solvent. If heating or pH adjustment enhances film formation and/or properties, this is done next. If a composite film or coating based on an emulsion is desired, a lipid material, and possibly a surfactant, is added. Next the mixture is heated to above the lipid melting point and then homogenized. Degassing is an important step to eliminate bubble formation in the final film or coating. Finally, the edible film or coating is formed by applying the prepared formulation to the desired coating or product surface and allowing the solvent to evaporate

- *Wrapping*: this method is obtained from cast films depending on firmness and flexibility for wrapping surface. It allows films to be cut to any size, and serves as an innovative and easy method for carrying and delivering a wide variety of ingredients such as flavoring, spices and seasoning that can later be used to cover foods. This method is especially useful when applied to highly spicy materials that need to be separated from the food products.

8. Preserving and protecting bioactive compounds through microencapsulation

Microencapsulation is a technique by which solid, liquid or gaseous active ingredients are packaged within a second material for the purpose of protecting or shielding the active compound from the surrounding environment. Thus the active compound is designated as the core material, whereas the surrounding material forms the shell. This technique can be employed in a diverse range of fields such as agricultural, chemical, pharmaceutical, cosmetics, printing and food industry [13].

Microcapsules can be classified on the basis of their size or morphology. Thus, microcapsules range in size from one micron; whereas, some microcapsules whose diameter is in the nanometer range are referred as nanocapsules to emphasize their smaller size. On the other hand, morphology microcapsules can be classified into three basic categories as mono-core (also called single-core or reservoir type), poly-core (also called multiple-core) and matrix types (Figure 1). Mono-core are microcapsules that have a single hollow chamber within the capsule; Poly-core are microcapsules that have a number of different sized chambers within the shell; and matrix type are microparticles that has the active compounds integrated within the matrix of the shell material. However, the morphology of the internal structure of a microparticle depends mostly on the selected shell materials and the microencapsulation methods that are employed [12-13].

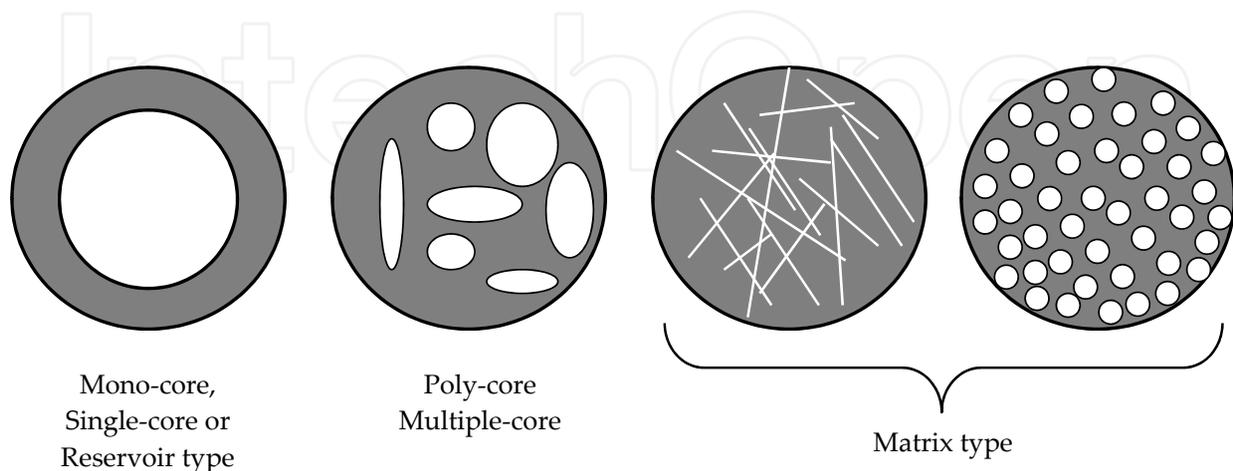


Figure 1. Morphology of microcapsules

Current trends of the consumers for eating healthy foods that preventing illness and to be low calories but rich in vitamins, minerals and other bioactive component have conducted to the researches and industrials to develop foods called "functional", where some ingredients to promote health are added. However simply adding ingredients to food products to improve nutritional value can compromise their taste, color, texture and aroma. Sometimes, they are slowly degraded and in consequence, lose their activity, or become hazardous by oxidation reactions. Active compounds can also react with components present in the food system, which may limit bioavailability. Microencapsulation is used to overcome all these challenges by providing viable texture blending, appealing aroma release, and taste, aroma and color masking. This technology enables to the food industries to incorporate minerals, vitamins, flavors and essential oils. In addition, microencapsulation can simplify the food manufacturing process by converting liquids to solid powder, decreasing production costs by allowing batch processing using low cost, powder handling equipment. Microcapsules also help at fragile and sensitive materials survive processing and packaging conditions and stabilize the shelf-life of the active compound.

On the other hand, applications of microencapsulations to foods have been increasing due to the protection of encapsulate materials of factors such as heat and humidity, allowing to maintain its stability and viability. The microcapsules help to food materials to withstand the conditions of processing and packing to improve taste, aroma, stability, nutritional value and appearance of their products. Some of the substances encapsulated have been fertilizers, oil of lemon, lipids, volatile flavors, probiotics, nutraceuticals, seeds of fruits like banana, grapes, guava, papaya, apple, blackberry, granadilla and citrus seeds. In this regard, the encapsulation offers great scope for conservation, germination and exchange of several fruit species, resulting in promising technique for the conservation, transport of transgenic plants and not seed-producing plants, lactase, colorants, enzymes, phytosterols, lutein, fatty acids, plant pigments, antioxidants, aromas and oleoresins, vitamins and mineral [14].

9. Pigments

Pigments are compounds very sensitive due to their instability in the presence of light, air, humidity and high temperatures therefore their use requires a chemical knowledge of their molecules and stability, in order to adapt them to the conditions of use during processing, packaging and distribution. One alternative for their use in the food industry is microencapsulation technology [80]. Carotenoids are used as dyes in food, beverages, cosmetics and animal feed, mainly poultry and fish. During the processing and storage, carotenoids can easily change in different isomers geometric and rust, this result in the reduction or loss of the dye and its biological properties. The main alternatives of applications to increase the stability of carotenoids and, allow its incorporation in hydrophilic environments, is the technique of microencapsulation by the method of spray called spray drying. In the same way, other pigments such as licopeno, lutein, enocianin, astaxantin, antocianins and pigments of nogal and urucú have also been encapsulated [14].

10. Vitamins

Both lipid-soluble (e.g. vitamin A, β -carotene, vitamins D, E and K) and water-soluble (e.g. ascorbic acid) vitamins can be encapsulated using various technologies. The most common reason for encapsulating these ingredients is to extend the shelf-life, either by protecting them against oxidation or by preventing reactions with components in the food system in which they are present. A good example is ascorbic acid (vitamin C), which is added extensively to a variety of food products as either an antioxidant or a vitamin supplement. Its application as a vitamin supplement is impaired by its high reactivity and, hence, poor stability in solution. It can degrade by a variety of mechanisms. For vitamin C encapsulation, both spray-cooling or spray-chilling and fluidized-bed coating can be used when the vitamins are added to solid foods, such as cereal bars, biscuits or bread. For application in liquid food systems, the best way to protect water-soluble ingredients is by encapsulation in liposomes. Liposomes are single or multilayered vesicles of phospholipids containing either aqueous-based or lipophilic compounds. Lipid-soluble vitamins such as vitamin A, β -carotene and vitamins D, E or K are much easier to encapsulate than water-soluble ingredients. A commonly-used procedure is spray-drying of emulsions [81].

11. Minerals

From a nutritional point of view, the iron is one of the most important elements, and its deficiency affects about one-third of the world's population. The best way to prevent this problem is through the iron fortification of foods. However, the bioavailability of iron is negatively influenced by interactions with food ingredients such as tannins, phytates and polyphenols. Moreover, the iron catalyses oxidative processes in fatty acids, vitamins and amino acids, and consequently alters sensory characteristics and decreases the nutritional value of the food. Microencapsulation can be used to prevent these reactions, although bioavailability should be rechecked carefully. The bioavailability of readily water-soluble iron salts such as FeSO_4 or ferrous lactate is higher than that of poorly water-soluble (e.g. ferrous fumarate) or water-insoluble (e.g. FePO_4) iron. Suitable encapsulation techniques depend on the water solubility of the compound. Liposome technology is the method of choice for iron fortification of fluid food products [81].

12. Polyunsaturated fatty acid

In recent years, have been developing a high consumer preference towards products that possess functional properties. Thus the addition of beneficial substances such as polyunsaturated fatty acids omega 3 and omega 6, to the daily diet of humans has increased significantly since that is associated with the prevention and treatment of heart disease, because they have antithrombotic effects. Also has been associated with inflammatory diseases, autoimmune arthritis and even cancer. However, rich in polyunsaturated fatty acids oils are susceptible to oxidative deterioration and acquire easily bad tastes and odors, also environmental factors as moisture, light and oxygen accelerate its degradation, what he

has done to the food and pharmaceutical industry to seek alternatives to prevent their deterioration. The microencapsulation has been an alternative to avoid the deterioration of oils because it can increase the oxidative stability of these and avoid the formation of products of oxidation of high molecular weight, in addition to mask unwanted flavors and aromas. It also gives some properties such as ease of handling and mixing, dispersion, and improvement of the consistency of the product during and after processing [82]. Advances in technologies of microencapsulation and the strategies used in its production have resulted in an increasing number of successful products fortified with omega-3 in the market [83], such as: dietary supplements, dairy, snacks, infant formulas and foods for babies, bakery products and beverages [84].

13. Probiotic bacteria

The microencapsulation has been successfully used to improve the survival of probiotic bacteria in dairy products such as cheese. Ozer et al. [85] demonstrated that colonies of *Lactobacillus ramosus* microencapsulated in a matrix of alginate maintained their viability over 48 h at pH 2, in comparison with free (without encapsulate) cells that were inactivated completely under the same conditions; Another example related to dairy products is the yogurt, in which Bifidobacteria are encapsulated to increase viability in this fermented beverage; Also the whey, the liquid product obtained during the preparation of the cheese can be dried by spraying for the production of whey powder and/or whey protein concentrates [14]. Bio-functional foods offer physiological health benefits and disease prevention over and above their nutritional contribution. Microencapsulation has become the recent tool used for protecting and delivering bio-actives in the development of bio-functional foods. Probiotic foods are by far the largest functional food market. They provide several health benefits including immune-stimulation. Viability, physiological and metabolic activity of these bio actives in a food product at the point of sale are important consideration for their efficacy, as they have to survive during shelf life of a food, transit through high acidic and alkaline conditions in the gastro-intestinal tract. Microencapsulation is an inclusion technique for entrapping a bio-functional nutrient or bio-active compound such as probiotic bacteria, folic acid and protease enzymes into a polymeric (gelled) matrix that may be coated by one or more semi-permeable polymers, by virtue of which the encapsulated substance become more stable than the free one [86].

14. Microencapsulation methods

Several encapsulation processes are based on making first droplets of the active compound (in gas, liquid or powder form) and these droplets are subsequently surrounded by the carrier material in a gas or liquid phase via different methods. Excellent reviews on the encapsulation processes have been published in the last years [12-14, 87-88]. For this reason, this section will only show the most commonly used methods in microencapsulation and the steps involved through a list (see Table 5).

Methods	Process steps	Morphology	Load (%)	Particle size (μm)
Spray-drying	Disperse or dissolve active in aqueous coating solution Atomize Dehydrate	Matrix	5-50	10-400
Fluid bed coating	Fluidize active powder Spray coating Dehydrate or cool	Reservoir	5-50	5-5,000
Spray-chilling/cooling	Disperse or dissolve active in heated lipid solution Atomize Cool	Matrix	10-20	20-200
Melt injection	Melt the coating Disperse or dissolve active in the coating Extrude through filter Cooling and dehydrating	Matrix	5-20	200-2,000
Emulsification	Dissolve active and emulsifiers in water or oil phase Mix oil and water phases under shear	Matrix	1-100	0.2-5,000
Preparation of emulsions with multilayers	Prepare o/w emulsions with lipophilic active in oil phase and ionic emulsifiers Mix with aqueous solution containing oppositely charged poly-electrolytes Remove excess of free poly-electrolytes (option) Repeat steps 2 and 3	Reservoir	1-90	0.2-5,000
Coacervation	Prepare o/w emulsions with lipophilic active in oil phase Mix under turbulent conditions Induce three immiscible phases Cool Crosslink (optionally)	Reservoir	40-90	10-800
Preparation of microspheres via extrusion or dropping	Dissolve or disperse active in alginate solution Drop into gelling bath	Matrix	20-50	200-5,000
Preparation of microspheres via emulsification	Emulsify water with biopolymer in oil phase	Matrix	20-50	10-1,000
Co-extrusion	Dissolve or disperse active in oil Prepare aqueous or fat coating Use an concentric nozzle, and press simultaneously the oil phase through the inner nozzle and the water phase through the outer one Drop into gelling or cooling bath	Reservoir	70-90	150-8,000
Encapsulation by rapid expansion of supercritical solution (RESS)	Create a dispersion of active and dissolved or swollen shell material in supercritical fluid Release the fluid to precipitate the shell onto the active	Matrix	20-50	10-400
Freeze- or vacuum drying	Dissolve or disperse active agent and carrier material in water Freeze the sample Drying under low pressure Grinding (option)	Matrix	Various	20-5,000
Preparation of nanoparticles	Various methods	Various	Various	0.1-1

Adapted from Zuidam and Shimoni [12]

Table 5. Overview of the most common microencapsulation processes

15. Advantages and disadvantages of the microencapsulation

According to Zuidam and Shimoni [12] the possible advantages and disadvantages of microencapsulated active compounds in the food industry could be:

Advantages

1. Superior handling of the active agent (e.g., conversion of liquid active agent into a powder, which might be dust free, free flowing, and might have a more neutral smell).
2. Immobility of active agent in food processing systems.
3. Improved stability in final product and during processing (i.e., less evaporation of volatile active agent and/or no degradation or reaction with other components in the food product such as oxygen or water).
4. Improved safety (i.e., reduced flammability of volatiles like aroma, no concentrated volatile oil handling).
5. Creation of visible and textural effects (visual cues).
6. Adjustable properties of active components (particle size, structure, oil- or water-soluble, color).
7. Off-taste masking.
8. Controlled release (differentiation, release by the right stimulus).

Disadvantages

1. Additional costs.
2. Increased complexity of production process and/or supply chain.
3. Undesirable consumer notice (visual or touch) of encapsulates in food products.
4. Stability challenges of encapsulates during processing and storage of the food product.

16. Conclusions and perspectives

Since, very good results have been obtained on enhancing overall quality, browning, oxidation and softening, shelf-life extension, control of decay, and nutraceutical benefits with a number of diverse additives and bioactive compounds incorporated into edible films and coatings designed for different foods, could be useful to consider that the use the edible films and coating as carrier of food additives and bioactive compounds represent a good alternative for food industry to improve the quality and safety of the food, as well as to offer new product to consumers.

Many companies and research institutes are looking for new ingredients with possible health benefits. Ingredients such as phytochemicals, wood-derived ingredients such as phytosterols, pro- and prebiotics, new types of carotenoids, trace minerals and polyphenols will be available in next the years. Microencapsulation will certainly play an important role in this process, although it will always make an ingredient more expensive to use and bioavailability should always be considered carefully.

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17. References

- [1] Martín-Belloso O, Soliva-Fortuny R, Oms-Oliu G (2006) Fresh-cut fruits. In: Hui Y.H, editor. Handbook of fruits and fruit processing. Iowa, USA: Blackwell Publishing. pp. 129-144.
- [2] Olivas G.I, Barbosa-Cánovas G.V (2005) Edible coatings for fresh-cut fruits. Crit. Rev. Food Sci. Nutr. 45(7-8): 657-670.
- [3] Min B, Ahn D.U (2005) Mechanism of lipid peroxidation in meat and meat products – A review. Food Sci. Biotechnol. 14(1): 152-163.
- [4] Fasseas, M.K, Mountzouris K.C, Tarantilis P.A, Polissiou M, Zervas G (2007) Antioxidant activity in meat treated with oregano and sage essential oils. Food Chem. 106: 1188-1194.
- [5] Cerqueira M.A, Lima A.M, Souza B.W.S, Texeira J.A, Moreira R.A, Vicente A.A, (2009) Functional polysaccharides as edible coating for cheese. J. Agric. Food Chem. 57: 1456-1462.
- [6] Gennadios A, Hanna M.A, Kurth L.B (1997) Application of edible coatings on meats, poultry and seafoods: A review. LWT. 30: 337-350.
- [7] Martín-Belloso O, Rojas-Graü M.A, Soliva-Fortuny R (2009) Delivery of flavor and active ingredients using edible films and coatings. In: Embuscado M.E, Huber K.C, editors. Edible films and coating for food applications. Chapter 10. New York: Springer Science + Business Media, LLC. pp. 295-313.
- [8] Raybaudi-Massilia R.M, Mosqueda-Melgar J, Tapia M.S (2010) Edible coatings as carriers of food additives on fresh-cut fruits and vegetables. Stewart Postharvest Rev. 3: 1-7.
- [9] Rojas-Graü M.A, Soliva-Fortuny R.C, Martín-Belloso O (2009) Edible coatings to incorporate active ingredients to fresh-cut fruits: a review. Trends Food Sci. Technol. 20:438-447.
- [10] Zhao Y (2010) Edible Coatings for enhancing quality and health benefits of berry fruits. In: Qian M.C, Rimando A.M, editors. Flavor and Health Benefits of Small Fruits. Chapter 18, Volume 1035. Washington DC, USA: ACS Symposium Series. pp 281-292.
- [11] Tapia M.S, Rojas-Graü M.A, Rodríguez F.J, Ramírez J, Carmona A, Martín-Belloso O (2007) Alginate- and gellan-based edible films for probiotic coatings on fresh-cut fruits. J. Food Sci. 72: E190-E196.
- [12] Zuidam N.J, Shimoni E (2010) Overview of microencapsulates for use in food products or processes and methods to make them. In: Zuidam N.J, Nedovic V.A, editors. Encapsulation technologies for active food ingredients and food processing. Chapter 2. New York: Springer Science + Business Media, LLC. pp. 3-29.

- [13] Dubey R, Shami T.C, Brasker-Rao K.U (2009) Microencapsulation technology and application. *Defence Sci. J.* 59(1): 82-95.
- [14] Parra-Huertas R.A (2011) Revisión: Microencapsulación de alimentos. *Rev. Fac. Nal. Agr. Medellín.* 63(2): 5669-5684.
- [15] Raybaudi-Massilia R.M, Mosqueda-Melgar J (2009) Factors affecting the incidence of pathogenic microorganisms of fresh-cut produce. *Stewart Postharvest Rev.* 4: 1-8.
- [16] Cutter C.N, Siragusa G.R (1997) Growth of *Brochothrix thermosphacta* in ground beef following treatments with nisin in calcium alginate gels. *Food Microbiol.* 14: 425-430.
- [17] Natrajan N, Sheldon B (2000) Inhibition of *Salmonella* on poultry skin using protein and polysaccharide-based films containing a nisin formulation. *J Food Prot.* 63(9): 1268-1272.
- [18] Millette M, Le Tien C, Smoragiewicz W, Lacroix M (2007) Inhibition of *Staphylococcus aureus* on beef by nisin-containing modified alginate films and beads. *Food Contr.* 18: 878-884.
- [19] Datta S, Janes M.E, Xue Q.G, Losso J, La Peyre J.F (2008) Control of *Listeria monocytogenes* and *Salmonella anatum* on the Surface of Smoked Salmon Coated with Calcium Alginate Coating containing Oyster Lysozyme and Nisin. *J. Food Sci.* 73(2): M67-M71.
- [20] Neetoo H, Ye M, Chen H (2010) Bioactive alginate coatings to control *Listeria monocytogenes* on cold-smoked salmon slices and fillets. *Int. J. Food Microbiol.* 136(3): 326-331.
- [21] Marcos B, Aymerich T, Monfort J.M, Garriga M (2008) High-pressure processing and antimicrobial biodegradable packaging to control *Listeria monocytogenes* during storage of cooked ham. *Food Microbiol.* 25: 177-182.
- [22] Oussalah M, Caillet S, Salmiéri S, Saucier L, Lacroix M (2006). Antimicrobial effects of alginate-based film containing essential oils for the preservation of whole beef muscle. *J. Food Prot.* 69: 2364-2369.
- [23] Oussalah M, Caillet S, Salmieri S, Saucier L, Lacroix M (2007) Antimicrobial effects of alginate-based film containing essential oils o *Listeria monocytogenes* and *Salmonella thyphimurium* present in bologna and ham. *J. Food Prot.* 70(4): 901-908.
- [24] Rojas-Graü M.A, Raybaudi-Massilia R.M, Soliva-Fortuny R.C, Avena-Bustillos R.J, McHugh T.H, Martín-Belloso O (2007) Apple puree-alginate edible coating as carrier of antimicrobial agents to prolong shelf life of fresh-cut apples. *Postharvest Biol. Technol.* 45: 254-264.
- [25] Raybaudi-Massilia R.M, Mosqueda-Melgar J, Martín-Belloso O (2008) Edible alginate-based coating as carrier of antimicrobials to improve shelf life and safety of fresh-cut melon. *Int. J. Food Microbiol.* 121: 313-327.
- [26] Raybaudi-Massilia R.M, Rojas-Graü M.A, Mosqueda-Melgar J, Martín-Belloso O (2008) Comparative study on essential oils incorporated into an alginate-based edible coating to assure the safety and quality of fresh-cut Fuji apples. *J. Food Prot.* 71: 1150-1161.
- [27] Olivás G.I, Mattinson D.S, Barbosa-Cánovas G.V (2007) Alginate coatings for preservation of minimally processed 'Gala' apples. *Postharvest Biol. Technol.* 45: 89-96.
- [28] Seol, K.H., Lim, D.G., Jang, A., Jo, C., y Lee, M. (2009). Antimicrobial effect of κ -carrageenan-based edible film containing ovotransferrin in fresh chicken breast stored at 5 °C. *Meat Science* 83(3): 479-483.

- [29] Jiang Z, Neetoo H, Chen H (2011) Efficacy of freezing, frozen storage and edible antimicrobial coating used in combination for control of *Listeria monocytogenes* on roasted turkey stored at chiller temperatures. *Food Microbiol.* 28(7): 1394-1401.
- [30] Jiang Z, Neetoo H, Chen H (2011) Control of *Listeria monocytogenes* on cold-smoked salmon using chitosan-based antimicrobial coatings and films. *J. Food Sci.* 76(1): M22-M26.
- [31] Dutta P.K, Tripathi S, Mehrotra G.K, Dutta J (2009) Perspectives for chitosan based antimicrobial films in food applications. *Food Chem.* 114(4): 1173-1182.
- [32] Ouattara B, Simard R.E, Piette G, Bejín A, Holler R.A (2000) Inhibition of surface spoilage bacteria in processed meats by application of antimicrobial films prepared with chitosan. *Int. J. Food Microbiol.* 62: 139-148.
- [33] Park S.L, Stan S.D, Daeschel M.A, Zhao Y (2006) Antifungal coatings on fresh strawberries (*Fragaria × ananassa*) to control mold growth during cold storage. *J. Food Sci.* 70(4): M202-M207.
- [34] Duan J, Park S, Daeschel M, Zhao Y (2007). Antimicrobial chitosan-lysozyme (CL) films and coatings for enhancing microbial safety of mozzarella cheese. *J. Food Sci.* 72(9): M355-M362.
- [35] Sangsuwan J, Rattanapanone N, Rachtanapun P (2008) Effect of chitosan/methyl cellulose films on microbial and quality characteristics of fresh-cut cantaloupe and pineapple. *Postharvest Biol. Technol.* 49: 403-410.
- [36] Ye M, Neetoo H, Chen H (2008) Control of *Listeria monocytogenes* on ham steaks by antimicrobials incorporated into chitosan-coated plastic films. *Food Microbiol.* 25(2): 260-268.
- [37] Ojagh S.M, Rezaei M, Razavi S.H, Hashem-Hosseini S.M (2010) Effect of chitosan coatings enriched with cinnamon oil on the quality of refrigerated rainbow trout. *Food Chem.* 120(1): 193-198.
- [38] Fajardo P, Martins J.T, Fuciños C, Pastrana L, Teixeira J.A, Vicente A.A (2010) Evaluation of a chitosan-based edible film as carrier of natamycin to improve the storability of Saloio cheese. *J. Food Eng.* 101(4): 349-356.
- [39] Siripatrawan U, Noipha S (2012). Active film from chitosan incorporating green tea extract for shelf life extension of pork sausages. *Food Hydrocolloids* 27(1): 102-108.
- [40] Franklin, N.B., Cooksey, K.D., y Getty, K.J.K. (2004). Inhibition of *Listeria monocytogenes* on the Surface of Individually Packaged Hot Dogs with a Packaging Film Coating Containing Nisin. *Journal of Food Protection* 67(3): 480-485
- [41] Nguyen T, Gidley M, Dykes G (2008). Potential of a nisin-containing bacterial cellulose film to inhibit *Listeria monocytogenes* on processed meats. *Food Microbiol.* 25(3): 471-478.
- [42] Santiago-Silva P, Soares N.F.F, Nóbrega J.E, Júnior M.A.W, Barbosa K.B.F, Volp A.C.P, Zerdas E.R.M.A, Würllitzer N.J (2009) Antimicrobial efficiency of film incorporated with pediocin (ALTA®2351) on preservation of sliced ham. *Food Contr.* 20: 85-89.
- [43] Sayanjali S, Ghanbarzadeh B, Ghiassifar S. 2011. Evaluation of antimicrobial and physical properties of edible film based on carboxy methyl cellulose containing potassium sorbate on some mycotoxigenic *Aspergillus* species in fresh pistachios. *LWT.* 44(4): 1133-1138.

- [44] Valencia-Chamorro S.A, Pérez-Gago M.B, del Río M.A, Palou L (2009) Effect of antifungal hydroxy propyl methyl cellulose (HPMC)–lipid edible composite coatings on postharvest decay development and quality attributes of cold-stored “Valencia” oranges. *Postharvest Biol. Technol.* 54(2): 72-79.
- [45] Durango A.M, Soares N.F.F, Andrade N.J (2006) Microbiological evaluation of an edible antimicrobial coating on minimally processed carrots. *Food Contr.* 17(5): 336-341.
- [46] Chiu P.E, Lai L.S (2010) Antimicrobial activities of tapioca starch/decolorized hsian-tsoa leaf gum coatings containing green tea extracts in fruit-based salads, romaine hearts and pork slices. *Int. J. Food Microbiol.* 139(1-2): 23-30.
- [47] Kang H.J, Jo C, Kwon J.H, Kim J.H, Chung H.J, Byun M.W (2007) Effect of a pectin-based edible coating containing green tea powder on the quality of irradiated pork patty. *Food Contr.* 18(5): 430-435.
- [48] Handley D, Ma-Edmonds M, Hamouz F, Cuppett S, Mandigo R, Schnepf M (1996) Controlling oxidation and warmed-over flavor in precooked pork chops with rosemary oleoresin and edible film. In: Shahidi F, editor. *Natural antioxidants chemistry, health effect, and application.* Champaign, USA: AOCS Press. pp. 311-318.
- [49] Ma-Edmonds M, Hamouz F, Cuppett S, Mandigo R, Schnepf M (1995) Use of rosemary oleoresin and edible film to control warmed-over flavor in precooked beef patties [abstract]. In: *IFT Annual Meeting Book of Abstracts.* Chicago, Illinois: Institute of Food Technologists. pp. 139. Abstract N° 50-6.
- [50] Hargens-Madsen M, Schnepf M, Hamouz F, Weller C, Roy S (1995) Use of edible films and tocopherols in the control of warmed-over flavor. *J. Am. Diet. Assoc.* 95: A-41.
- [51] Wu Y, Weller C.L, Hamouz F, Cuppett S, Schnepf M (2001) Moisture loss and lipid oxidation for precooked ground-beef patties packaged in edible starch-alginate-based composite films. *J. Food Sci.* 66(3): 486-493.
- [52] Atarés L, Pérez-Masiá R, Chiralt A (2011). The role of some antioxidants in the HPMC film properties and lipid protection in coated toasted almonds. *J. Food Eng.* 104(4): 649-656.
- [53] Song Y, Liu L, Shen H, You J, Luo Y (2011) Effect of sodium alginate-based edible coating containing different anti-oxidants on quality and shelf life of refrigerated bream (*Megalobrama amblycephala*). *Food Contr.* 22(3-4): 608-615.
- [54] Lin D, Zhao Z (2007) Innovations in the development and application of edible coatings for fresh and minimally processed fruits and vegetables. *CRFSFS.* 6: 60-75.
- [55] Ahvenainen R (1996) Improving the shelf life of minimally processed fruit and vegetables. *Trends Food Sci. Tech.* 7: 179-187.
- [56] Friedman, M. Food browning and its prevention: Overview. *Journal of Agricultural and Food Chemistry* 1996: 44(3):631–652.
- [57] Baldwin E.A, Nisperos M.O, Chen X, Hagenmaier R.D (1996) Improving storage life of cut apple and potato with edible coating. *Postharvest Biol. Technol.* 9: 151-163.
- [58] Brancoli N, Barbosa-Cánovas G.V (2000) Quality changes during refrigerated storage of packaged apple slices treated with polysaccharide films. In: Barbosa-Cánovas G.V, Gould G.W, editors. *Innovations in food processing.* Pennsylvania: Technomic Publishing Co. pp. 243-254.

- [59] Lee J.Y, Park H.J, Lee C.Y, Choi W.Y (2003) Extending shelf-life of minimally processed apples with edible coatings and antibrowning agents. *LWT*. 36: 323-329.
- [60] Bico S.L.S, Raposo M.F.J, Morais R.M.S.C, Morais A.M.M.B (2009) Combined effects of chemical dip and/or carrageenan coating and/or controlled atmosphere on quality of fresh-cut banana. *Food Contr*. 20: 508-514.
- [61] Rojas-Graü M.A, Tapia M.S, Rodríguez F.J, Carmona A.J, Martín-Belloso O (2007) Alginate and gellan based edible coatings as support of antibrowning agents applied on fresh-cut Fuji apple. *Food Hydrocolloids* 21: 118-127.
- [62] Rojas-Graü M.A, Tapia M.S, Martín-Belloso O (2008) Using polysaccharide-based edible coatings to maintain quality of fresh-cut Fuji apples. *LWT*. 41: 139-147.
- [63] Oms-Oliu G, Soliva-Fortuny R, Martín-Belloso O (2008) Edible coatings with antibrowning agents to maintain sensory quality and antioxidant properties of fresh-cut pears. *Postharvest Biol. Technol*. 50: 87-94.
- [64] García E, Barret D.M (2002) Preservative treatments for fresh cut fruits and vegetables. In: Lamikanra O, editor. *Fresh-Cut Fruits and Vegetables*. Florida: CRC Press. pp. 267-304.
- [65] Harker F.R, Redgwell R.J, Hallett I.C, Murray S.H, Carter G (1997) Texture of fresh fruit. *Horticultural Rev*. 20: 121-224.
- [66] Soliva-Fortuny R.C, Martín-Belloso O (2003) New advances in extending the shelf life of fresh-cut fruits: a review. *Trends Food Sci. Technol*. 14: 341-353.
- [67] Oms-Oliu G, Soliva-Fortuny R, Martín-Belloso O (2008) Using polysaccharide-based edible coatings to enhance quality and antioxidant properties of fresh-cut melon. *LWT*. 41: 1862-1870.
- [68] Tapia M.S, Rojas-Graü M.A, Carmona A, Rodríguez F.J, Soliva-Fortuny R, Martín-Belloso O (2008) Use of alginate and gellan-based coatings for improving barrier, texture and nutritional properties of fresh-cut papaya. *Food Hydrocolloids*. 22: 1493-1503.
- [69] Mei Y, Zhao Y (2003) Barrier and Mechanical Properties of Milk Protein-Based Edible Films Containing Nutraceuticals. *J. Agr. Food Chem*. 51(7): 1914-1918.
- [70] Elliott J.G (1998) Application of antioxidant vitamins in foods and beverages. *Food Technol*. 53: 46-48.
- [71] Mei Y, Zhao Y, Yang J, Furr H.C (2002) Using edible coating to enhance nutritional and sensory qualities of baby carrots. *J. Food Sci*. 67(5): 1964-1968.
- [72] Hernández-Muñoz P, Almenar E, Ocio M.J, Gavara R (2006) Effect of calcium dips and chitosan coatings on postharvest life of strawberries (*Fragaria x ananassa*). *Postharvest Biol. Technol*. 39: 247-253.
- [73] Han C, Zhao Y, Leonard S.W, Traber M.G (2004) Edible coatings to improve storability and enhance nutritional value of fresh and frozen strawberries (*Fragaria x ananassa*) and raspberries (*Rubus ideaus*). *Postharvest Biol. Technol*. 33(1): 167-178.
- [74] Duan J, Cherian G, Zhao Y (2010). Quality enhancement in fresh and frozen lingcod (*Ophiodon elongates*) fillets by employment of fish oil incorporated chitosan coatings. *Food Chem*. 119(2): 524-532.

- [75] Naidu A.S, Bidlack W.R, Clemens R.A (1999) Probiotic spectra of lactic acid bacteria (LAB). *Crit. Rev. Food Sci. Nutr.* 38: 113-126.
- [76] Giancone T (2007) Hydrocolloid-based edible films: composition-structure-properties relationship. Thesis Doctoral. Università degli Studi di Napoli Federico II. Department of Food Science. Available: http://www.fedoa.unina.it/1669/1/Giancone_Sienze_Tecnologie.pdf. Accessed 2012 Feb 10.
- [77] Gontard N, Guilbert S (1999) Bio-packaging: technology and properties of edible and/or biodegradable material of agricultural origin. In: Mathlouthi M, editor. *Food packaging and preservation*. Chapter 9. Gaithersburg, USA: Aspen Publisher, Inc. pp. 159-181.
- [78] Pavlath A.E, Orts W (2009). Edible films and coatings: Why, what, and how?. In: Embuscado M.E, Huber K.C, editors. *Edible films and coatings for food applications*. Chapter 1. New York: Springer Science + Business Media, LLC. pp. 1-24.
- [79] Chien P.J, Sheu F, Yang F.H (2007) Effects of edible chitosan coating on quality and shelf life of sliced mango fruit. *J. Food Eng.* 78: 225-229.
- [80] Parize A, Rozone T, Costa I, Fávere V, Laranjeira M, Spinelli A, Longo E (2008) Microencapsulation of the natural urucum pigment with chitosan by spray-drying in different solvents. *African J. Biotechnol.* 7(17): 3107-3114.
- [81] Schrooyen P, Meer R, Kruif C (2001) Microencapsulation: its application in nutrition. *Proc. Nutr. Soc.* 60(4): 475-479.
- [82] Quispe-Arpasi D, Matos-Chamorro A, Quispe-Condori S (2011) Microencapsulación de aceites y su aplicación en la industria de alimentos. I Congreso Nacional de Investigación, Lima, Perú. November 2-4, 2011. Available: <http://papiros.upeu.edu.pe/bitstream/handle/123456789/180/CIn33Articulo.pdf?sequence=1> Accessed 2012 April 10.
- [83] Smith J, Charter E (2010) *Functional food product development*. United Kingdom: Blackwell Publishing Ltd. 505 p.
- [84] Sanguansri L, Augustin M (2007) Microencapsulation and delivery of Omega-3 fatty acids. In: Shi J, editor. *Functional food ingredients and nutraceuticals: Processing technologies*. Florida, USA: Taylor & Francis. pp. 297-327.
- [85] Ozer B, Avni H, Senel E, Atamer M, Hayaloglu A (2009) Improving the viability of *Bifidobacterium bifidum* BB-12 and *Lactobacillus acidophilus* LA-5 in white-brined cheese by microencapsulation. *Int. Dairy J.* 19(1): 22-29.
- [86] Kailasapathy K, Madziva H, Anjani K, Seneweera S, Phillips M (2006) Recent trends in the role of micro encapsulation in the development of bio-functional foods. XIVth International Workshop on Bioencapsulation, Lausanne, USA. Available: http://impascience.eu/bioencapsulation/340_contribution_texts/2006-10-05_O7-1.pdf. Accessed 2012 April 10.
- [87] Gibbs B.F, Kermasha S, Alli I, Mulligan C.N (1999) Encapsulation in the food industry: A review. *Int. J. Food Sci. Nutr.* 50: 213-224.
- [88] Desobry S, Debeaufort F (2011) Encapsulation of flavors, nutraceuticals, and antibacterials. In: Baldwin E.A, Hagenmaier R, Bai J, editors. *Edible coatings and films to improve food quality*. 2nd Edition. Chapter 11. Boca Raton, USA: CRC Press. pp. 333-372.