

Antimicrobial Coatings for Ensuring Safety of Fresh Produces

Sean X. Liu*

Cereal Products and Food Science Research Unit, National Center for Agricultural Utilization Research, US Department of Agriculture,
1815 N. University Street, Peoria, IL 61604, USA
Correspondence: * Sean.Liu@ARS.USDA.GOV

ABSTRACT

Safety of fresh produce has been a perennial issue for the industry in the US despite tightening up regulations and implementing good manufacturing practice. The diversity of crops and labor-intense operations in the fresh produce production created a unique set of contamination routes that are not common in other food productions. New technologies and procedures have been developed to minimize the occurrences of in-production and shipping/distribution contaminations; however, in order to be effective, these proactive measures have to be implemented and practices consistently, which is not totally reassuring given the inevitability of human errors or occasional incompetence. As a consequence, in recent years, many researchers have been looking into development of passive protection of fresh produce from contamination through antimicrobial coatings of fresh produces or antimicrobial packaging materials that are used in fresh produce packing. This review critically examines the current technologies and developments in antimicrobial coatings and antimicrobial food packaging materials as a food safety tool for fresh produce producers.

Keywords: active packaging; chitosan, edible films, food safety, fruits and vegetables

CONTENTS

INTRODUCTION.....	73
ANTIMICROBIAL FOOD PACKAGING MATERIALS	74
EDIBLE FILMS OR COATINGS WITH INTRINSIC ANTIMICROBIAL PROPERTY	75
EDIBLE FILMS OR COATINGS CONTAINING ANTIMICROBIAL AGENTS	76
Sensory issues with edible packaging films or coatings	77
Regulatory issues.....	77
CONCLUDING REMARKS	77
DISCLAIMER	77
REFERENCES.....	77

INTRODUCTION

Recent outbreaks of microbiologically contaminated fresh produces have heightened consumer concerns about safety of fresh produces, which generally are not cooked prior to consumption and sometimes not even thoroughly washed after taking home by consumers. These concerns have from time to time depressed the demand of a fresh produce that is recalled due to suspected contamination. The economic toll as a result of recall as well as human suffering as a result of food poisoning is substantial. Current remedy to the problem focuses on sound management practices during growing season and during post-harvesting processing. This proactive remedy of fending off microbial contamination of fresh produces works in theory and in most practical cases; however, there are many post-harvesting processing steps that could lead to contamination if the procedures of processing and workers' hygiene are not strictly followed (review of approaches to improving shelf life of fresh produces can be found elsewhere; for example, a 1996 paper by Ahvenainen). In the United States, majority of fresh produces are produced by a handful of states in the west region of the country or imported from South America. Even in the U.S., agricultural workers who tender crops and perform post-harvesting processing are mainly migrant workers (documented or undocumented) from Latin America with high turnover rate. Their on-the-job training of food safety

and handling is not always complete or even adequate. As such, there is always a potential danger that something somewhere could go wrong with devastating consequence.

The vulnerability of fresh produces to microbial contamination has generated a lot of interests in developing passive protection schemes against microbial contaminations among researchers and food processors alike. At fundamental level, the safety issue of fresh produces is parallel to the safety issue of automobiles. It is true that seatbelt has saved countless lives but not all drivers and their passengers wear them all the time. The advent of airbags provides a passive means of protecting drivers and passengers in the case that the seatbelts are not wore or not sufficient to counter the impact of car collisions. The equivalent of "airbags" in cars to fresh produce safety is introduction of anti-microbial agents for fresh produces, either directly coated onto food or as a part of food packaging materials.

Unlike other food items, fresh produces continue to respire and transpire after harvesting; oxygen is used and carbon dioxide and ethylene are produced as metabolism products when fresh produces are in storage (Hardenburg *et al.* 1986). Processing of fresh produces, even minimal processing, could increase the respiration activity 1.2-7.0-fold (Ahvenainen 1996). As are result, many fresh produces are commonly packaged to provide effective physical barrier to different environmental hazards such as oxygen, relative humidity that could reduce the shelf life of a fresh produce

product. These packaging materials of fresh produces can also be used as the platform for incorporation of antimicrobial agents.

This paper provides a critical review of current research and development of edible anti-microbial coatings and non-edible anti-microbial packaging materials relevant to the fresh produce industry in recent decades.

ANTIMICROBIAL FOOD PACKAGING MATERIALS

Food packaging materials provide more than just a barrier that wraps a food item in a neat container that does not interact with food itself; it also serves as barriers to undesirable environmental parameters such as oxygen, moisture and water vapor while maintaining mechanical integrity of the package for transportation and visual presentation. The most common packaging materials for foods and particularly fresh produces are plastics. Not all plastics are the same in terms of providing barrier properties; different materials function differently in terms of barrier properties. For example, polyethylene is used mainly for sealing and as a water vapor barrier; polypropylene provides good mechanical properties and water vapor barrier; polyamide offers aroma- and oxygen-barrier with stiffness; EVOH excels as high oxygen barrier; EVA is excellent for sealing; while mLLDPE presents good optical and mechanical properties. In the case of preventing ethylene from accumulating around fresh fruits, none of the above materials excels; there are several commercial products that utilize either ethylene adsorbing layer in a multi-layered packaging material or ethylene-adsorbing sachet in the package to slow down ripening of fresh fruits. Although polymers still dominate food packaging material market, there is urgent push for developing bio-based food packaging materials. The benefit of uses of bio-based materials for food packaging is two-fold: it reduces amount of plastics that end up in landfill or incinerators; it also reduces the use of petroleum products. The key to success of bio-based packaging materials lies in the cost-structure and mechanical and barrier properties of the biodegradable materials. Majority of these bio-based packaging materials suffer from deficiency either in mechanical properties or properties of water vapor, oxygen and chemical barrier. Current research on bio-based materials for food packaging has been expertly and succinctly reviewed by Siracusa *et al.* (2008).

However, as far as prevent fresh produces from microbial contamination is concerned, none of these materials themselves alone, whether plastics or bio-based materials (with the exception of chitosan-based materials), can accomplish the task; antimicrobial agents or materials with native antimicrobial properties have to be incorporated into the packaging. Incorporating antimicrobial agents into food package materials or development of antimicrobial packaging films has been researched for a number of years. It is a part of new packaging technology trend called Active Packaging. Active packaging is designed to perform certain beneficial functions other than merely an inert barrier (López-Rubio *et al.* 2004). These beneficial functions, as was referred in the beginning of this paragraph, include providing barriers to oxygen and moisture migrations and scavenging of undesirable gases and fluids in the food matrix as well as inhibiting microbial spoilages through incorporation of antimicrobial agents that are either bound or incorporated into packaging materials. The main thrust of this technology is to deploy antimicrobial packaging materials to prevent microbial contamination of food in the package (Ghosh *et al.* 1977; Vojdani and Torres 1990; Rico-Pena and Torres 1991; Weng and Hotchkiss 1992, 1993; Chen *et al.* 1996; Han and Floros 1998; Devlieghere *et al.* 2000; Han 2000; Ouattara *et al.* 2000; Cooksey 2001; Kim *et al.* 2003; Chung *et al.* 2003; Sebt *et al.* 2003a, 2003b; Lee *et al.* 2004a; Jofré *et al.* 2007; Lagaron *et al.* 2007; Chawengkijwanich and Hayata 2008; Chollet *et al.* 2008; Guiga *et al.* 2008; Rodríguez *et al.* 2008; Gutiérrez *et al.* 2009). The antimicrobial agents in the packaging materials

are generally non-toxic; some are bio-based food-related materials; they are approved for food or pharmaceutical uses or considered as Generally Recognized As Safe (GRAS) by U.S. FDA. The incorporation of non-toxic antimicrobial agents into packaging materials can inhibit or retard surface microbial growth on food as the agent is leached or released from the packaging material into the surface of the food product. The antimicrobial agent needs to reach adequate concentration on the food surface to inhibit or retard microbial growth since microbial contamination of most foods occurs primarily at the surface due to post-harvesting handling. These embedded agents work best where the packaging material touches the surface of food product. Thus, vacuum packaging of foods is ideal mode of packaging for an antimicrobial packaging material as the material is in intimate contact with the surface of the food product. There are other possible ways of ensuring delivery of adequate concentration of the antimicrobial agent on the surface of the food or inner structures of food matrices such as employment of a volatile solvent or other carrier media as well as relying on Brownian motion or molecular diffusion due to concentration gradient or structural peculiarity.

Although published results from the research efforts have showed the technology to be promising, the actual applications of the modified packaging materials are lacking outside of Japan. There are concerns about the safety and efficacy of antimicrobial packaging materials for prolonging shelf life of food. In order to make antimicrobial agents incorporated into packaging materials effective, they need to be coated on the surface of the packaging material that directly contacts food and release slowly to ensure long-lasting effect, or in the case of antimicrobial agent in the matrix of the packaging material, the agent needs to be easily released from the packaging material matrix and preferably, the release kinetics of the antimicrobial agent ensures long, steady release rate of the agent over time with a dosage that inhibits pathogenic or spoiling microorganisms. These are not easy tasks to accomplish with a food packaging system like fresh produces that are not liquid or of neat shape like a slab of beef steak.

The antimicrobial agents that are coated on or “incorporated” into polymeric or paper materials in many published articles and patents are mainly food grade additives that has been used for generations: sodium benzoate, benzoic acid, propionic acid, potassium sorbate, sorbic acid, lacticin, pectin, ethylenediaminetetraacetic acid (EDTA), and enzymes (Chien *et al.* 2007; Coma 2008; Sivarooban *et al.* 2008). More newly emerging antimicrobial agents, primarily from food sources, are getting increasing attention: nisin (a bacteriocin), triclosan, fatty-acids, chitin, chitosan, grape seed extract, spices/essential oils and certain components of essential oils or spices (Chung *et al.* 2003; Kim *et al.* 2003; Limjaroen *et al.* 2003; Sebt *et al.* 2003a; Srinivasa *et al.* 2003; Lee *et al.* 2004b; Salmieri and Lacroix 2006; Arfa *et al.* 2007; Jofré *et al.* 2007; Rodríguez *et al.* 2007; López *et al.* 2007; Coma 2008; Chollet *et al.* 2008; Guiga *et al.* 2008; Rodríguez *et al.* 2008; Gutiérrez *et al.* 2009; Mayachiew *et al.* 2009). Nanoparticles are also increasingly tested for antimicrobial properties (Chawengkijwanich and Hayata 2008); silver nanoparticles have been used commercially in food packaging and composite nano-structured calcium silicate has been suggested to be used as carrier for silver ion to inhibit microbial growth on foods (Johnston *et al.* 2008). Other possible antimicrobial agents that potentially can be used for antimicrobial food packaging are disinfectants. Disinfectants are widely used for inactivating a wide array of harmful microorganisms in food operations and they are used for disinfecting fresh produces. The challenge will be how one can incorporate these disinfectants into food packaging materials with appropriate concentration without compromising the mechanical integrity of the materials as well sensory attributes of fresh produces these packaging materials contain. Titanium dioxide nanoparticle coated oriented-polypropylene film has been tested and it showed that the film caused *E. coli* to inactivation on

cut lettuce under UV light (Chawengkijwanich and Hayata 2008). It is an interesting development but more work needs to be done to address efficacy issue. It should be emphasized that all these studies on novel antimicrobial agents coated on or “incorporated” into plastic or paper packaging materials were not aiming at fresh produces; it is safe to say that most these technologies, if eventually commercialized, will be difficult if not impossible to use on fresh produces, particularly leafy vegetables as most of them require direct contact between food surfaces and packaging materials coated with antimicrobial agents. Volatile components from essential oils may be exception to this generalization; however, there is reasonable concern of the odor from these volatile components that may impart an undesirable sensory attribute to some fresh produces. Furthermore, the cost factor of these antimicrobial packaging materials may relegate the use of them to limited fresh produces. In contrast, edible antimicrobial coatings or films (see the following sections of this article), either based on chitosan/chitin or other food grade materials, may provide a better protection to food safety of fresh produces.

EDIBLE FILMS OR COATINGS WITH INTRINSIC ANTIMICROBIAL PROPERTY

Among all candidates of edible packaging films or coatings, chitosan-based films are unique – they possess intrinsic antimicrobial property. When applied to food surfaces, chitosan-based films can be used directly or in conjunction with other antimicrobial agents as edible antimicrobial packaging materials. This distinctive versatility of chitosan-based films, along with their favorable mechanical properties, increases their range of applications and antimicrobial potency in food packaging. The prospective of chitosan-based food-grade antimicrobial food packaging materials presents an exciting opportunity for the fresh produce industry. Chitosan is a bio-based material that has intrinsic antimicrobial properties as well as the potential of being used as platform materials for antimicrobial food packaging or edible antimicrobial food coating due to its good film-forming properties (Shahidi *et al.* 1999). Chitosan is a polysaccharide derived from chitin found in shells of shrimp and other shellfish and enjoys GRAS status. The positively charged molecule is used in the U.S. as a dietary supplement and is marketed for its claimed ability to increase HDL cholesterol, enhance immune systems, and increase fat elimination from the digestive tract. Chitosan is also widely researched for biomedical science development including drug delivery carriers and surface wound dressing materials (Agnihotri *et al.* 2004). Chitosan, being cationic at neutral or basic condition, is insoluble in water. In acidic condition, the amino groups of chitosan undergo protonation and chitosan becomes soluble. This unique property can be explored for controlled release and development of nano-scale or microscale or even thin-film coating of chitosan systems. Chitosan has been proven to be antimicrobial and can absorb heavy metals, thus making it an excellent candidate for active packaging and edible antimicrobial coating of minimally processed foods (Dutta *et al.* 2008).

There are several proposed mechanisms for intrinsic antimicrobial properties of chitosan and its derivatives (Rabea *et al.* 2003). Interactions between positively charged chitosan molecules and negatively charged microbial cell membranes leads to the leakage of proteinaceous and other intracellular components of the cells (Fernandez-Saiz *et al.* 2009). Chitosan is also believed to function as a chelating agent that selectively combines with trace metal (that is why chitosan is also used in removing heavy metals from wastewater) and thus inhibits the microbial growth and in the case of fungal contamination, the production of mycotoxins (Young *et al.* 1982; Cuero *et al.* 1991). Other possible mechanisms include chitosan binding water molecules therefore denying activities of enzymes of pathogens and chitosan binding DNA and inhibition of mRNA synthesis by penetrating towards the nuclei of the microorganisms

(Sudarshan *et al.* 1992). The bactericidal and fungicidal properties of chitosan vary with the species of microorganisms, the molecular structure and weight of chitosan, the degree of polymerization, the chemical or nutritional composition of substrates as well as environmental conditions. Chitosan is more effective in inhibiting fungal contamination in fresh produces; for example, El Ghaouth *et al.* (1992a) has proven that strawberries coated with chitosan had 60% reduced decay of the fruits by the fungi after 14 days of storage. Not all bacterial food pathogens can be inhibited by chitosan. Even among bacterial species that can be inhibited by chitosan, the molecular weight of chitosan has various effects on antimicrobial activity of chitosan (Zheng and Zhu 2003). *E. coli* and other Gram-negative bacteria are more susceptible to lower-molecular-weight chitosan attacks while Gram positive bacteria are more inhibited by chitosan with high molecular weight. The range of molecular weight of chitosan that is believed to be effective in inhibiting fungal and bacterial contamination in food is 10,000–100,000 (Dutta *et al.* 2008). The concentration of chitosan in inhibiting microorganisms varies with microbial species and the minimal inhibitory concentrations of chitosan and its derivatives for different bacterial culture were tabulated by Shahidi *et al.* (1999). However, the optimal biocide property and film-forming conditions of chitosan continue to be researched. Bordenave *et al.* (2007) found that instead of forming a chitosan film, chitosan penetrated deeply into paper; this may not be surprising since both cellulose fibers of paper and chitosan are hydrophilic. Hydrophilicity of chitosan depends on molecular weight and acidity. The antimicrobial properties of chitosan are also subject to the moisture and acidity of the environment. For example, Beverly *et al.* (2008) has demonstrated that the acetic chitosan edible film is more effective in controlling *L. monocytogenes* on the surface of ready-to-eat roast beef than the lactic acid chitosan film. The biocide groups of chitosan will lose as organic acid evaporation in a wet condition (Lagaron *et al.* 2007). In a typical packaged food matrix, presence of moisture on the surface of the food will cause biocide groups to release rapidly – this may or may not be desirable depending on type of food and desired shelf life (Lagaron *et al.* 2007). It might not be an issue for fresh produce since the point of microbial contamination is believed to occur during the post-harvesting prior to packaging. For packaged foods that require extended biocide release, controlled migration of moisture from foods to chitosan film is needed.

The antimicrobial properties of chitosan have not gone unnoticed by the food industry. Chitosan has been proposed to use as edible films and coatings for extending shelf life of foods for many years (Shahidi *et al.* 1999; Dutta *et al.* 2008). They are mainly used as environmentally-friendly and sometimes edible alternative or supplement to petroleum-based plastic films dominated in the food industry. Chitosan can be either coated onto a common packaging material or coated directly on food surfaces. The film-forming property of chitosan and its derivatives is generally good and their films are tough and durable. N,O-carboxymethylchitin film has been approved for use as food wraps in North America (Davis *et al.* 1988).

Unlike plastics, chitosan-based films have moderate water permeability thus they are excellent food packaging materials for extending shelf life of fresh produce and foods with higher water activity (Bordenave *et al.* 2007). It is also observed that chitosan based films showed to be a very good barrier to permeation of oxygen (Wong *et al.* 1992). Earlier studies on extension of the storage life and better control of decay of strawberries, peaches, pears, kiwifruits, tomatoes, cucumbers, and bell peppers have proven the effectiveness of chitosan films; the results are probably a combination of inhibition of fungal attacks, reduction in respiration rates, and slowing of ripening due to decrease in ethylene and carbon dioxide evolution (El Ghaouth *et al.* 1991a, 1991b, 1992a, 1992b; Du *et al.* 1997; Durango *et al.* 2006).

The chitosan-based antimicrobial film can be applied to food surface as an edible protecting film by dipping and spraying. The antimicrobial edible film can also reduce the moisture loss, delay respiration, and slow discoloration due to exposure to sunlight and/or oxygen. Chitosan sometimes are combined with cellulose derivatives such as hydroxy propyl methyl cellulose (HPMC), a promising edible antimicrobial film carrier, to produce edible films with better protection against microbial contamination and damages caused by environmental conditions such as moisture, oxygen, and respiration (Miller *et al.* 2004).

In order to make chitosan-based edible films or food packaging materials, plasticizers such as polyols are often added to the film form solution. Fatty acid and other hydrophobic food-grade ingredients are sometimes used to reduce the water vapor permeability of the film (Vargas *et al.* 2006). Chemical modification such as cross-linking of chitosan can also reduce water permeability (Min *et al.* 2006). These ingredients and plasticizers alter not only mechanical properties of the film but also barrier properties of the film (Srinivasa *et al.* 2007). A number of studies on chitosan-based coatings of sliced fresh produces have been conducted showing varying degree of antimicrobial capabilities (Durango *et al.* 2006; Vargas *et al.* 2006; Chien *et al.* 2007; Vargas *et al.* 2009).

Enhancing chitosan-based edible or non-edible food packaging films or coatings are possible with additional GRAS antimicrobial agents or chemically modification. Pranoto *et al.* (2005a, 2005b) incorporated garlic oil, potassium sorbate and nisin into the chitosan film and observed increased antimicrobial activities with sacrificing mechanical properties of chitosan films. Belalia *et al.* (2008) noticed an increase in antimicrobial activities after chitosan was chemically modified to produce quaternary ammonium salt.

As an edible material, chitosan has potentials to use as base material for edible antimicrobial coatings or films for fresh produce packaging; it can also be used as coating material on the surface of paper or plastic. The only downside of application of chitosan and its derivatives is the availability of the material (chitin) and thus relative high price of purified material of chitosan.

EDIBLE FILMS OR COATINGS CONTAINING ANTIMICROBIAL AGENTS

An edible film or coating is a thin layer of film prepared from food or other food-grade materials that acts both a barrier to environmental hazards and microbial contamination or inhibition, thus protecting foods from contamination and extending longer shelf life. Edible films or coatings have been used for extending shelf life of foods since ancient times. Beeswax has been used by Chinese to protect oranges and apples since 12th or 13th century (Park 1999); it is still being used on fresh fruits despite the drawback of wax as edible coating for apples or pears as it interfere with normal respiration of these fruits when ripening (Smock 1940). Edible coatings of foods provide regulatory functions to migrations of moisture, oxygen, carbon dioxide, lipid, and aroma/flavor (Penã and Torres 1991; de Moura *et al.* 2009). In the case of fresh produces, the permeabilities of oxygen, carbon dioxide, and moisture in these edible films are the most important factors to consider when developing an edible films/coatings for fresh produces as living tissues of fresh produces respire in the packages. Edible films or coatings can serve as carriers of food additives. Antioxidants are sometimes added to edible films to delay oxidative damages to fresh produce. Common antioxidants are used in edible films or coatings are ascorbic acid (vitamin C), ascorbyl palmitate, butylated hydroxyanisole, butylated hydroxytoluene, citric acid, propyl gallant, and tocopherols, a.k.a., vitamin E (Cuppert 1994). N-acetylcysteine is used in edible films as anti-browning additive (Rojas-Graü *et al.* 2007). Another important aspect of edible film development is to incorporate antimicrobial agent into the edible films to inhibit microbial growth on the surface of fruits and vege-

tables, a predominant cause of food spoilage. The amount of antimicrobials and antioxidant additives to edible films or coatings is small as films or coatings impregnated with these additives contact directly on the surface of fresh produces where microbial spoilage occurs. All additives used in edible films or coatings are GRAS status or from food components with chemical or biochemical modifications. The most commonly used antimicrobials used in edible films or coatings are sodium benzoate, benzoic acid, propionic acid, potassium sorbate, sorbic acid, trisodium phosphate, lactic acid, lauric acid, benzoic acid, pediocin, nisin, lactacin, ethylenediaminetetraacetic acid (EDTA), chitosan, green tea powder, grape seed extract, spices/essential oils or their components, thiosulfonates, imazali, conalbumin, isothiocyanates, benomyl, silver, hyptylparaben, and enzymes (Cutter 2006; Chien *et al.* 2007; Kang *et al.* 2007; Coma 2008; Sivarooban *et al.* 2008). Although these antimicrobial agents are relatively small in quantity in edible films and considered safe to consume, they should be sparsely used in edible films or coatings as health-conscious consumers are wary of any additive that is not considered "natural." Prediction and controlled release of additives in edible films or coatings are strongly recommended (Guillard *et al.* 2009).

Edible coatings or films are prepared by casting solutions of carrier material(s), antimicrobial agents, plasticizer, modifying agents in different combinations and compositions. Coatings or films may consist of single or multiple layers in either dry or wet form. The coatings or films are formed by the following mechanisms: (1) simple coacervation, (2) complex coacervation, (3) gelation or thermal coagulation, and drying of film-form solution (Gontard *et al.* 1992; Guilbert *et al.* 1996). All these formed films or coatings need to meet a set of functional requirements in addition to antimicrobial activity; moisture barrier, gas barrier (O₂ and CO₂), color and appearance, and mechanical properties. Many edible films or coatings are rigid when dried thus some sorts of plasticizers have to be added to improve the film flexibility. It is possible that plasticizer or other additives will alter the barrier properties of the film as well release mechanism or capability of retention of the antimicrobial agent.

Considerable efforts have to devote to studies of the migration of antimicrobial agents in food packaging materials and foods, sometimes in model gel; these antimicrobial agents are mostly organic acids such as sorbic acid, a popular model agent. Several studies have also tried to investigate controlled release of macromolecules from edible films or coatings in food systems or in model systems, which is far more challenging than describing release mechanisms of organic acids or ions. Mattisson *et al.* (2000) studied diffusivities of antimicrobial lysozyme in gels and in liquids using holographic laser interferometry. Hirota *et al.* (2000)'s study was a more fundamental research on the effect of charge on protein (myoglobin) diffusion in hydrogels. Both studies found that protein diffusion in the gels – food simulants (agarose, a neutrally charged gel and κ-Carrageenan, a negatively charged gel) is pH dependent. This dependency is not unexpected as pH affects charges on protein surface, which ultimately influences intermolecular diffusion of the proteins in the gels or in liquids. Similarly, Sebti *et al.* (2003b) conducted a study that used Fick's second law to determine the apparent diffusion coefficient of nisin in agarose gel and found that the apparent diffusion coefficient of nisin incorporated into a cellulose matrix in the gel is smaller than that of free nisin in solution in the same gel because of the desorption step of nisin from cellulose matrix. This is not a new issue for antimicrobial food packaging field, whether the underlying packaging material is edible or not. Desorption of antimicrobial agent from a substrate (bio-based or synthetic) may determine the efficacy of antimicrobial activity on food surfaces. It is no doubt that incorporation of antimicrobial agents in packaging substrates reduces the apparent diffusion coefficients of these agents; this is not necessarily all negative – it can be explored for controlled release for a long, more sus-

taining antimicrobial activities. More researches on migration of antimicrobial agents in edible films or coatings are urgently needed.

Sensory issues with edible packaging films or coatings

The contribution of edible films with antimicrobial agents to sensory profile of underlying fresh produces should not be ignored. This is an issue that has received very little attention from the researchers of edible antimicrobial films or coatings; there are exceptions to this generalization (see Roller and Seedhar 2002; Rojas-Graü *et al.* 2007). As fresh produces are likely to be consumed fresh, the impact of edible antimicrobial films or coatings could be a challenge for the development of edible antimicrobial packaging for fresh produces. Edible films or coatings used as antimicrobial agent carriers usually have little or taste or smell as a lot of polysaccharides or proteins are bland and contain little volatiles. Some naturally-derived essential oils, such as garlic oil, however, have strong aromas that may or may not go well with certain fresh produces given the low threshold of our olfactory sense about these essential oils. There are some anecdotes of evidence that the essential oils used in edible films affects sensory profiles of some fresh cut fruits (Rojas-Graü *et al.* 2007) although others claimed that the added antimicrobial agents did not affect the sensory profiles of foods (Eswaranandam *et al.* 2006; Rabaudi-Massilia *et al.* 2008). Thus, sensory implications of development of edible antimicrobial films or coatings are needed to be included in all research and development of new edible food packaging materials.

Regulatory issues

Once an antimicrobial agent is added to edible films or coatings, one or more issues immediately arise: is the antimicrobial agent is of natural food or food additives? Depending on the answer to this question, one or more governmental regulatory agencies could get involved; there could be certain limitation on quantity of these agents that can be used in fresh produces. In general, since all components of edible films or coatings are consumed along with fresh produces, they should be at minimum of food grade and do not have toxic effects. Different countries and regional single market such as European Community (EU) have their own specific regulations on food manufacturing and food additives used in food products. In the USA, federal Food and Drug Administration (FDA) regulates almost all final food products and food additives that used in food matrices and U.S. Department of Agriculture regulates "organic" labeling of fresh produces, dairy products or others. What would happen if edible antimicrobial film is applied to organic produces? Can they still be claimed "organic" even the edible film is not of "organic" origin or the antimicrobial agent is not synthetic but neither is "organic" certified? Additionally, federal Centers for Disease Control and Prevention (CDC) could get involved in food poisoning cases. The Food Allergen Labeling and Consumer Protection Act of 2004 (FALCPA) in the USA brings home another critical issue in development of edible antimicrobial packaging materials: food allergens. Many edible films or coatings developed are derived from wheat and milk or peanut proteins; they are allergic to many people. Some vegetables such as those from the legume family also cause allergic reaction in some people. Clearly, there is labeling requirement for these edible film-coated fresh produces to ensure food safety.

CONCLUDING REMARKS

Traditional or novel food packaging materials coated with non-toxic antimicrobial agents are promising active food packaging materials for suppression of microbial growth on food surfaces. There are many ways of incorporating anti-

microbial agents into packaging materials. Several of these researches have resulted in commercial products. It is expected that more researches will be conducted in this field as researchers and companies are seeking food packaging materials with antimicrobial properties thus rendering food they contain longer shelf life.

Chitin and chitosan receive wide interests in food packaging research both as bio-based packaging materials with antimicrobial properties and antimicrobial agents incorporated into other packaging material substrates. They have a strong presence in research topics of edible films or coating for ensuring food quality and safety. It appears to be the most promising bio-based food packaging materials that provide good barrier and antimicrobial properties. However, chitosan-based materials have limited availability due to their source in the shell fish industry; worse, the potential chitosan based food packaging materials will compete with the biomedical industry, the environmental industry, and nutritional supplement industry. Bacterial, fungal or insect based chitin and chitosan, though potentially enormous, are still in the laboratory stage. This will all hinder the commercialization of chitosan-based food packaging in the market.

Edible films or coatings have a unique place in preventing microbial contamination of fresh produces. They have already been used to some extent in fruits and certain fruit-type vegetables; mostly these coatings/films are used as a physical barrier to migration of gases that affect the quality of and shelf life of fresh produces. Antimicrobial agents of non-toxic nature can be ready to apply to many edible films or coatings; however, how well these edible antimicrobial films or coatings can inhibit microbial spoilage in real world situation still is a grand challenge. There is anecdote evidence that suggests some fresh produces can be easily applied edible antimicrobial films or coatings; but most are difficult to predict based on current research activities – it is discouraging to know that there is lack of research on incorporation of edible antimicrobial films or coatings into packaging of leafy vegetable or other fresh produce with rough surface morphology. Some "new" natural antimicrobial agents such as essential oils present a unique practical problem for using on fresh produces due to its overwhelming odor at very low threshold.

Like all new technologies, many issues related to food safety, toxicity, sensory attributes, consumer preferences, and regulatory considerations need to be carefully examined before any of these technologies can be commercialized. For example, there is a concern that those bio-based polymers used for food packaging could interfere with physiological functions or metabolism in the human body (Shahidi *et al.* 1999). More research is needed to address these issues before large scale adoption of edible food packaging based on natural biopolymers.

DISCLAIMER

Mention of trade names or commercial products in this article is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

REFERENCES

- Agnihotri SA, Makkikarjuna NN, Aminabhavi TM (2004) Recent advances on chitosan-based micro- and nanoparticles in drug delivery. *Journal of Controlled Release* **100**, 5-28
- Ahvenainen, R (1996) New Approaches in improving the shelf life of minimally processed fruit and vegetables. *Trends in Food Science and Technology* **7**, 179-187
- Arfa AB, Preziosi-Belloy L, Chalier P, Gontard N (2007) Antimicrobial paper based on a soy protein isolate or modified starch coating including carvacrol and cinnamaldehyde. *Journal of Agricultural and Food Chemistry* **55**, 2155-2162
- Belalia R, Grelier S, Benaissa M, Coma V (2008) New bioactive biomaterials based on quaternized chitosan. *Journal of Agricultural and Food Chemistry* **56**, 1582-1588

- Beverly RL, Janes ME, Prinyawiwatwala W No, HK** (2008) Edible chitosan films on ready-to-eat roast beef for the control of *Listeria monocytogenes*. *Food Microbiology* **25**, 534-537
- Bordenave N, Grelier S, Pichavant F, Coma V** (2007) Water and moisture susceptibility of chitosan and paper-based materials: structure-property relationships. *Journal of Agricultural and Food Chemistry* **55**, 9479-9488
- Chawengkijwanich C, Hayati Y** (2008) Development of TiO₂ powder-coated food packaging film and its ability to inactivate *Escherichia coli in vitro* and in actual tests. *International Journal of Food Microbiology* **123**, 288-292
- Chen M, Yeh GH, Chiang B** (1996) Antimicrobial and physicochemical properties of methylcellulose and chitosan films containing a preservative. *Journal of Food Processing and Preservation* **20**, 379-390
- Chien P-J, Sheu F, Yang F-H** (2007) Effects of edible chitosan coating on quality and shelf life of sliced mango fruit. *Journal of Food Engineering* **78**, 225-229
- Chollet E, Swesi Y, Degraeve P, Sebti I** (2008) Monitoring nisin desorption from a multi-layer polyethylene-based film coated with nisin loaded HPMC film and diffusion in agarose gel by an immunoassay (ELISA) method and a numerical modeling. *Innovative Food Science and Emerging Technologies* **10**, 208-214
- Chung D, Papadakis SE, Yam KL** (2003) Evaluation of a polymer coating containing triclosan as the antimicrobial layer for packaging materials. *International Journal of Food Science and Technology* **38**, 165-169
- Coma V** (2008) Bioactive packaging technologies for extended shelf life of meat-based products – review. *Meat Science* **78**, 90-103
- Cooksey K** (2001) Antimicrobial food packaging materials. *Additives for Polymers* **8**, 6-10
- Cuerti RG, Osuji G, Washington A** (1991) *N*-carboxymethyl chitosan inhibition of aflatoxin production: role of zinc. *Biotechnology Letters* **13**, 441-444
- Cuppert SL** (1994) Edible coatings as carriers of food additives, fungicides and natural antagonists. In: Krochta JM, Baldwin EA, Nisperos-Carriedo MO (Eds) *Edible Coatings and Films to Improve Food Quality*, Technomic Publishing Co., Lancaster, USA, pp 121-137
- Cutter CN** (2006) Opportunities for bio-based packaging technologies to improve the quality and safety of fresh and further processed muscle foods. *Meat Science* **74**, 131-142
- Davis DH, Elson CM, Hayes ER** (1988) *N*,*O*-Carboxymethyl chitosan, a new water soluble chitin derivative. *Fourth International Conference on Chitin and Chitosan*, Trondheim, Norway, pp 467-475
- de Moura MR, Aouada FA, Avena-Bustillos RJ, McHugh TH, Krochta JM, Mattoso L** (2009) Improved barrier and mechanical properties of novel hydroxypropyl methylcellulose edible films with chitosan/tripolyphosphate nanoparticles. *Journal of Food Engineering* **92**, 448-453
- Devlieghere F, Vermeiren L, Jacobs M, Debevere J** (2000) The effectiveness of hexamethylenetetramine-incorporated plastic for the active packaging of foods. *Packaging Technology and Science* **13**, 117-121
- Du J, Gemma H, Iwahori S** (1997) Effect of chitosan coating on the storage of peach, Japanese pear, and kiwifruit. *Journal of the Japanese Society of Horticulture Science* **66**, 15-22
- Durango AM, Soares NFF, Andrade NJ** (2006) Microbiological evaluation of an edible antimicrobial coating on minimally processed carrots. *Food Control* **17**, 336-341
- Dutta PK, Tripathi S, Mehrotra GK, Dutta J** (2008) Perspectives for chitosan-based antimicrobial films in food applications. *Food Chemistry* **114**, 1173-1182
- El Ghaouth A, Arul J, Ponnampalam R, Boulet M** (1991a) Chitosan coating effect on storability and quality of fresh strawberries. *Journal of Food Science* **56**, 1618-1620
- El Ghaouth A, Arul J, Ponnampalam R, Boulet M** (1991b) Use of chitosan coating to reduce water loss and maintain quality of cucumber and bell pepper fruits. *Journal of Food Processing and Preservation* **15**, 359-368
- El Ghaouth A, Arul J, Asselin A, Benhamou N** (1992a) Antifungal activity of chitosan on two postharvest pathogens of strawberry fruits. *Phytopathology* **82**, 398-402
- El Ghaouth A, Ponnampalam R, Castaigne F, Arul J** (1992b) Chitosan coating to extend the storage life of tomatoes. *HortScience* **27**, 1016-1018
- Eswaranandam S, Hettiarachchy NS, Meullenet JF** (2006) Effect of malic and lactic acid incorporated soy protein coatings on the sensory attributes of whole apple and fresh-cut cantaloupe. *Journal of Food Science* **71**, S307-S313
- Fernandez-Saiz P, Lagarón JM, Ocio MJ** (2009) Optimization of the biocide properties of chitosan for its application in the design of active films of interest in the food area. *Food Hydrocolloids* **23**, 913-921
- Ghosh KG, Srivasta AN, Nirmala N, Sharma TR** (1977) Development of application of fungistatic wrappers in food preservation. Part II. Wrappers made by coating process. *Journal of Food Science and Technology* **14**, 261-264
- Gontard N, Guilbert S, Cuq JL** (1992) Edible wheat gluten films: influences of the main process variables on film properties using response surface methodology. *Journal of Food Science* **57**, 190-195
- Guiga W, Galland S, Peyrol E, Degraeve P, Carnet-Pantiez A, Sebti I** (2008) Antimicrobial plastic film: physico-chemical characterization and nisin desorption modeling. *Innovative Food Science and Emerging Technologies* **10**, 203-207
- Guilbert N, Gontard N, Gorris GM** (1996) Prolongation of the shelf life of perishable food products using biodegradable films and coatings. *LWT - Food Science and Technology* **29**, 10-17
- Guillard V, Issoufov V, Redl A, Gontard N** (2009) Food preservative content reduction by controlling sorbic acid release from a superficial coating. *Innovative Food Science and Emerging Technologies* **10**, 108-115
- Gutiérrez L, Sánchez C, Batlle R, Nerín C** (2009) New antimicrobial active package for bakery products. *Trends in Food Science and Technology* **20**, 92-99
- Han JH, Floros JD** (1998) Simulating diffusion model and determining diffusivity of potassium sorbate through plastics to develop antimicrobial packaging films. *Journal of Food Processing and Preservation* **22**, 107-122
- Han J** (2000) Antimicrobial food packaging. *Food Technology* **54**, 56-65
- Hardenburg RE, Watada AE, Wang CY** (1986) The commercial storage of fruits, vegetables, and florist and nursery stock. *USDA Agriculture Handbook No. 66* (revised)
- Hirota N, Kumaki Y, Narita T, Gong JP, Osada Y** (2000) Effect of charge on protein diffusion in hydrogels. *Journal of Physical Chemistry* **104**, 9898-9903
- Jofré A, Garriga M, Aymerich T** (2007) Inhibition of *Listeria monocytogenes* in cooked ham through active packaging with natural antimicrobials and high-pressure processing. *Journal of Food Protection* **70**, 2498-2502
- Johnston JH, Borrmann T, Tankin D, Cairns M, Crindrod J, Mcfarlane A** (2008) Nano-structured composite calcium silicate and some novel applications. *Current Applied Physics* **8**, 504-507
- Kang HJ, Jo C, Kwon JH, Kim JH, Chung HJ, Byun MW** (2007) Effect of a pectin-based edible coating containing green tea powder on the quality of irradiated pork patty. *Food Control* **18**, 430-435
- Kim Y-M, An D-S, Park H-J, Lee D** (2003) Properties of nisin-incorporated polymer coatings as antimicrobial packaging materials. *Packaging Science and Technology* **15**, 247-254
- Lagarón JM, Fernandez-Saiz P, Ocio M** (2007) Using ATR-FTIR spectroscopy to design active antimicrobial food packaging structures based on high molecular weight chitosan polysaccharide. *Journal of Agricultural and Food Chemistry* **55**, 2554-2562
- Lee C, Park H, Lee D** (2004) Influence of an antimicrobial packaging on kinetics of spoiling microbial growth in milk and orange juice. *Journal of Food Engineering* **65**, 527-531
- Lee CH, An DS, Lee SC, Park HJ, Lee DS** (2004) A coating for use as an antimicrobial and antioxidative packaging material incorporating nisin and α -tocopherol. *Journal of Food Engineering* **62**, 323-329
- Limjaroen P, Ryser E, Lockhart H, Harte B** (2003) Development of a food packaging coating material with antimicrobial properties. *Journal of Plastic Films and Sheeting* **19**, 95-109
- López-Rubio A, Almenar E, Hernández-Muñoz P, Lagarón JM, Catalá R, Gavara R** (2004) Overview of active polymer-based packaging technologies for food applications. *Food Review International* **20**, 357-387
- López P, Sánchez C, Batlle R, Nerín C** (2007) Development of flexible antimicrobial films using essential oils as active agents. *Journal of Agricultural and Food Chemistry* **55**, 8814-8824
- Mattisson C, Roger P, Jönsson B, Axelsson A, Zacchi G** (2000) Diffusion of lysozyme in gels and liquids a general approach for the determination of diffusion coefficients using holographic laser interferometry. *Journal of Chromatography* **743**, 151-167
- Mayachiew P, Devahastin S, Mackey BM, Niranjana K** (2009) Effects of drying methods and conditions on antimicrobial activity of edible chitosan films enriched with galangal extract. *Food Research International* (accepted)
- Min F-L, Huang C-T, Liang H-F, Chen M-C, Chiu Y-L, Chen C-H, Sung H-W** (2006) Physicochemical, antimicrobial, and cytotoxic characteristics of a chitosan film cross-linked by a naturally occurring cross-linked agent, aglycone geniposidic acid. *Journal of Agricultural and Food Chemistry* **54**, 3290-3296
- Miller H, Grelier S, Pardon P, Coma V** (2004) Antimicrobial and physicochemical properties of chitosan-HPMC-based films. *Journal of Agricultural and Food Chemistry* **52**, 6585-6591
- Ouatara B, Simard RE, Piette G** (2000) Diffusion of acetic and propionic acids from chitosan-based antimicrobial packaging films. *Journal of Food Science* **65**, 768-773
- Park HJ** (1999) Development of advanced edible coatings for fruits. *Trends in Food Science and Technology* **10**, 254-260
- Penã DCR, Torres JA** (1991) Sorbic acid and potassium sorbate permeability of an edible methylcellulose-palmitic acid films: water activity and pH effects. *Journal of Food Science* **56**, 497-499
- Pranoto Y, Salokhe VM, Rakshit SK** (2005) Physical and antimicrobial properties of alginate-based edible film incorporated with garlic oil. *Food Research International* **38**, 267-272
- Pranoto Y, Rakshit SK, Salokhe VM** (2005) Enhancing antimicrobial activity of chitosan films by incorporating garlic oil, potassium sorbate and nisin. *LWT - Food Science and Technology* **38**, 859-865
- Rabea EI, Badawy ME T, Stevens CV, Smagghe G, Steurbaut W** (2003) Chitosan as antimicrobial agent: applications and mode of action. *Biomacromolecules* **4**, 1457-1465
- Raybaudi-Massilia RM, Mosqueda-Melgar J, Martin-Belloso O** (2008) Edi-

- ble alginate-based coating as carrier of antimicrobials to improve shelf-life and safety of fresh-cut melon. *International Journal of Food Microbiology* **121**, 313-327
- Rico-Pena DC, Torres JA** (1991) Sorbic acid and potassium sorbate permeability of an edible methylcellulose-palmitic acid film: water activity and pH effect. *Journal of Food Science* **56**, 497-499
- Rodríguez A, Nerin C, Batlle R** (2007) The use of natural essential oils as antimicrobial solutions in paper packaging. Part II. *Progress in Organic Coatings* **60**, 33-38
- Rodríguez A, Nerin C, Batlle R** (2008) New cinnamon-based active paper packaging against *Rhizopus stolonifer* food spoiling. *Journal of Agricultural and Food Chemistry* **56**, 6364-6369
- Rojas-Graü MA, Raybaudi-Massilia RM, Soliva-Fortuny RC, Avena-Bustillos RJ, McHugh TH, Martín-Belloso O** (2007) Apple-puree-alginate edible coating as carrier of antimicrobial agents to prolong shelf-life of fresh-cut apples. *Postharvest Biology and Technology* **45**, 254-264
- Rojas-Graü MA, Avena-Bustillos RJ, Olsen K, Friedman M, Henika P, Martín-Belloso O, Pan Z, McHugh TH** (2007) Effects of plant essential oils and oil compounds on mechanical barrier and antimicrobial properties of alginate-apple puree edible films. *Journal of Food Engineering* **81**, 634-641
- Roller S, Seedhar M** (2002) Carvacrol and cinnamon acid inhibit microbial growth in fresh-cut melon and kiwifruit at 4°C and 8°C). *Letter of Applied Microbiology* **35**, 390-394
- Salmieri S, Lacroix M** (2006) Physicochemical properties of alginate/poly-caprolactone-based films containing essential oils. *Journal of Agricultural and Food Chemistry* **54**, 10205-10214
- Sebti I, Delves-Broughton J, Coma V** (2003a) Physicochemical properties and bioactive of nisin-containing cross-linked hydroxypropylmethylcellulose films. *Journal of Agricultural and Food Chemistry* **51**, 6468-6474
- Sebti I, Ripoche A, Carnet D, Blanc D, Saure, R, Coma V** (2003b) Controlled diffusion of an antimicrobial peptide from a biopolymer film. *Transactions IChemE* **81**, 1099-1104
- Shahidi F, Arachchi JKV, Jeon Y-J** (1999) Food applications of chitin and chitosan. *Trends in Food Science and Technology* **10**, 37-51
- Siracusa V, Rocculi P, Romani S, Dalla Rosa M** (2008) Biodegradable polymers for food packaging: a review. *Trends in Food Science and Technology* **19**, 634-643
- Sivarooban T, Hettiarachchy NS, Johnson MG** (2008) Physical and antimicrobial properties of grape seed extract, nisin, and EDTA incorporated soy protein edible films. *Food Research International* **41**, 781-785
- Sleeth RB, Furgal HP** (1965) Method of coating freeze-dried meat. U.S. Patent 3,165,416
- Smock RM** (1940) Some additional effects of waxing apples. *American Society of Horticultural Science* **37**, 448-452
- Srinivasa PC, Ramesh MN, Kumar KR, Tharanathan RN** (2003) Properties and adsorption studies of chitosan-polyvinyl alcohol blend films. *Carbohydrate Polymer* **53**, 431-438
- Srinivasa PC, Ramesh MN, Tharanathan RN** (2007) Effect of plasticizers and fatty acids on mechanical and permeability characteristics of chitosan films. *Food Hydrocolloids* **21**, 1113-1122
- Sudarshan NR, Hoover DG, Knorr D** (1992) Antimicrobial action of chitosan. *Food Biotechnology* **6**, 257-272
- Vargas M, Albors A, Chiralt A, González-Martínez C** (2006) Quality of cold-stored strawberries as affected by chitosan-oleic acid edible coatings. *Postharvest Biology and Technology* **41**, 164-171
- Vargas M, Chiralt A, Albors A, González-Martínez C** (2009) Effect of chitosan-based edible coatings applied by vacuum impregnation on quality preservation of fresh-cut carrot. *Postharvest Biology and Technology* **51**, 263-271
- Vojdani F, Torres JA** (1990) Potassium sorbate permeability of methylcellulose and hydroxypropyl methylcellulose coatings: effect of fatty acids. *Journal of Food Science* **55**, 841-846
- Weng Y, Hotchkiss JH** (1992) Inhibition of surface molds on cheese by polyethylene containing the antimicrobial imazalil. *Journal of Food Protection* **55**, 367-369
- Weng Y, Hotchkiss JH** (1993) Anhydrides as antimicrobial agents added to polyethylene films for food packaging. *Packaging Technology and Science* **6**, 123-128
- Wong DWS, Gastineau FA, Gregorski KA, Tillin SJ, Pavlath AE** (1992) Chitosan-lipid films: microstructure and surface energy. *Journal of Agricultural and Food Chemistry* **40**, 540-544
- Young DH, Kohle H, Kauss H** (1982) Effect of chitosan on membrane permeability of suspension cultured *Glycine max* and *Phaseolus vulgaris* cells. *Plant Physiology* **70**, 1449-1454
- Zheng LY, Zhu JF** (2003) Study of antimicrobial activity of chitosan with different molecular weight. *Carbohydrate Polymers* **54**, 527-530