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Review

Nanobiomaterials in antimicrobial food packaging

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ABSTRACT

Post-harvest losses of fresh fruits and vegetables are more serious. Therefore, many strategies have been developed to decrease the fruit losses. New bio-based materials have been exploited to develop edible and biodegradable packaging as a big effort to extend shelf life and improve quality of food while reducing packaging waste. Unfortunately, the use of edible and biodegradable polymers has been limited because of problems related to performance and processing. The application of nanotechnology to these polymers may open new possibilities for improving not only the properties but also the cost-price-efficiency. The addition of nanoparticles as an antimicrobial agent, are also an option to modify positively the properties of nanobiobased packaging. The main objectives of this review are to evaluate the capabilities of nanoparticles filled bio based packaging to preservation and extended shelf life of fresh fruits as a new approach.

Keywords: *packaging, nanotechnology, nanocomposite, biopolymer, antimicrobial.*

INTRODUCTION

Today, there has been a significant increase in the production and usage of non-biodegradable materials or plastics as food packaging material. Non-biodegradable materials usually are manufactured from petroleum products and cause waste disposal problems. In order tomeet the increasing demand for sustainability and environmental safety, a great deal of research has been focused on the development of food packaging materials that speedily degrade and are fully mineralized in the environment. Biopolymers have been a preferred alternative to be researched and developed into eco-friendly food packaging materials due to its biodegradability. Utilized food packaging materials created from biopolymers can be placed into bio-waste collection for further composting, resulting in organic by-products such as carbon dioxide (CO_2) and water (H_2O). Unfortunately, there are drawbacks to the use of biopolymers as food packaging materials including poorer mechanical, thermal, and barrier properties in comparison to the conventional non-biodegradable materials made from petroleum. Due to this, much effort was made in research to improve the properties of biopolymers as exemplified in the use of the nanocomposite concept. The incorporation of nanofillers such as silicate, clay, and titanium dioxide (TiO_2) into biopolymers may not only improve the biopolymers' mechanical and barrier properties but also offer other functions and applications in food packaging as an antimicrobial agent, biosensor, and oxygen scavenger. The bio-nanocomposite can be active food packaging whereby the food packaging can interact with food in various ways by releasing beneficial compounds such as antimicrobial agent, antioxidant agent, or by eliminating some detrimental elements such as oxygen or water vapor. The bio-nanocompositecan also be made into smart food packaging which can use some mechanisms to register and transmit information regarding the quality or safety of the food as well as identify if the property of the packaged food has microbial contamination or the date expired. . The development of antimicrobial bio-nanocomposite materials for food packaging is not only crucial to lessen environmental problems but also improve the antimicrobial activity of the food packaging materials. The main objectives of this review are to evaluate the capabilities of nanoparticles filled bio based packaging to preservation and extended shelf life of fresh fruit as a new approach (Othman, 2014).

FOOD PACKAGING

Traditional food packaging is meant for mechanical supporting of otherwise non-solid food, and protecting food from external influences (Emamifar, 2011). The fundamental roles of food packaging are as follows: to protect food products from external influences and damage, be a container for the food, and offer consumers ingredient and nutritional information. Traceability, convenience, and tamper indication are secondary functions of ever-increasing importance. In addition, the objective of food packaging is to contain food in a cost-effective fashion which fulfils industry requirements and consumer desires, upholds food safety standards, and minimizes the environmental impact. However, there are other issues to consider in packaging technology including energy and material costs, greater social and environmental awareness and strict protocols on the use of pollutants and disposal of municipal solid waste. Packaging provides protection from three major classes of external influences: chemical, biological, and physical. Chemical protection minimizes compositional changes triggered by environmental influences such as exposure to gases (typically oxygen), moisture (gain or loss), or light (visible, infrared, or ultraviolet). Biological protection provides a barrier to microorganisms (pathogens and spoiling agents), insects, rodents, and other animals, thereby preventing disease and spoilage. Physical protection shields food from mechanical damage and includes cushioning against the shock and vibration encountered during distribution (Marsh &Bugusu, 2007). Packaging has allowed us to have a wide variety of foods year round that would not be possible without the protection of the package. Foods now have a longer shelf life, resulting in less loss due to spoilage. Packaging also provides for convenience with products that can be heated in the package and products that can be purchased as single-serve items. Demand for quality food has driven packaging innovation, and innovations in packaging have helped to create new food categories and added convenience (Risch, 2009).

NOVEL FOOD PACKAGING

New trends and demands in the global food supply chain present new challenges, which create an increasing demand for safer foods. As a consequence, longer shelf life is demanded. At the same time numerous small niche production sites are appearing with a very different set of food safety challenges. This calls for new research in food packaging as an instrument to ensure a safer global food supply. Packaging technologies such as modified atmosphere packaging and antimicrobial active packaging based on volatile (essential oils and alcohols) or non-volatile components (chitosan and nisin) have been developed as a result of these demands. The increase in crude oil prices and the growing environmental concerns have led to increased research on alternative packaging materials from renewable or bio-derived resources. Food products have specific micro-organisms associated with them. However, changes in environmental condition in the package can leave products vulnerable to other micro-organisms. Therefore there is need for detailed studies on the interplay among products, related micro-organisms and packaging materials. Such knowledge will form the foundation for the development of better preservation methods for new and existing products. Novel and advanced polymeric materials are being developed for enhanced food packaging. The development of these materials is based on conventional polymer science methods, as well as newer technologies including active and intelligent packaging, biopolymers, nanotechnology and nanocomposites (Giuseppe et al., 2010).

ACTIVE AND INTELLIGENT FOOD PACKAGING

New food packaging technologies are developing as a response to consumer demands or industrial production trends towards mildly preserved, fresh, tasty and convenient food products with prolonged shelf-life and controlled quality. In addition, changes in retailing practices or consumers' lifestyles, pose major challenges to the food packaging industry and act as the causative factor for the development of new and improved packaging concepts which lengthen shelf-life while upholding and monitoring food safety and quality. Innovations in packaging were up to now limited mainly to a small number of commodity materials such as barrier materials (new polymers, complex and multilayer materials) with new designs, for marketing purposes. However, food packaging has no longer just a passive role in protecting and marketing a food product. New concepts of active and intelligent packaging are due to play an increasingly important role by offering numerous and innovative solutions for extending the shelf-life or maintain, improve or monitor food quality and safety. Next to these, numerous other concepts such as ethanol emitters (*e.g.* for bakery products), ethylene absorbers (*e.g.* for climacteric fruits), carbon dioxide emitters/ absorbers, time/ temperature and oxygen indicators *etc*. have been developed. In a general way, the field has been extended largely as a series of niche markets owing to the current approach of packaging industries looking at it in terms of new market opportunities. Introduction of active and intelligent packaging can extend the shelf life of food or improve its organoleptic properties and thus prevent food losses. According to the FDA report of 2011, about 1.3 billion tons of food is thrown away every year. Every year only Europe produces 89 million tons of wasted food, and the average rubbish thrown by European household constitutes 20–30% of food purchased. New packaging solutions allow to improve the economic aspect. The interest in active and intelligent packaging is successively increasing.Active packaging in which subsidiary constituents have been deliberately included in or on either the packaging material or the package headspace to enhance the performance of the package system However, active packaging means maintaining and extending product shelf life by adding the incorporation of certain additives into packaging film or within packaging containers . Active packaging, allows packages to interact with food and the environment and play a dynamic role in food preservation. In contrast to traditional packaging, active and intelligent packaging may change the composition and organoleptic characteristics of food, provided that the changes are consistent with the provisions for food. Besides the released substances will be allowed to be used as food additives. The principles behind active packaging are based either on the intrinsic properties of the polymer used as packaging material itself or on the introduction (inclusion, entrapment *etc.*) of specific substances inside the polymer. Besides, active packaging refers to the incorporation of certain additives into packaging systems (whether loose within the pack, attached to the inside of packaging materials or incorporated within the packaging materials themselves) in order to preserve or lengthen product quality and shelf-life. Packaging may be considered active when it it has a role in food preservation other than just offering a static barrier to external conditions. On the other hand, active packaging has been defined as packaging, which 'changes the condition of the packed food to extend shelf-life or to improve safety or sensory properties, while maintaining the quality of packaged food'. The development of a whole range of active packaging systems, some of which may have applications in both new and existing food products, is fairly new. Active packaging includes additives or 'freshness enhancers' that can participate in a host of packaging applications and by doing so, enhance the preservation function of the primary packaging system. Intelligent packaging that contains an external or internal indicator to provide information about aspects of the history of the package and/or the quality of the food intelligent packaging (also described as smart packaging) is packaging that in some way senses and informs the manufacturer, retailer and consumer of the state of some properties of the food it encloses or the environment in which it is kept . Although distinctly different from the concept of active packaging, features of intelligent packaging can be used to check the effectiveness and integrity of active packaging systems. Intelligent packaging devices are capable of sensing and providing information about the function and properties of packaged food and can provide assurances of pack integrity, tamper evidence, product safety and quality, and are being utilized in applications such as product authenticity, anti-theft and product traceability. Intelligent packaging devices include sensors, time-temperature indicators, gas sensing dyes, microbial growth indicators, physical shock indicators, and numerous examples of tamper proof, anti-counterfeiting and anti-theft technologies (Dobrucka&Cierpiszewski, 2014).

ANTIMICROBIAL FOOD PACKAGING

Antibacterial agents are of significance to a number of industrial sectors including environmental, food, synthetic textiles, packaging, healthcare, medical care, as well as construction and decoration. They can be generally classified into two types, organic and inorganic. Organic antibacterial materials are often less stable particularly at high temperatures and/or pressures compared to inorganic antibacterial agents. This leads to potential problems for the product creation. Thus, inorganic materials such as metal and metal oxides have attracted a great deal of attention in the past decade due to their ability to endure harsh process conditions (Zhang et al., 2007). Antimicrobial packaging is a type of active packaging (Appendini& Hotchkiss, 2002). Antimicrobial food packaging materials have to prolong the lag phase and decrease the growth rate of microorganisms in order to lengthen shelf life and maintain product quality and safety. The increasing consumer demand for fresh, convenient, and safe food products as well as the need to package foods in a flexible manner for transportation and storage signals a bright future for AM Packaging. However, more data must be collected through research on the chemical, microbiological and physiological effects of these systems on the packaged food, especially on the issues of nutritional quality and human safety. Thus far, research on AM packaging has focused mainly on the development of various methods and model systems rather than its efficiency in actual food preservation. Further research is crucial in the identification of the types of foods which can most benefit from AM packaging materials (Suppakul et al., 2003). Antimicrobial packaging can take several forms including:

1. Supplimenting packages with sachet pads containing volatile antimicrobial agents.
2. Assimilation of volatile and non-volatile antimicrobial agents directly into polymers.
3. Coating or adsorbing antimicrobials onto polymer surfaces.
4. Restriction of antimicrobials in polymers by ion or covalent linkages.
5. Usage of polymers which are characteristically antimicrobial (Appendini& Hotchkiss, 2002).

Among the active packaging applications, the addition of antimicrobials as a means of extending the bacterial lag phase, slowing the growth rate of micro-organisms and maintaining food quality and safety is receiving substantial attention. . Direct addition of antimicrobials (organic acids or their respective acid anhydrides, spice extracts, chelating agents, metals, enzymes, bacteriocins, etc.) might initiate some loss of activity because of leaching into the food matrix and cause a cross-reaction with other food components such as lipids or proteins. Therefore, the use of packaging films containing antimicrobial agents could be more efficient by a controlled migration of the compound into the food, not only allowing for initial inhibition of undesirable micro-organisms, but also residual activity over time, during the transport and storage of food during distribution (Mauriello et al., 2005) Packaging with combinations of more than one incorporated antimicrobial have also been investigated. For example, it is theorized that compounds active against Gram-positive bacteria (i.e. lysozyme) combined with chelating agents (i.e. EDTA) can target Gram-negative bacteria. Addition of EDTA to edible films containing nisin or lysozyme, however, had little inhibition effect on E. coli and Salmonella typhimurium. The justification for incorporating antimicrobials into the packaging is to prevent surface growth in foods where a large portion of spoilage and contamination occurs. For instance, intact meat from healthy animals is essentially sterile and spoilage occurs primarily at the surface. This approach can reduce the addition of larger quantities of antimicrobials that are usually incorporated into the bulk of the food. Many antimicrobials are incorporated at 0.1-5% w/w of the packaging material, particularly films. Antimicrobials may be incorporated into melting polymers i or by solvent compounding. Thermal polymer processing methods such as extrusion and injection molding may be used with thermally stable antimicrobials such as metals (Appendini& Hotchkiss, 2002). Antimicrobials that cannot tolerate the processing temperatures of the polymers are often coated onto the materials, which require surface functionalisation to improve the adhesion of the coatings. Surface immobilization requires the presence of functional groups on both the antimicrobial and the polymer and also spacer molecules that bond the polymer surface to the active agent (Radheshkumar&Münstedt, 2006).

BIODEGRADABLE FOOD PACKAGING

One of the challenges facing the food packaging industry in its efforts to produce biobased primary packaging is to match the durability of the packaging with product shelf-life. The biologically based packaging material must remain stable without changes of mechanical and/or barrier properties and must function properly during storage until disposal. Subsequently, the material should biodegrade efficiently. Environmental conditions conducive to biodegrationmust be avoided during storage of the food product, whereas optimized conditions for biodegration must exist after discarding. The most important parameters for controlling stability of the biologically based packaging material are appropriate water activity, pH, nutrients, oxygen, storage time, and temperature. Thus, dry products may be safely stored for extended periods, whereas moist foods would have limited storage periods. Prior to using biobased materials for primary food packaging, the effects on food quality and food safety must be examined (Petersen et al., 1999). In the recent years, bio-based and biodegradable products have raised great interest since sustainable development policies tend to expand with the decreasing reserve of fossil fuel and the growing concern for the environment. These polymers bring a significant contribution to the sustainable development in view of the wider range of disposal options with minor environmental impact. As a result, the market of these environmentally friendly materials is in rapid expansion, 10–20 % per year.

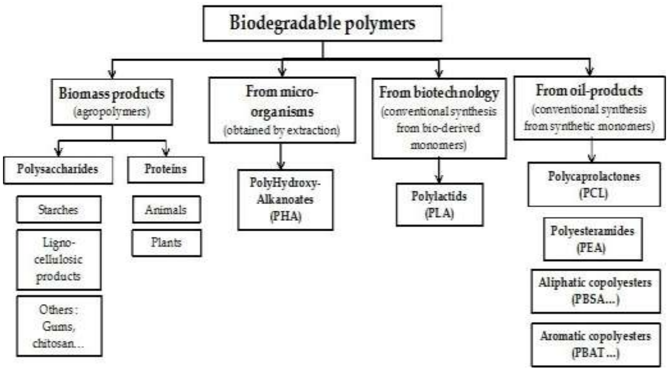

Figure 1. Clasification of the main biodegradable polymers (Avérous&Pollet, 2012).

Classification of the main biodegradable polymers are included (Figure 1): (i) polymers from biomass such as agro-polymers from agro-resources (e.g., starch or cellulose), (ii) polymers obtained by microbial production such as the polyhydroxyalkanoates (PHAs), (iii) polymers conventionally and chemically synthesized from monomers obtained from agro-resources, e.g., the polylactic acid (PLA), and (iv) polymers obtained from fossil resources. Only the first three categories (i–iii) are obtained from renewable resources. Biodegradable polymers can further classified as agropolymers (category i) and biodegradable polyesters or biopolyesters (categories ii–iv), which can be synthesized from fossil resources but main productions are obtained from renewable resources It could be further classified these biodegradable polymers into two main categories: the agro-polymers (category i) and the biodegradable polyesters or biopolyesters (categories ii–iv) (Avérous&Pollet, 2012). Main biodegradable polymers will be discussed in more detail in the following sections of the present review.

Polysaccharides

Polysaccharides are the most abundant macromolecules in the biosphere. These complex carbohydrates constituted of glycosidic bonds are often one of the main structural elements of plants and animals exoskeleton. The polysaccharides presented in this review are starch, cellulose, chitin and chitosan.

Starch

Among the natural polymers, starch is of interest. It is regenerated from carbon dioxide and water by photosynthesis in plants. Owing to its complete biodegradability, low cost and renewability, starch is considered as a promising candidate for developing sustainable materials. It is mainly composed of two homo polymers of D-glucose: amylase, a mostly linear α- D (1, 4′)-glucan and branched amylopectin, having the same backbone structure as amylose but with many α-1, 6′-linked branch points. Evidently, starch is hydrophilic. The available hydroxyl groups on the starch chains potentially exhibit reactivity specific for alcohols. In other words, they can be oxidized and reduced, and may participate in the formation of hydrogen bonds, ethers and esters. Starch has different proportions of amylose and amylopectin ranging from about 10–20% amylose and 80–90% amylopectin depending on the source. Amylose is soluble in water and forms a helical structure. Starch occurs naturally as discrete granules since the short branched amylopectin chains can form helical structures which crystallize. Starch granules display hydrophilic properties and strong inter-molecular association through hydrogen bonding formed by the hydroxyl groups on the granule surface. Owing to its hydrophilicity, the internal interaction and morphology of starch will be readily changed by water molecules, and thereby its glass transition temperature (Tg). Furthermore, the dimension and mechanical properties of it is depending on the water content. Tg of native starch can be as low as 60 to 80°C when the weight fraction of water is in the range 0.12 to 0.14, which allows starch to be successfully injection moulded to obtain thermoplastic starch polymers in the presence of water. On the other hand, the hydrophilicity of starch can be used to improve the degradation rate of some degradable hydrophobic polymers. Starch is totally biodegradable in a wide variety of environments. It can be hydrolyzed into glucose by microorganism or enzymes, and then metabolized into carbon dioxide and water. It is worth noting that carbon dioxide will recycle into starch again by plants and sunshine. Starch itself is poor in process ability, also poor in the dimensional stability and mechanical properties for its end products. Therefore, native starch is not used directly. To improve the properties of starch, various physical or chemical modifications of starch such as blending, derivation and graft copolymerization have been investigated (Lu et al., 2009).

Cellulose

Many polymer researchers are of the opinion that polymer chemistry had its origins with the characterization of cellulose. Cellulose differs in some respects from other polysaccharides produced by plants, the molecular chain being very long and consisting of one repeating unit (Cellobiose). Naturally, it occurs in a crystalline state. From the cell walls, cellulose is isolated in microfibrils by chemical extraction. In all forms, cellulose is a very highly crystalline, high molecular weight polymer, which is infusible and insoluble in all but the most aggressive, hydrogen bond-breaking solvents such as N-methylmorpholine-N-oxide. Because of its infusibility and insolubility, cellulose is usually converted into derivatives to make it more process able. All of the important derivatives of cellulose are reaction products of one or more of the three hydroxyl groups, which are present in each glucopyranoside repeating unit, including: (1) ethers, e.g. methyl cellulose and hydroxyl-ethyl cellulose; (2) esters, e.g. cellulose acetate and cellulose xanthate, which is used as a soluble intermediate for processing cellulose into either fibre or film forms, during which the cellulose is regenerated by controlled hydrolysis; and (3) acetals, especially the cyclic acetal formed between the C2 and C3 hydroxyl groups and butyraldehyde. Cellulose acetate is currently used in high volume applications ranging from fibres, to films, to injection moulding thermoplastics. It has the physical properties and relatively low material costs that have tended to exclude other biodegradable polymers from being widely accepted in the marketplace. Cellulose molecules has amphiphilic character and has a high density of hydroxyl groups and consists of a chain of β-(1→4)-linked glucose residues (Gabor &Ovidiu, 2012; Rein et al., 2012).

Chitin and chitosan

Chitin and chitosan belong to the important family of linear polysaccharides consisting of varying amounts of β -(1→4)-linked 2-acetamido-2-deoxy- β -D-glucopyranose (GlcNAc) and 2-amino-2-deoxy- β -D-glucopyranose (GlcN) units. Chitin samples contain a high content of GlcNAc units; hence, they are insoluble in water and common organic solvents. On the other hand, they dissolve only in solvents such as N, N dimethylacetamide, hexafluoroacetone or hexafluoro-2-propanol. When the degree of N-acetylation (defined as the average number of N-acetyl-D-glucosamine units per 100 monomers expressed as a percentage) is less than 50%, chitin becomes soluble in aqueous acidic solutions (pH < 6.0) and is called chitosan. This means that the term "chitosan" denotes a group of fully and partially deacetylated chitins but a rigid nomenclature. The degree of N de acetylation between chitin and chitosan has not been determined. Some authors consider that chitosan is a polysaccharide containing at least 60% GlcN residues. According to the nomenclature proposed by the European Chitin Society (EUCHIS), chitin and chitosan should be categorized on the basis of their insolubility and solubility in 0.1 M acetic acid; the insoluble material is called chitin, whereas the soluble one is chitosan. Chitin and chitosan have great economic value because of their flexible biological activities such as biocompatibility, biodegradability, non-toxicity and adsorptive abilities, as well as chemical applications, mainly in the medical and pharmaceutical industries. Chitin, being a highly insoluble and chemically rather unreactive material, has far fewer applications in comparison to chitosan. By chemically modifying the primary amino and free hydroxyl groups of chitin and chitosan, their solubility in water and organic solvents can be improved, thus increasing their range of biomedical applications. The range of chitin/chitosan molecular weight is extensive (from several to more than thousands of kDa) and is thus divided into three categories: low-molecular-weight chitosan (LMWC), medium-molecular-weight chitosan (MMWC) and high-molecular-weight chitosan (HMWC). As a result of increasing MW, some physicochemical and biological properties of chitin/chitosan and its solutions change, which determines the bioactivity of the material (Kumirska et al., 2011).

Proteins

Proteins can be defined as natural polymers able to form amorphous three-dimensional structures stabilized mainly by noncovalent interactions. The term ''protein'' comes from the Greek, proteios, for ''primary, first and foremost''. The functional properties of these materials are highly dependent on structural heterogeneity, thermal sensitivity, and hydrophilic behavior of proteins. Numerous vegetable and animal proteins are commonly used as biodegradable polymers.

Zein

Zein is the alcohol-soluble protein of corn and is categorized as a prolamine. Prolaminezein and commercial zein are not the same material. It is the main protein of corn.In the kernel, zein is located in protein bodies ≈1 μm in size. Biologically, zein is a mixture of proteins varying in molecular size and solubility. These proteins can be separated by differential solubilities and their related structures into four distinct types: α, β, γ, and δ. Alpha-zein is by far the most abundant, accounting for ≈70% of the total. The next most majorzein is gamma-zein, contributing ≈20% to the total. Alpha-Zein can be extracted using only aqueous alcohol, whereas the other zeins need a reducing agent in the solvent to be extracted. Zein solubilized from whole kernel corn without using a reducing agent will consist of mixtures of aggregates and monomers. Zein that is extracted without using reducing agents is known as native zein (Lawton, 2002).

Wheat Gluten

Wheat gluten is a general term for water-insoluble proteins of wheat flour which is composed of a mixture of polypeptide molecules, considered to be globular proteins. Cohesiveness and elasticity of gluten give integrity to wheat dough and facilitate film formation. Wheat gluten contains the prolamine and glutelin fractions of wheat flour proteins, typically referred to as gliadin and glutenin, respectively. While gliadin is soluble in 70% ethanol, glutenin is not. Although insoluble in natural water, wheat gluten dissolves in aqueous solutions of high or low pH at low ionic strength (Bourtoom, 2008). Gliadins and glutenins are present in almost equivalent quantities in wheat gluten and have comparable amino acid compositions, with high concentrations in glutamine and proline. The amount, size distribution, and macromolecular architecture of glutenins and gliadins greatly influence the rheological, processing, mechanical, and physicochemical properties of the gluten.

Casein

Casein is the principal protein aceous component of milk, where it accounts for about 80% of the total protein inventory. Casein is interesting due to its numerous uses in the food, drug, and cosmetic industries as well as to its importance as an investigation material for clarifying essential questions regarding the chemistry of protein. In fact it is part of a group called phosphoproteins,. It includes four individual gene product components denoted αs1-, αs2-, β- and κ-casein, which differ in primary construction and type and degree of posttranslational modification. These four casein types are fundamentally different in their molecular weights as follows: αs1-casein (MW= 23 KD, ~38.49%), αs2-casein (MW 25KD, ~10.06%), β-casein (MW 24KD, ~38.74%), κ-casein (MW19KD, ~12.57%) (Wang, et al., 2013). There are trace amounts of gamma-casein occurring naturally on account of limited proteolysis of β-casein by plasmin. The principal casein components have several genetic variants and contain variable numbers of phosphoseryl residues. Kappa-Casein contains only one phosphoseryl residue, and it is also glycosylated. Another unique attribute of caseins is the large amount of propyl residues, especially in beta-casein, which greatly affect the composition of caseins, because the proline residues disrupt the formation of alpha-helical and beta-sheet. In addition, all casein proteins have different hydrophobic and hydrophilic zones along the protein chain. Alpha-s-Caseins are the main casein proteins containing 8-10 seryl phosphate groups, while beta-casein contains about 5phosphoserine residues, and it is more hydrophobic than alpha-s-caseins and kappa-casein. Because alpha-s-caseins and beta-caseins are highly phosphorylated, they are very susceptible to the concentration of calcium salts, that is, they will precipitate with excess calcium ions. Unlike other caseins, kappa-caseins are glycoproteins, and they have only one phosphoserine group. Therefore, they are stable in the presence of calcium ions, and they exhibit a main function in protecting other caseins from precipitation. High temperature (above 120ºC) can cause denaturation of casein It causes the casein to gradually become insoluble. Furthermore, it is sensitive to pH and will precipitate at its isoelectric pH (Phadungath, 2005).

Whey proteins

Whey protein is the name commonly applied to milk serum proteins. If the casein is removed from skim milk by some precipitation method, such as the addition of mineral acid, there remains in solution a group of proteins which are called milk serum proteins. As long as they are not denatured by heat, they are not precipitated at their isoelectric points. They are however usually precipitated by polyelectrolytes such as carboxymethyl cellulose. Technical processes for recovery of whey proteins often make use of such substances or of a combination of heat and pH adjustment. When milk is heated, some of the whey proteins denaturate and form complexes with casein, thereby decreasing the ability of the casein to be attacked by rennet and to bind calcium. Curd from milk heated to a high temperature will not release whey as ordinary cheese curd does, due to the smaller number of casein bridges within and between the casein molecules. Whey proteins in general, and alpha-lactalbumin in particular, have very high nutritional values. Their amino acid composition is very close to that which is regarded as a biological optimum. Whey protein derivatives are widely used in the food industry (Bylund, 1995).

Microbial polymer

PolyHydroxy-Alkanoates

In nature, prokaryotic microorganisms react to sudden increases in essential nutrients in their usually hostile environment by storing important nutrients for survival during prolonged period of starvation. PHAs are one such storage compound. PHAs are usually produced when carbon sources are in abundance. The carbon sources are integrated, changed into hydroxyalkanoate (HA) compounds and finally polymerized into high molecular weight PHAs and stored as water insoluble granules in the cell cytoplasm. PHAs are an excellent storage composed because their presence in the cytoplasm, even in large quantities does not disturb the osmotic pressure of the cell. PHA granules can be observed as retractile granules under phase contrast light microscope. Among them, the classification based on monomer size, which refers to the number of carbon atoms in the HA monomer, and the type of polymer is the most common and will be described in detail here. The main enzyme in PHA synthesis, i.e., the PHA synthase, determines the sizes of the monomers that are accepted as substrates for the polymerization reaction. Several types of PHAs have been identified to possess commercial value, i.e., poly(3-hydroxybutyrate) [P(3HB)] that can be blended with starch and also with synthetic plastics to make the latter partially degradable; poly(3-hydroxybutyrate-co-3-hydroxyvalerate [P(3HB-co 3HV)] because of its hardness and excellent processability; poly(3-hydroxybutyrate-co-4-hydroxybutyrate [P(3HB-co-4HB)] because of its superior mechanical properties and bioabsorbability; poly(3-hydroxybutyrate-co-3-hydroxyhexanoate [P(3HB-co-3HHx)] that exhibits close resemblance to polypropylene (PP) and low-density polyethylene (LDPE). PHAs are polymers of carbon, oxygen and hydrogen. R refers to the length of the side chain whereas x refers to the size of the alkyl group. Both R and x determines the type of HA monomer unit. When the total number of carbon atoms in the HA monomer unit is equal to or less than five, the HA is known as a short chain-length (SCL) monomer and the PHAs resulting from the polymerization of SCL monomers are known as SCL PHA. When the total number of carbon atoms in the HA monomer unit is at least six and up to 14, the HA is known as a medium chain-length (MCL) monomer and the PHAs resulting from the polymerization of MCL monomers are known as MCL PHA (Loo &Sudesh, 2007).

Biotechnological polymers

Polylactic acid

Lactic acid (2-hydroxypropionic acid) is the basic building block for PLA. It is a very water-soluble, three-carbon chiral acid that is naturally occurring and is generally appear in the L(-) form. It is used as an acidulate in foods, as a structure block for biodegradable polymers (PLA), and is converted to esters and used as a green solvent for metal cleaning, paints, and coatings. Polylactic acid (PLA) is a solid thermoplastic polymer that can be semi crystalline or totally amorphous, depending on the stereo purity of the polymer backbone. L(-)-lactic acid (2-hydroxy propionic acid) is the natural and most common form of the acid, but D(+)-lactic acid could be synthesized by microorganisms or through racemization. D-lactic acid units are incorporated into L-PLA to optimize the crystallization kinetics for specific making processes and applications. It may be the polymer with the wide area of applications because of its ability to be stress crystallized, thermally crystallized, impact modified, filled, copolymerized, and processed in most polymer processing equipment. It can be mold into transparent films, fibers, or bottles, like PET. PLA also has great organoleptic characteristics and is excellent for food packaging applications. The starting material for the final polymer, lactic acid, is made by a fermentation process using 100% annually renewable resources. The polymer will also rapidly degrade in the environment and its by-products have very low toxicity .They were converted to carbon dioxide and water (Henton et al., 2005).

Petrochemical polymers

Polycaprolactone

Polycaprolactone (PCL) was one of the primary polymers synthesized by the Carothers group in the early 1930s. It became commercially available following efforts to identify synthetic polymers that could be degraded by microorganisms. PCL can be prepared by either ring opening polymerization of ε-caprolactone using a variety of anionic, cationic and co-ordination catalysts or via free radical ring-opening polymerization of 2-methylene-1-3 dioxepane. PCL is a hydrophobic, semi-crystalline polymer; its crystallinity decrease with increasing molecular weight. The good solubility of PCL, its low melting point (59–64 ◦C) and exceptional blend-compatibility has stimulated extensive research into its potential application in the biomedical field. The appeal of these biopolymers was related to numerous advantages over other biopolymers in use at that time. These included tailorable degradation kinetics and mechanical properties, ease of shaping and manufacture enabling appropriate pore sizes conducive to tissue in-growth. Functional groups could also be added to exhibit the polymer more hydrophilic, adhesive, or biocompatible. Because PCL degrades at a slower rate than polyglycolide (PGA), poly d,l-lactide (PDLA) and its copolymers and was therefore originally used in drug-delivery devices that stay active for over 1 year and in slowly degrading suture materials (Woodruff &Hutmacher, 2010).

BIODEGRADATION

The word ‘bio-plastic’ is used confusingly. In our understanding, however, bio-plastics consist of either biodegradable plastics (*i.e.,* plastics produced from fossil materials) or bio-based plastics (*i.e.,* plastics synthesized from biomass or renewable resources) (Tokiwa et al., 2009). There is a global research effort to develop biodegradable polymers as a waste management option for polymers in the environment. Biodegradation (i.e. biotic degradation) is a chemical degradation of materials (i.e. polymers) triggered by the action of microorganisms such as bacteria, fungi and algae. The best definition of a biodegradable polymer is “a degradable polymer wherein the primary degradation mechanism is through the action of metabolism by microorganisms.” Biodegradation is considered a type of degradation involving biological activity. Biodegradation is the major mechanism of loss for most chemicals released into the environment. This process refers to the degradation of polymers by living microorganisms to produce degraded products. The most major organisms in biodegradation are fungi, bacteria and algae. Natural polymers are degraded in biological systems by oxidation and hydrolysis. Biodegradable materials degrade into biomass, carbon dioxide and methane. In the case of synthetic polymers, microbial utilization of its carbon backbone as a carbon source is required. Main bacteria in the biodegradation process include, *Bacillus* (capable of producing thick-walled endospores that are resistant to heat, radiation and chemical disinfection), *Pseudomonas*, *Klebsiella*, *Actinomycetes*, *Nocardia*, *Streptomyces*, *Thermoactinomycetes*, *Micromonospora*, *Mycobacterium*, *Rhodococcus*, *Flavobacterium*, *Comamonas*, *Escherichia*, *Azotobacter* and *Alcaligenes* (some of them can accumulate polymer up to 90% of their dry mass). The most important factors that affected on microorganism growth is temperature. Other important factors are sources of carbon and nitrogen, and pH. Fungi active in the biodegradation process are *Sporotrichum*, *Talaromyces*, *Phanerochaete*, *Ganoderma*, *Thermoascus*, *Thielavia*, *Paecilomyces*, *Thermomyces*, *Geotrichum*, *Cladosporium*, *Phlebia*, *Trametes*, *Candida*, *Penicillium*, *Chaetomium*, and *Aerobasidium*. If oxygen is present, aerobic biodegradation happen and carbon dioxide is produced. If there is no oxygen, an anaerobic degradation happen and methane is produced. When conversion of biodegradable materials or biomass to gases (like carbon dioxide, methane, and nitrogen compounds), water, salts, minerals and residual biomass occurs, this process is called mineralization. Mineralization is carried out when all the biodegradable materials or biomass are used and all the carbon is converted to carbon dioxide. Biodegradable materials have the proven ability to decompose in the most common environment where the material is disposed within one year through natural biological processes into non-toxic carbonaceous soil, water or carbon dioxide. The chemical structure (responsible for functional group stability, reactivity, hydrophylicity and swelling behaviour) is the most important factor affecting the biodegradability of polymeric materials. (Leja&Lewandowicz, 2010). Schema of polymer degradation under aerobic and anaerobic conditions is presented in Figure 2.

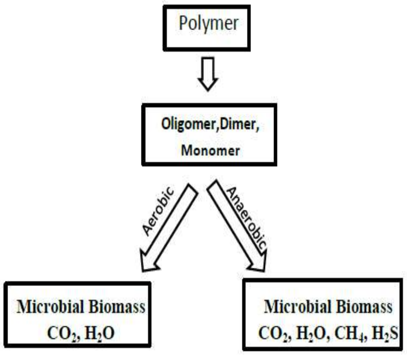

Figure 2. Scheme of polymer degradation under aerobic and anaerobic conditions (Leja&Lewandowicz, 2010).

APPLICATION OF NANOTECHNOLOGY IN FOOD INDUSTRY

Nanotechnology is the generation and use of materials and compostions at the nanometer gauge .It suggests a board range of chance for the development of innovative products and applications in food systems. Nanotechnology and nanomaterials are a natural part of food processing and conventional foods, because the characteristic properties of many foods rely on nanometer sized components, for example, nanoemulsions and foams. Recent technological developments have led the way for the manufacture of nanoparticles to be added to food. These could be finely divided forms of existing ingredients, or completely new chemical structures. While many definitions for nanotechnology exist, the National Nanotechnology Initiative calls it “nanotechnology” only, if it involves all of the following:

1) Research and technology development at the atomic, molecular or macromolecular levels, in the length scale of approximately 1-100 nanometer range

2) Creating and using structures, devices and systems that have novel properties and functions because of their small and/or intermediate size

3) Ability to control or manipulate on the atomic scale.

Nanotechnology applications in the food sector are on the increase globally and are expected to grow speedily in the future. The types of material produced can be at the nanoscale in one dimension (very thin coatings), two dimensions (nanowires) or three dimensions (nanoparticles, such as very fine powder preparations). In food and agricultural systems nanotechnologies cover many aspects, such as food security, packaging materials, disease treatment, delivery systems, bioavailability, and new tools for molecular and cellular biology and new materials for pathogen detection. Nanotechnology recently introduced in the food packaging industry can possibly provide solutions to food packaging problems such as short shelf life (Emamifar et al., 2010). Nanoparticles have many potential for using in agro-food production i.e. processing of pesticides, fertilizers, food additives, cosmetics, feed (e.g. vitamins), packaging and textiles. Food undergoes a variety of postharvest and processing-induced modifications that affect its biological and biochemical makeup, so nanotechnology developments in the fields of biology and biochemistry could also influence the food industry. Systems with structural features in the nanometer length range could affect aspects from food safety to molecular synthesis. Nanotechnology focuses on the characterization, fabrication, and manipulation of biological and nonbiological structures smaller than 100 nm. Structures on this scale have been exhibited to have unique and new functional properties. Using of nanotechnology in the food industry are beginning to have a bearing on the important aspects of food and associated industries from food safety to the molecular synthesis of new food products. Examples of nanotechnology as a tool for achieving further advancements in the food industry are as follows:

1- Increased security of manufacturing, processing, and shipping of food products through sensors for pathogen and contaminant detection

2- Devices to maintain historical environmental records of a particular product and tracking of individual shipments

3- Systems that provide integration of sensing, localization, reporting, and remote control of food products (smart/intelligent systems) and that can increase efficacy and security of food processing and transportation

4- Encapsulation and delivery systems that carry, protect, and deliver functional food ingredients to their specific site of action. Application matrix of nanotechnology in food science and technology are presented in Figure 3.

It can be stated that there are a great number of potential applications of nanotechnology within the food industry. Applications in food packaging are consideredhighly promising because they can modify the safety and quality of food products. This includes intelligent packaging, which is reactive to the environment and active packaging, which is able to interact with the food product (Ozimek et al., 2010; Weiss et al., 2006).

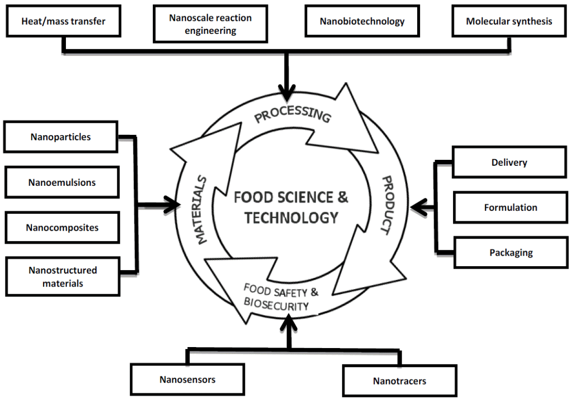

Figure 3. Application matrix of nanotechnology in food science and technology (Weiss et al., 2006).

ANTIMICROBIAL PROPERTIES OF BIO-NANOCOMPOSITES

Nanocomposite materials have been widely studied for approximately the past 20 years. This new generation of nanostructured hybrid materials, constitutes a new class of materials named nanocomposites. Today, polymer nanocompositesare defined as polymers containing fillers with at least one dimension smaller than 100 nm. Contrary to traditional composites, polymer nanocomposites generally involve low content of well-dispersed nanofillers. The main reason is that it is not necessary to fill the polymer with high quantities of filler to achieve high mechanical properties. Particularly, the nanoparticles are generally considered as excellent opportunities for the development of high-performance multifunctional composites. The components of a nanocomposite material can be constituted of inorganic/inorganic, inorganic/organic or organic/organic sources. The recent resurgence of interest in nanocompositesis related to several reasons. Due to the high specific surface area, nanocomposites present a large volume of interfacial matrix material (interphase) with properties different from those of the bulk polymer. The same dispersion of nanoparticles resulted in large matrix-filler interfacial area, changing the molecular mobility, relaxation behavior and ensuing thermal and mechanical properties. Bionanocomposites are bio-based nanocomposites. Actually, they represent an emerging group of nanostructured hybrid materials. Expanding the concept of biocomposites to the nanostructured hybrid materials, bionanocompositescan be defined in the following two ways. It could designate nanocomposites as materials made from renewable nanoparticles and petroleum-derived polymers. However, nanocomposites derived from biopolymers and synthetic or inorganic nanofilers also come under bionanocomposites. Bio-nanocomposites open an opportunity for the use of novel, high performance, light weight green nanocomposite materials making them to replace conventional nonbiodegradable petroleum-based plastic packaging materials. Biopolymers have been considered as a potential environmentally-friendly substitute for the use of non-biodegradable and non-renewable plastic packaging materials. Biopolymer packaging materials also may serve as gas and solute barriers and complement other types of packaging by improving the quality and extending the shelf-life of foods. Furthermore, biopolymer packaging materials are excellent vehicles for incorporating a wide variety of additives, such as antioxidants, antifungal agents, antimicrobials, colors, and other nutrients. Several concerns must be addressed prior to commercial use of biobased primary packaging materials. These concerns include degradation rates under various conditions, changes in mechanical properties during storage, potential for microbial growth, and release of harmful compounds into packaged food products. In reality, relatively poor mechanical and high hydrophilic properties with poor processibility of those biopolymer-based packaging materials are causing a major limitation for their industrial use. Antimicrobial function of some nanoparticles or nanocomposite materials has long been recognized and exploited in the field of various industries including packaging sector for their antimicrobial activity as growth inhibitors, antimicrobial agents, antimicrobial carriers or antimicrobial packaging films. Nanocomoposite antimicrobial systems are particularly effectivebecause of the high surface-to-volume ratio and increase surface reactivity of the nano-sized antimicrobial agents, making them able to inactivate microorganisms more completely than their micro- or macro-scale counterparts. Metal ions (silver, copper, gold, platinum), metal oxide (TiO_2, ZnO, MgO), organically modified nanoclay (quaternary ammonium modified MMT, Ag-zeolite), natural biopolymers (chitosan), natural antimicrobial agents (nisin, thymol, carvacrol, isothiocyanate, antibiotics), enzymes (peroxidase, lysozyme), and synthetic antimicrobial agents (quaternary ammonium salts, EDTA, propionic, benzoic, sorbic acids), generally used to manufacture antimicrobial nanocompositematerials . Typical biodegradable polymers and inorganic or antimicrobial materials investigated for the preparation of bio-nanocompositesare presented in Table 1. Combinations of more than one antimicrobials incorporated into packaging materials have also been investigated. Antimicrobial packaging is another area with high potential for applying bio-nanocomposite technology to control undesirable microorganisms on foods by means of the incorporation of active molecules such as antimicrobial compounds in or coated onto the packaging materials.

Table 1. Typical biodegradable polymers and inorganic or antimicrobial materials investigated for the preparation of bio-nanocomposites(Rhim et al.2013).

Biodegradable polymers	Antimicrobial materials
Starch or thermoplastic starch (TPS)	Clay (e.g. Montmorillonite (MMT))
Chemically modified cellulose (e.g., cellulose acetate (CA) and cellulose acetate butyrate (CAB))	Organically modified nanoclay (quaternary ammonium modified MMT, Ag-zeolite)
Polylactic acid (PLA)	Metal ions (e.g. silver, copper, gold, platinum)
Polycaprolactone (PCL)	Metal oxide (e.g. TiO2, ZnO, MgO)
Polyhydroxyalkanoate (PHA)	Natural biopolymers (e.g. chitosan)
Polyhydroxybutyrate (PHB)	Natural antimicrobial agents (e.g. nisin, thymol, carvacrol, isothiocyanate, antibiotics)
Poly(butylene succinate) (PBS)	Enzymes (peroxidase, lysozyme)
	Synthetic antimicrobial agents (quaternary ammonium salts, EDTA, propionic acid, benzoic acid, sorbic acid)

Antimicrobial food packaging materials must lengthen the lag phase and lessen the growth rate of microorganisms in order to extend shelf life and maintain product quality and safety (Emamifar et al. 2011).Bio-nanocomposites with antimicrobial function are highly useful to minimize the growth of post-processing contaminant microorganisms, extending shelf life of food and maintaining product quality and safety. Nanoparticles or nanocomposite materials have been investigated for antimicrobial activity as growth inhibitors, antimicrobial agents, antimicrobial carriers or antimicrobial packaging films. The major potential food applications for antimicrobial bio-nanocomposite films include meat, fish, poultry, bread, cheese, fruits, and vegetables. Other active packaging applications for bionanocomposites that are expected to find increased attention in the future include antioxidant releasing films, color containing films, light absorbing/regulation systems, anti-fogging and anti-sticking films, susceptors for microwave heating, gas permeable/breathable films, bioactive agents for controlled release, and insect repellant packages. Intelligent or smart packaging is intended to monitor and provide information about the quality of the packaged food or its surrounded environment to predict or decide the safe shelf life. The intelligent/smart packaging may respond to environmental conditions, alert a consumer to contamination of pathogens, detect harmful chemicals or degradation products caused by food deterioration, indicate food quality, and initiate self-healing. The control and manipulation of nano-sized clay platelets made it possible for the creation of smart materials, by combining the wide type of properties provided by the clay with the functionality of organic components. The intelligent packaging application of nanocomposite is mainly based on the function of package to provide information about keeping the product quality such as package integrity (leak indicator), time–temperature history of the product (time–temperature indicators (TTI)), and tracing the origin of the packaged food products (nano barcodes or radio frequency identification (RFID). High barrier nanocoatings consisting of hybrid organic–inorganic nanocomposite coatings by sol–gel process are being developed for oxygen-diffusion barriers for plastics such as PET. The coatings are produced through atmospheric plasma technology using dielectric barrier discharges. The coatings have been reported to be very efficient at keeping out oxygen and retaining carbon dioxide, and can rival traditional active packaging technologies such as oxygen scavengers (Siqueira et al, 2010; Rhim et al., 2013).

APPLICATIONS OF ANTIMICROBIAL POLYMER BIONANOCOMPOSITES FOR FRESH STRAWBERRY PACKAGING

Strawberries are a highly perishable fruit with a short postharvest life mainly due to fungal decay. The shelf-life of fresh strawberries at low temperatures (0–4 ◦C) is usually around 5 days. Therefore, many strategies have been developed to decrease the strawberry losses. As a case study a laboratory trial by Emamifar&Asharfi, 2014, was conducted to producing antimicrobial bionanocomposite films at different concentrations (1%,3%, 5%, and 7%; w/w of total solid) containing ZnO. Therefore, Emamifar&Asharfi, 2014, evaluate the capabilities of ZnO nanoparticles filled starch films, to be used as cap of packaging, to preservation and extended shelf life of strawberry as a new approach. The mean initial population immediately after packaging was determined to be 140 cfu/g for mold and yeast and 700 cfu/g for total aerobic bacteria in fresh strawberries. Variations in population of mold and yeast and total aerobic bacteria are shown in Figures. 4 and 5, respectively. In all packaging, the mean population of mold and yeast has increased after 4 days storage, whereas the mean population of total aerobic bacteria except for packages containing 5% and 7% nano-ZnO has increased after 4 days storage. In these samples a significant decrease ($p<0.01$) in total count was measured in compared to the other packages and pure starch packages (control) during 4 days storing.

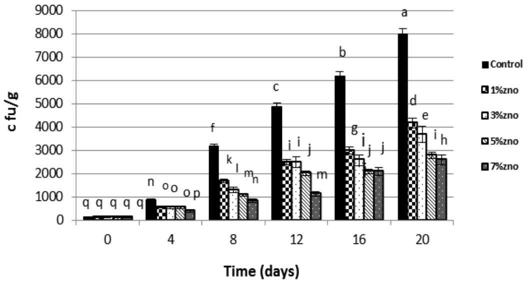

Figure 4. Effect of packaging containing ZnO nanoparticles on the population of mold and yeast during 20 days of storage at 4°C (Emamifar&Asharfi, 2014)

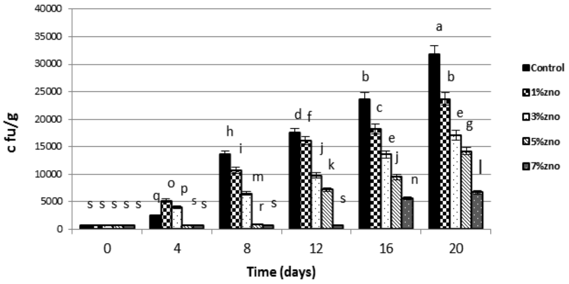

Figure 5. Effect of packaging containing ZnO nanoparticles on the population of mesophile bacteria during 20 days of storage at 4°C (Emamifar&Asharfi, 2014).

It appears that antibacterial activity of nanocomposite containing nano-ZnO is significantly ($p<0.05$) higher than antifungal activity of them. Sawai&Yoshikawa, 2004 shows that ZnO ,CaO and, MgO powder have a good antimicrobial activity against board spectrum microorganisms but antimicrobial activity of ZnO is very weak against *Saccharomyces cerevisiae* and other fungi compared with bacteria. According to Figure 4, the level of mold and yeast population increased to 3200 cfu/g after 8 days of storage in control packages which is higher that of in packages containing 1% (1700 cfu/g), 3% (1300 cfu/g), 5% (1100 cfu/g), and 7% (850 cfu/g) nano-ZnO. Moreover, according to Figure 5, the level of total aerobic bacteria population increased to 13600 cfu/g after 8 days of storage in control packages which is higher that of in packages containing 1% (10700 cfu/g), 3% (6500 cfu/g) 5% (900 cfu/g), 7% (760 cfu/g). However, by increasing the time of storage up to 20 days, microbial population in strawberry increased in all packages but packaging of strawberries that were covered by the nanocomposite containing nano-ZnO indicated a significantly less loading level than control. No significant differences were observed in total aerobic bacteria populations between packages containing 7%, nano-ZnO up to 8 days of storage. However, it seems that packages containing 7% and 5% nano-ZnO have significantly ($p<0.05$) the best antimicrobial activity compared with other nanocomposites up to 20 days storage of fresh strawberry at 4°C . It can be concluded that, by increasing the concentration of nano-ZnO up to 7% the antimicrobial activity of film was increased (Zhang et al., 2008; Zvekić et al., 2011).

CONCLUSIONS

The postharvest sector includes all points in the value chain from production in the field to the food being placed on a plate for consumption. Significant amounts of the food produced in developing countries are lost after harvest. The causes of post-harvest losses, which some estimates suggest could range from 15 to as high as 50 percent of what is produced, are manifold. Therefore, many strategies have been developed to improve losses throughout the process of postharvest. Packaging is an essential component of the food system and plays a critical role in containing, protecting and preservation food and other agro-industrial raw materials from field to the end user. Packaging technology must therefore balance food protection with other issues, including energy and material costs, greater social and environmental awareness, and stringent rules on pollutants and disposal of municipal solid waste (MSW). Proper waste management is crucial in the protection of human health and the environment and the preservation of natural resources. EPA's guidelines for solid waste management stress the use of a hierarchical, integrated management approach involving source reduction, recycling, composting, combustion, and landfilling. Nanotechnology recently introduced in the food packaging industry can provide potential solutions to food packaging challenges such as short shelf life. Therefore, application of nanobiocomposites containing antimicrobial agent in food packaging are the new concepts and technologies required to advance the development of new packaging materials. Using bio-based packaging materials not only increased the shelf life of foods (fruits and vegetables), but also improved their safety, quality and biodegradability. However, antimicrobial bionanocomposite packaging are new ecofriendly food packaging that included natural or synthetic biopolymers based on incorporation of antimicrobial agents into polymer-based nanocomposites, have led to the development of versatile antimicrobial materials that are useful for a wide variety of packaging, biomedical and general use applications.

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Conflicts of Interest

The authors declare no conflict of interest.