ANTIMICROBIAL PACKAGING WITH NATURAL COMPOUNDS – A REVIEW

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ABSTRACT. Background: Packaging problems are an integral part of logistics and the implementation of packaging significantly affects the effectiveness of logistics processes, as a factor which increases the safety and the quality of products being transported. Active packaging is an area of technology needed to meet the requirements of the contemporary consumer. Active packaging creates additional opportunities in systems for packing goods, as well as offering a solution in which the packaging, the product and surroundings interact. Furthermore, active packaging allows packaging to interact with food and the environment and play a dynamic role in food preservation. The main role of antimicrobial packaging is to inhibit the growth of microorganisms that reduce the quality of the packaged product.

Methods: The application of natural antimicrobial agents appears to be safe for food products. Also, these compounds have potential applications as a natural preservative in the food packaging industry. This study presents some antibacterial agents, namely chitosan, nisin and pectins.

Results and conclusion: Natural substances used in active packaging can eliminate the danger of chemical substances migrating to food.

Key words: logistics, antimicrobial packaging, natural antibacterial agents.

INTRODUCTION

The principal function of packaging is protection from heat, light, the presence or absence of moisture, oxygen, pressure, enzymes, microorganisms, insects, dirt and dust particles, gaseous emissions, and other things which might damage the product [Brody et al. 2001]. Antimicrobial packaging is a promising form of active packaging. Active packaging refers to the incorporation of certain additives into packaging film or within packaging containers, with the aim of maintaining and extending product shelf life [Day 1989]. Moreover, active packaging, sometimes referred to as interactive packaging, is intended to sense internal or external environmental change and to respond by changing its own properties and hence the internal package environment [Brody et al., 2001].

Antimicrobial agents may be incorporated directly into packaging materials for slow release into the food surface or may be used in vapour form [Wilson 2007]. Antimicrobials reduce the growth rate and maximum population of microorganisms (spoilage and pathogenic) by extending the lag phase of microbes or inactivating them [Quintavalla, Vicini 2002]. The use of packaging films containing antimicrobial agents could be more efficient than the direct addition of these compounds into food. The nature of the antimicrobial agents is very diverse [organic acids, enzymes, bacteriocins, fungicides, natural extracts, ions, ethanol etc.] as well as
the nature of the materials into which they are included, such as papers, plastics, metals or combinations of these materials [Dainelli et al., 2008].

ANTIMICROBIAL AGENTS

According to Nilsson et al. [2000] antimicrobial packaging technology that has the potential to modify headspace atmosphere creates an effective hurdle against bacterial cells. An antimicrobial in gaseous form is evenly distributed in the headspace of a packaging structure and functions more effectively compared to antimicrobials in solid or liquid forms. The most commonly used antimicrobial gas in food application is CO₂, which inhibits both Gram-positive and Gram-negative microorganisms. Also, an alternative method of generating antimicrobial gas would be the removal of molecular O₂ from the headspace of a packaging structure. Molecular O₂ encourages the growth of obligate aerobic, facultative anaerobic, and microaerophilic microorganisms and causes the development of off flavor and off-color, as well as the loss of nutritional components of packaged food products [de Kruijf et al. 2002].

Antimicrobial effects which are achieved by adding antimicrobial agents into the packaging system or using antimicrobial polymeric materials generally function in one of three ways: release, absorption or immobilisation [Han 2003]. Also, the choice of an antimicrobial agent depends primarily on its activity against the targeted microorganisms [Han, 2000 2005]. Many other factors, however, need to be considered when designing antimicrobial packaging systems, such as specific activity, resistance of microorganisms, controlled release mechanisms, the chemical nature of foods and antimicrobials, storage and distribution conditions, film/container casting process conditions, physical and mechanical properties of antimicrobial packaging materials, organoleptic characteristics and toxicity of antimicrobials, and corresponding regulations [Han 2000]. All these factors should be carefully considered according to the corresponding regulations in order to design an effective active packaging [Sadaka et al. 2014]. As a result, there are many antimicrobial agents that exist and are widely used. Antimicrobial agents have different activities and affect different microorganisms. This is due to the characteristic antimicrobial mechanisms and also to the various physiologies of microorganisms. Therefore three groups of antimicrobial agents can be used: chemical agents, natural agents, and probiotics. Antibacterial agents of natural origin include chitosan, nisin and pectins.

CHITOSAN

Chitosan is natural antimicrobial polymer obtained by deacetylation of chitin obtained commercially from shrimp and crab shell. Also, in literature chitosan has been reported as an antibacterial agent against a wide variety of microorganisms [Entsar et al. 2003, Wu et al. 2005]. The antimicrobial mechanism for chitosan is related to interactions of the cationic chitosan with the anionic cell membranes, increasing membrane permeability and eventually resulting in rupture and leakage of the intracellular material. Moreover, it has been reported that bulk chitosan and its nanoparticles are ineffective at pH<6, probably because of the absence of protonated amino groups [Qi et al. 2004]. The applications of chitosan in food packaging are mainly justified by their antimicrobial and antifungal activities against pathogenic and spoilage microbes. Chitosan-based active films allow food preservation to be extended and the use of chemical preservatives to be reduced [Aider 2010]. According to Rabea et al. [2003], chitosan exhibits greater antimicrobial activity than chitin, due to the greater number of free amino groups, which respond to the antimicrobial activity upon protonation. Cellulose and chitosan are the two most abundant biorenewable natural materials and have shown great promise for their application in food packaging. These naturally-occurring polymers have the ability to form films and moderate oxygen and moisture permeability [Byun et.al. 2012, Mi et al. 2006, Pereda et al 2011, Souza et al. 2010, Vargas et al. 2011]. Yu et al. [2013] prepared packaging films from water-soluble chitosan [N,O-carboxymethyl chitosan, NOCC] and cellulose [methylcellulose, MC]. They prepared active
Antimicrobial packaging with caffeic acid which was incorporated into the composite films in fixed and releasable types. The caffeic acid-incorporated films showed 20-fold increases in antioxidant activity and 6-fold increases in antibacterial activity as compared to the caffeic acid-free composite films. The releasable caffeic acid could be continuously released from the composite films and showed a significant inhibitory effect on lipid oxidation of menhaden oil-in-water emulsion. The chitosan antifungal action was evaluated against Alternaria alternata, Aspergillus niger, and Rhizopus oryzae [Ziani et al. 2009]. Cruz-Romero et al. [2013] presented antimicrobial activity of chitosan, organic acids and nano-sized solubilisates for potential use in smart antimicrobially-active packaging for potential food applications. Abdou et al. [in press] demonstrated that a plasma pretreated polypropylene surface treated with chitosan could impart antibacterial and antifungal properties, since chitosan has been shown to possess efficient antimicrobial abilities. One of the most promising active bio-films is the one based on chitosan combined with different materials, such as plant and animal proteins, polysaccharides and antimicrobial peptides [bacteriocin] such as nisin and divergicin, a new class of bacteriocin produced by Carnobacterium divergens [Tahiri et al. 2004, Tahiri et al. 2009]. Moreover, Elsabee et al. [2008] studied tomato packing in bags of transparent polypropylene film coated with twelve non-nanoscale alternating layers of chitosan and pectin, in view of a new concept of active packaging for fruit preservation. Chitosan and pectin can interact at pH 5.6. However the gel behaviour depends upon the degree of esterification of the pectin. In fact the polyelectrolyte complex formation requires ionized carboxylate 015groups of pectin and protonated amino groups of chitosan [Lehr et al. 1992].Wang et al. [2015] prepared antibacterial packaging film containing chitosan/poly[vinyl alcohol] [Nisin-CS/PVA]. The experimental results showed antimicrobial activity films against S. aureus which may have potential as an active film in food packaging. Duran et al. [2016] presented the use of nisin, natamycin, pomegranate and grape seed extracts in chitosan coating to extend the shelf life of strawberries. Antimicrobial agents were added to chitosan at a concentration of 1% w/v. The results showed that all coatings have a good effect on the quality of strawberries during the storage period.

**NISIN**

Nisin is an antimicrobial peptide with 34 amino acids and a molecular weight of 3.5 kDa, produced by strains of Lactococcus lactissubsp. lactis [Mulders et al. 1991]. Moreover, is the most common bacteriocin, tested for many applications. This peptide has antimicrobial properties, especially against the food-borne pathogens Listeria monocytogenes, Staphylococcus aureus or Bacillus cereus [Brewer et al. 2002, Lopez-Pedemonte et al. 2003]. Due to its effect on the important Gram positive food-borne pathogens, many studies have focused on the incorporation of nisin into various kinds of films made of cellulose, nylon, whey protein isolate, hydroxypropyl methylcellulose, zein etc. and their use as nisin delivery system packaging films to reduce undesirable bacteria in foodstuffs [Chollet et al. 2008, Coma et al. 2001, Gadang et al. 2008, Ko et al. 2001, Kristo et al. 2008, Natrajan Sheldon 2000, Neetoo et al. 2008, Nguyen et al. 2008, Teerakarn et al. 2002]. Studies have also been published by a number of authors on the use of nisin as an antimicrobial in a wide variety of food products [Delves-Broughton et al. 1996]. Nguyen et al. [2008] prepared nisin-containing bacterial cellulose film to inhibit Listeria monocytogenes on processed meats. This cellulose film containing nisin was developed and used in a proof-of-concept study to control Listeria monocytogenes and total aerobic bacteria on the surface of vacuum-packed frankfurters. Additionally, many studies have shown that nisin may be efficiently incorporated into cellulose-based packaging films and used for controlling pathogens in food products [Ming et al. 1997, Scannell et al. 2000, Franklin et al. 2004, Luchansky & Call 2004].

Ercolini et al. [2010] studied the spoilage-related microbial populations of beef and to investigate the effect of nisin-activated antimicrobial packaging on the development of beef spoilage at low temperatures.
The combination of chill temperatures and antimicrobial packaging proved to be effective in enhancing the microbiological quality of beef cuts by inhibiting LAB, carnobacteria and Brochothrix thermosphacta in the early stages of storage and by reducing the loads of Enterobacteriaceae. Economou et al. [2009] presented the effect of nisin and EDTA treatments in increasing the shelf-life of raw poultry products stored under modified atmosphere packaging at 4°C. Neetoo et al. [2008] used nisin-coated plastic films to control Listeria monocytogenes on vacuum-packaged cold-smoked salmon. The effect of storage temperature, nisin concentration on low-density polyethylene [LDPE] film, and inoculation level on the growth and survival of Listeria monocytogenes was investigated. At 4°C [low and high inoculum levels] and 10 °C [low inoculum level], it was found that the degree of inactivation or growth inhibition of Listeria monocytogenes was directly related to the concentration of nisin. The fact that nisin delayed the growth of Listeria monocytogenes populations in smoked salmon at both low and high inoculum levels show that nisin might be used to control post-processing contamination of Listeria monocytogenes in cold-smoked salmon. Cao-Hoang et al. [2010] presented the effectiveness of the nisin-coated sodium caseinate films against Listeria innocua in cheese during storage at refrigerated temperatures, indicating that combining nisin into sodium caseinate films is a promising method to enhance the safety and extend the shelf life of processed cheeses.

PECTIN

Pectin is a natural polysaccharide, poly [1,4-galacturonic acid], which is obtained from the cell walls of terrestrial plants and exhibits polyanionic behaviour [Farris et al. 2011, May 1990]. It is now known that pectin is a major component of the plant cell wall and the most complex macromolecule in nature [Voragen et al., 2009]. Pectin is also a high-molecular weight, biocompatible, non-toxic, and anionic natural polysaccharide extracted from cell walls of higher plants [Zouambia et al. 2009]. Gopi et al. [2014] extracted pectin from banana peel. The experimental results revealed that the hydroxyapatite HAP nanoparticles synthesized in the presence of an optimized concentration of pectin are pure, low crystalline, spherical and discrete particles with reduced size. Ravishankar et al. [2012] studied films based on pectin and apple, carrot or hibiscus. The films were treated with carvacrol or cinnamaldehyde and their antimicrobial activity was tested against Listeria monocytogenes on contaminated ham and bologna.

Physical or chemical modification of pectin can lead to new products with significant functional properties. The applications of pectin have been also extended greatly from food and food additives to various fields, such as drug delivery [Mishra et al. 2008, Souto-Maior et al. 2010], antithrombotic agents [Cipriani et al. 2009], and mucoadhesive [Sharma, Ahuja 2011] and antimicrobial substances in materials for packaging food. Moreover, Tripathi et al. [2010] developed an antimicrobial chitosan/poly [vinyl alcohol]/pectin ternary film for food packaging applications. It exhibited significant antimicrobial activity against various pathogenic bacteria such as Escherichia coli, Staphylococcus aureus, Bacillus subtilis, Pesudomonas and Candida albicans. Pectin is a general term for a group of valuable natural polysaccharides extracted from edible plant material where they occur as structural materials. Its main sources are citrus peel and apple pomace [Stasse-Wolthuis et al. 1980, Thibault, Ralet 2001]. Gorrasi et al. [2012] prepared films containing pectins obtained from apples. The films showed antimicrobial activity, indicating the potential application of prepared complexes in the packaging field and the potential usage of pectin-antimicrobials as coating agents for a wide number of packaging polymers. The antimicrobial activity of pectin edible films incorporated with nisin and its combination with treatment of ionizing radiation was used to control Listeria monocytogenes on a ready-to-eat [RTE] turkey meat by Jin et al. [2009]. The combination of irradiation with pectin film containing nisin resulted in a 3.95 log CFU/cm² reduction at 1 kGy and a 5.35 log CFU/cm² reduction at 2 kGy; indicating a synergistic effect on Listeria viability on the surface of RTE turkey meat. In the same year, Jin et al. [2009] tested pectin-poly lactide [PLA] composite films.
treated with nisin against Listeria monocytogenes. Alves et al. [2011] developed models of composite films based on commercial pectin and carrageenan, containing organically modified nanoclays. The barrier properties to water vapor and CO$_2$ of a polymeric matrix composed of kappa-carrageenan and pectin [66.7% kappa-carrageenan], with the inclusion of nanoclays, was studied. The effect of particle content of the films on water vapor permeability [WVP] was dependent on the driving force applied.

**SUMMARY**

Traditional food packaging is designed to mechanically support otherwise non-solid food, and to protect food from external influences. Antimicrobial packaging appears to be one of the most promising applications of active food packaging technology.

Natural compounds that do not have any significant medical or environmental impact could potentially serve as effective alternatives to conventional antibacterial or antifungal agents. Therefore, the preparation of antimicrobial materials with natural substances is a huge challenge to researchers and producers of packaging food.

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**Ergebnissen und Fazit:** Die natürlichen antimikrobiellen, in den aktiven Verpackungen verwendeten Substanzen beseitigen zugunsten der Verbraucher die Gefahr der Migration von chemischen Stoffen zu Lebensmitteln.

**Codewörter:** Logistik, antibakterielle Verpackungen, natürliche antibakterielle Substanzen

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