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Antimicrobial packaging films with a sorbic acid based coating

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Abstract

Following the enhancing demand for packaged raw food products with extended shelf-life, intensive efforts have been made developing active packaging materials with incorporated antimicrobials. With regard to technical problems in the production of antimicrobial films containing sorbic acid, the aim of this study was to evaluate the feasibility of packaging films coated with a lacquer containing sorbic acid to inhibit the growth of contaminating microorganisms on food surfaces. The antimicrobial efficacy was determined applying the test strains *Escherichia coli* DSM 498, *Listeria monocytogenes* DSM 15675 and *Saccharomyces cerevisiae* DSM 70449 according to the Japanese Industrial Standard Method JIS Z 2801:2000. Storage tests were performed with Gouda cheese and pork loin inoculated with the test strain *E. coli* and covered with the antimicrobial films and reference films respectively. In both storage experiments the results of the viable cell count showed a significant inhibitory effect on *E. coli* due to the antimicrobial properties of the developed packaging films, whereas there was no reduction of viable cells on the surface of the reference samples. These findings affirm that antimicrobial packaging films with a sorbic acid based coating are promising to provide a significant contribution to the quality and safety of packaged food.

Keywords: active packaging; sorbic acid; antimicrobial film; antimicrobial efficacy

1. Introduction

Concerning food quality, consumers requirements are on a high level: Food products ready-to-eat, pre-portioned, remaining fresh and retaining their attractive appearance for days are requested on the market and following retailers and distributors are demanding products that stay fresh and especially microbiologically safe for a long time. Therefore simple packaging with a passive barrier layer is often...
insufficient. Active packaging materials equipped with antimicrobials have been developed in order to increase the shelf-life of food products and reduce a potential microbial risk. Especially in the last ten years there has been done a lot of research to develop new films with antimicrobial properties [1].

In 2009 a new EU regulation on active and intelligent packages has been established that facilitates the application of this new packaging technology in the European Union [2].

At the Fraunhofer Institute for Process Engineering and Packaging IVV, Germany, a new antimicrobial packaging film has been developed. First trials have shown that this active film is very effective to protect food surfaces from the growth of various bacteria, yeasts and moulds [3]. The active antimicrobial applied in this film is sorbic acid as a conventionally used preservative that represents no health or allergenic hazard [4, 5]. It is approved according to food law for many different kinds of foods [6]. As there are still technological problems in the production of films with incorporated sorbic acid, the antimicrobial is embedded in a lacquer on top of the packaging film. The lacquer enables a successive release of low amounts of sorbic acid to the surface of the packaged food. In this way a protection from microbial contamination of the packaged food at the contact area could be provided. Thus longer shelf-life and safer foods can be achieved.

The objective of this study was to determine the antimicrobial activity of this active packaging film against various food contaminating microorganisms according to the Japanese Industrial Standard Method JIS Z 2801:2000 [7] and to study its antimicrobial efficacy on artificially contaminated foodstuffs like meat and cheese.

2. Materials & Methods

2.1. Film preparation

Polyvinyl acetate was dissolved in ethyl acetate by stirring at room temperature. After the polyvinyl acetate was solved, sorbic acid was added to the lacquer. The lacquer was applied by a lab bench coater on a common polyethylene packaging film and a polyethylene polyamide composite film respectively. The lacquer was dried by air.

2.2. Test for antimicrobial efficacy

The antimicrobial efficacy of the developed active films was investigated via the Japanese Industrial Standard Method JIS Z 2801:2000 [7].

The applied test strains were E. coli DSM 498, Listeria monocytogenes DSM 15675 and Saccharomyces cerevisiae DSM 70449. All test strains were obtained from “Deutsche Sammlung von Mikroorganismen und Zellkulturen GmbH, Braunschweig, Germany (DSM)”. For each test strain an aliquot of an overnight culture was added to a 500-fold diluted nutrient broth in order to adjust a concentration of at least 1 x 10⁵ colony forming units per ml (cfu/mL).

Triplicates of each test film were cut to a size of 5 x 5 cm, inoculated with 400 μL of the test inoculum and covered with a sterile PET-film. Two uncoated films were used in triplicates; one for the determination of the initial cell count directly after inoculation, the other was treated according to the test samples as a reference. All samples were incubated for 24 hours at the specific growing temperature of the germs (E. coli and Listeria monocytogenes at 37 °C, Saccharomyces cerevisiae at 25 °C) and 90 % relative humidity. A colony count after 24 h showed the number of the surviving cells.

2.3. Storage test of Gouda cheese

Pre-sliced and packed Gouda cheese was cut to a size of 7 x 7 cm and inoculated with 1.8 x 10³ cfu/cm² of E. coli DSM 498 using a clean bench. Finally they were covered with the lacquered
polyethylene-film. For reference samples, inoculated cheese slices were covered with the according lacquered polyethylene-film without sorbic acid. The cheese samples were stored in a sterile petri-dish at 8 °C up to 28 days. The count of *E. coli* was monitored as a function of time. After homogenizing the cheese with Ringer dissolution, the viable count of *E. coli* was determined by pour-plate method using violet red bile agar (VRB) after 0, 1, 3, 7, 14, 21 and 28 days of storage. Each inoculation and the colony count were done in triplicates.

2.4. Storage test of pork

Pork loin was cut into pieces of 5 x 5 cm and a thickness of approx. 2 cm using a clean bench. These samples were inoculated with 1.3 x 10³ cfu of *E. coli* DSM 498 and finally vacuum packed with the lacquered composite film of polyethylene and polyamide. For reference samples, inoculated meat pieces were packaged with the according non-lacquered composite film. The samples were stored at 8 °C up to 7 days. The count of *E. coli* was monitored as a function of time. The viable count of *E. coli* was determined by pour-plate method using violet red bile dextrose agar (VRBD) after 0, 1, 3 and 7 days of storage. Each inoculation and the colony count were done in triplicates.

2.5. Calculation of error bars

The error bars in the following figures (results and discussion) show standard deviation based on 3-fold measurements.

3. Results & Discussion

3.1. Antimicrobial efficacy

Investigating the antimicrobial efficacy of the developed film, for *E. coli* DSM 498 a count reduction of 6.5 log, for *Listeria monocytogenes* DSM 346 a count reduction of 5.6 log and for *Saccharomyces cerevisiae* DSM 70449 a count reduction of 5.4 log was observed in comparison to the reference samples (Fig. 1). These findings prove that the developed antimicrobial film protects effective against pathogenic food-infecting microorganisms like *E. coli* and *Listeria monocytogenes* as well as food spoiling contaminants like yeasts.

![Fig. 1. Antimicrobial activity of an antimicrobial film with a sorbic acid based coating and a reference film without sorbic acid according to JIS Z 2801:2000 [8]](image_url)
3.2. Storage test of Gouda cheese

*E. coli* is known as a faecal contaminant of all kinds of foods, but a contamination could also be affected by the raw milk [9]. Therefore this germ was chosen for the storage test of cheese. To study the effect of the developed antimicrobial packaging film, Gouda cheese was inoculated with 1.8 x 10³ cfu/cm² of *E. coli* DSM 498. While on the reference samples, covered with a lacquered polyethylene-film without sorbic acid, the viable count of *E. coli* stayed nearly the same during the storage time of four weeks, the viable *E. coli*-count on the cheese slices covered with the active film was reduced by approx. one log cycle to one tenth of the initial count (Fig. 2). In summary a positive effect of the antimicrobial film on the microbiological safety of cheese after contamination could be demonstrated.

![Graph showing bacterial count of E. coli DSM 498 on Gouda cheese](image)

Fig. 2. Development of the bacterial count of *E. coli* DSM 498 in a period of 28 days at 8 °C on inoculated Gouda cheese covered with an antimicrobial film with a sorbic acid based coating and a reference film without sorbic acid [8]

3.3. Storage test of pork

Freshly slaughtered meat, which is nearly sterile, generally becomes contaminated with microorganisms during subsequent cutting and processing stages e.g. by feces, hides and skins, germs on tools, hands or rinsing water [10, 11]. So it is not random that especially surfaces of meat pieces can get contaminated with pathogens like *E. coli*.

To prove the antimicrobial effect of the active film on meat, pork loin pieces were inoculated with 1.3 x 10³ cfu/piece of *E. coli* DSM 498. Fig. 3 shows, that a count reduction to one forth of the initial inoculum could be observed, while the bacterial count of *E. coli* on the reference stayed nearly the same after 7 days of storage. As also found in the case of the Gouda cheese, a positive effect of the antimicrobial film on the microbiological status of the meat samples after contamination could be demonstrated.
Fig. 3. Development of the bacterial count of *E. coli* DSM 498 in a period of 7 days at 8 °C on inoculated pork loin packaged in an antimicrobial film with a sorbic acid based coating and a reference film without sorbic acid [8]

4. Conclusion

The antimicrobial packaging film developed at Fraunhofer IVV could be shown to be efficient against various food-contaminating microorganisms like *E. coli*, *Listeria monocytogenes* and the yeast *Saccharomyces cerevisiae*. The antimicrobial efficacy against *E. coli* could be observed with a standardized test method as well as via application on food samples like artificially contaminated cheese and meat. This antimicrobial film is able to prevent and even reduce the growing of pathogens such as *E. coli* on food surfaces. In this way the sorbic acid containing lacquer coating is a promising technique to contribute significantly to the quality and safety of packaged food.

References


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