

Article

Antimicrobial Double-Layer Coating Prepared from Pure or Doped-Titanium Dioxide and Binders

Ran Li ^{1,2}, Tony Z. Jin ¹, Zengshe Liu ³ and LinShu Liu ^{1,*} 

¹ Eastern Regional Research Center, Agricultural Research Service, U.S. Department of Agriculture, 600 E. Mermaid Lane, Wyndmoor, PA 19038, USA; ran.li2@wsu.edu (R.L.); tony.jin@ars.usda.gov (T.Z.J.)

² State Key Laboratory of Hollow Fiber Membrane Materials and Processes, Tianjin Polytechnic University, No. 399 Binshuixi Road, Xiqing District, Tianjin 300387, China

³ National Center of Agricultural Utilization Research, Agricultural Research Service, U.S. Department of Agriculture, 1815 N. University Street, Peoria, IL 61604, USA; kevin.liu@ars.usda.gov

* Correspondence: Linshu.liu@ars.usda.gov; Tel.: +1-215-233-6486

Received: 27 October 2017; Accepted: 17 January 2018; Published: 20 January 2018

Abstract: Fruit and vegetable containers with microbe-free surfaces can be made by coating with titanium dioxide (TiO₂) particles or nonmetal (C, N, B, F) doped-TiO₂ particles, using wear resistant polymers, such as zein, and paint, as the binders and to form a continuous binding phase. The doped-TiO₂ powders absorb visible light radiation, and thus possess a higher antibacterial effect than non-modified TiO₂ particles in environmental conditions. The study also presents a double-layer coating to use less TiO₂ particles in coating, while achieving higher antimicrobial activity. Containers with microbe-free surfaces can stop cross-contamination from infected workers or spoiled/decayed/contaminated fruits or vegetables, and thus are expected to be able to reduce the risk from microbiological contamination of fruits and vegetables during harvest in fields, and postharvest storage or transportation.

Keywords: microbe-free surface; nonmetal-doped titanium dioxide; produce

1. Introduction

The fruit agribusiness contributes more than \$9 billion annually to the US economy. About 59% of apple and 37% of other non-citrus fruits (such as peach, plum, grape, and cherry, etc.) are consumed by Americans without processing [1]. Eating fresh produce improves quality of life, and fresh fruits and vegetables are important components of a healthy and balanced diet to protect from and fight against a wide range of illnesses, such as diabetes, cancers, high blood pressure, obesity, and cardiovascular diseases. With an increase in the awareness of advantages of fresh foods over processed foods, and through the encouragement of government health agencies and social organizations, the consumption of fresh fruits and raw vegetables has increased in industrial countries and post-industrial countries [2,3]. Meanwhile, the risk of digestive diseases associated with the consumption of produce has also increased [4,5].

Fruit and vegetable contamination can occur in the field or at the postharvest stage. Foodborne pathogens can establish themselves on growing fruits and vegetables from contaminated irrigation water, flies, insects, and soil, or from raw animal manure, sewage, etc. The contaminants can be amplified by the proliferation of the pathogens, or by cross-contamination from infected workers, agricultural materials, or equipment used during harvest and storage, such as containers or shelves [6–8]. Containers are used for the collection of fruits and vegetables in fields, transportation, and storage in warehouses and retail rooms. Containers and shelves, either made from cellulosic materials (such as cardboards, baskets, and boxes from bamboo or wood), plastics, or metals, seldom

are disinfected or rarely have self-sterilizing functions. They may provide an additional route for transmission (cross-contamination) of pathogens and spoilage organisms once they are contaminated.

Titanium dioxide (TiO₂) has a strong killing effect on all kinds of bacteria under UV irradiation, while metal (Cu, Co, Ni, Cr, Mn, Mo, Nb, V, Fe, Ru, Au, Ag, Pt)-doped-TiO₂ and nonmetal (N, S, C, B, P, I, F)-doped-TiO₂ exhibits high antibacterial activity, even under visible light [9–14]. Due to their chemical stability, non-toxicity, and capacity to be repeatedly used without losing activity, these inorganic compounds have demonstrated usefulness as disinfection or sterilization reagents for water and air purification, self-clean solid surfaces, and food protection.

We present here a double-layer coating consisting of a binding phase to cover the solid surfaces with a topping dress of TiO₂ or doped-TiO₂ particles. Both of the coating materials were tested for antibacterial activity under UV and visible light radiation, and for durability.

2. Materials and Methods

2.1. Materials

TiO₂ particles (rutile, particle size <5 μm; 99.9%), tetraethyl ammonium tetra-fluoroborate [(CH₃CH₂)₄NBF₄, TEATFB, 99%], Zein (95%), and ethanol (200 proof) were purchased from Sigma-Aldrich-Fluka (St Louis, MO, USA). Petri dish was from VWR (85 × 10 mm, *D* × *H*; West Chester, PA, USA). Paint (KILZ Latex 2[®]; Masterchem Industries, LLC, Imperial, MO, USA) was purchased from a local HomeDepot store (Wyncote, PA, USA).

Escherichia coli O157:H7 Oklahoma was obtained from the culture collection of the U.S. Department of Agriculture, Agricultural Research Service, Eastern Regional Research Center (Wyndmoor, PA, USA). Tryptic soy broth (TSB) was from Remel, Inc. (Lenexa, KS, USA). Tryptic Soy Agar (TSA) was from Difco/Becton Dickinson (Sparks, MD, USA). Butterfield's phosphate buffer (pH 7.2) was from Hardy Diagnostics (Santa Maria, CA, USA). Deionized (D.I.) water was prepared by ion exchange (Millipore; Billerica, MA, USA).

2.2. Preparation and Characterization of Doped-TiO₂ Particles

C, N, B, F-codoped TiO₂ particles were prepared as described by Pelaez and Im et al. [10,15]. Briefly, TiO₂ powders and TEATFB were mixed at an appropriated ratio and then dried at 110 °C followed by calcination at 600 °C for 2 h under vacuum at the heating rate of 10 °C/min. The resultant powders were characterized for the element concentration and the electronic state of the elements of the materials using an X-ray photoelectron spectrometer (K-AlphaTM + XPS; ThermoFisher Scientific, Waltham, MA, USA).

2.3. Coating Preparation

Single-layer coating and double-layer coating were prepared in the present study (Figure 1). Zein and commercially available paint were chose as binders. Zein has demonstrated the properties that are necessary to form tough, glossy, hydrophobic, greaseproof coatings that were resistant to microbial attack [16], which were found to be useful in a variety of applications in bioplastics, coating and adhesive materials, laminated board and substitutes for shellac, carriers of bioactives, and pharmaceuticals [17–22]. Using zein as a binder, the coating materials were made by mixing TiO₂ or doped-TiO₂ in zein/ethanol solution (85% ethanol; zein content, 2.5%, *w/v*). Briefly, 25, 50, or 250 mg TiO₂ or doped-TiO₂ powders were dispersed in 100 mL of zein/ethanol solution, in which the ratios of TiO₂/zein were 1, 2, and 10 wt %, respectively.

For single-layer coating, 10 mL of each TiO₂/zein solution was evenly distributed across a petri dish followed by being air-dried on bench at room temperature. Petri dish coated with 10 mL zein solution was also prepared.

For double-layer TiO₂/zein coating, 8 mL of zein solutions were coated on the surface of a petri dish to form first layer, when the first layer was 80% dried (about 3 h), 2 mL of 10% TiO₂/zein or doped-TiO₂/zein suspension were spread on the surfaces of the first zein layer to form the second layer.

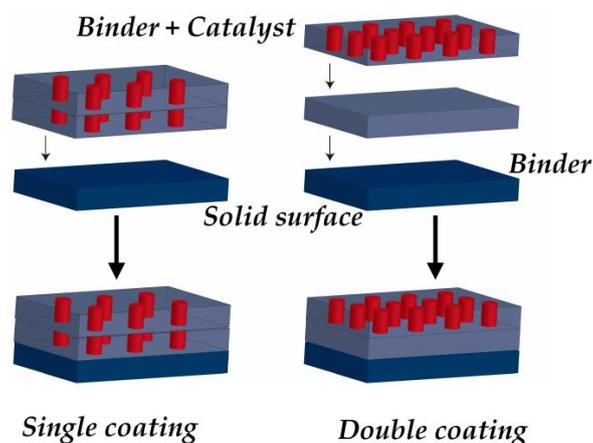


Figure 1. Preparation of single and double layer coating.

To test the durability of coating, coating layers were made on metal plates ($D \times H$, 25 × 2 mm; 1.5 mL for each), using the mixture of TiO₂ particles (6%) with KILZ Latex. The paint has been used popularly in the decoration and protection of house, furniture, automobile, ship, and many others that may contact with air, water, foods, and hazards, etc. The painting was firmly adhered to material surfaces after drying. Based on information we have so far, no findings of residual KILZ Latex in food have been reported.

2.4. Antimicrobial Activity Test

Fluorescent light (FL) has a broad range of wavelength that covers UV wave length (300–380 nm) and visible light length (400–800 nm). A fluorescent light installed inside of biohood (40 W, 80 cm above the samples) was used for light irradiation. The illumination density was 11 $\mu\text{mol}/\text{s}\cdot\text{m}^2$, as measured by a photometer with Quantum sensor (Li-COR, Lincoln, NE, USA).

The tests were carried out as reported previously [23]. Briefly, prior to the inoculum preparation, *E. coli* O157:H7 cells were grown in TSB aerobically at 37 °C for 16–18 h. Serial 10-fold dilutions were performed in sterile 0.1% peptone water and inoculated into TSB so as to achieve a population of ca. 4 log colony forming unit (CFU/mL).

5 mL of inoculum was evenly distributed on each coated petri dish, which was enough to cover the surface of the petri dish. Petri dishes without any coating served as controls. Samples were evaluated in three different ways: under FL irradiation for 18 h, under FL irradiation for 15 h followed by placing in dark for 3 h, and under FL irradiation for 3 h followed by placing in dark for 15 h. Specimens, 100 μL of the inoculated TSB in petri dishes, were sampled at hour 3 of the treatment and at the end of experiment (18 h), which was followed by a set of dilution with sterile Butterfield's phosphate buffer (pH 7.2), surface plated (100 μL per plate and three plates per dilution) onto TSA. Plates were incubated at 37 °C for 24 h before counting CFU. After first sampling, inoculum left on each petri dish was decanted and each petri dish was washed with 10 mL sterile water once, and placed in biohood with fan on for 10 min. Then each dish was poured with 10 mL TSA (50 °C) and incubated at 37 °C for 24 h. Each experiment was conducted in triplicate.

2.5. Durability of TiO₂ Coating

The durability of TiO₂ coatings was examined using metal disks coated with TiO₂/paint formulations. Each coated disk (there were four disks for each coating) was placed in a 3.0 L beaker

with 2.0 L water and stirred at 1000 rpm for 1 min at room temperature with the aid of mechanical stirring (Cole-Parmer digital mixer; Cole-Parmer, Vernon Hills, IL, USA). The distance from the tip of the stirrer to the bottom of the beaker was 10 cm. The disks were washed up to 5 times to simulate the repeated use of a food container. The metal disks were moved out from the water and air-dried, and submitted to UV irradiation for 10 min to eliminate possible microbial contamination. The disks thus treated were surface-inoculated with 50 μ L of *E. coli* inoculum. The inoculated disks were placed in biohood under regular fluorescent lamp as described above for 3 or 18 h. At 3 or 18 h, each disk was swept with a wet cotton tip across the entire sample surface in three different directions (0° , 45° , and 90°), put the swab tip off into a test tube containing 10 mL of peptone water, and vortexed in the tube for 1 min. Specimens were serially diluted and surface plated onto TSA. Plates were incubated at 37°C for 24 h before counting CFU.

2.6. Statistical Analysis

Antimicrobial experiments were conducted in triplicate. Data points were expressed as the mean \pm standard deviation. Data were analyzed using analysis of variance from SAS version 9.1 software (SAS Institute, Cary, NC, USA). Duncan's multiple range tests were used to determine the significant difference of mean values. Unless stated otherwise, significance was expressed at 5% level.

3. Results and Discussion

3.1. Doped-TiO₂ Preparation

The results of elemental analysis of pure and doped-TiO₂ powders were shown in Table 1. In contrast to the non-treated TiO₂ powders, the non-metal elements of boron, carbon, nitrogen, and fluorine were detected with the doped samples. The concentrations of the 4 elements in the prepared samples reduced in the sequence of C > F > N > B, which matches the order in the starting material TEATFB. In a previous publication [15], Lee et al. reported the order of C > B > F > N found in doped-TiO₂ powders that were prepared by the same method as ours. The authors claimed that the volatilization of F and N during calcination was the cause of the reduction of the amounts measured for F and N. However, we found it only occurred when the calcination temperature up to 1000 $^\circ\text{C}$.

Table 1. Atomic concentrations (%).

Sample	B	C	N	O	F	Ti
Non-treated TiO ₂	0.0	0.0	0.0	68.2	0.0	30.8
Modified TiO ₂	0.4	17.8	0.6	56.0	1.2	22.5

The XPS spectra of the Ti 2p levels of pure TiO₂ and doped-TiO₂ were shown in Figure 2. The binding energy of the core level in doped sample indicated the successful incorporation of dopants into the lattice, confirming the results shown in Table 1. We measured the N 1s XPS spectra of the prepared codoped-TiO₂ samples. A significant peak at around 400 eV was recorded (Figure 3). This finding is consistent with that reported previously, where peaks at around 400 eV were assigned to N 1s for doped-TiO₂ powders prepared from TiO₂ and TEATFB and for N-doped anatase TiO₂ made by the atmospheric-pressure plasma-enhanced nanoparticles synthesis (APPENS) process operated under normal temperature [24]. We also measured the 1s XPS spectra of C, B, and F in the prepared codoped-TiO₂ samples, recorded a peak of 285.6 eV for C–Ti, a peak at 191.7 eV for B–Ti, and a peak around 684.5 eV for F–Ti, as well as another peak of 685 eV in the “shoulder” of asymmetrical line for F (standing for F–Ti–O bond). These data were very close to that shown in [15], where 402 eV was assigned to N–Ti, 191 and 187.5 eV to B–Ti, 285 eV to C–Ti, and 685 eV to F–Ti. Similar results were also published by other laboratories [24–28]. By further measuring the ultraviolet-diffuse reflectance spectra (data not shown), it is easy to calculate the minimum energy required to activate the electron

in the doped-TiO₂ samples that fell into the range of 2.98–3.06 eV, which is less than 3.2 eV for TiO₂. This reduction can be attributed to the charge-transfer transition between the dopants and the TiO₂ conduction or valence band, as has been suggested [29].

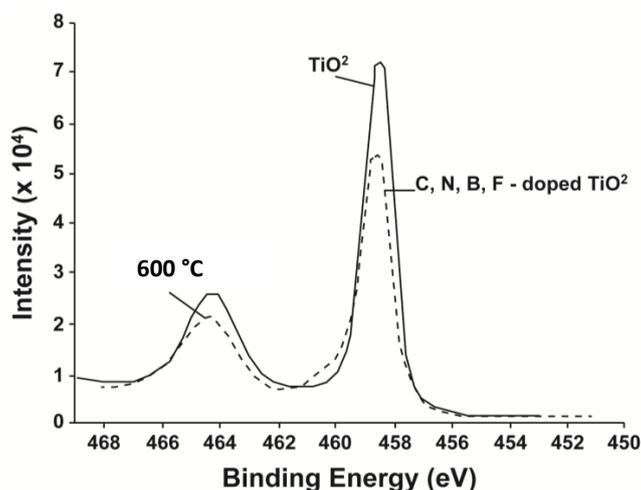


Figure 2. Ti 2p XPS spectra of doped and non-treated TiO₂ samples.

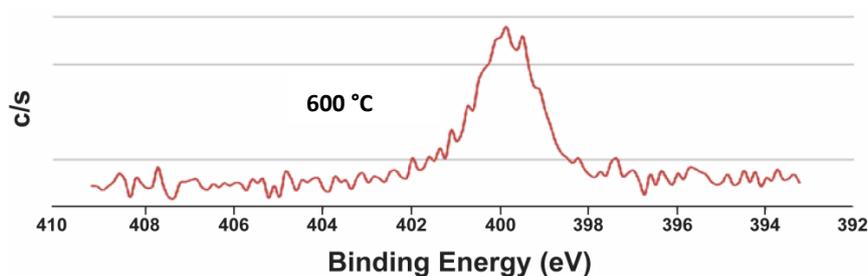


Figure 3. XPS spectrum of N 1s within C, F, N, B-codoped-TiO₂ sample.

3.2. Microbe-Free Surfaces

In the present study, zein and paint were used as binders to form thin films containing TiO₂ or doped-TiO₂ powders for surface coating. First, we examined the morphology of obtained films; then, we investigated their antimicrobial activities.

Figure 4 shows the surface morphology of TiO₂ powders, C, N, B, F-doped-TiO₂ powders, neat zein films, single-layer zein films with of TiO₂ powders 2% and 10%, double-layer zein coating with 8 mL neat zein in the bottom, and 2 mL 10% TiO₂ particles on the top. The thermal treatment process of TiO₂ with (CH₃CH₂)₄NBF₄ (Figure 4A,a) at 600 °C slightly changed the appearance of TiO₂ particles (Figure 4B,b), but the particle size remained at the same level. The surfaces of neat zein film appeared as a glossy and uniform material without pores (Figure 4C,c). The incorporation of TiO₂ particles resulted in a rough morphology (Figure 4D–F,d–f); TiO₂ particles were randomly distributed within the matrix phase. The number of particles in a defined area increased with the increase in TiO₂ content. Both samples of E and F had similar morphologies, showing similar TiO₂ content on their surfaces.

The antibacterial activity of zein/TiO₂ films was first evaluated by measuring the populations of *Escherichia coli* 0157:H7 on the films with different TiO₂ contents that pre-coated on petri dishes. The experiments were carried out under fluorescent light irradiation. As shown in Figure 5, no significant changes in *E. coli* 0157:H7 populations were observed in the first three hours of irradiation, regardless of whether TiO₂ powders were added or how they were added. A significant decrease in

E. coli populations was observed after 18 h on TiO₂/zein films. The cell numbers of *E. coli* inoculated on zein films (Sample #0) were less than those on non-coated petri dishes (controls), indicating *E. coli* suffered from growth on zein films as compared with the controls. It has been reported that zein films are resistant to bacterial attack [30]. The incorporation of TiO₂ powders further suppressed bacterial growth. As the ratio of TiO₂/zein increased from 0% (sample #0) to 1% (sample #1) and 2% (sample #2), the number of *E. coli* decreased 1 log successively. For the zein films with 10% TiO₂ content (sample #3), no viable *E. coli* cells were detected (<1 log CFU/mL), indicating a bacteria-free surface created at high dose TiO₂.

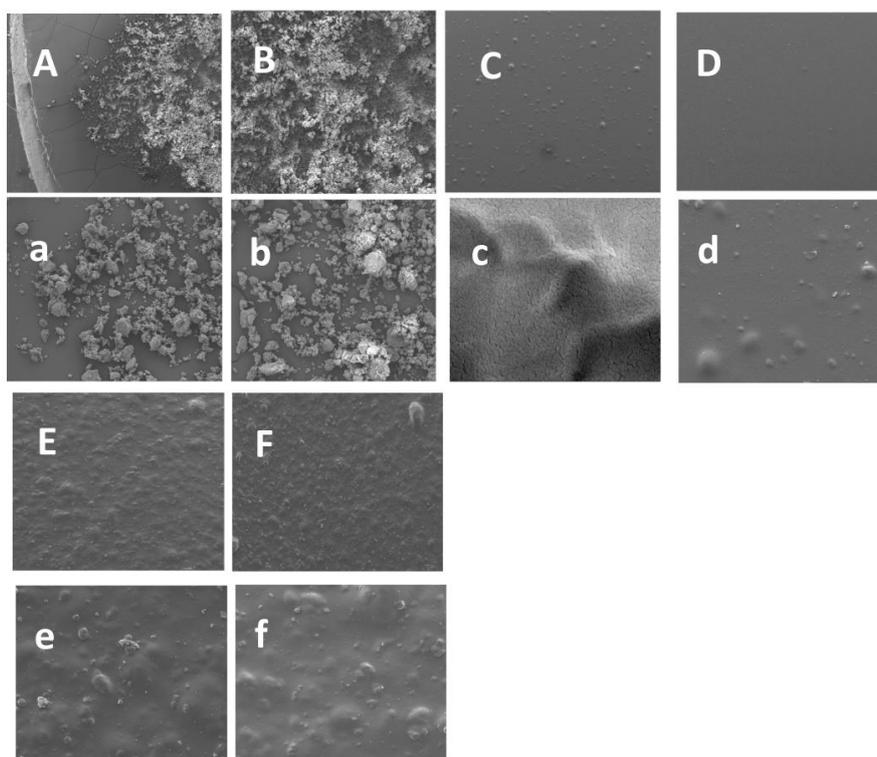


Figure 4. SEM topography of TiO₂ (A,a), doped-TiO₂ powders (B,b), neat zein films (C,c), single-layered zein films incorporated with doped-TiO₂ powders at 2% (D,d) and 10% (E,e), and double-layered film containing doped-TiO₂ powders of 2%. Field width: 5.5 mm (A–F) and 275 μm (a–f).

The antimicrobial activity of double-layer coating was then compared with the single-layer one. As shown in Figure 5, sample #2⁺ was a double-layer film with a neat zein layer in the bottom and a TiO₂/zein layer on the top. The double-layer sample #2⁺ contained the same amount of TiO₂ powders (2%) as sample #2 has, while the TiO₂ content in the top layer was equal to that of entire sample #3 (10%). These results indicate the antibacterial activity of TiO₂/zein films is determined by the amount of TiO₂ embedded on the surfaces. For the double-layer films, the coating materials consist of a neat binder layer in bottom and another layer containing titanium dioxide powders on the top (Figure 1). While the more concentrated TiO₂ powders on the top layer function more efficiently in deactivating microbes, the bottom layer, supposedly, should be able to facilitate the coating to anchor to the solid surfaces.

Florescent lamps are commonly used in warehouses and retail stores; they emit a narrow range of UV light (300–380 nm) in combination with a wide window of visible light (400–800 nm). Containers and shelves are usually exposed under visible light irradiation or UV irradiation for irregular time periods. To evaluate the effect of light irradiation on the antimicrobial activities of TiO₂ coatings, Petri dishes coated with single-layer of 2% TiO₂/zein were irradiated under two different conditions prior to antimicrobial test: one group was exposed to light for 15 h followed by placing in dark for 3 h;

another group was exposed to light for 3 h followed by placing in dark for 15 h. The Petri dishes coated with doped-TiO₂/zein formulation were included in the second group and tested in comparison with TiO₂/zein coatings. As shown in Figure 6, sample #2 (group 1) had significantly lower *E. coli* populations than sample #2⁻ (group 2 coated with TiO₂/zein) after 18 h, indicating that longer time exposure to light enhanced the antimicrobial activity of the coatings. That could be attributed to the narrow UV window provided by the FL. However, sample #2⁺⁺ (coated with codoped-TiO₂/zein formulation) reduced much more *E. coli* cells than both sample #2 and sample #2⁻, and even sample #3 when compared under the same experimental conditions (Figure 5, black column). This could be attributed to the visible light irradiation that provides minimal energy to create electrons and promotes the electrons in the codoped-TiO₂ to travel from valence band to the conduction band. The energy generated by visible light irradiation is not strong enough to activate pure TiO₂ samples [31–33].

Data shown in Figures 5 and 6 indicated that the *E. coli* survival number was not significantly reduced after three hours of FL irradiation. Similar results were reported previously [34]. Probably, at the beginning of irradiation, the active species were generated, and UV deleterious effect began to attack the membrane but did not sufficiently overcome the self-defense and auto repair mechanisms of the species or cause serious damage to the bacteria to cause cell death; in this case, the injured bacterial cells can be recovered if enough nutrition supply is guaranteed. Under the longer irradiation, the anti-stress enzymes were no longer able to protect the bacterial membrane against oxidation; hence, the *E. coli* survival number decreased significantly.

Interestingly, codoped TiO₂ coating (2⁺⁺) showed more bacterial reduction than non-codoped TiO₂ samples after 15 h in the dark (Figure 6). Wu et al. investigated the efficiency of photocatalytic inactivation *E. coli* under visible light irradiation using titanium dioxide nanoparticles codoped with N and Ag [35]. They also observed that the inactivation of *E. coli* in the dark after irradiation. It could be due to the biocidal properties of doped elements. The mechanism needs to be further investigated.

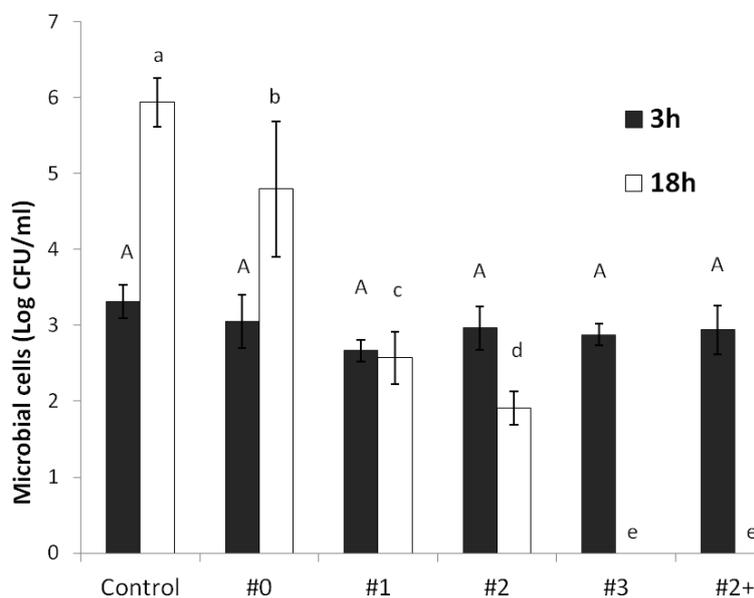


Figure 5. Effect of TiO₂ on growth of *E. coli* O157:H7. The bacterial populations on the surfaces of the films were determined after 3 (black column) and 18 h (white column) under FL irradiation. Control: no coating. Each tested Petri dish coated with 10 mL TiO₂/zein formulation. The ratio of TiO₂/zein in sample #0: 0 (neat zein); sample #1: 1%; sample #2: 2%; sample #3: 10%; sample #2⁺: double-layer film, 8 mL neat zein in bottom and 2 mL of sample #2 on top. Error bars represent the standard deviation of the mean. Data having a same uppercase (3 h) or lowercase (18 h) letter are not significantly different ($p > 0.05$). No viable *E. coli* cells were detected (<1 log CFU/mL) for samples #3 and #2⁺ at 18 h.

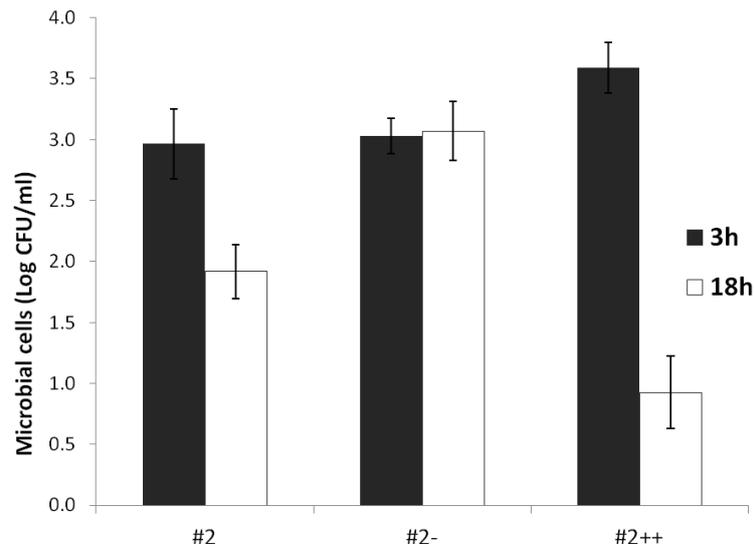


Figure 6. Effect of light irradiation on growth of *E. coli* O157:H7 on zein/TiO₂ coatings (2%). Sample #2: under light for 15 h and then in dark for 3 h; sample #2⁻: under light for 3 h and then in dark for 15 h; sample #2⁺⁺: replacing TiO₂ with codoped-TiO₂, under light for 3 h and then at dark for 15 h. After 3 h (black column) and 18 h (white column) of irradiation, fractions were taken, processed for bacterial survival. Error bars represent the standard deviation of the mean.

Figure 7 shows the residual *E. coli* cells on the films after washing. Sample #2⁺ had no colonies, Sample #2 had 11 colonies, Sample #2⁻ had 140 colonies, and Sample #0 had over 300 colonies. It clearly demonstrated that the films coated with TiO₂/zein did not only inhibit or reduce *E. coli* on their surfaces as shown in Figures 5 and 6 but also increased the resistance to bacterial attachment and prevented biofilm forming on their surfaces.

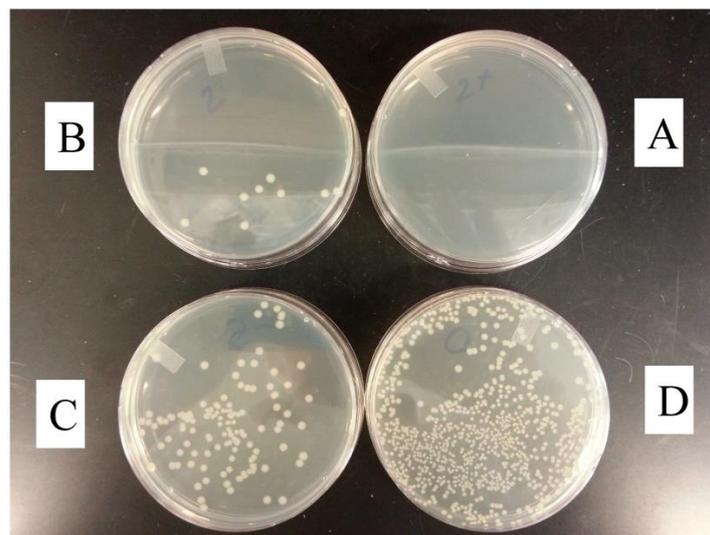


Figure 7. *E. coli* O157:H7 colonies on the surface of coated petri dishes after washing. Each petri dish coated with 10 mL zein formulations. (A) Sample #2⁺: bilayered film, 8 mL neat zein in bottom and 2 mL TiO₂/zein (2%) on top (under light for 15 h and then in dark for 3 h); (B) Sample #2: TiO₂/zein (2%, under light for 15 h and then in dark for 3 h); (C) Sample #2⁻: TiO₂/zein (2%, under light for 3 h and then in dark for 15 h); (D) Sample #0: coating with neat zein (under light for 15 h and then in dark for 3 h).

Durability is another important consideration in coating material applications. We examined the durability of TiO₂ coating using metal plate coated with TiO₂/paint. Figure 8 shows the residual *E. coli* cells on the metal disks coated with a single layer TiO₂/paint and washed 0 to 5 times. For samples exposed to light for 3 h (Figure 8a), there were no significant differences between controls (no coating) and coated samples without TiO₂; both samples had 3–3.5 log CFU/mL *E. coli* cells on the surfaces. Samples coated with TiO₂ had approximated 2 log CFU/mL, the coating reduced over 90% of *E. coli* on the disk surfaces regardless of the time length of water washing; for samples exposed to light for 18 h (Figure 8b), the residual *E. coli* cells were not detectable by the current method (<1.0 log CFU/mL) and achieved more than 99.7% reduction. Again, there were no significant differences observed for the antimicrobial activity among samples with different water-washing times.

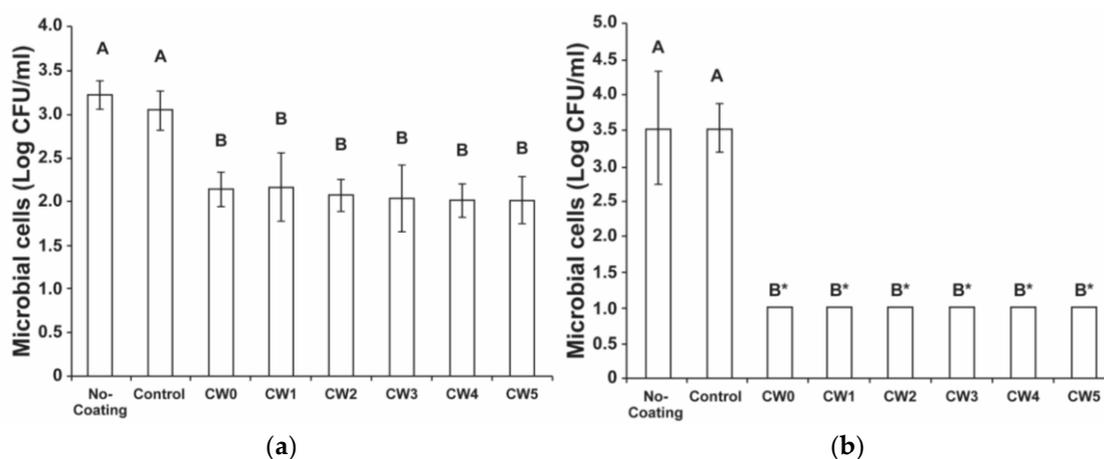


Figure 8. Survival of *E. coli* O157:H7 on the surface of metal disks. Sample disks under light for 3 h (a) and 18 h (b). No-coating: a disk without any coating; Control: coated disk without TiO₂; CW0-5: Disk coated with codoped-TiO₂ being washed in water for 0 to 5 times; *: under detectable level (<1.0 log/CFU); Error bars represent the standard deviation of the mean; Data having a common letter are not significantly different ($p > 0.05$).

4. Conclusions

We have developed a formulation consisting of zein or normal paint as binders embedded with titanium dioxide powders as active component. The two-step coating procedure allows more active titanium dioxide powders to concentrate on coating surfaces; thus, higher anti-microbial activity can be achieved. Containers and frameworks, either made from paper, or plastic, or metal coated with TiO₂/zein (or TiO₂/paint), especially with codoped-TiO₂, are expected to be able to reduce the risk of the microbiological contamination of fruits and vegetables during harvest and postharvest; hence, they could be used in farms, food producers, warehouses, and retail-house.

Acknowledgments: The authors gratefully acknowledge the technical support of Joseph Uknalis.

Author Contributions: L.S.L. conceived and designed the experiments; R.L., T.Z.J., and Z.L. performed the experiments; R.L., T.Z.J., Z.L., and L.S.L. analyzed the data; R.L., T.Z.J., and L.S.L. wrote the paper.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. USDA Document. Available online: <https://www.ers.usda.gov/data-products> (accessed on 18 January 2018).
2. Wells, H.F.; Buzby, J.C. *Dietary Assessment of Major Trends in U.S. Food Consumption, 1970–2005*; Economic Information Bulletin No. (EIB-33); U.S. Department of Agriculture Economic Research Service: Washington, DC, USA, 2008.
3. USDA Document. Available online: <https://www.choosemyplate.gov> (accessed on 18 January 2018).

4. Beuchat, L.R.; Nail, B.V.; Adler, B.B.; Clavero, M.R.S. Efficacy of spray application of chlorinated water in killing pathogenic bacteria on raw apples, tomatoes, and lettuce. *J. Food Prot.* **1998**, *61*, 1305–1311. [[CrossRef](#)]
5. Outbreaks of *Escherichia coli* O157:H7 infection associated with drinking unpasteurized apple cider—October 1996. Update on emerging infections from the Center for Disease Control and Prevention. *Ann. Emerg. Med.* **1997**, *29*, 645–646.
6. Lynch, M.F.R.; Tauxe, V.C.; Hedberg, W. The growing burden of foodborne outbreaks due to contaminated fresh produce: Risks and opportunities. *Epidemiol. Infect.* **2009**, *137*, 307–315. [[CrossRef](#)]
7. Sapers, G.; Solomon, E.; Matthews, K.R. *The Produce Contamination Problem: Causes and Solutions*; Academy Press: Burlington, MA, USA, 2009.
8. Langholz, J.A.; Jay-Russell, M.T. Potential role of wildlife in pathogenic contamination of fresh produce. *Hum. Wildl. Interact.* **2013**, *7*, 140–157.
9. Augugliaro, V.; Bellardita, M.; Loddo, V.; Palmisano, G.; Palmisano, L.; Yurdakal, S. Overview on oxidation mechanism of organic compounds by TiO₂ in heterogeneous photocatalysis. *J. Photochem. Photobiol. C Photochem. Rev.* **2012**, *13*, 224–245. [[CrossRef](#)]
10. Pelaez, M.; Nolan, N.; Pillai, S.C.; Seery, M.K.; Falaras, P.; Kontos, A.G.; Dunlop, P.S.M.; Hamilton, J.W.J.; Byrne, J.A.; O'shea, K.; et al. A review on the visible light active titanium dioxide photocatalysts for environmental applications. *Appl. Catal. B Environ.* **2012**, *15*, 331–349. [[CrossRef](#)]
11. Kedziora, A.; Trek, W.S.; Kepinski, L.; Bulga-Ploskonsa, G.; Doroszkiewicz, W. Synthesis and antibacterial activity of novel titanium dioxide doped with silver. *J. Sol-Gel Sci. Technol.* **2012**, *62*, 79–86. [[CrossRef](#)]
12. Roy, A.; Parveen, A.; Koppalkar, A.R.; Prasad, A. Effect of nano-titanium dioxide with different antibiotics against methicillin-resistant *Staphylococcus Aureus*. *J. Biomater. Nanotechnol.* **2010**, *1*, 37–41. [[CrossRef](#)]
13. Balek, V.; Li, D.; Šubrt, J.; Večerníková, E.; Hishita, S.; Mitsuhashi, T.; Haneda, H. Characterization of nitrogen and fluorine co-doped titania photocatalyst: Effect of temperature on microstructure and surface activity properties. *J. Phys. Chem. Solids* **2007**, *68*, 770–784. [[CrossRef](#)]
14. Li, D.; Haneda, H.; Hishita, S.; Ohashi, N. Visible-light-driven N-F-Codoped TiO₂ photocatalysts. I. synthesis by spray pyrolysis and surface characterization. *Chem. Mater.* **2005**, *17*, 2588–2595. [[CrossRef](#)]
15. Im, J.S.; Yun, S.-M.; Lee, Y.-S. Investigation of multielemental catalysts based on decreasing the band gap of titania for enhanced visible light photocatalysis. *J. Colloid Interface Sci.* **2009**, *336*, 183–188. [[CrossRef](#)] [[PubMed](#)]
16. Corradini, E.A.; de Medeiros, S.; Carvalho, E.; Curvelo, A.J.F.; Mattoso, H.L.C. Mechanical and morphological characterization of starch/zein blends plasticized with glycerol. *J. Appl. Polym. Sci.* **2006**, *101*, 4133–4139. [[CrossRef](#)]
17. Parris, N.; Coffin, D.R. Composition factors affecting the water vapor permeability and tensile properties of hydrophilic zein films. *J. Agric. Food Chem.* **1997**, *45*, 1596–1599. [[CrossRef](#)]
18. Lawton, J.W. Zein: A history of processing and use. *Cereal Chem.* **2002**, *79*, 1–18. [[CrossRef](#)]
19. Liu, L.S.; Fishman, M.L.; Hicks, K.B.; Kende, M.; Ruthel, G. Pectin/Zein Beads for Potential Colon-Specific Drug Delivery: Synthesis and in vitro Evaluation. *Drug Deliv.* **2006**, *13*, 417–423. [[CrossRef](#)] [[PubMed](#)]
20. Torres-Giner, S.; Gimenez, E.; Lagaron, J.M. Characterization of the morphology and thermal properties of zein prolamine nanostructures obtained by electrospinning. *Food Hydrocoll.* **2008**, *22*, 601–614. [[CrossRef](#)]
21. Oh, Y.K.; Flanagan, D. Diffusional properties of zein membranes and matrices. *Drug Dev. Ind. Pharm.* **2010**, *36*, 497–507. [[CrossRef](#)] [[PubMed](#)]
22. Zhang, B.; Luo, Y.; Wang, Q.A. Development of silver-zein composites as a promising antimicrobial agent. *Biomacromolecules* **2010**, *11*, 2366–2375. [[CrossRef](#)] [[PubMed](#)]
23. Li, W.; Liu, L.S.; Jin, T.Z. Antimicrobial activity of allyl Isothiocyanate used to coat biodegradable composite films as affected by storage and handling conditions. *J. Food Prot.* **2012**, *75*, 2234–2237. [[CrossRef](#)] [[PubMed](#)]
24. Chen, C.; Bai, H.; Chang, S.; Chang, C.; Den, W. Preparation of N-doped TiO₂ photocatalyst by atmospheric pressure plasma process for VOCs decomposition under UV and visible light sources. *J. Nanopart. Res.* **2006**, *9*, 365–375. [[CrossRef](#)]
25. Grey, I.E.; Li, C.; MacRae, C.M.; Bursill, L.A. Boron Incorporation into Rutile. Phase Equilibria and Structure Considerations. *J. Solid State Chem.* **1996**, *127*, 240–247. [[CrossRef](#)]
26. Park, H.; Choi, W. Effects of TiO₂ Surface Fluorination on Photocatalytic Reactions and Photoelectrochemical Behaviors. *J. Phys. Chem. B* **2004**, *108*, 4086–4093. [[CrossRef](#)]

27. Giannakopoulou, T.; Todorova, N.; Trapalis, C.; Vaimakis, T. Effect of fluorine doping and SiO₂ under-layer on the optical properties of TiO₂ thin films. *Mater. Lett.* **2007**, *61*, 4474–4477. [[CrossRef](#)]
28. Yu, J.; Zhao, X.; Zhao, Q.; Wang, G. Preparation and characterization of super-hydrophilic porous TiO₂ coating films. *Mater. Chem. Phys.* **2001**, *68*, 253–259. [[CrossRef](#)]
29. Mi, Y.Y.; Wang, S.J.; Chai, J.W.; Pan, J.S.; Huan, C.H.A.; Feng, Y.P. Effect of nitrogen doping on optical properties and electronic structures of SrTiO₃ film. *Appl. Phys. Lett.* **2006**, *89*, 231922. [[CrossRef](#)]
30. Serpone, N. Is the band gap of pristine TiO₂ narrowed by anion- and cation-doping of titanium dioxide in second-generation photocatalysts? *J. Phys. Chem. B* **2006**, *110*, 24287–24293. [[CrossRef](#)] [[PubMed](#)]
31. Zaleska, A. Doped-TiO₂: A Review. *Recent Pat. Eng.* **2008**, *2*, 157–164. [[CrossRef](#)]
32. Grabowska, E.; Reszcyńska, J.; Zaleska, A. Mechanism of phenol photodegradation in the presence of pure and modified-TiO₂: A review. *Water Res.* **2012**, *46*, 5453–5471. [[CrossRef](#)] [[PubMed](#)]
33. Liou, J.W.; Chang, H.H. Bactericidal effects and mechanisms of visible light-responsive titanium dioxide photocatalysts on pathogenic bacteria. *Arch. Immunol. Ther. Exp.* **2012**, *60*, 267–275. [[CrossRef](#)] [[PubMed](#)]
34. Chen, F.; Yang, X.; Wu, Q. Photocatalytic Oxidation of *Escherichia coli*, *Aspergillus niger*, and Formaldehyde under Different Ultraviolet Irradiation Conditions. *Environ. Sci. Technol.* **2009**, *43*, 4606–4611. [[CrossRef](#)] [[PubMed](#)]
35. Wu, P.; Xie, R.; Imlay, K.; Shang, J.K. Visible-Light-Induced Bactericidal Activity of Titanium Dioxide Codoped with Nitrogen and Silver. *Environ. Sci. Technol.* **2010**, *44*, 6992–6997. [[CrossRef](#)] [[PubMed](#)]



© 2018 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<http://creativecommons.org/licenses/by/4.0/>).