Furman University Furman University Scholar Exchange

Chemistry Publications

Chemistry

2020

A proposed mechanism to induce multi-layer polydiacetylenecoated filter color response to bacteria

Timothy Hanks Furman University

Follow this and additional works at: https://scholarexchange.furman.edu/chm-publications

Part of the Biochemistry Commons, Materials Chemistry Commons, and the Polymer Chemistry Commons

Recommended Citation

Hanks, Timothy, " A proposed mechanism to induce multi-layer polydiacetylene-coated filter color response to bacteria " (2020). *Chemistry Publications*. 7. https://scholarexchange.furman.edu/chm-publications/7

This Article (Journal or Newsletter) is made available online by Chemistry, part of the Furman University Scholar Exchange (FUSE). It has been accepted for inclusion in Chemistry Publications by an authorized FUSE administrator. For terms of use, please refer to the FUSE Institutional Repository Guidelines. For more information, please contact scholarexchange@furman.edu.

Contents lists available at ScienceDirect

Results in Chemistry

journal homepage: www.elsevier.com/locate/rechem

A proposed mechanism to induce multi-layer polydiacetylene-coated filter color response to bacteria

Yueyuan Zhang ^a, Paul L. Dawson ^{a,*}, Tzuen-Rong Tzeng ^b, Timothy W. Hanks ^c, Julie K. Northcutt ^a, William T. Pennington ^d

^a Food, Nutrition, and Packaging Sciences Department, Clemson University, Clemson, SC, USA

^b Department of Biological Sciences, Clemson University, Clemson, SC, USA

^c Department of Chemistry, Furman University, Greenville, SC, USA

^d Department of Chemistry, Clemson University, Clemson, SC, USA

ARTICLE INFO

Article history: Received 5 June 2020 Accepted 27 July 2020 Available online xxxx

Keywords: Polydiacetylene Filter PDA PCDA pH Bacteria Salmonella Typhimurium E. coli

ABSTRACT

The purpose of this study was to develop a multi-layer diacetylene (PDA)-coated filter and to verify the proposed color response mechanism of PDA-coated filters to bacteria. Unlike other reports which immobilize PDA liposomes on solid support or create monolayer Langmuir films, multi-layer PDA-coated filters were generated by directly evaporating organic solvents. Different from incorporating functional headgroups or inserting phospholipids into PDA, we reported that bacterial growth can trigger the color change of PDA sensors without any modification or phospholipid insertion. The mechanism that pH change from bacteria metabolites lead to color change of PDA filter was proposed and verified by carefully designed dextrose-free medium with phenol red. Further, culturing *Salmonella Typhimurium, E. coli, L. innocua* and *M. luteus* with PDA-coated filters on phenol red agar in absence of dextrose verified the potential of applying PDA-coated filter for bacterial detection, specifically amine-producing bacteria. Thus, PDA-coated filters may be a useful tool for food safety and shelf life applications. © 2020 The Authors. Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license

(http://creativecommons.org/licenses/by-nc-nd/4.0/).

and ribbons [11,12] have also been observed when supplemented with phospholipids or modified with headgroups. Examples of

substrate-based forms are self-assembled monolayers [13], multi-

layer coatings on surfaces [14], fibers [15], nanotubes [16], and

liposome-immobilized on a solid support [17]. Currently, most re-

ported PDA-based sensors for biological application were in liquid-

based form especially liposomes [18,19] due to the relative ease of

PDA modification. Reported substrate-based PDA sensors for biolog-

ical application have focused on immobilizing liposome on glass [20]

or embedded in semi-solid substrates such as agar [20-22] or sodium

alginate [23]. Substrate-based sensors such as paper-based PDA sen-

sors are mostly reported for chemical detection [14,24] but seldom

reported for biological applications. In this study, we generated a

substrate-based PDA sensor, multi-layer PDA on filter by directly

evaporating an organic solvent containing PDA followed by

tional headgroups to enhance sensor specificity and sensitivity. Target

biomolecules or chemical species interact with the incorporated func-

tional headgroups that change the conformation of the PDA backbone and therefore trigger color change of the biosensor. Nagy et al. [25] reported incorporating Gal- α 1,4-Gal functional headgroups into PDA

nanoparticles for detection of Shiga-like toxin-producing E. coli O157:

Development of PDA-based biosensors focus on incorporating func-

1. Introduction

Polydiacetylenes (PDAs) are highly conjugated polymers assembled from monomers containing two conjugated diacetylenes (DAs). DAs undergo photopolymerization via a 1,4-addition to form a repeating double-single-triple bond pattern along the carbon backbone. The highly-conjugated material takes on an intense blue appearance and has rather strict geometrical arrangement [1]. Stimuli such as heat [2], pH change [3], mechanical stress [4] and chemical solvents [5] have been reported to lead to an obvious color change of PDAs from blue to a red/pink color. This color change can be observed easily by the naked eye. It has been widely accepted that the color change of PDA is highly related to PDA geometry and comes from the conformational change of the PDA backbone [6]. Considering the chromatic properties of PDAs, it serves well in applications for visual detection of biological or chemical substances.

Liquid-based and substrate-based forms are typical forms of PDA-based sensors. A well-known example of liquid-based PDA is self-assembled liposomes or so-called vesicles. Self-assembled, non-spherical structures such as flat sheets [7], tubules [8,9], helices [10],

* Corresponding author. E-mail address: pdawson@clemson.edu (P.L. Dawson).

https://doi.org/10.1016/j.rechem.2020.100065

2211-7156/© 2020 The Authors. Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

photopolmerization.







H7. Similarly, Wu et al. [18] reported peptide-functionalized PDA liposomes for bacterial lipopolysaccharide detection. Antibodies [26] and aptamers [27] also have been reportedly conjugated to PDA for E. coli detection. Besides headgroup modification, other proposed strategy to apply PDA-based biosensors is inserting phospholipid molecules into PDA structure to mimic the cell membrane. Inserted phospholipid molecules serve as a recognition element for biological material detection. This recognition will change the conformation of PDA backbone and therefore trigger color change of PDA-based biosensors. Silbert et al. [16] reported bacteria-secreted membrane-active compounds triggered color changes in phospholipid containing PDA nanoparticles. Instead of incorporating functional headgroups or inserting phospholipid into PDA, we report that culturing bacteria can trigger the color change of PDA sensors without any modification or phospholipid insertion. Mechanism of PDA color response to culturing bacteria was proposed and verified by varying pH of bacteria metabolite-supernatant and incubated PDA-coated filters in pH adjusted supernatant with analysis and discussion. The potential of utilizing this PDA-coated filter for bacteria detection was further explored.

2. Material and methods

2.1. Materials

Mixed Whatman[™] cellulose ester membrane filters (GE Healthcare Life Science, Buckinghamshire, UK) (white with grid) with pore size of 0.45um and a diameter of 47 mm were coated with 10,12-pentacosadiynoic (PCDA) (GFS Chemicals, Columbus, OH). Isopropyl alcohol (BDH, Radnor, PA) was used to dissolve PCDA and coat the filters. Mineralight® lamp Model UVGL-58 (Upland, CA) was used as UV light to polymerize PCDA. *Salmonella Typhimurium 14028, Escherichia coli HB101, Listeria innocua and Micrococcus luteus* were the test bacteria in this study. Tryptic soy broth (TSB), BBL phenol red broth (composition: pancreatic digest of casein, sodium chloride and phenol red), dextrose, and agar were purchased from Becton Dickinson (Sparks, MD). A pH meter (Orion[™] model 420A, Thermo Scientific, Beverly, MA) was used to measure supernatant pH and a Samtian 43. × 43.4 × 43.4 cm photo box with LED light source was used for photography and a 6.3 cm wide rubber paint roller was used to coat the filters with PDCA.

2.2. Coating PCDA on filters

PCDA was dissolved in isopropyl alcohol at a concentration of 3 mg/ ml, by stirring for 1 h followed by filtering through a 0.45 um filter to eliminate aggregates then stored at 4 °C until use. The PCDA stock solution was further diluted to 1.5 mg/ml with isopropyl alcohol and was "painted" on the surface of the mixed cellulose ester membrane filter with a small rubber paint roller. The membrane filter was air-dried for 2 min followed by exposure to UV at 254 nm for 1 min to obtain the multi-layer blue color poly-PCDA coated membrane filter (PDA-coated filter). PDA-coated filter discs for use on agar were generated using a hole punch. Scheme of coating PCDA on filters was shown in Fig. 1. A HunterLab UltraScan® Pro dual-beam spectrophotometer was used to characterize the PDA-coated filters. Five spots (area view diameter 4.826 mm) on each PDA-coated filter were randomly selected and scanned using a D65 light source to obtain reflectance to transmittance (%) data for PDA-coated filters.

2.3. Effect of dextrose supplement on bacteria metabolite and color response of PDA-coated filters

Phenol red broth with and without dextrose was used as a direct indicator to monitor pH change due to bacterial growth. Dextrose at a 5% concentration was used to supply phenol red broth with dextrose. *S*. Typhimurium, *E. coli, L. innocua* and *M. luteus* were cultured in phenol red broth without and with dextrose at 37 °C with mixing at 60 rpm for 16 h then centrifuged at 3000g for 5 min to harvest the suspension. The pH of suspension was measured and recorded. PDA-coated filter discs were added to suspensions and kept at 37 °C with mixing at 60 rpm for 2 h. PDA-coated discs were then taken from metabolitesupernatant and photos were taken using iPhone 6 s in Samtian photo box (LED light source at maximum) to record the color change of PDA-coated filter discs.

2.4. Effect of metabolites on pH and color response of PDA-coated filters

Furthermore, the pH of another set metabolite-supernatant was adjusted to 7 with HCl (0.1 N) for phenol red broth without dextrose and adjusted to pH = 8 with NaOH (0.1 N) for phenol red broth with dextrose to investigate pH effect on PDA-coated filter discs. PDA-coated filter discs were then added to metabolite-supernatants and kept at 37 °C with mixing at 60 rpm for 2 h. PDA-coated discs were then taken from metabolite-supernatant and photos were taken using iPhone 6 s in Samtian photo box to record the color changes of PDA-coated filter discs.

2.5. Application of PDA-coated filters for bacteria detection

Phenol red in the absence of dextrose was used to culture *S*. Typhimurium, *E. coli*, *L. innocua* and *M. luteus*. The pH of phenol red agar without dextrose was adjusted to pH 7.2 before autoclaving. *Salmonella* Typhimurium and *E. coli* were cultured in TSB overnight for 16 h then centrifuged at 3000 g for 5 min and washed twice with phosphate buffered saline (PBS) and then adjusted to $\sim 10^8$ cells/ml by diluting with phosphate buffer. Cell concentration was estimated by light transmission at 600 nm adjusting O.D. = 0.5 and verified by the plate count method. One ml of 10^8 cells/ml was added to 99 ml of PBS followed by filtering through PDA-coated filters. After filtration, PDA-coated filters were transferred to phenol red agar and incubated at 37 °C for 22 h after which plates were photographed.

3. Results and discussion

3.1. Characterization of PDA-coated filters

PDA-coated filters displayed blue color with a relatively uniform coating based on visual inspection. Reflectance to transmittance spectrum of PDA-coated filters quantified color of PDA-coated filters as shown in Fig. 2 and can be used as a measurement of reproducibility of PDA-coated filters. Average CIE L*a*b* values for all filters used in the experiment were L* = 51.53 ± 3.95 , a* = 1.36 ± 1.48 , b* =



Fig. 1. Scheme of coating PCDA on filters and small discs cut from larger filter.



Fig. 2. a. Reflectance to transmittance spectrum of PDA-coated filters. b. PDA-coated filter. c. PDA-coated filter discs.

 30.83 ± 1.34 with Chroma calculated as 30.8 ± 2.0 . The amount of PDA coated on the filters was difficult to quantify because of the coating method; however, the reflectance to transmittance spectrum and average CIEL*a*b* values of PDA-coated filters provided a measurement for estimating the consistency of PDA-coatings.

3.2. Effect of dextrose supplement on bacteria metabolites and color response of PDA-coated filters

Phenol red indicates the pH change of the growth medium due to bacteria metabolites, changing color from orange to magenta (alkaline) or yellow (acid) as shown in Fig. 3 Metabolite-supernatant. Culturing bacteria in broth overnight and then utilizing metabolite-supernatant without cells for testing ensured harvesting all metabolites from bacteria growth as well as eliminated the possibility of PDA-coated filter interacted directly with bacterial cells. Control group of noninoculated phenol red broth and phenol red broth with dextrose did not trigger color transition of PDA-coated filter discs (Fig. 3 Control). This result indicated that the medium itself did not trigger the color transition of PDA-coated filter discs. Moreover, the color change of PDA-coated filter discs from blue to pink was observed after being treated with metabolite-supernatant of phenol red without dextrose but not metabolite-supernatant of phenol red with dextrose (Fig. 3 PDA-coated filter discs). *Linnocua* cultured in phenol red broth without dextrose is an exception and can be explained by *Linnocua* cannot grow without dextrose supplement in phenol red broth and therefore no



Fig. 3. Effect of dextrose on bacteria metabolite and color response of PDA-coated filter discs. Columns with the same label originated from the same sample. (P = Phenol red broth, PD = Phenol red broth with dextrose; + indicated color change, - indicated no color change.) (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

metabolites were produced in supernatant. These observations revealed that bacteria metabolites from growing on phenol red broth without dextrose triggered the color change of PDA-coated filter discs while metabolites which come from growing on phenol red broth with dextrose supplement cannot trigger color change of PDA-coated filter discs. Comparing PDA-coated filter discs incubated in phenol red broth with and without dextrose supplement, we observed that bacteria produce different metabolites during growth when in supply or in absent of dextrose. Those metabolites vary pH of supernatant and this pH change may play an important part in color transition of PDA-coated filter discs.



Fig. 4. Effect of metabolite-supernatant pH adjustment on color response of PDA-coated filter discs. (n = 2) (**a**) pH adjustment for phenol red supernatant with dextrose from acid to pH = 8 (**b**) pH adjustment for phenol red supernatant without dextrose from alkaline to pH = 7. Rows with the same label originated from the same sample. (PD = Phenol red supernatant with dextrose, PD.A = Phenol red supernatant with dextrose adjusted, P = Phenol red supernatant, P.A = Phenol red supernatant adjusted; + indicated color change, - indicated no color change). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

3.3. Effect of pH on color response of PDA-coated filters

Metabolites due to bacterial growth increased the pH of phenol red broth without dextrose and decreased the pH of phenol red broth with dextrose. To investigate pH effect on color response of PDAcoated filter discs, metabolite-suspension pH was adjusted from acid to alkaline and from alkaline to neutral. Color change of PDA-coated filter disc was observed when adjust the metabolite-solution pH in phenol red broth with dextrose from acidic to pH 8 (Fig. 4, PD.A column). This phenomenon indicated that pH can trigger color change of PDAcoated filter discs. Adjustment of the metabolite-solution pH in phenol red broth from alkaline pH to pH 7 did not trigger a color change in PDA-coated filter discs (Fig. 4, P.A column). This observation eliminated the possibility of other metabolites which cannot change pH of supernatant trigger color response of PDA-coated filters since these metabolites were maintained in supernatant when adjusting pH to neutral but color change of PDA-coated filter was not observed. In summary, these observations supported the theory that bacteria metabolites which can increase the pH of supernatant induce color change of PDA-coated filter and implied that PDA-coated filter may be applied for amine producing bacteria detection.

3.4. Mechanism for color change of PDA-coated filters

Microorganisms frequently change the pH of their habitat by producing acidic or basic metabolic waste products [28]. With dextrose supplementation, microorganisms can catabolize carbohydrates into pyruvate and similar intermediates and then enter the tricarboxylic acid (TCA) cycle pathway to produce energy [28]. Metabolites from carbohydrate are acidic which decrease the microbial habitat pH. In absence of dextrose supplement, microorganisms utilize proteins, peptides or amino acids as an energy source. Decarboxylation of amino acid will produce CO₂ and amine products which increases the microbial habitat pH. It has been reported that Salmonella and Escherichia spp. are capable of amine production [29,30]. This pH change caused by bacteria growth was verified in this experiment by culturing Salmonella Typhimurium, E. coli, Linnocua and M.luteus in phenol red broth with or without dextrose supplement. The mechanism of bacteria metabolites increase environment pH and therefore lead to color change of PDA-coated filter was proposed based on pH adjustment observations and analysis. The proposed mechanism was further supported by the fact that it has been well known that alkaline pH triggers the color change of PDA liposome and PDA monolayers. Possible explanation is that alkaline pH results in the ionization of the carboxyl group of PDA in PDA coated filters and the consequences of ionization are breakdown of hydrogen-bonding network in PDA and electrostatic repulsion between adjacent carboxylate groups [12,31]. Stress created from electrostatic repulsion accumulates in the PDA backbone and eventually twists the PDA backbone. This twist of the PDA backbone causes the color change of multi-layer PDA coated filters from blue to pink.

3.5. Application of PDA-coated filters for bacteria detection

To investigate the possibility of applying PDA-coated filters for bacteria detection, *S.* Typhimurium, *E. coli, L. innocua* and *M. luteus* were filtered through PDA-coated filters and cultured on phenol red agar without dextrose. Fig. 5 shows the top of the plate which is the PDAcoated filter and the bottom of the plate which is the phenol red agar. Consistent with growing bacteria in broth, bacteria changed the color of PDA coated filters to pink, while the phenol red indicator was turned from orange to magenta (Fig. 5 P column) which indicated an increase of medium pH. Time difference for obvious color change was observed for *Salmonella* Typhimurium in 7 h and *E. coli*, *M.luteus* in 10 h. Colonies



Fig. 5. Color response of PDA-coated filters and phenol red indicator to bacteria growth. Columns with the same label originated from the same agar plate (Top = top view of plates showing the PDCA filter. Bottom = bottom view of plates showing the phenol red agar without dextrose. $P \cdot C =$ Control group of phenol red without dextrose, P = Phenol red without dextrose). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

of *M. luteus* formed are yellow and therefore color change of PDA-coated filter for *M.luteus* is not as obvious as *S.* Typhimurium and *E. coli*. Colonies of L. *innocua* was not observed on PDA-coated filter while phenol red did not change color. This observation means that *Linnocua* cannot grow on phenol red without dextrose medium and therefore did not change color of PDA-coated filters. The color of PDA-coated filter and phenol red indicator of control group remained unchanged during 22 h culturing, which eliminated other possible reasons for color change of PDA-coated filters such as temperature, PBS buffer, and environment (Fig. 5 P·C and PD.C column). These observations verified that combining with dextrose absence medium, PDA-coated multilayer filter can be applied for *Salmonella* Typhimurium and *E. coli* detection.

4. Conclusions

In this study, coating of PDA on the surface of membrane filters was achieved by evaporation of organic solvent. Degree of coating was quantified by measuring color of PDA-coated filter using a spectrophotometer which at the same time ensured only PDA-coated filters with good reproducibility utilized in this experiment. Color change of PDA-coated filters from blue to pink was observed concurrent with the alkaline pH induced from bacteria metabolism. Adjustment of supernatant pH verified that alkaline pH from bacteria metabolites leads to color change of PDA-coated filter to cultured bacteria was proposed and the potential of applying PDA-coated filter for bacteria detection was supported by testing on *Salmonella* Typhimurium, *E. coli, L.innocua* and *M.luteus* on phenol red without dextrose agar. Advantages of using this strategy include concentrating bacteria in sampling, visual detection and the potential to quantify bacteria on the filter.

Credit roles

Yueyuan Zhang, Investigation, Methodology, Validation, Visualization, Writing - original draft; Writing - review & editing. Paul L. Dawson, Investigation, Methodology, Validation, Writing - original draft; Writing - review & editing. Project administration; Resources, Supervision Tzuen-Rong Tzeng, Methodology, Writing - review & editing Timothy W. Hanks, Methodology, Validation, Writing - original draft; Writing - review & editing. Julie K. Northcutt, Methodology, Validation, Writing - original draft; Writing - review & editing. William T. Pennington, Methodology, Validation, Writing - original draft; Writing - review & editing.

Funding sources

Technical Contribution No. 6821 of the Clemson University Experiment Station. This material is based upon work supported by NIFA/ USDA, under project number Enhancing Microbial Food Safety by Risk Analysis SC-1700553. Food Safety from the Farm to the Fork: Integrated Strategies and Innovations for South Carolina SC-1700535.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgment

The author would like to thank Ian Cummings for providing *E. coli HB101*.

References

 H. Menzel, S. Horstmann, M. Mowery, M. Cai, C. Evans, Diacetylene polymerization in self-assembled monolayers: influence of the odd/even nature of the methylene spacer, Polymer 41 (2000) 8113–8119.

- [2] J. Huo, Z. Hu, G. He, X. Hong, Z. Yang, S. Luo, X. Ye, Y. Li, Y. Zhang, M. Zhang, H. Chen, High temperature thermochromic polydiacetylenes: design and colorimetric properties, Appl. Surf. Sci. 423 (2017) 951–956.
- [3] S.J. Kew, E.A. Hall, pH response of carboxy-terminated colorimetric polydiacetylene vesicles, Anal. Chem. 78 (7) (2006) 2231–2238.
- [4] R.W. Carpick, D.Y. Sasaki, A.R. Burns, First observation of mechanochromism at the nanometer scale, Langmuir 16 (3) (2000) 1270–1278.
- [5] S. Dolai, S.K. Bhunia, S.S. Beglaryan, S. Kolusheva, L. Zeiri, R. Jelinek, Colorimetric polydiacetylene–aerogel detector for volatile organic compounds (VOCs), ACS Appl. Mater. Interfaces 9 (3) (2017) 2891–2898.
- [6] M. Schott, The colors of polydiacetylenes: a commentary, J. Phys. Chem. B 110 (2006) 15864–15868.
- [7] Y. Su, J. Li, L. Jiang, Chromatic immunoassay based on polydiacetylene vesicles, Colloids Surf. B: Biointerfaces 38 (2004) 29–33.
- [8] J. Schnur, R. Price, P. Schoen, P. Yager, J. Calvert, J. Georger, A. Singh, Lipid-based tubule microstructures, Thin Solid Films 152 (1987) 181–206.
- [9] A. Plant, D. Benson, G. Trusty, Probing the structure of diacetylenic phospholipid tubules with fluorescent lipophiles, Biophys. J. 57 (1990) 925–933.
- [10] G. Wang, R.I. Hollingsworth, Easily accessible uniform wide-diameter helical, cylindrical, and nested diacetylene superstructures that can be metallized and oriented in magnetic fields, Langmuir 15 (1999) 6135–6138.
- [11] G. Wang, R.I. Hollingsworth, Offsetting the tubule-forming tendency of chiral diacetylene-containing lipids: planar strips, ribbons, and liposomes from a diacetylenic lipid analog of a thermophilic bacterium, Adv. Mater. 12 (2000) 871–874.
- [12] Q. Cheng, M. Yamamoto, R.C. Stevens, Amino acid terminated polydiacetylene lipid microstructures: morphology and chromatic transition, Langmuir 16 (2000) 5333–5342.
- [13] T. Kim, R.M. Crooks, M. Tsen, L. Sun, Polymeric self-assembled monolayers. 2. Synthesis and characterization of self-assembled polydiacetylene mono-and multilayers, J. Am. Chem. Soc. 117 (1995) 3963–3967.
- [14] T. Eaidkong, R. Mungkarndee, C. Phollookin, G. Tumcharern, M. Sukwattanasinitt, S. Wacharasindhu, Polydiacetylene paper-based colorimetric sensor array for vapor phase detection and identification of volatile organic compounds, J. Mater. Chem. 22 (2012) 5970–5977.
- [15] N. Moazeni, A.A. Merati, M. Latifi, M. Sadrjahani, S. Rouhani, Fabrication and characterization of polydiacetylene supramolecules in electrospun polyvinylidene fluoride nanofibers with dual colorimetric and piezoelectric responses, Polymer 134 (2018) 211–220.
- [16] Heo, J.M., Son,Y., Han,S., Ro, H.J., Jun, S., Kundapur, U., Noh, J. and Kim, J.M., 2019. Thermochromic polydiacetylene nanotube from amphiphilic macrocyclic diacetylene in aqueous solution. Macromolecules, 52(11), pp.4405–4411.
- [17] F. Jannah, J.H. Kim, J.W. Lee, J.M. Kim, J.M. Kim, H. Lee, Immobilized polydiacetylene lipid vesicles on polydimethylsiloxane micropillars as a surfactin-based label-free bacterial sensor platform, Front. Mater. 5 (2018) 57.
- [18] J. Wu, A. Zawistowski, M. Ehrmann, T. Yi, C. Schmuck, Peptide functionalized polydiacetylene liposomes act as a fluorescent turn-on sensor for bacterial lipopolysaccharide, J. Am. Chem. Soc. 133 (25) (2011) 9720–9723.
- [19] J. Park, S.K. Ku, D. Seo, K. Hur, H. Jeon, D. Shvartsman, H.K. Seok, D.J. Mooney, K. Lee, Label-free bacterial detection using polydiacetylene liposomes, Chem. Commun. 52 (68) (2016) 10346–10349.
- [20] S. Seo, J. Lee, E.J. Choi, E.J. Kim, J.Y. Song, J. Kim, Polydiacetylene liposome microarray toward influenza a virus detection: effect of target size on turn-on signaling, Macromol. Rapid Commun. 34 (9) (2013) 743–748.
- [21] L. Silbert, I.B. Shlush, E. Israel, A. Porgador, S. Kolusheva, R. Jelinek, Rapid chromatic detection of bacteria by use of a new biomimetic polymer sensor, Appl. Environ. Microbiol. 72 (11) (2006) 7339–7344.
- [22] D. Meir, L. Silbert, R. Volinsky, S. Kolusheva, I. Weiser, R. Jelinek, Colorimetric/fluorescent bacterial sensing by agarose-embedded lipid/polydiacetylene films, J. Appl. Microbiol. 104 (3) (2008) 787–795.
- [23] J.S. Kauffman, B.M. Ellerbrock, K.A. Stevens, P.J. Brown, W.T. Pennington, T.W. Hanks, Preparation, characterization, and sensing behavior of polydiacetylene liposomes embedded in alginate fibers, ACS Appl. Mater. Interfaces 1 (6) (2009) 1287–1291.
- [24] F. Bai, Z. Sun, P. Lu, H. Fan, Smart polydiacetylene nanowire paper with tunable colorimetric response, J. Mater. Chem. 22 (30) (2012) 14839–14842.
- [25] J.O. Nagy, Y. Zhang, W. Yi, X. Liu, E. Motari, J.C. Song, J.T. Lejeune, P.G. Wang, Glycopolydiacetylene nanoparticles as a chromatic biosensor to detect Shiga-like toxin producing Escherichia coli 0157: H7, Bioorg. Med. Chem. Lett. 18 (2) (2008) 700–703.
- [26] B.A. Pindzola, A.T. Nguyen, M.A. Reppy, Antibody-functionalized polydiacetylene coatings on nanoporous membranes for microorganism detection, Chem. Commun. 8 (2006) 906–908.
- [27] Y.W. Zhao, H.X. Wang, G.C. Jia, Z. Li, Application of aptamer-based biosensor for rapid detection of pathogenic Escherichia coli, Sensors 18 (8) (2018) 2518.
- [28] L.M. Prescott, J.P. Harley, D.A. Klein, Microbiology, Wm. C. Brown Publishers, 1990 143–160 pp123.
- [29] Y. Özogul, F. Özogul, Biogenic Amines Formation, Toxicity, Regulations in Food, 2019.
- [30] J.P. Viala, S. Meresse, B. Pocachard, A.A. Guilhon, L. Aussel, F. Barras, Sensing and adaptation to low pH mediated by inducible amino acid decarboxylases in Salmonella, PLoS One 6 (7) (2011).
- [31] N. Mino, H. Tamura, K. Ogawa, Photoreactivity of 10, 12-pentacosadiynoic acid monolayers and color transitions of the polymerized monolayers on an aqueous subphase, Langmuir 8 (2) (1992) 594–598.

Paul L. Dawson is a Professor in the Department of Food, Nutrition and Packaging Sciences that studies ways to improve food safety and quality. His research has encompassed many food products from the farm, food production facilities and in the laboratory. He works directly with the industry through the Cooperative Extension Service and funded research projects. Paul grew up in Salisbury, MD completing a B.S. in 1977 at Salisbury University. After two years of post-baccalaureate work. Perdue Inc. employed him for 1.5 years in the Products Research Area. Paul then obtained a MS degree at University of Florida in Poultry Science studying processing effects on poultry meat quality with Dr. Doug Janky. Paul then attended North Carolina State University completing his Food Science doctorate in 1989 studying various treatments on the lipid fractions of mechanically deboned poultrv meat under the guidance of Dr. Brian Sheldon. After a two-year post-doctorate at NC State treatments to reduce microbial growth on carcasses and eggs, Paul came to Clemson University's Food Science Department and was promoted to Full Professor in 2001. Dawson has authored/coauthored 113-refereed scientific publications, 18 technical book chapters, two books and has over 250 popular media interviews in the field of food microbiology, antimicrobial active packaging and food quality.

Julie K. Northcutt is a Professor in the Department of Food, Nutrition and Packaging Sciences at Clemson University, where she has worked since 2008. From 2014 to 2018, Dr. Northcutt served as the Extension Program Team Leader for the Food Safety and Nutrition Program (43 County Agents, 5 Extension Associates and 4 Faculty Specialists). Prior to Joining the faculty at Clemson, she was a Research Food Technologist promoted to Lead Scientist for the United States Department of Agriculture (2001–2008). While working at the USDA, Dr. Northcutt led a team of research scientists working on pathogen reduction strategies during commercial processing of food. From 1995 to 2001, Dr. Northcutt was an Assistant Professor, promoted to Associate Professor at the University of Georgia. Her research interest includes food microbiology (Salmonella, Listeria, generic Escherichia coli and Campylobacter spp.) and food chemistry (quality, shelf-life and product formulation), especially as they relate to commercial food manufacturing. Dr. Northcutt received her Ph.D. in Food Technology with a minor in Biochemistry from North Carolina State University. She received a Master of Science

degree in Food Science and Human Nutrition, and a Bachelor of Science degree in Biochemistry from Clemson University.

Timothy W. Hanks received a B.S. in Chemistry from the South Dakota School of Mines and Technology and a PhD in Organic Chemistry from Montana State University. After working as a Postdoctoral Research Associate at the University of Minnesota, he moved to South Carolina, joining the Chemistry Department at Furman University in 1990. He is currently Professor of Chemistry and Department Chair at Furman and also an Adjunct Professor in the Chemistry Department at Clemson University. He has held positions as a Visiting Professor in the Clemson School of Materials Science, and Visiting Scientist at Départment du Recherche Fondamentale sur la Matiére Condensée, Commissariat á l'Energie Atomique in Grenoble, France. In 2011 he spent six months as a Fulbright Senior Scholar and Visiting Scientist at the Intelligent Polymer Research Institute at the University of Wollongong, Australia.

William T. Pennington is an Alumni Distinguished Professor and Chair of the Department of Chemistry at Clemson University. He received a B.A. degree in chemistry from Hendrix College in 1977, a Ph.D. in inorganic chemistry in 1983 from the University of Arkansas, with A.W. Cordes, and was a postdoctoral research associate with I.C. Paul and D.Y. Curtin at the University of Illinois for two years after graduate school. Before joining the faculty at Clemson, he worked for two years at Molecular Structure Corporation. His two main areas of research involve the study and application of chromic polydiacetylenes chemico- and biosensors, and the investigation of halogen bonding as a reliable solid-state synthon for crystal design applications.

Yueyuan Zhang received her Master of Science in Food Science from Clemson University in 2014. Currently, she is a Ph.D. candidate in Department of Food, Nutrition, and Packaging Sciences under supervision of Dr. Paul Dawson at Clemson University, SC, USA. Her research focuses on developing polydiacetylene-based biosensors for bacteria detection.