Carbohydrate Polymers 172 (2017) 352–364

Contents lists available at ScienceDirect

Carbohydrate Polymers

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

Mining marine shell wastes for polyelectrolyte chitosan anti-biofoulants: Fabrication of high-performance economic and ecofriendly anti-biofouling coatings

Reda F.M. Elshaarawy[a,b,*], Fatma H.A. Mustafa[c], Lasse van Geelen[d], Amaal E.A. Abou-Taleb[e], Hermine R.Z. Tadros[e], Rainer Kalscheuer[d,**], Christoph Janiak[b,*]

[a] Faculty of Science, Suez University, Suez, Egypt
[b] Institut für Anorganische Chemie und Strukturchemie, Heinrich-Heine Universität Düsseldorf, Düsseldorf, Germany
[c] Marine Chemistry Laboratory, Marine Environment Division, National Institute of Oceanography and Fisheries (NIOF), Suez, Egypt
[d] Institute for Pharmaceutical Biology and Biotechnology, Heinrich-Heine-University Düsseldorf, Düsseldorf, Germany
[e] Marine Chemistry Laboratory, Marine Environment division, NIOF, Alexandria, Egypt

ARTICLE INFO

Article history:
Received 31 March 2017
Received in revised form 16 May 2017
Accepted 18 May 2017
Available online 24 May 2017

Keywords:
Marine biofouling
Biofilm
Polyelectrolyte chitosan Schiff base
Anti-fouling coating
Environmental parameters

ABSTRACT

Banning organotins as antifouling biocides in 2003 was the starting point for many researchers to search for novel economic and environmentally-friendly anti-fouling biocides. In our present contribution, we have successfully functionalized a natural biopolymer, chitosan (CS), isolated from marine wastes with polyelectrolyte brushes akin to ionic liquids. These antifouling biopolymers anchoring polyelectrolyte brushes were *in vitro* assessed for their ability to eradicate or inhibit the *Staphylococcal*/*Escherichia* biofilms. Moreover, these anti-fouling candidates were incorporated into the matrix of commercial paint to formulate antifouling coatings which were subjected to a field static immersion test in the Mediterranean Sea in comparison to a standard antifoulant, Diuron®. The obtained results revealed the prevention of biofilms along with a promising anti-fouling performance. So the new polyelectrolyte chitosan architectures may offer promising anti-foulants additives for biofouling coating applications.

1. Introduction

Over few years ago, due to the invasion of the biofouling phenomenon which act as a destructive environmental and economic problem (Eshet et al., 2011; Krishnan, Weinman, & Ober, 2008; Liu, Singh, & Liu, 2013), substantial efforts have been devoted for tailoring surfaces with minimum fouling propensity which represents one of the outstanding challenges in today's surface science (Nurioglu, Esteves, & de With, 2015). Bacterial adhesion to a substrate and growth of biofilm are crucial steps in the fouling protocol, which can ultimately allow colonization by macrofouling organisms (Chambers, Stokes, Walsh, & Wood, 2006). For example, *Staphylococcus aureus* (*S. aureus*) biofilm growth was reported to be the major step in the biofouling of fabric and medical devices (Hou et al., 2007; Schlag, Nerz, Birkenstock, Altenberend, & Götz, 2007; Zhu et al., 2007), furthermore, its pronounced role in marine bio-fouling was reported, as well (Rajalakshmi, Fathima, Rao, & Nair, 2014; Shikuma & Hadfield, 2010). To address surface biofouling while maintaining bulk integrity of different materials, metal-based (such as Cu(I) complexes and organotin) antifouling coatings had been developed (Krishnan et al., 2008) and used for several decades. However, their negative environmental impacts and toxicity toward non-target aquatic organisms prompted the International Maritime Organization (IMO) (Cui, Teo, Leong, & Chai, 2014; Fusetani, 2011) to recently ban their utilization. Thus, it remains one of the most outstanding challenges to develop environmentally-friendly antifoulants with synergistic biofilm eradication efficacy, which could be further incorporated into the matrix of commercial paints to fabricate safe ecofriendly antibiofouling coatings for diverse applications.

To date, the biopolymer chitosan (CS), a polysaccharide composed of 2-amino-2-deoxy-β-D-glucose (β-D-Glc-NH_2) and 2-

* Corresponding authors at: Institut für Anorganische Chemie und Strukturchemie, Heinrich-Heine Universität Düsseldorf, Düsseldorf, Germany.
** Corresponding author.
E-mail addresses: reda_shaarauy@yahoo.com, reda_elshaarawi@science.suez.edu.eg, Reda.El-Shaarawy@uni-duesseldorf.de (R.F.M. Elshaarawy), proffatema@yahoo.com (F.H.A. Mustafa), Rainer.Kalscheuer@hhu.de (R. Kalscheuer), janiak@uni-duesseldorf.de (C. Janiak).

http://dx.doi.org/10.1016/j.carbpol.2017.05.059
0144-8617/

acetamido-2-β-D-glucose (β-D-Glc-NHAc) units, may offer an ideal promising antifouling coating candidate due to its broad-spectrum of pharmacological effects including antibacterial, antifungal and anti-algal efficacy (Choi et al., 2001; No, Park, Lee, & Meyers, 2002; Rhoades & Roller, 2000; Roller & Covill, 1999; Sudarshan, Hoover, & Knorr, 1992) along with excellent anti-fouling (Abiraman, Kavitha, Rengasamy, & Balasubramanian, 2016; Roux, Krieg, Yeates, & Breytenbach, 2005; Xu, Pranantyo, Neoh, Kang, & Fu, 2016) performance. Furthermore, chitosan is part of a green chemistry strategy as it is extracted mainly from marine wastes (e.g. shrimp shell) using a very simple and economic protocol (*i.e* economically attractive) which open wide windows for it in many application such as enantioseparation (Feng et al., 2017; Wang, Xia, Chena, Huangb, & Bai, 2017), drug delivery (Islam & Ferro, 2016) and several biomedical applications (Ahmed & Ikram 2016; Rinaudo, 2006) and membrane technology (Sanjari & Asghari, 2016), as well.

Notably, imidazolium, pyridinium and quaternary ammonium ionic liquids (ILs)-based architectures demonstrated significantly higher antimicrobial efficacies (Carson et al., 2009; Docherty & Kulpa, 2005; Elshaarawy & Janiak, 2014a, 2014b; Elshaarawy, Mostafa, Refaee, & El-Sawi, 2015) against Gram-positive/-negative bacteria, fungi and algae (Alberto, Rossato, Alves, Alves, & Braga, 2011; Pernak, Sobaszkiewicz, & Mirska, 2003). These remarkable biocidal activities could be attributed to the strong electrostatic interaction with the anionic microbial cell wall (Sauvet, Fortuniak, Kazmierski, & Chojnowski, 2003) followed by disruption of the cytoplasmic membrane (Gottenbos, van der Mei, Klatter, Nieuwenhuis, & Busscher, 2002; Hazziza-Laskar, Nurdin, Helary, & Sauvet, 1993). Recently, much attention has thus been paid to polymeric ionic liquids (PILs) or polyelectrolytes (PEs) due to their amazing features including high thermal and mechanical stabilities, inherent conductivity (Nakajima & Ohno, 2005), excellent electrochemical properties (Azzaroni, Moya, Farhan, Brown, & Huck, 2005), biocompatibility (Lu, Yan, & Texter, 2009) and catalytic efficacy (Jia et al., 2010).

So and according to recently reported works (Elshaarawy & Janiak, 2016; Chen et al., 2015), it strongly plausible that the modulation of chitosan (CS) with PILs grafts is an effective way to improve aqueous solubility and pharmacological performance, as well. Inspired by the aforementioned facts and as a continuation of our research program oriented for designing and fabrication of pharmacological (Elshaarawy & Janiak, 2014a,b; Elshaarawy et al., 2015; Elshaarawy, Mokbel, & El-Sawi, 2016; Elshaarawy, Refaee, & El-Sawi, 2016; Elshaarawy, Eldeen, & Hassan, 2017) and antifouling (Elshaarawy & Janiak, 2016; Elshaarawy, Mustafa, Herbst, Farag, & Janiak, 2016; Elshaarawy et al., 2016) candidates, we intend herein to use a natural biopolymer chitosan (CS) (see Scheme S1) as a raw material for fabrication of novel polyelectrolyte-grafted chitosan Schiff bases (PECSBs) with anticipated anti-biofilms and anti-biofouling performance looking forward for developing new economically attractive, ecofriendly and promising antifouling coatings.

2. Experimental section

Instrumentation, materials, extraction of chitin from shrimp shells, partial deacetylation of chitin for preparation of CS, fabrication of low molecular weight CS (LMWCS) and the preparation details of a series of ionic liquids-based aldehydes were described in the electronic supplementary information (ESI†).

2.1. Synthesis of polyelectrolyte chitosan Schiff bases (PECSB1-4)

Generally, 2 g of LMWCS was dissolved in 200 mL of a mixed solution of 2% aqueous acetic acid/ethanol under stirring at room temperature for 30 min. Ionic liquids-based aldehydes (**3**a-c, **4**) (equivalent to molar N-content in LMWCS, half molar in case of **4**) was dissolved in EtOH (30 mL) and the solution was added to the chitosan solution over a period of 30 min at 50 °C under vigorous stirring. After stirring for further 24 h at the same temperature, the reaction mixture was concentrated under reduced pressure to give oily residue which diluted with excessive amount of ethylacetate (AcOEt) and ultrasonically irradiated for 3 h to solidify these obtained oily products. The isolated solids were filtered and then washed with 30:70, 20:80, and 0:100 EtOH:AcOEt mixtures, sequentially. Finally, the desired products **PECSB1-4** were dried at 35 °C under vacuum for 24 h. Samples of the isolated solids were characterized as follows;

2.1.1. Poly-(5-(N,N,N-trimethylammonium chloride)-3-methylsalicylidene) chitosan (PECSB1)

Pale yellow powder, yield (3.16 g). FTIR (KBr, cm^{-1}): 3448 (vs, br, $\nu_{(O-H+NH2)}$), 3178 (m, br, $\nu_{(N-H)}$), 1663 (s, sh, $\nu_{(C=O)acetyl}$), 1632 (vs, sh, $\nu_{(C=N)azomethine}$), 1558 (m, sh, amide II), 1374 (m, sh, amide III), 1280 (m, sh, $\nu_{(Ar-O)}$), 1073 (m, sh, $\nu_{(C-O-C)str}$), 894 (m, sh, $\nu_{(C-O-C)}$, β-glycosidic linkage). ^{1}H NMR (600 MHz, 1% $CD_3COOD/D_2O)_{60\,°C}$ δ (ppm):11.42 (s, 2H), 9.08 (d, *J* = 11.1 Hz, 2H), 8.29 (s, 1H), 7.57 (s, 2H), 7.24 (s, 2H), 5.61-5.38 (m, 4H), 4.78 (s, 4H), 4.28-4.10 (m, 4H), 4.02-3.81 (m, 4H), 3.82 (br, s, 4H), 3.56 (t, *J* = 7.2 Hz, 4H), 3.44 (t, *J* = 6.9 Hz, 4H), 3.37 (s, 9H), 3.33 (s, 9H), 3.28-3.05 (m, 4H), 2.77 (t, *J* = 7.8 Hz, 4H), 2.12 (s, 3H). ^{13}C NMR (151 MHz, 1% $CD_3COOD/D_2O)_{60\,°C}$ δ (ppm): 177.23, 163.72, 159.51, 138.25, 136.12, 135.15, 133.42, 130.30, 129.06, 128.34, 128.31, 127.59, 127.48, 126.51, 126.08, 125.63 124.22, 110.12, 89.89, 83.65, 81.44, 80.61, 79.99, 76.55, 75.33, 72.36, 71.23, 69.04, 68.89, 66.19, 63.28, 56.91, 55.70, 53.67, 52.48, 52.41 and 24.66.

2.1.2. Poly-N-(5-(1,2-dimethylimidazolium chloride)-3-methylsalicylidene) chitosan (PECSB2)

Yellow powder, yYield (3.36 g). FTIR (KBr, cm^{-1}): 3429 (vs, br, $\nu_{(O-H+NH2)}$), 3178 (m, br, $\nu_{(N-H)}$), 1663 (vs, sh, $\nu_{(C=O)acetyl}$), 1632 (vs, sh, $\nu_{(C=N)Azomethine}$), 1549 (m, sh, amide II), 1367 (m, sh, amide III), 1281 (m, sh, $\nu_{(Ar-O)}$), 1154 (s, sh, $\nu_{(H-C=C+H-C=N)bend}$, Im), 1072 (m, sh, $\nu_{(C-O-C)str}$), 896 (m, sh, $\nu_{(C-O-C)}$, β-glycosidic linkage), 768 (m, sh, Im). ^{1}H NMR (600 MHz, 1% $CD_3COOD/D_2O)_{60\,°C}$ δ (ppm): 11.28 (s, 1H), 11.09 (s, 1H), 9.18 (d, *J* = 10.8 Hz, 2H), 8.36 (s, 1H), 7.98 (dd, *J* = 8.6, 2.5 Hz, 2H), 7.91 (d, *J* = 8.1, 2H), 7.88–7.76 (m, 2H), 7.45–7.34 (m, 2H), 5.88 (s, 2H), 5.85 (s, 2H), 5.24 (d, *J* = 5.8, 4H), 4.47–4.25 (m, 6H), 4.13-4.01 (m, 4H), 3.95 (s, 6H), 3.84 (t, *J* = 6.9 Hz, 6H), 3.59 (br, s, 6H), 3.03 (s, 6H), 2.75 (t, *J* = 8.0 Hz, 4H), 2.22 (s, 6H), 1.92 (s, 3H). ^{13}C NMR (151 MHz, 1% $CD_3COOD/D_2O)_{60\,°C}$ δ (ppm): 176.48, 168.36, 162.48, 160.10, 158.19, 152.66, 145.11, 141.98, 138.23, 136.84, 133.16, 130.25, 129.03, 128.51, 128.22, 127.96, 127.53, 126.78, 126.34, 124.71, 109.98, 91.88, 81.51, 77.22, 75.51, 73.45, 71.10, 68.34, 62.06, 58.38, 56.41, 53.48, 50.71, 43.78, 31.21, 24.54, 20.01, 18.21 and 13.16.

2.1.3. Poly-N-(5-(2,4-dimethylpyridinium chloride)-3-methylsalicylidene) chitosan (PECSB3)

Orange yellow powder, yield (3.40 g). FTIR (KBr, cm^{-1}): 3438 (vs, br, $\nu_{(O-H+NH2)}$), 3200 (m, br, $\nu_{(N-H)}$), 1659 (vs, sh, $\nu_{(C=O)acetyl}$), 1631 (vs, sh, $\nu_{(C=N)Azomethine}$), 1563 (m, sh, amide II), 1355 (m, sh, amide III), 1279 (m, sh, $\nu_{(Ar-O)}$), 1158 (s, sh, $\nu_{(H-C=C+H-C=N)bend}$, Py), 1065 (m, sh, $\nu_{(C-O-C)str}$), 890 (m, sh, $\nu_{(C-O-C)}$, β-glycosidic linkage), 742 (m, sh, Py). ^{1}H NMR (600 MHz, 1% $CD_3COOD/D_2O)_{60\,°C}$ δ (ppm): 11.33 (s, 1H), 9.10 (d, *J* = 7.8 Hz, 2H), 8.98 (d, *J* = 10.1 Hz, 2H), 8.34 (s, 1H), 7.90 (s, br, 2H), 7.75 (d, *J* = 7.6 Hz, 2H), 7.45–7.34 (m, 2H), 7.21–7.07 (m, 2H), 5.99 (s, 4H), 5.55 (d, *J* = 6.1, 4H), 4.46–4.24 (m, 6H), 4.15-4.00 (m, 4H), 3.88 (t, *J* = 6.9 Hz, 6H), 3.74 (s, br, 4H), 3.56 (s, br, 4H), 3.28–3.01 (m, 4H), 2.79 (s, 6H), 2.51 (s, 6H), 2.26 (s, 6H), 1.95 (s, 3H). ^{13}C NMR (151 MHz, 1% $CD_3COOD/D_2O)_{60\,°C}$ δ (ppm):