Chemistry–A European Journal
Research Article
doi.org/10.1002/chem.202203764
Chemistry Europe
European Chemical Societies Publishing
www.chemeurj.org

Reduction-triggered Disassembly of Organic Cationic Nanorods Produces a Cysteine Protease Mimic (Miravet et al.)

Diego Navarro-Barreda,[a] César A. Angulo-Pachón,[a] Francisco Galindo,[a] and Juan F. Miravet*[a]

Abstract: The emergence of catalytic activity associated with a disassembly process is reported, reminiscent of complex biological systems. A cystine derivative with pendant imidazole groups self-assembles into cationic nanorods in the presence of the cationic surfactants cetylpyridinium chloride (CPC) or cetyltrimethylammonium bromide (CTAB). Disulfide reduction triggers nanorod disassembly and the generation of a simple cysteine protease mimic, which shows a dramatically improved catalytic efficiency in the hydrolysis of *p*-nitrophenyl acetate (PNPA).

Stimuli-responsive surfactants are fascinating molecules for constructing chameleonic aggregates in aqueous media, which can modulate properties such as the surface activity or the solubilization/entrapment of actives.[1] Furthermore, in a biomedical context, responsive amphiphiles permit the preparation of nanocarriers for targeted delivery, as is the case of stimuli-responsive liposomes.[2]

Additionally, the regulation of catalysis in supramolecular systems is an exciting area of research related to Systems Chemistry[3] and studies on the origin of life.[4] Stimuli-regulated catalytic activity has been reported several times upon self-assembly of various structures such as an amino-acid derivative,[5] peptides,[6–8] an amphiphile with a 1,4,7-triazacyclononane unit,[9] helices derived from benzene-1,3,5-tricarboxamide[10] or porphyrins.[11] Regulation of catalytic activity in those systems can be achieved with different stimuli like temperature,[5] pH,[6,8,11] light,[7,9] or salts.[10] The hydrolysis of nitrophenyl esters is often the benchmark reaction tested in these systems,[6–9] although other reactions have been assayed, such as aldol-type,[5,11] and hydrosilylation.[10]

We have recently studied reduction-sensitive molecular nanoparticles formed by an anionic bolaamphiphile that contains a disulfide unit as a linker.[12] In drug delivery, introducing disulfide moieties is a common approach for preparing reduction-sensitive nanoparticles, which release their load upon reaction with glutathione.[13] L-Cystine, the dimeric form of L-cysteine, represents an accessible and convenient building block for preparing reduction-sensitive materials such as molecular gels,[14] peptide-based micelles,[15] and nanogels.[16]

Cetyltrimethylammonium bromide (CTAB) forms cationic spherical micelles in water with a critical micelle concentration (CMC) value below 1 mM (reported values depend on the ionic strength of the medium and the methodology used for its determination).[17] A transition from spherical to rod-like micelles occurs for CTAB concentrations above ca. 0.2 M..[18,19] However, if chloride counterions are present (CTAC), only spherical micelles are formed in that range of concentrations.[20,21] Notably, the addition of anions, such as the extensively studied case of sodium salicylate, draws diluted solutions of CTAB, in the millimolar range, towards the formation of micellar nanorods.[22,23] A similar behaviour has been reported for the related cationic surfactant cetylpyridinium (CPC stands for the corresponding chloride salt used in this work).[24]

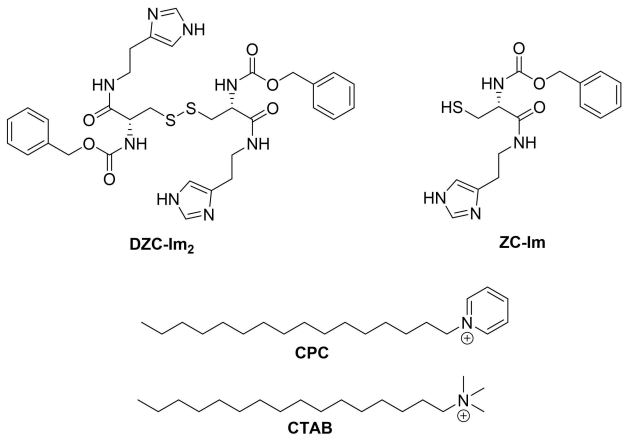
Here we report on nanorod formation by co-assembly of CTAB or CPC with a red-ox sensitive, imidazole-pending cystine derivative (**DZC-Im$_2$**, Scheme 1). Reduction of the cystine unit to cysteine results in nanorod disassembly and generates an esterase-like organic catalyst (Scheme 2).

Following our interest in developing molecular gels[25] and nanoparticles[12] sensible to reduction by glutathione, L-cystine was used as a scaffold to prepare the compound **DZC-Im$_2$** (Scheme 1). The amine groups of cystine were Z-protected as described in the literature,[26] and the carboxylic acid units were condensed with histamine (see Scheme S1 and Figures S1–S4 for NMR analysis).

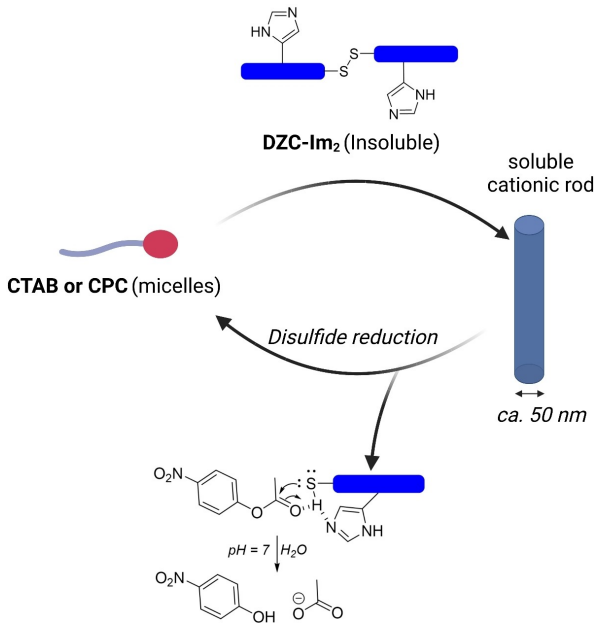
The ionizable imidazole groups introduced in this way were initially aimed to provide solubility around neutral pH. However, a ^{1}H NMR study of the solubility of **DZC-Im$_2$** at different pH values revealed that above pH 6, neutral, insoluble species predominate in the system (Figure S5). However, the addition of cationic surfactants CTAB or CPC permitted complete

[a] *Dr. D. Navarro-Barreda, Dr. C. A. Angulo-Pachón, Prof. Dr. F. Galindo, Prof. Dr. J. F. Miravet*
Department of Inorganic and Organic Chemistry
Universitat Jaume I
Av. Vicent Sos Baynat, s/n 12071 12071Castelló de la Plana (Spain)
E-mail: miravet@uji.es

Supporting information for this article is available on the WWW under https://doi.org/10.1002/chem.202203764



Scheme 1. Structure of the cysteine derivative and cationic surfactants used in this work.



Scheme 2. Pictorial representation of rod formation, its reductive disassembly and the catalytic process studied.

solubilization of **DZC-Im$_2$** at pH 7, indicating the incorporation of this molecule in the cationic micelles formed by the surfactants. For example, a 0.50 mg mL^{-1} (0.7 mM) clear and homogenous solution of **DZC-Im$_2$** was prepared at pH 7 (0.1 M HEPES buffer) in the presence of 1.25 mg mL^{-1} of CPC (3.7 mM) or CTAB (3.4 mM). It is to be noted that the concentration of the surfactants in those samples is well above their CMC values, which are reported to be ca. 1 mM or lower depending on the conditions.[17,24]

Dynamic light scattering (DLS) analysis of solutions of CTAB and CPC (1.5 mg/mL, HEPES 0.1 M) showed poor quality correlograms indicating that the amount of micelles at this concentration is not high enough to obtain good DLS data (Figure S6). However, upon incorporating **DZC-Im$_2$**, the corresponding nanoparticles, **DZC-Im$_2$@CTAB** and **DZC-Im$_2$@CPC**, were formed, showing by DLS a good correlogram and a relatively narrow and symmetrical diameter distribution (Figure 1). Dynamic Light Scattering showed that the Z-average diameters were 232+/−4 nm (PdI = 0.245) and 24+/−38 nm (PdI = 0.250) for the samples with CTAB and CPC, respectively. It has to be noted that these values correspond to apparent diameters calculated for the diffusion of spherical particles. However, as shown below, the system is constituted by nanorods.

CTAB and CPC were used in this study as they represent two of the most common and available cationic surfactants. In addition, a common anionic surfactant like SDS was also assayed but did not afford a micelle to rod morphological change, which, as mentioned previously, is a particular characteristic of different CTAB or CPC systems.

The ζ-potential, directly related to the surface charge of the nanoparticles, was determined by electrophoretic mobility (see Figure S6) and found to be +49 and +44 mV, respectively, for **DZC-Im$_2$@CTAB** and **DZC-Im$_2$@CPC** (Figure S7). ζ-Potential values higher than 30 mV are associated with good colloidal stability,[27] as was the case for the **DZC-Im$_2$@surfactant** samples, which remained clear solutions for several days.

The shape of the objects could be observed by electron microscopy (Figure 2). Straight nanorods with a monodisperse diameter of 50 nm were observed for **DZC-Im$_2$@CPC** and **DZC-Im$_2$@CTAB** samples. These nanorods are considerably wider than the reported cylindrical micelles formed by CPC or CTAB, which have diameters below 10 nm.[28] The length of the rods is in the range of 200–1000 nm with an average value of 499 nm (standard deviation is 210 nm, see the histogram in Figure S9). Such nanostructures are reminiscent of the self-assembled fibrillar networks formed by L-cystine derivatives reported by Menger and coworkers.[26] In the present case, it could be argued that compared to conventional self-assembled fibrillar networks, the coaggregation of **DZC-Im$_2$** with the cationic surfactants limits the length of the straight fibres and avoids the cross-linking that would originate fibrillar networks and gelation. Instead, discrete colloidal nanorods are formed.

As expected, the nanorods are responsive to the presence of reducing agents. The fragmentation of **DZC-Im$_2$** was achieved by reductive treatment with tris(2-

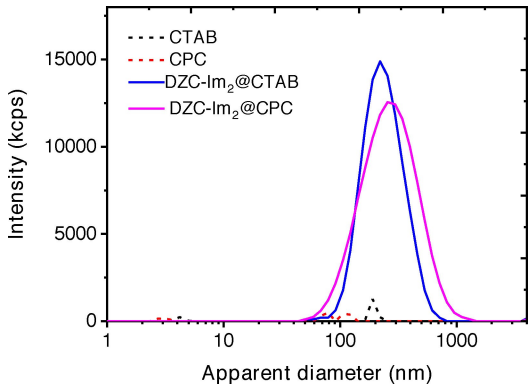


Figure 1. DLS size distribution curves.

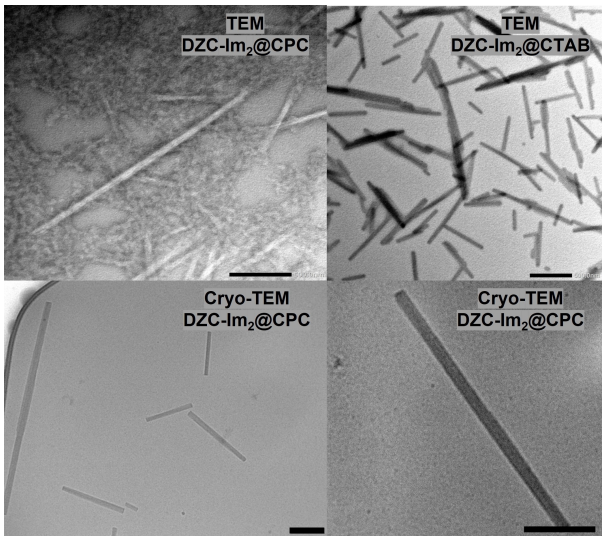


Figure 2. TEM and cryo-TEM images of the nanorods formed by **DZC-Im$_2$@CTAB** and **DZC-Im$_2$@CPC** (0.1 M HEPES, pH 7.0; uranyl acetate was used as a staining agent for the TEM images). Scale bars: 500 nm (for TEM images) and 200 nm (for Cryo-TEM images).

carboxyethyl)phosphine (TCEP),[29] affording **ZC-Im** (Scheme 1). DLS analysis revealed a dramatic decrease in the intensity of dispersed light, ascribable to particle disassembly 5 h after the addition of TCEP (Figure 3 and Figure S9). Furthermore, after reductive treatment, DLS revealed the dispersion of light by objects of a diameter of 1–3 nm, which can be ascribed to the formation of micelles, which were not detected before the reaction with TCEP (Figure S10).

The presence of imidazole units in **DZC-Im$_2$** prompted us to evaluate its potential catalytic activity as a hydrolase and the role that aggregation may play in catalysis. Although compound **DZC-Im$_2$** was poorly soluble in water, it could be dispersed upon ultrasonication into a homogeneous suspension. Catalytic (hydrolase-like) self-assembled nanostructures with imidazole groups have been formed previously using a

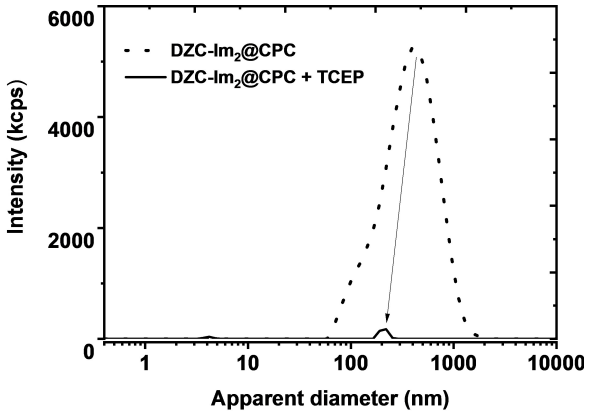


Figure 3. Variation of the intensity of scattered light measured by DLS for a sample **DZC-Im$_2$@CPC** upon addition of TCEP (2.0 mM) after 5 h at 37 °C.

cyclodipeptide,[30] hydrogelators[31,32] and peptide amphiphiles..[33–35]

The study of the hydrolysis of *p*-nitrophenyl acetate (PNPA), a common benchmark reaction to asses hydrolase-like activity,[36] was carried out at 25 °C, pH 7.0 (HEPES 0.1 M), and the reaction progress was monitored by the UV-Vis absorbance of p-nitrophenol species formed. The results were adjusted to pseudo-first-order kinetics (see Equation (3); k_{bkgd} and k_{cat} are, respectively, the rate constants for the uncatalyzed and imidazole-derivative catalyzed reaction).

$$\text{rate} = (k_{bkgd} + k_{cat}\,[\text{DZC-Im}_2])\,[\text{PNPA}] \quad (1)$$

$$k_{\text{DZC-Im2}} = k_{bkgd} + k_{cat}\,[\text{DZC-Im}_2] \quad (2)$$

$$\text{rate} = k_{\text{DZC-Im2}}\,[\text{PNPA}] \quad (3)$$

The kinetic experiments revealed that the dispersed aggregates of **DZC-Im$_2$** (0.14 mM) present an enhanced activity compared to the background hydrolysis ($k_{\text{DZC-Im2}}/k_{bkgd} = 5.5$). As shown in Figure 4, Upon adding CPC (0.74 mM), the formation of **DZC-Im$_2$@CPC** nanorods resulted in a higher catalytic activity ($k_{\text{DZC-Im2@CPC}}/k_{bkgd} = 10$) which could be tentatively ascribed to a higher number of solvent-exposed imidazole groups, compared to the case of insoluble dispersed aggregates of **DZC-Im$_2$**. Similar results were obtained for the system with CTAB (see Figure S9) with $k_{\text{DZC-Im2@CTAB}}/k_{bkgd} = 10$.

The hydrolysis of PNPA was dramatically accelerated, one order of magnitude, upon reductive treatment and consequent disassembly of **DZC-Im$_2$@CPC** nanorods ($k_{\text{DZC-Im2}}/k_{bkgd} = 111$; $k_{\text{DZC-Im2@CPC}}/k_{\text{DZC-Im2}} = 12$; Figure 4). Control experiments revealed that the TCEP alone did not affect the hydrolysis rate. Similar results were obtained with CTAB (see Supporting Information, $k_{\text{DZC-Im2}}/k_{bkgd} = 128$; $k_{\text{DZC-Im2@CTAB}}/k_{\text{DZC-Im2}} = 12$). The reduction of the disulfide groups of **DZC-Im$_2$** produces two molecules of **ZC-Im**

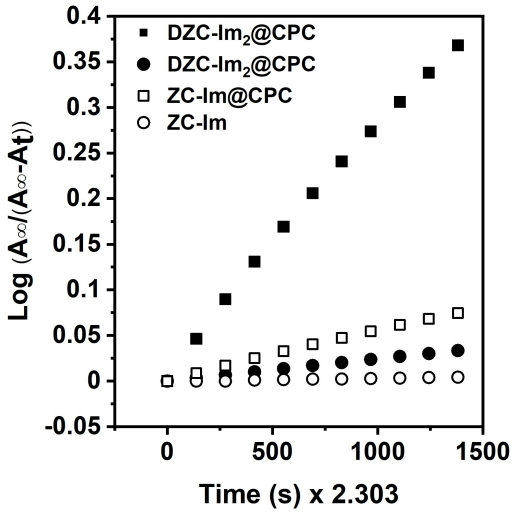


Figure 4. Pseudo-first-order plots for PNPA hydrolysis at 25 °C, pH 7 in 0.1 M HEPES buffer. Slope equals $k_{\text{ZC-Im}}$ (squares), $k_{\text{DZC-Im2@CPC}}$ (circles), $k_{\text{DZC-Im2}}$ (triangles) and k_{bkgd} (inverted triangles). A_∞ and A_t, respectively, stand for the absorbance measured at the end of the reaction and at a given time.

(Scheme 1), which present a thiol and an imidazole group. Notably, the presence of these two functional groups makes of **ZC-Im** a simple cysteine protease mimic. Indeed, **ZC-Im** was easily isolated by chromatography and characterized after the reduction of the parent compound with TCEP in water.

The catalytic activity in **ZC-Im** and **ZC-Im@CPC** was assessed by dissolving this compound in the buffer under the same conditions assayed above (Figure 4; similar results were obtained for **ZC-Im@CTAB**, see Figure S7). The catalytic activity of **ZC-Im** in the hydrolysis reaction was almost negligible ($k_{ZC\text{-}Im}/k_{bkgd} = 1.3$) but was significantly increased in the presence of the CPC ($k_{ZC\text{-}Im@CPC}/k_{bkgd} = 23$). A rationale for this result is that the presence of the CPC micelles permits achieving a high effective concentration for both the catalyst and PNPA, a hydrophobic substrate, upon their incorporation into the micelles. This effect is well-known and reported in different cases of micellar catalysis.[37] Although **ZC-Im@CPC** catalyzes PNPA hydrolysis, the efficiency is lower than that of **DZC-Im2@CPC** after reduction with TCEP, despite being the same catalyst. In this regard, it can be argued that a critical issue is a difference in water solubility at pH = 7 between **DZC-Im$_2$** (< 0.1 mM as determined by NMR) and **ZC-Im** (2 mM). Therefore, in the system **DZC-Im$_2$@CPC**, a quantitative inclusion of **DZC-Im$_2$** molecules in the rods is expected, and reductive treatment presumably affords micelles containing most of the formed compound **ZC-Im**. However, upon dissolution of previously prepared **ZC-Im** in micellar medium, considering its solubility in water, probably only a fraction of the molecules is loaded in the micelles.

One exciting possibility of this system is the regeneration of the nanorods by oxidation of the thiol **ZC-Im** back to disulfide **DZC-Im$_2$**. This reaction works easily using iodine without surfactants in water, affording a precipitate of **DZC-Im$_2$**. Unfortunately, the reaction is incompatible with the presence of CPC or CTAB, producing the fast precipitation of the surfactant with all the assayed oxidants (Iodine with catalytic iodide, hexacyanoferrate (III) and hydrogen peroxide-catalytic iodine). The precipitation of cationic surfactants in the presence of different anions has been reported previously.[38]

Cysteine proteases are widely studied enzymes involved in the hydrolysis of peptides and proteins[39] and are implicated in various disease processes.[40] The active site of the cysteine proteases presents a cysteine and a histidine moiety. It has been proposed that the histidine's imidazole group facilitates deprotonation of the SH group of cysteine, generating a thiolate, which would act as a nucleophile towards the carbonyl of the peptidic bond.[41]

Performing kinetic experiments at different concentrations of **DZC-Im$_2$** permitted the obtention of k_{cat} values [see Equation (2)]. The results in Table 1 reveal a 10-fold increase in the catalytic constant upon reducing **DZC-Im$_2$@CPC** nanorods with TCEP. Similarly, a 6-fold increase was measured for **DZC-Im$_2$@CPC**. It has to be noted that the k_{cat} values are one order of magnitude higher than that of imidazole at the same pH (0.13 M^{-1} s^{-1})[36] and comparable to those reported for some amino acid and peptide derivatives at the same pH, with values ranging from 1.8 to 10.6 M^{-1} s^{-1}.[42–45]

Table 1. Kinetic constants of PNPA hydrolysis at pH 7 and 25 °C.[a]

Catalyst	k_{cat} [M^{-1} s^{-1}]
DZC–Im2@CPC	0.2
DZC–Im2@CPC + TCEP	2.1
DZC–Im2@CTAB	0.2
DZC–Im2@CTAB + TCEP	1.4

[a] Estimated error is lower than 5% in all the cases.

Therefore, the results indicate that the simple molecule **ZC-Im**, presenting vicinal thiol and imidazole units, could be regarded as a cysteine protease mimic. Only a few examples of such mimics have been reported in the literature, such as zwitterions produced from imidazole-thiol pairs,[46] peptides with different arrangements of the cysteine and histidine units,[47] and blends of cyclic dipeptides bearing histidine or cysteine.[48]

In conclusion, the results presented here constitute, up to our knowledge, an unprecedented example of how a single stimulus (reducing agent) simultaneously triggers the disassembly of nanoobjects (nanorods in this case) and originates the emergence of catalytic activity. It could be stated that this relatively simple system mimics the behaviour of much more complex biological systems, such as the cyclic AMP-dependent protein kinase. The activity of this omnipresent enzyme is controlled by cAMP-promoted dissociation, which results in two phosphorylating active units. In the case reported here, the dimeric disulfide catalytic precursor is solubilized with the help of the cationic surfactants affording nanorods which constitute a stealth catalyst to be activated in a reducing environment.

Acknowledgements

The authors acknowledge the financial support from the Spanish Ministry of Science and Innovation, co-funded by the European Regional Development Fund of the European Union (grants RTI2018-096748-B-I00) and Universitat Jaume I (grant UJI-B2018-30).

Conflict of Interest

The authors declare no conflict of interest.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Keywords: cysteine protease · enzyme mimic · self-assembled rods · stimuli-responsive nanoparticles

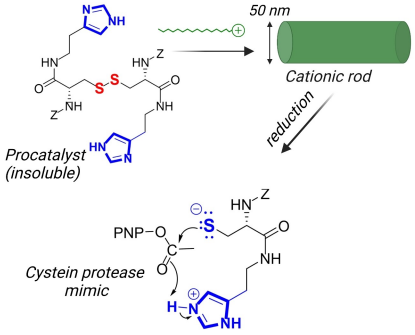
[1] P. Brown, C. P. Butts, J. Eastoe, *Soft Matter* **2013**, *9*, 2365–2374.

[2] Y. Lee, D. H. Thompson, *Wiley Interdiscip. Rev. Nanomed. Nanobiotechnol.* **2017**, *9*, DOI 10.1002/wnan.1450.
[3] E. Mattia, S. Otto, *Nat. Nanotechnol.* **2015**, *10*, 111–119.
[4] S. Mann, *Angew. Chem. Int. Ed.* **2013**, *52*, 155–162; *Angew. Chem.* **2013**, *125*, 166–173.
[5] F. Rodríguez-Llansola, B. Escuder, J. F. Miravet, *J. Am. Chem. Soc.* **2009**, *131*, 11478–11484.
[6] C. Zhang, R. Shafi, A. Lampel, D. MacPherson, C. G. Pappas, V. Narang, T. Wang, C. Maldarelli, R. V. Ulijn, *Angew. Chem.* **2017**, *129*, 14703–14707; *Angew. Chem. Int. Ed.* **2017**, *56*, 14511–14515.
[7] Y. Zhao, B. Lei, M. Wang, S. Wu, W. Qi, R. Su, Z. He, *J. Mater. Chem. B* **2018**, *6*, 2444–2449.
[8] M. Díaz-Caballero, S. Navarro, M. Nuez-Martínez, F. Peccati, L. Rodríguez-Santiago, M. Sodupe, F. Teixidor, S. Ventura, *ACS Catal.* **2021**, *125*, 618–626.
[9] C. Z. J. Ren, P. Solís Muñana, J. Dupont, S. S. Zhou, J. L. Y. Chen, *Angew. Chem. Int. Ed.* **2019**, *58*, 15254–15258; *Angew. Chem.* **2019**, *131*, 15398–15402.
[10] Y. Li, X. Caumes, M. Raynal, L. Bouteiller, *Chem. Commun.* **2019**, *55*, 2162–2165.
[11] A. Arlegui, P. Torres, V. Cuesta, J. Crusats, A. Moyano, *Euro. J. Org. Chem.* **2020**, *2020*, 4399–4407.
[12] D. Navarro-Barreda, B. Bedrina, F. Galindo, J. F. Miravet, *J. Colloid Interface Sci.* **2022**, *608*, 2009–2017.
[13] Y. Ding, Y. Dai, M. Wu, L. Li, *Chem. Eng. J.* **2021**, *426*, 128880.
[14] J. P. Wojciechowski, A. D. Martin, P. Thordarson, *J. Am. Chem. Soc.* **2018**, *140*, 2869–2874.
[15] Y. Dai, Z. Jiang, J. Li, M. Wang, C. Liu, W. Qi, R. Su, Z. He, *J. Mater. Chem. B* **2020**, *8*, 1944–1951.
[16] M. Mackiewicz, J. Romanski, P. Krug, M. Mazur, Z. Stojek, M. Karbarz, *Eur. Polym. J.* **2019**, *118*, 606–613.
[17] B. Naskar, A. Dan, S. Ghosh, V. K. Aswal, S. P. Moulik, *J. Mol. Liq.* **2012**, *170*, 1–10.
[18] T. Imae, R. Kamiya, S. Ikeda, *J. Colloid Interface Sci.* **1985**, *108*, 215–225.
[19] A. Desai, D. Varade, J. Mata, V. Aswal, P. Bahadur, *Colloids Surf. A* **2005**, *259*, 111–115.
[20] J. Ulmius, B. Lindman, G. Lindblom, T. Drakenberg, *J. Colloid Interface Sci.* **1978**, *65*, 88–97.
[21] V. K. Aswal, P. S. Goyal, *Phys. Rev. E – Stat. Physics, Plasmas, Fluids, Relat. Interdiscip. Top.* **2003**, *67*, 8.
[22] Z. Lin, J. J. Cai, L. E. Scriven, H. T. Davis, *J. Phys. Chem.* **1994**, *98*, 5984–5993.
[23] V. K. Aswal, P. S. Goyal, P. Thiyagarajan, *J. Phys. Chem. B* **1998**, *102*, 2469–2473.
[24] L. Abezgauz, K. Kuperkar, P. A. Hassan, O. Ramon, P. Bahadur, D. Danino, *J. Colloid Interface Sci.* **2010**, *342*, 83–92.
[25] D. Navarro-Barreda, C. A. Angulo-Pachón, B. Bedrina, F. Galindo, J. F. Miravet, *Macromol. Chem. Phys.* **2020**, *221*, 1–6.
[26] F. M. Menger, K. L. Caran, *J. Am. Chem. Soc.* **2000**, *122*, 11679–11691.
[27] D. J. Pochapski, C. Carvalho Dos Santos, G. W. Leite, S. H. Pulcinelli, C. V. Santilli, *Langmuir* **2021**, *37*, 13379–13389.
[28] K. R. Francisco, M. A. da Silva, E. Sabadini, G. Karlsson, C. A. Dreiss, *J. Colloid Interface Sci.* **2010**, *345*, 351–359.
[29] J. A. Burns, J. C. Butler, J. Moran, G. M. Whitesides, *J. Org. Chem.* **1991**, *56*, 2648–2650.
[30] Y. Chen, Y. Yang, A. A. Orr, P. Makam, B. Redko, E. Haimov, Y. Wang, L. J. W. Shimon, S. Rencus-Lazar, M. Ju, P. Tamamis, H. Dong, E. Gazit, *Angew. Chem. Int. Ed.* **2021**, *60*, 17164–17170.
[31] A. M. Garcia, M. Kurbasic, S. Kralj, M. Melchionna, S. Marchesan, *Chem. Commun.* **2017**, *53*, 8110–8113.
[32] N. Singh, M. P. Conte, R. V. Ulijn, J. F. Miravet, B. Escuder, *Chem. Commun.* **2015**, *51*, 13213–13216.
[33] M. O. Guler, S. I. Stupp, *J. Am. Chem. Soc.* **2007**, *129*, 12082–12083.
[34] Z. Huang, S. Guan, Y. Wang, G. Shi, L. Cao, Y. Gao, Z. Dong, J. Xu, Q. Luo, J. Liu, *J. Mater. Chem. B* **2013**, *1*, 2297–2304.
[35] M. Bélières, N. Chouini-Lalanne, C. Déjugnat, *RSC Adv.* **2015**, *5*, 35830–35842.
[36] A. Lombardo, *J. Chem. Educ.* **1982**, *59*, 887–888.
[37] G. La Sorella, G. Strukul, A. Scarso, *Green Chem.* **2015**, *17*, 644–683.
[38] W. Selig, *Mikrochim. Acta* **1979**, *72*, 373–381.
[39] H. H. Otto, T. Schirmeister, *Chem. Rev.* **1997**, *97*, 133–171.
[40] R. Leung-Toung, Y. Zhao, W. Li, T. Tam, K. Karimian, M. Spino, *Curr. Med. Chem.* **2006**, *13*, 547–581.
[41] J. W. Keillor, R. S. Brown, *J. Am. Chem. Soc.* **1992**, *114*, 7983–7989.
[42] N. Singh, M. P. Conte, R. V. Ulijn, J. F. Miravet, B. Escuder, *Chem. Commun.* **2015**, *51*, 13213–13216.
[43] N. Liu, S. B. Li, Y. Z. Zheng, S. Y. Xu, J. S. Shen, *ACS Omega* **2022**, *16*, 29.
[44] A. J. Burton, A. R. Thomson, W. M. Dawson, R. L. Brady, D. N. Woolfson, *Nat. Chem.* **2016**, *8*, 837–844.
[45] Z. Huang, S. Guan, Y. Wang, G. Shi, L. Cao, Y. Gao, Z. Dong, J. Xu, Q. Luo, J. Liu, *J. Mater. Chem. B* **2013**, *1*, 2297–2304.
[46] K. I. Skorey, R. S. Brown, *J. Am. Chem. Soc.* **1985**, *107*, 4070–4072.
[47] M. J. Heller, J. A. Walder, I. M. Klotz, *J. Am. Chem. Soc.* **1977**, *99*, 2780–2785.
[48] A. J. Kleinsmann, B. J. Nachtsheim, *Org. Biomol. Chem.* **2019**, *18*, 102–107.

Manuscript received: December 2, 2022
Accepted manuscript online: February 20, 2023
Version of record online: ■■, ■■

RESEARCH ARTICLE

The triggered disassembly of nanorods produces an effective enzyme mimic, a process related to complex biological systems as cyclic AMP-dependent protein kinase. An insoluble cysteine protease mimic precursor is incorporated into soluble cationic rods in the presence of cationic surfactants. Reduction of the disulfide unit provokes rod disassembly and generation of a cysteine protease mimic, producing an extraordinary acceleration of PNPA hydrolysis.



*Dr. D. Navarro-Barreda, Dr. C. A. Angulo-Pachón, Prof. Dr. F. Galindo, Prof. Dr. J. F. Miravet**

1 – 6

Reduction-triggered Disassembly of Organic Cationic Nanorods Produces a Cysteine Protease Mimic (Miravet et al.)