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Simultaneous Dyeing and Antibacterial Finishing of Nylon 6 Fabric Using Reactive Cationic Dyes

R. Farouk, Y.A. Youssef, A.A. Mousa and H.M. Ibrahim

Textile Research Division, National Research Centre, Dokki, Giza, Egypt. P.O. Box, 12622

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Abstract: A new bifunctional reactive cationic dye containing bis (monochlorotriazine/quaternary ammonium) reactive cationic system was synthesized. This dye and its monofunctional analogue were studied for simultaneous dyeing and antibacterial finishing of nylon fabric using exhaust method. Relatively high exhaustion and total dye fixation were achieved at pH 5 and 100°C for both dyes. The bifunctional dye exhibited higher levels of exhaustion and fixation efficiency compared to the monofunctional dye. Antibacterial activities of the dyed samples at different concentrations of both dyes were studied against gram positive (*Staphylococcus aureus*) and gram negative (*Escherichia coli*) bacteria. The dyed samples for both dyes are durable to washing and exhibited antibacterial activity after 10 times wash. The presence of the additional quaternary ammonium group in the bifunctional reactive cationic dye promotes a higher antibacterial activity on nylon 6 fabric compared to the nylon fabric dyed with the monofuncional dye. The dyeings exhibited good to high fastness properties.

Key words: Antibacterial activity • Dyeing • Nylon 6 fabric • Quaternary ammonium group • Reactive cationic dyes

INTRODUCTION

In the recent years, the most prominent research and development in the field of functional dyes and their textile applications have attracted much attention. These dyes could bring more concerns to unify the traditional textile dyeing and functional finishing processes into one process. As an attempt to impart antibacterial finishing simultaneously with dyeing, two novel monoazo acid dyes based on N-ester-1, 8-naphthalimide have been synthesized and applied to nylon 6. These dyes showed good build up and antibacterial activity against Gram-positive and Gramnegative bacteria [1]. Also, some monoazo disperse dyestuffs have been synthesized by the reaction of sulphanilamidodiazonium chloride derivatives with indan-1, 3- dione and applied to wool and nylon 6.6 fabrics. These dyes gave excellent dyeing properties with good biological activities towards different microorganisms [2]. Moreover, the antibacterial

activity of two new disazo direct dyes containing 4, 4'-diaminostilbene-2, 2'-disulfonic acid 4. 4'-diaminobenzanilide as bis-diazomium components and 4-chlorosalicylanilide as coupling component have been investigated on cotton fabric [3]. Other researchers have demonstrated that the cationic dyes, containing quaternary ammonium residues, can be effectively introduced into textile materials to achieve simultaneous coloration and antimicrobial function. The presence of a chemically bonded quaternary ammonium group enables the biostatic activity of the dyes because of the positive charge at the N-atom inflicts a variety of detrimental effects on microbes, including damage to cell membranes, denaturation of proteins and disruption to the cell structure [4]. To increase the effectiveness and the durability of the antimicrobial finish of these dyes, research has been oriented toward synthesis of cationic reactive dves by chemical binding of a quaternary ammonium group to the reactive group of the dye which can be

chemically bonded with the substrate during the application process. For example, an antibacterial reactive cationic dye was synthesized by reacting aminoanthraquinone with cyanuric chloride, 3-dimethylamino-1-propanol and lauryl chloride; this dye was applied to cotton fabric without salt addition. The dye exhibited adequate antimicrobial activities against Gram positive and Gram negative bacteria at a concentration of 10 ppm [5]. Different studies previously reported about reactive cationic dyes containing quaternary ammonium compound but without examining their antibacterial activities [6-10].

In the interest of understanding the role of quaternary ammonium compound in the reactive dye molecule, the present work describes the synthesis of new bifunctional reactive cationic dye containing bis (monochlorotriazine) and bis(quaternary ammonium group) reactive cationic system [Bis(MCT/QA) dye 1] compared with monofunctional reactive cationic dye having one quaternary ammonium group and monochlorotriazine reactive group [MCT/QA dye 2]. The dyes were applied to nylon 6 fabric and their dyeing, colour fastness and antibacterial properties were assessed.

Bifunctional bis (MCT/QA) reactive cationic dye 1

Monofunctional MCT/QA reactive cationic dye 2

Expermintal

Materials: Single jersey knitted nylon 6 fabric, 114 g/m², 135 d/30f was obtained from El-Shourbagy Co., Egypt. Before dyeing the fabric was treated with a solution containing 2g/l nonionic detergent (Sera® Wash M-RK, DyStar, Egypt) at 80°C for 1h, thoroughly washed in water and air dried at room temperature. 4-Aminoacetanilide and m-toluidine were obtained from Fluka Chemie AG. Cyanuric chloride, 4, 4'-diaminobenzanilide and 3-aminoacetanilide were obtained from Aldrich. All other chemicals and solvents used in this study were of laboratory reagent grade. Two bacterial strains were obtained from the Faculty of women for Art, Science & Education, Ain Shams University, Cairo, Egypt. Staphylococcus aureus (S. aureus) as Gram-positive (g+ve) bacteria and Escherichia coli (E. coli) as Gram-

negative (g-ve) bacteria were selected, as they are the most frequent bacteria in the wound infection to represent g+ve and g-ve bacteria. Fresh inoculants for antibacterial assessment were prepared on nutrient broth at 37°C for 24 hours.

Methods

Synthesis of 3-aminophenyltrimethylammonium Salt: A solution of 3-aminoacetanilide (9.29 g; 0.06 mole; 97%) in water/ carbon tetrachloride mixture (1:1), was supplied with dimethylsulphate. The reaction mixture was stirred at 70 -75°C for 10h and controlled by TLC. After the reaction was completed, carbon tetrachloride was isolated from the reaction mixture using separating funnel. 5 ml of concentrated sulphuric acid was added drop wise to the solution at room temperature, then the reaction

refluxed for 2h to produce a solution of 3-aminophenyltrimethylammonium methosulphate, which was used directly without isolation.

Synthesis of the Bifunctional bis (MCT/QA) Reactive Cationic Dye 1: The synthesis of bifunctional bis (MCT/QA) reactive cationic dye 1 was described as following:

The first step was a condensation reaction of 3-aminophenyl trimethylammonium methosulphate with cyanuric chloride. An equivalent amount of 3-aminophenyltrimethylammonium salt solution (0.02 mole), adjusted at pH 7, was added drop wise over 30 min to a freshly prepared suspension of cyanuric chloride (3.88 g, 0.02 mole, 95%) in acetone (25ml) and crushed ice (15-20g). The reaction mixture was stirred at 0-5°C for 3h while controlling the pH at 4-5 using aqueous 2M sodium carbonate solution.

The second step was a diazotization/coupling reaction of 4, 4'-diamino-benzanilide and m-toludine. A solution of *m*-toludine (2.26 ml; 0.02 mole; 95%), HCl (2.4 ml; 37%) and 25 ml water was coupled at 0-5°C for 3 hours with the diazonium salt of 4, 4'-diaminobenzanilide (2.30g, 0.01 mole, 99%), which was prepared and bis-diazotised by the method previously described [11]. Then pH was adjusted to 4 by using 2M sodium carbonate aqueous solution, the reaction was stirred under these conditions for a further 2h. The pH of the resultant dye solution was adjusted to 7-7.5 and the precipitated dye was filtered off and dried in a vacuum oven at 40°C.

This dye (4.64 g, 0.01 mole) was dissolved in acetone, then added in portions to the cyanurated 3-aminophenyltrimethylammonium salt over 45 min. The reaction mixture was stirred at 30°C for 5h and maintaining the pH at 5-5.5, in order to complete the second condensation reaction. The precipitated reactive cationic dye was then filtered off and dried at room temperature under vacuum. The dye was characterized by wavelength of maximum absorption (λ_{max}) in 90% aqueous DMF = 407.2 nm and its chemical structure ($C_{53}H_{57}N_{17}S_2O_9Cl_2$) was confirmed by elemental analysis, FT-IR and ¹HNMR spectroscopic analysis.

Elemental analysis, Calc. (%): C, 52.56; H, 4.74; N, 19.66; S, 5.29. Found (%): C, 52.41; H, 4.62; N, 19.41; S, 5.16. IR (v/cm⁻¹): 3393 (NH), 2924, 2847 (–C-H), 1570 (N=N), 1485, 1395 (C-N), 797 (C-Cl).

¹H NMR: δH ([²H₆] DMSO)/ppm: 2.64 (6H, *s*, CH_{3 (m-toluidine)}), 3.10-3.42 (18H, *hidden*, CH_{3 (quaternary)}), 6.17-6.52 (4H, m, *Ph* H), 7.57-7.95 (10H, m, *Ph* H), 8.09 (1H, s, NH_{amide}), 10.41 (s, NH).

Synthesis of the Monofunctional MCT/QA Reactive Cationic dye 2: 4-Aminoacetanilide (3.79 g, 0.025 mole, 99%) was diazotized by the method previously described [12]. The resulting diazonium solution was coupled with a solution of m-toludine (2.82 ml; 0.025 mol; 95%), HCl (3.1 ml; 37%) and 27 ml water at 0-5°C for 3 hours. Then pH was adjusted to 4 using 2M sodium carbonate aqueous solution, the reaction was stirred under these conditions for a further 2 h. The pH of the resultant dye solution was adjusted to 7-7.5, then the precipitated dye was filtered off and dried in a vacuum oven at 40°C. This dye (5.37g, 0.02 mole) was dissolved in acetone, then added to a neutral solution of cyanurated 3-aminophenyltrimethyl ammonium salt, prepared as described above. The reaction mixture was stirred at 30°C for 5h and maintaining the pH at 5-5.5, in order to complete the second condensation reaction. The precipitated dye was filtered off and dried at room temperature under vacuum. The dye was characterized by wavelength of maximum absorption (λ_{max}) in 90% aqueous DMF = 399.8 nm.

The structure of the dye **2** (C₂₈H₃₂N₉SO₅Cl) was confirmed by elemental analysis, FT-IR and ¹HNMR.

Elemental analysis, Calc. (%): C, 52.37; H, 5.02; N, 19.63; S, 4.99. Found (%): C, 52.07; H, 4.98; N, 19.43; S, 4.75. IR (v/cm⁻¹): 3402 (NH), 2921, 2853 (–C-H), 1571 (N=N), 1487, 1401 (C-N), 795 (C-Cl).

¹H NMR: δH([²H₆]DMSO)/ppm: 2.06 (3H, *s*, CH_{3(amide)}), 2.85 (3H, *s*, CH_{3 (m-toluidine)}), 3.38-3.84 (9H, *hidden*, CH_{3 (quatemary)}), 6.48-6.54 (2H, m, *Ph* H), 7.48-7.79 (5H, m, *Ph* H), 7.91 (1H,

s, NH_{amide}), 10.24 (s, NH).

Dyeing Procedure: Nylon 6 fabric was dyed using the reactive cationic dyes **1** and **2** in an Ahiba dyeing machine at a liquor ratio 40:1. The dyebath was prepared at room temperature at a range of dye concentrations (0.25, 0.5, 1.0, 1.5, 2.0 and 3.0% owf) and the pH was adjusted to 4, 5, 6, 7 and 8 using 0.1 mol sodium carbonate solution and diluted acetic acid solution. Dyeing was started at 40°C, then the temperature was raised to 100°C over 70 min. Dyeing was continued at the desired temperature for a further 60 min (130 min total dyeing time)

for studying the effect of pH and dye concentration. To study the effect of dyeing time, dyeings were examined at intervals up to total dyeing time of 160 min. After dyeing, all the dyed samples were rinsed with water and air dried.

Measurements

Instruments: The infrared (IR) spectra were recorded on a Nexus 670 FT-IR Spectrometer (KBr; Thermo Nicolet, USA). The 'HNMR spectra were recorded on a JEOL 500 MHz spectrometer (Japan) using TMS as an internal standard and the chemical shift values are expressed in δ ppm and J values given in Hz. The wavelength of maximum absorption (λ_{max}) was measured on a Shimadzu UV-2401PC UV/Vis spectrophotometer (Shimadzu, Japan).

Dye Exhaustion: The uptake of dye by nylon 6 fabric was measured by sampling the dyebath before and after dyeing. The dye concentration (g/l) of the dyebath, diluted 25-fold with 94% aqueous DMF, was measured on a Shimadzu UV- 2401PC UV/VIS spectrophotometer at λ_{max} using a calibration curve previously obtained using known dye concentrations in 90% aqueous DMF. The percentage of dyebath exhaustion (%*E*) was calculated using Eq. 1 [13]:

$$\%E = \left[1 - \binom{C_2}{C_1}\right] \times 100 \tag{1}$$

where C_1 and C_2 are the concentrations of dye in the dyebath before and after dyeing, respectively.

Dye Fixation: The dye fixation ratio (%F), the percentage of exhausted dye chemically bound on the fibre, was measured by refluxing the dyed samples in 50% aqueous DMF (liquor ratio 20:1) for 10 min to extract the unfixed dye. This procedure was repeated until the extract was clear. The concentration of the extract was then measured spectrophotometrically at (λ_{max}) and the dye fixation ratio calculated using Eq. 2 [13]:

$$\%F = \frac{\left(C_1 - C_2 - C_3\right)}{\left(C_1 - C_2\right)} \times 100 \tag{2}$$

where C_3 is the concentration of extracted dye.

From the dyebath exhaustion (E) and dye fixation ratio (F), the total dye fixation (T), which is the percentage of dye chemically bound relative to the total amount of dye used, was calculated for all dyeings using Eq. 3 [13].

$$\%T = \frac{(\%E \times \%F)}{100} \tag{3}$$

Antibacterial Test Method: The antibacterial activity of the samples was determined against the tested bacteria by disk diffusion method on an agar plate [14].

Fastness Testing: The dyed samples, after washing-off using 2 g/l nonionic detergent (Sera® Wash M-RK, DyStar, Egypt) at 80°C for 15 min, were tested by the standard ISO methods [15]. The tests were as follows: Colour fastness to washing, ISO 105-C02 (1989); fastness to rubbing, ISO 105-X12 (1987); fastness to perspiration, ISO 105-E04 (1989); and fastness to light (ISO 105-B02 (1988).

RESULTS AND DISCUSSIONS

Characterization of the Reactive Cationic Dyes: The dye structure was confirmed by using FT-IR, ¹H-NMR spectroscopy. Infrared absorbance bands at 3393 (dye 1) and 3402 (dye 2) are assigned to the stretching vibration of N–H groups. Also, both dyes are characterized by C–H stretching vibrations appear at 2847, 2924 cm⁻¹ for dye 1 and at 2921, 2853 cm⁻¹ for dye 2. The presence of quaternary ammonium salt is confirmed by new absorption bands at 1485, 1395 for dye 1 and at 1487, 1401cm⁻¹ for dye 2, which assignable to C-N groups. The ¹H-NMR spectra showed multiple peaks in the region 6.17-7.95 ppm in case of dye 1 and 6.48-7.79 ppm for dye 2 assigned to aromatic protons. Additionally, the peak appeared at 8.09 ppm (dye 1) and at 7.91 ppm (dye 2) can be attributed to the amide protons.

Effect of Dyeing pH: A series of dyeings was carried out for nylon 6 fabric at 100°C by varying the dyebath pH from 4 to 8 using 2% owf dye concentration. The dye exhaustion and total fixation yield for reactive cationic dyes 1 and 2 under these conditions were evaluated. The results obtained in Fig. 1 clearly show that the variation of pH has a marginal effect on the dye exhaustion values and total fixation yield for both dyes. This reflects that both dyes are almost independent to dyeing pH and have high resistance to the variations of dyebath pH conditions. From the results we can notice a slight increase in exhaustion and fixation values, achieved for both dyes at pH 5. This pH is slightly acidic, so the extent of protonated amino end group relative to amino group is decreased. The difference in structure-reactivity

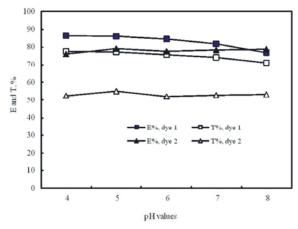


Fig. 1: Exhaustion (E%) and total fixation (T%) of reactive cationic dyes 1 and 2 on nylon 6 fabric at different pH values.

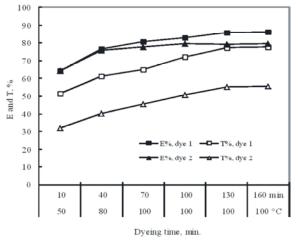


Fig. 2: Effect of dyeing time on the exhaustion and total fixation of dyes 1 and 2 (2% owf) on nylon 6 fabric.

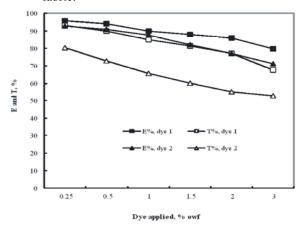


Fig. 3: Exhaustion and total fixation at different concentrations of reactive cationic dyes 1 and 2 on nylon 6 fabric.

between the bifunctional dye 1 and the monofunctional dye 2 leads to higher exhaustion and fixation values achieved using dye 1, due to the presence of the bis (MCT) reactive system in dye 1 instead of the MCT reactive group in dye 2. As well as, the affinity of dye 1 towards nylon fabric is much higher than dye 2 due to the presence of bis (QA) group, which may increases the ionic interaction between the dye cationic groups and the carboxylic-end groups of the fiber.

Effect of Dveing Time: The extent of exhaustion and total fixation yield was investigated for both dyes 1 and 2, at different dyeing times using 2% owf dye concentration, at pH 5 and 100°C. Fig. 2 shows that the dyeing exhaustion for both dyes is significantly increased within the initial dyeing time (30 min) and exhibited relatively high exhaustion values for both dyes within the remaining dyeing time, depending on the dye affinity to the fiber. It is also clear that the total fixation yield of the dyes increases gradually as the dyeing proceeds. Along the range of dyeing time studied, dye 1 exhibited higher exhaustion and fixation efficiency than dye 2. This indicates that the bifunctional bis (MCT/QA) reactive cationic system in dye 1 imparts high substantivity and fixation efficiency compared to the monofunctional MCT/QA reactive cationic system in dye 2.

Effect of Dye Concentration: The study was extended to investigate the dyeing behaviour of both dyes at different dye concentrations (0.25, 0.5, 1, 1.5, 2, 3% owf). The extent of the exhaustion and total fixation yield of dyes 1 and 2 on nylon fabric was examined at pH 5 and 100°C. The results given in Fig. 3 clearly show that at low dye concentrations the extent of exhaustion values achieved using the bifunctional dye 1 were slightly higher than those of the monofunctional but at higher dye concentrations, the bifunctional dye 1 exhibited much better extent of exhaustion than the monofunctional dye 2. It is also clear that the bifunctional dye 1 offers much higher extent of total dye fixation than dye 2, over the range of dye concentration studied. This indicates the potential dye affinity and fixation of the bifunctional bis (MCT/QA) dye 1. For further confirmation to this potentiality, the study was extended to evaluate the antibacterial and colour fastness properties for both bifunctional monofunctional dyes at different dye concentrations as mentioned below.

Table 1: Antibacterial activity of the dyed nylon fabric with reactive cationic dyes 1 and 2 against *S. aureus* and *E. coli*.

		Zone of inhibition (diameter in mm)						
Dye	Dye conc.,% owf	Staphylococcus aureus	Escherichia coli					
1	1	11.0	9.5					
	2	17.0	14.5					
	3	24.0	19.5					
2	1	6.0	4.5					
	2	12.5	13.5					
	3	15.5	16.0					

Table 2: The reduction% for the antibacterial activity of the dye solution of reactive cationic dyes 1 and 2

	Reduction%					
Dye	S. aureus	E. coli				
1	99.90	62.26				
2	72.87	40.54				

Table 3: Effect of washing durability of the dyed fabrics on antibacterial activity of dyes 1 and 2

		Zone of inhibition (diameter in mm)						
Dye	Wash cycle	Staphylococcus aureus	Escherichia coli					
1	0	17.0	14.5					
	1	12.0	11.5					
	5	12.0	9.5					
	10	9.5	8.0					
2	0	12.5	13.5					
	1	12.0	10.0					
	5	11.5	10.0					
	10	7.0	5.0					

Antibacterial Activities of the Reactive Cationic Dyes

Effect of Dye Concentration: For assessment of antibacterial activity, samples dyed with reactive cationic dyes 1 and 2 were subjected to disk diffusion susceptibility test method. The effect of dye concentration (1, 2 and 3% owf) on antibacterial activity was studied and the results are listed in Table 1. The zone of inhibition (diameter) was recorded in each case. The results of undyed samples show clear growth of bacteria under them with no zone of inhibition, indicating that the undved fabric by itself does not inhibit bacterial activity. The investigated dyes inhibit bacterial growth as is evident from the absence of growth under all dyed samples. It was observed that as the dve concentration increases the zone of inhibition increases almost linearly. The increase is much larger for dye 1 than dye 2 as shown in Table 1. From the clear zone of inhibition obtained, it is

apparent that these dyes are bactericidal in nature and not bacteriostatic. The antibacterial activity of the bifunctional dye 1, for all the dyed samples, is more than that of the monofunctional dye 2 because of the presence of bis (QA) groups in its molecular structure. These results are in agreement with the previous studies which explained that the positive electrical charges allow dye molecules to be adsorbed readily onto microbial surfaces and then penetrate the cell membrane, followed by destruction of cell membranes and leakage of cell inclusion body. Simultaneously, bacterial enzyme systems are destroyed, causing bacteria death [16, 17]. The results also showed that the dyed samples give better antibacterial efficacy against S. aureus than E. coli. This may be due to that the cell wall of E. coli (Gram-negative) is more intricate than the cell wall of S. aureus (Gram-positive) thus antibacterial dyes adsorb more readily onto the surfaces of the S. aureus [16, 18]. The results obtained are also confirmed by the reduction% for the antibacterial activity of the dye solution as shown in Table 2.

Effect of Washing Durability of the Dyed Samples:

The effect of washing durability on the antibacterial activity of the dyed samples using 2% owf dye concentration for dyes 1 and 2 was studied and the results are shown in Table 3. The dyed samples were washed off using 2g/l nonionic detergent for 15 min at 80°C for frequent times up to maximum ten times wash. The results clearly show that the dyed samples for both dyes are durable to washing and the samples still have acceptable antibacterial activity after 10 times wash. The bifunctional dve 1 shows higher washing durability on antibacterial activity than the monofunctional dye 2. The bifunctional bis(MCT) reactive system allow the dye molecule to be covalently bonded to the fibre along with its bis(QA) groups causing better antibacterial efficacy than the monofunctional system in dye 2.

Fastness Properties: The fastness properties of the reactive cationic dyes 1 and 2 on nylon 6 at 1 and 2% owf dye concentrations, liquor ratio 40:1, pH 5 at 100°C for both dyes are given in Table 4. The results show that the fastness properties of the dyed samples of dye 1 for washing and perspiration are slightly better than those of dye 2 depending on the percentage of dye fixed. The rubbing fastness properties for dye 2 are slightly better. The light fastness was identical for both dyes. This seems reasonable as the dyes under investigation have the same chromophoric system.

Table 4: Fastness properties of reactive cationic dyes 1 and dye 2 on nylon 6 fabric

		Fastness to perspiration											
		Fastness	s to rubbing	Wash fastness		Alkaline		Acidic					
Dye	Dye conc. (% owf)	Dry	Wet	Alt ¹	SC ²	SW ³	Alt	SC	SW	Alt	SC	SW	Light
1	1	3-4	3-4	4-5	5	4-5	4-5	5	4-5	4-5	5	4-5	5
	2	3	3-4	4-5	5	4-5	4-5	5	4-5	4-5	5	4-5	5
2	1	4	4	4-5	4-5	4	4-5	4-5	4	4-5	4-5	4	5
	2	3-4	4	4-5	4-5	3-4	4-5	4	3-4	4-5	4	3-4	5

¹Alt = alteration; ²SC = staining on cotton; ³SW = staining on wool

CONCLUSION

A new antibacterial bifunctional reactive cationic dye containing bis (MCT/QA) reactive cationic system was synthesized for simultaneous dyeing and antibacterial finishing for nylon 6 fabric with high application performance, compared with reactive cationic dye having monofunctional MCT/QA reactive cationic system. The dyes were applied at different dyeing conditions. Better exhaustion and total dye fixation were achieved at pH 5 and at 100°C for both dyes. The dyes possess high resistance to the variations of dyeing pH conditions. Because of the bis (OA) groups in dve 1, it leads to better substantivity to nylon fabric coupled with higher exhaustion and fixation efficiency of being based on bis (MCT) reactive system compared to the monofunctional reactive cationic dye 2. Even more important, the additional quaternary ammonium group in dye 1 makes such model of reactive cationic dyes show higher antibacterial efficacy compared monofunctional analogue. The satisfactory dyeing performance and good antibacterial properties on nylon fabric should lead to design of novel antibacterial reactive cationic dyes with good application properties.

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