



Synthesis and spectral properties of new fluorescent hydrazone disperse dyes and their dyeing application on polyester fabrics



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ABSTRACT

A new series of solid state fluorescent disperse dyes based on hybrid structure of pyrrolinone ester (PES)/ coumarin moiety was prepared and fully characterized using spectroscopic analysis such as ¹HNMR, MS and IR spectra. The coumarin derivative (7- amino-4- methyl coumarin) was initially prepared from amino phenol via multi steps condensation reactions and its diazonium salt coupled with the pyrrolinone ester to form the hydrazone dyes. ¹HNMR data of the prepared dyes exhibited a pure hydrazone form as a mixture of *E*, *Z* isomers. These dyes showing a significant emission in solid state as well as on the dyed fabric. The dyeing application on polyester fabrics was investigated which showing that these dyes show high affinity to polyester fabric, and minor change in the dye affinity by changing pH even under alkaline conditions was observed.

1. Introduction

Fluorescent disperse dyes are used for coloration of synthetic fibers, particularly when aesthetic or functional reasons are required. The most investigated and commercially important groups are coumarins based dyes [1, 2, 3, 4]. Coumarin dyes exhibit intense fluorescence on substitution of various functional groups at different positions [5, 6, 7]. The fluorescence properties of coumarin derivatives can be altered by substituting different functional groups like amino, hydroxy and methoxy at the 7th position. These derivatives have been found to be the most fluorescent molecules among the coumarin family members. 7-Aminocoumarin molecules have been used to synthesize optical brighteners [8] and disperse dyes for the textile coloration [9, 10, 11]. Modification of the electron-accepting ability of the substituent at the 3rd position improves the fluorescence properties of the coumarin derivatives. Moreover, the incorporation of another heterocyclic moiety either as a substituent group or as a fused component into coumarin alters the properties of the parental material, and the resulting compounds may generally exhibit promising or even unprecedented properties. Arylhydrazones dyes was used as an efficient disperse dyes for high temperature polyester dyeing which showing a high affinity with high color strength [12]. In this context, alkyl 4,5-dihydro-5-oxo-2-aryl (1H)pyrrole-3-carboxylate (pyrrolinone ester) is known as very important heterocyclic intermediate for the preparation of different category of colorants [13,

14, 15, 16, 17, 18, 19, 20]. The presence of active methylene group at position 4 in pyrrolinone ester enhances the electrophilic substitution reaction with different electrophiles such as nitriles, aldehydes and diazonium salts [21, 22]. The diazo-coupling reaction with pyrrolinone ester was previously reported in details which forming a pure hydrazone dyes as a mixture of *E* and *Z* isomer and the more abundant isomer was usually *Z* isomer in all previously reported derivatives [13, 14]. In this work we are presenting the synthesis and application of new derivatives of solid state fluorescence hydrazone dyes based on pyrrolinone ester which in conjugated with 7-amino-4-methyl coumarin derivative. The photo-physical study such as absorption and emission spectra in solution and as polycrystalline form will be studied in details. The dyeing application on polyester fabrics of the prepared dyes will be investigated at a wide range of pH, in particular alkaline pH dyeing of polyester. The effect of fluorescence character of the dye into the brightness of dyed fabric will be also discussed.

2. Experimental

2.1. Materials and chemicals

Methyl chloroformate 98%, 3-amino phenol, ethyl acetoacetate, ethyl benzoate, diethylcarbonate 97% and 4-bromoacetophenone 98% were purchased from Sigma Aldrich (Germany) and used directly without any

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further purification. All solvents used (dioxan, dimethylsulfoxide, dimethylformamide, acetonitrile, acetic acid and acetone) were of spectroscopic grade. 100% polyester fabric (91 g/m², yarn count: dtex 44) was supplied by El-Mahalla El-Kobra Company, Egypt. The fabrics were scoured before application in an aqueous bath containing 2 g.L⁻¹ of nonionic detergent at 80 °C for 30 min to remove any processing oils and impurities. Thin-layer chromatography (TLC) was performed by a Kieselgel 60 F254 (Merck, Darmstadt, Germany), for observation of reaction progress and the purity of the prepared intermediates and synthesized dyes.

2.2. Equipment

Melting point was recorded using Sturat SMP30 (UK). The elemental analysis was measured using EA 1108 FISONs elemental analyzer (Germany). Nuclear Magnetic Resonance (NMR) spectra were recorded on a Bruker-spectroscopy DTX 400 ultrashield in hexadeuterateddimethylsulphoxide (DMSO-d₆) using tetramethylsilane (TMS) as internal reference and the chemical shifts (δ) are given in. The UV/vis absorption spectra were recorded on a ShimadzuUV2401 spectrophotometer at the wavelength of maximum absorption ($\lambda_{\max}$) in a range of solvent with different polarity. Mass spectra were measured using Thermo-Scientific™ Fleet LCQ™ ion trap (Germany). Samples were dissolved in MeCN and analyzed by direct infusion at a flow rate of 5 mL/min. Mass spectra were recorded in the range of 50–1000 m/z using electrospray ionization (ESI) and the negative ions as well as positive ion were recorded. Helium 0.5 mL/min was used as a carrier gas. Pressure and flow rate of nitrogen was 10 psi and 5 L/min. Ion source temperature was 275 °C, an ionization voltage was from 2.6 to 2.7 kV.

FT/IR spectra were measured on JASCO FT/IR-4700 spectrometer. Steady-state fluorescence spectra were measured using a JASCO FP-8300 spectrofluorometer (JASCO Inc., USA), the emission spectra was measured in different environment such as polycrystalline, in solution and on the dyed fabric.

2.3. Synthesis of the dye intermediates

7-Amino-4-methyl coumarin was prepared as previously reported method [23, 24]. While different derivatives of pyrrolinone ester (PES) were prepared by the same procedure as described in our previous work [15, 16, 18].

2.4. General procedure for dye preparation

The dyes 1–3 were prepared by diazo-coupling reaction between the diazonium salt of 7-amino-4-methyl coumarin with alkyl 4,5-dihydro-5-oxo-2-aryl (1H) pyrrole-3-carboxylate derivatives (pyrrolinone ester PES), the diazotization of 7-amino-4-methyl coumarin was carried out as follow.

7-Amino-4-methyl-coumarine (0.01 mol) was dissolved in concentrated hydrochloric acid (7.5 mL), (15 mL) acetic acid and stirred until amine was completely dissolved then the mixture cooled (external cooling) to the temperature 0–5 °C. Sodium nitrite solution (0.01 mol) in water (10 mL) was then added dropwise with vigorous stirring during 30 min at 0–5 °C. The reaction mixture was kept at 0 °C for further 30 min. Using iodide-starch paper the excess of nitrous was destroyed by adding traces of sulfamic acid till the blue color on iodide-starch paper disappears. The solution of the diazonium compound was used for next coupling reaction.

(0.01 mol) of PES derivatives were dissolved in 100 mL of ethanol under reflux, then cooled down to 0 °C. The previously prepared diazonium salt was added to the PES dispersion portion wise during 20 min and the mixture was vigorously stirred at 0 °C for 4 h and the reaction mixture was stand overnight and the temperature was allowed to increase till room temperature.

The mixture was filtrated off and the dye cake was washed by water

and dried.

2.4.1. Ethyl-4-(2-(4-methyl-2-oxo-2H-chromen-7-yl)hydrazono)-5-oxo-2-phenyl-4,5-dihydro-1H-pyrrole-3-carboxylate (dye 1)

Preparative yield 85%; purified by recrystallization from ethanol, mp = 258–261 °C.

¹H-NMR (400 MHz, DMSO-d₆, δ , ppm) Z isomer: 1.21 (3H, t, ³J = 7.1 $\pm$ 0.2 Hz, CH₃); 2.38 (3H, s, CH₃); 4.16 (2H, q, ³J = 7.1 $\pm$ 0.2 Hz, CH₂); 6.17–7.71 (9H, m, aromatic protons); 11.42 (1H, s, –CONH–); 13.09 (1H, s, –NHN =).

¹H-NMR (400 MHz, DMSO-d₆, δ , ppm) E isomer: 0.86 (3H, t, ³J = 7.1 $\pm$ 0.2 Hz, CH₃); 2.38 (3H, s, CH₃); 4.04 (2H, q, ³J = 7.1 $\pm$ 0.2 Hz, CH₂); 6.17–7.71 (9H, m, aromatic protons); 11.36 (1H, br. s, –CONH–); 12.98 (1H, s, –NHN =) as shown in supplementary Figure S1.

IR (ATR, $\nu_{\max}$ /cm⁻¹): 3484 (NH, br), 3142 (N–H), 3049 (C–H, aromatic), 2976 (C–H, aliphatic), 1695 (C=O, ester), 1609 (C=O, amide), 1545 (C=N), 1385, 1208 (C–N), 1092 (C–O) as shown in supplementary Figure S7.

MS analysis, M = 417.41 Positive-ion MS: m/z 416.33 [M–H], 100 %; m/z 370.62 [M–H–C₂H₅OH]⁺ as shown in supplementary Figure S4.

Elemental analysis: Calculated (C₂₃H₁₉N₃O₅): C (66.18%), H (4.59%), N (10.07%), and O (19.16%); Found: C (66.25%), H (4.67%), N (10.11%), and O (19.19%)

2.4.2. Methyl-2-(4-bromophenyl)-4-(2-(4-methyl-2-oxo-2H-chromen-7-yl)hydrazono)-5-oxo-4,5-dihydro-1H-pyrrole-3-carboxylate (Dye 2)

Preparative yield 77.5%; purified by recrystallization from ethanol, mp = 295–298 °C.

¹H-NMR (400 MHz, DMSO-d₆, δ , ppm) Z isomer: 2.36 (3H, s, CH₃); 3.68 (3H, s, CH₃); 6.17–7.69 (8H, m, aromatic protons); 11.47 (1H, s, –CONH–); 12.98 (1H, s, –NHN =).

¹H-NMR (400 MHz, DMSO-d₆, δ , ppm) E isomer: 2.36 (3H, s, CH₃); 3.68 (3H, s, CH₃); 6.17–7.69 (8H, m, aromatic protons); 11.37 (1H, s, –CONH–); 12.95 (1H, s, –NHN =) as shown in supplementary Figure S2.

IR (ATR, $\nu_{\max}$ /cm⁻¹): 3475 (NH, br), 3220 (N–H), 3070 (C–H, aromatic), 2986 (C–H, aliphatic), 1690 (C=O, ester), 1605 (C=O, amide), 1535 (C=N), 1382, 1219 (C–N), 1081 (C–O) as shown in supplementary Figure S8.

MS analysis, M = 482.28. Positive-ion MS: m/z 483.08 [M + H], 100 %; m/z 450.17 [M–CH₃OH]⁺, Negative-ion MS: m/z 480.17 [M–H][–] as shown in supplementary Figure S5.

Elemental analysis: Calculated (C₂₂H₁₆BrN₃O₅): C (54.79%), H (3.34%), N (8.17%), and O (16.59%). Found: C (54.91%), H (3.40%), N (8.21%), and O (16.66%).

2.4.3. Ethyl-4-(2-(4-methyl-2-oxo-2H-chromen-7-yl)hydrazono)-5-oxo-2-(thiophen-2-yl)-4,5-dihydro-1H-pyrrole-3-carboxylate (Dye 3)

Preparative yield 85%; purified by recrystallization from ethanol, mp = 268–271 °C.

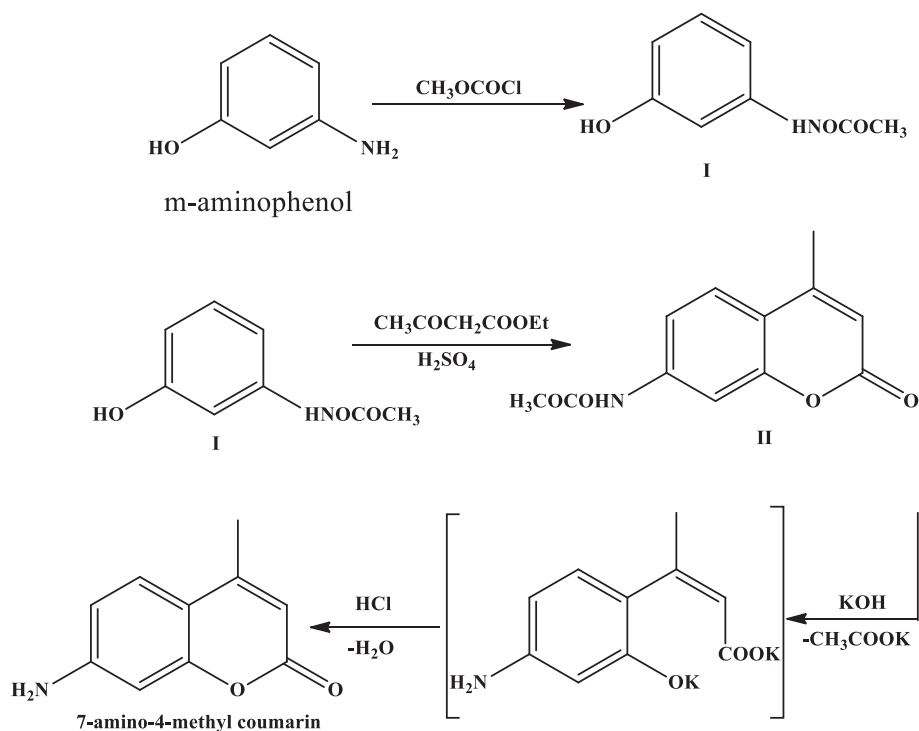
¹H-NMR (400 MHz, DMSO-d₆, δ , ppm) Z isomer: 1.38 (3H, t, ³J = 7.1 $\pm$ 0.2 Hz, CH₃); 2.34 (3H, s, CH₃); 4.15 (2H, q, ³J = 7.1 $\pm$ 0.2 Hz, CH₂); 6.17–7.71 (7H, m, aromatic protons); 11.52 (1H, s, –CONH–); 12.96 (1H, s, –NHN =).

¹H-NMR (400 MHz, DMSO-d₆, δ , ppm) E isomer: 1.05 (3H, t, ³J = 7.1 $\pm$ 0.2 Hz, CH₃); 2.34 (3H, s, CH₃); 4.05 (2H, q, ³J = 7.1 $\pm$ 0.2 Hz, CH₂); 6.16–7.92 (7H, m, aromatic protons); 11.38 (1H, br. s, –CONH–); 12.58 (1H, s, –NHN =) as shown in supplementary Figure S3.

IR (ATR, $\nu_{\max}$ /cm⁻¹): 3668 (NH, br), 3137 (N–H), 3079 (C–H, aromatic), 2979 (C–H, aliphatic), 1705 (C=O, ester), 1670 (C=O, amide), 1557 (C=N), 1388, 1218 (C–N), 1063 (C–O) as shown in supplementary Figure S9.

MS analysis, M = 423.44. Positive-ion MS: m/z 422.50 [M–H], 100 %; m/z 376.42 [M–H–C₂H₅OH] as shown in supplementary Figure S6.

Elemental analysis: Calculated (C₂₁H₁₇N₃O₅S): C (59.57%), H (4.05%), N (9.92%), and O (18.98%). Found: C (59.79%), H (4.13%), N (10.01%), and O (19.00%).



Scheme 1. The synthetic routes for the synthesis of 7-amino-4-methyl coumarin (AMC).

2.5. General procedure of dyeing application

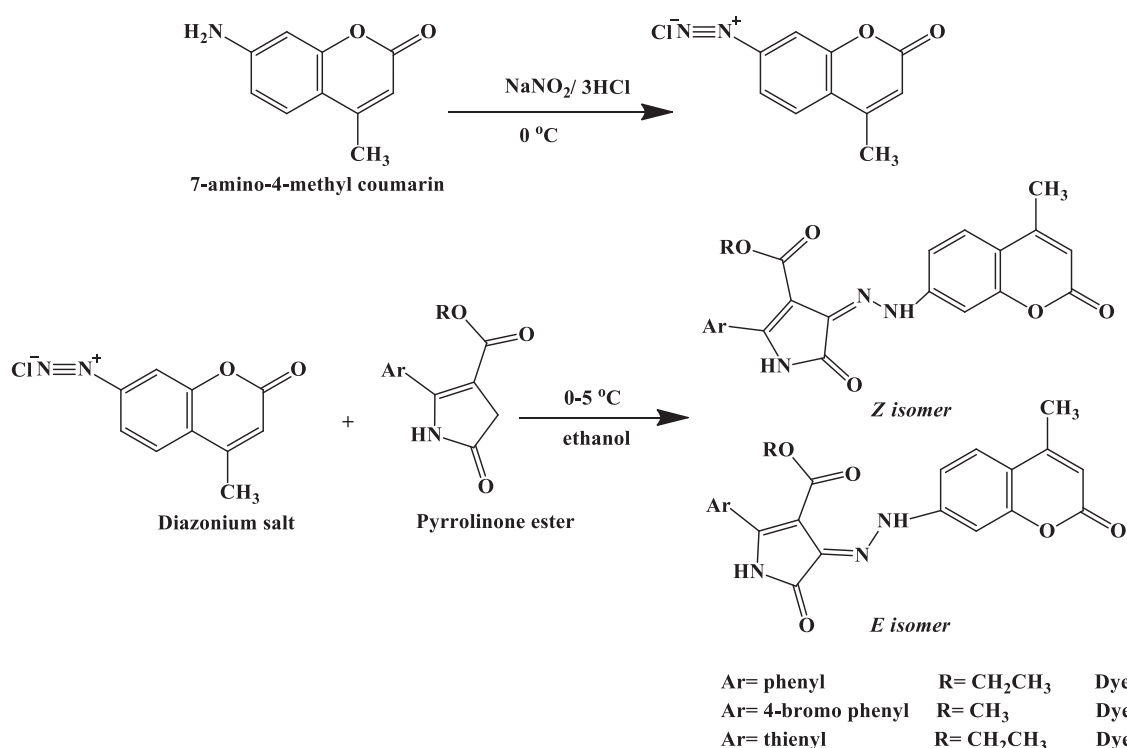
2.5.1. Preparation of dye dispersion

For satisfactory dispersion in the dyebath, the synthesized dyes were initially finished by mortar milling of the desired amount of dye powder in the presence of an anionic dispersing agent (Sera Gal P-LP; DyStar, Egypt) in a 1:1 ratio with a few drops of *N,N*-dimethylformamide, and

then the resulting paste containing a certain % owf dye was mixed with water. The dye solution was ultra-sonicated for 10 min to obtain fine dispersion and then added to the dyebath.

2.5.2. Dyeing procedure

Dyeing was conducted using the previously prepared dye solution of dye 1–3 at the required depth (0.5, 1, 2 and 3 % owf). The pH of the bath



Scheme 2. Synthesis of solid state fluorescent disperse dyes 1–3.

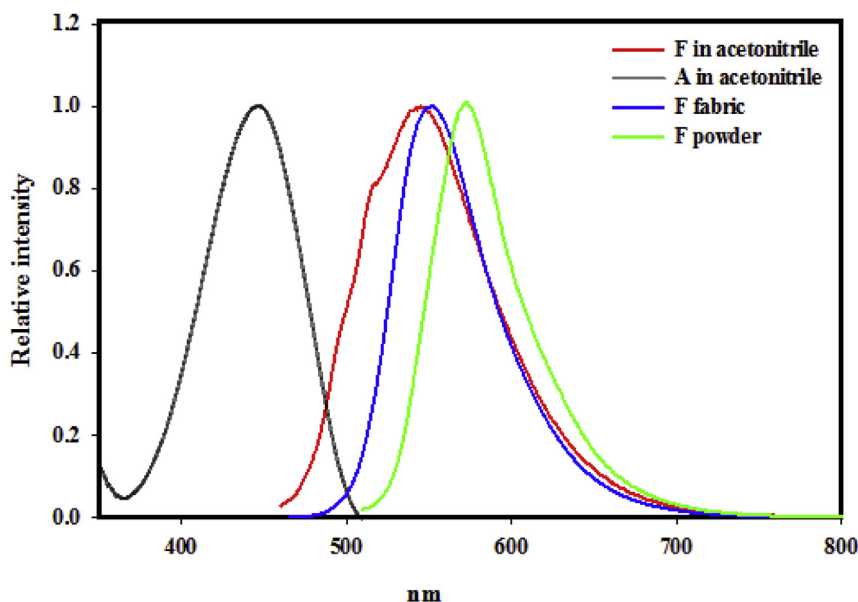


Fig. 1. The absorption (A) and emission (F) spectra in solution, solid state and on the dyed fiber for Dye 1.

was adjusted to 4–5 using 1% diluted acetic acid solution at 40 °C. Dyeing of polyester samples (5 g) was performed by raising the temperature to 120 °C at a rate of 2.5 °C/min and holding at this temperature for 60 min. The dyed samples were then removed, thoroughly rinsed with water, washed off using 2 g.L⁻¹ nonionic detergent (Sera Wash M-RK; DyStar) at a liquor ratio of 50:1 and at 60 °C for 15 min, and then rinsed in cold running water and allowed to dry in the open air.

2.5.3. Color measurements

The relative color strength (K/S) of dyed fabrics was measured by the light reflectance technique using Kubelka-Munk Eq. (1) [25]. The reflectance (R) of the dyed fabrics was measured on Shimadzu UV/Vis spectrophotometer.

$$K/S = \frac{(1-R)^2}{2R} \quad (1)$$

where, R = Decimal fraction of the reflection of the dyed fabric
K = Absorption coefficient, and S = scattering coefficient

2.5.4. Fastness testing

A specimen of dyed polyester fabrics of 1% owf depth of shade were tested according to ISO standard test methods. The wash fastness test was assessed in accordance with the standard method ISO 105-C06 B2S (4 g.L⁻¹ ECE detergent, 1 g.L⁻¹ sodium perborate, 25 steel balls) at 50 °C for 30 min and at a liquor ratio of 50:1. Fastness to acidic and alkaline perspiration was determined with a perspirometer set at specific pressure, temperature and time in accordance with standard method ISO 105-E04. The sublimation fastness test was carried out using a fixometer at 180 and 210 °C in accordance with ISO 105-P01. The light fastness test was assessed using a xenon arc lamp test in accordance with ISO 105-B02.

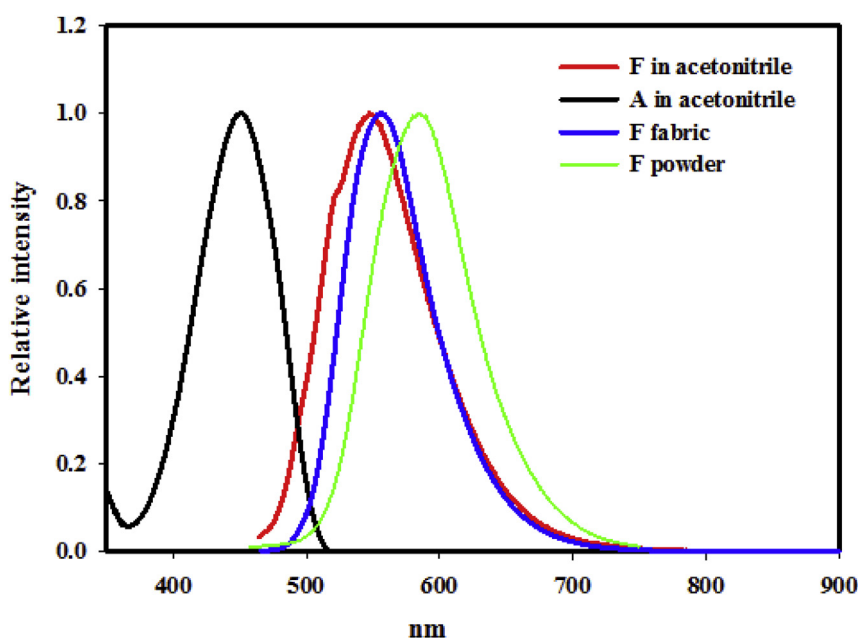


Fig. 2. The absorption (A) and emission (F) spectra in solution, solid state and on the dyed fiber for Dye 2.

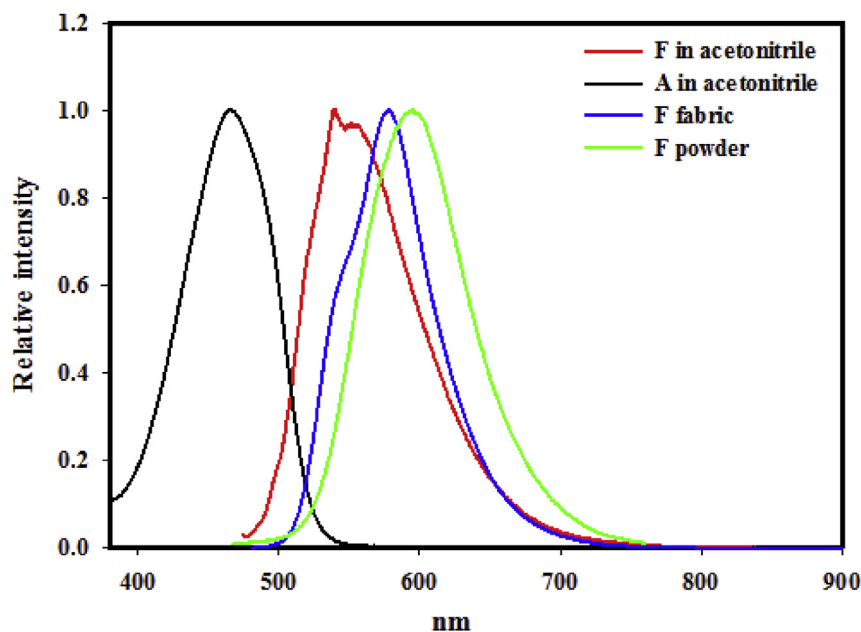


Fig. 3. The absorption (A) and emission (F) spectra in solution, solid state and on the dyed fiber for Dye 3.

Table 1

The absorption, fluorescence maxima and molar absorptivity coefficient for dyes 1-3.

Dye	Absorption λ_{max} nm (acetonitrile)	Emission λ_{max} nm (acetonitrile)	Emission λ_{max} nm (powder)	Emission on the dyed fabrics λ_{max} nm	ϵ_{max} [L.mol ⁻¹ .cm ⁻¹]
1	447	546 (514 sh)	572	551	34140
2	452	548 (519 sh)	580	557	31690
3	454	540, 555	592	565	31720

sh (shoulder vibrionic emission band).

3. Results and discussions

3.1. Synthesis of dye derivatives

The amino coumarin derivatives (7-amino -4-methyl coumarin) AMC was successfully synthesized by multi-step reaction starting by the masking of amino group in *m*-amino phenol by the acylation using methoxycarbonyl chloride which formed *m*-(N-methoxy carbonyl amino) phenol (I) in excellent yield >90%. 7-(N-methoxy carbonyl-amino)-4-methyl coumarin (II) was formed by the condensation with acetoacetic ester in sulfuric acid. The methoxycarbonyl was easily hydrolyzed using strong basic condition to obtain 7-amino-4-methyl coumarin as shown in Scheme 1.

The preparation of the new fluorescent disperse dyes 1-3 based on pyrrolinone ester derivatives (PES) as a hybrid structure with coumarin

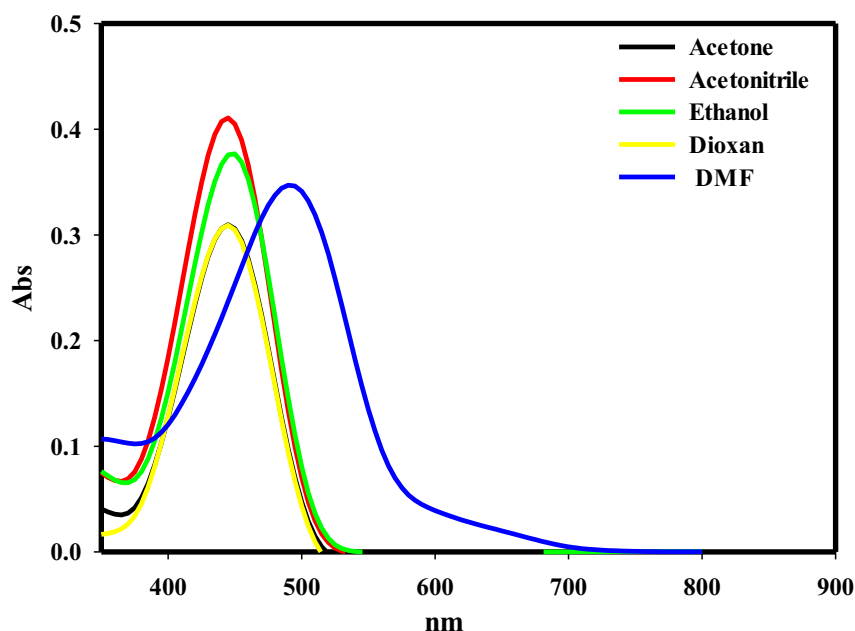


Fig. 4. Effect of solvent polarity in absorption spectra for Dye 1.

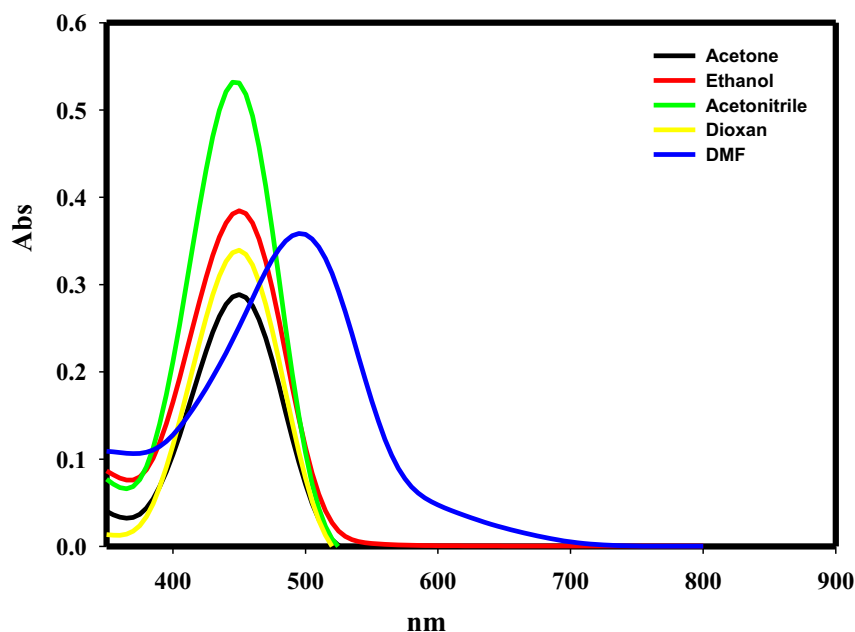


Fig. 5. Effect of solvent polarity in absorption spectra for Dye 2.

derivatives was described on Scheme 2. The synthesis of the dyes 1–3 was basically depend on azo-coupling on the active methylene group in PES. The dyes formed was fully characterized which showed that, the dye structure was formed in pure hydrazone form as known of such pyrrolinone ester based hydrazone dyes [13, 14, 16, 19].

The HNMR data which confirmed the prepared dye structures was showing a significant *E, Z* isomers mixture in the tested samples due to the presence of the exocyclic C=N bond and this results was clear from the appearing of two sets of protons signal of aliphatic terminal ester group –COOR as well as the –NHCO and NH hydrazone. The HNMR signals for the *Z* isomer appear at higher δ values and it constitutes in more than 80% of the isomer mixture in all dye derivatives which agree with our previously reported results for such hydrazone based pyrrolinone ester [13, 14, 19].

3.2. Spectral properties of the prepared dyes 1–3

3.2.1. UV/vis absorption spectra

A dilute solution of the prepared dyes 1–3 (1×10^{-5} M) in acetonitrile was prepared and absorption spectra was recorded at room temperature. the absorption spectra of all dyes was showing a significant HOMO-

LUMO sharp absorption band without any vibronic band in the range between 447 to 454 nm as shown in Figs. 1, 2, and 3. The terminal aryl group in position two shows a minor effect of the absorption spectra as summarized in Table 1, Dye 3 showed higher bathochromic absorption spectra comparing with dye 1 by 7 nm due to the effect of the thienyl group as expected. The molar absorptivity coefficient was calculated and showing good color strength of the prepared dyes which was higher than $30000 \text{ L mol}^{-1} \cdot \text{cm}^{-1}$.

Solvatochromic effect on the absorption spectra was studied. Different solvent with different dielectric constant such as dioxan, acetone, acetonitrile, ethanol and *N, N*-diethylformamide (DMF) was used as protic and non-protic solvent. The effect of solvent polarity on the absorption spectra using dioxan, acetone, acetonitrile, ethanol not showing any significant change in absorption spectra while in case of DMF a significant bathochromic shift (60 nm) comparing with other solvents in absorption spectra was observed. This bathochromic shift is not mainly associated with the high dielectric constant of DMF; both of DMF and acetonitrile have very close values. Meanwhile the bathochromic shift of dye absorbance in DMF may be attributed to the hydrogen bonding formation and also to the alkalinity character of DMF [26] (see Figs. 4 and 5).

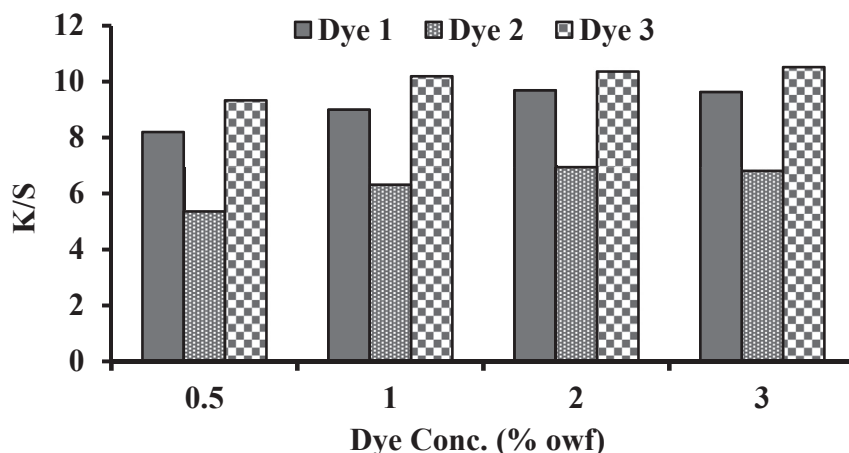


Fig. 6. Effect of dye concentration on dye uptake for dyes 1–3 (dyeing temperature 130°C , dyeing time 30 min, and pH 4.5).

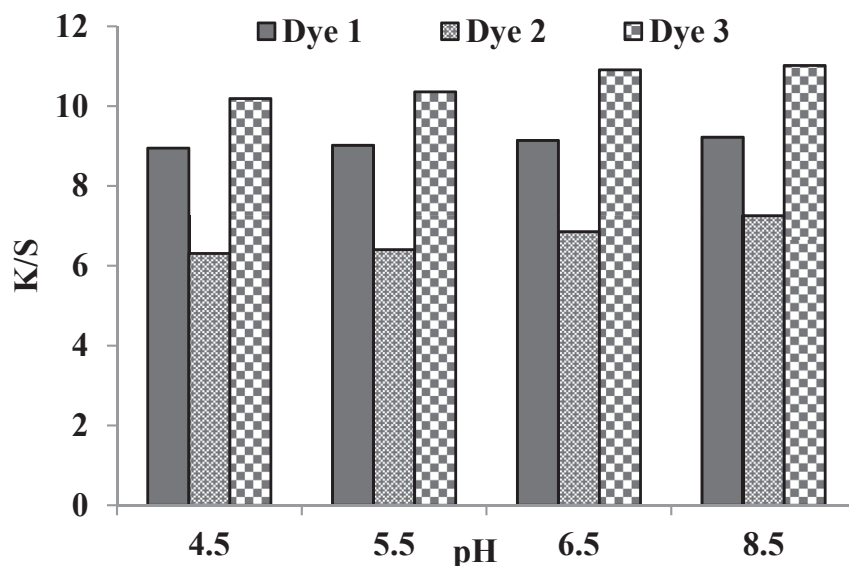


Fig. 7. Effect of dyeing pH on the dye uptake (dye concentration 1% owf, dyeing temperature 130 °C, and dyeing time 30 min).

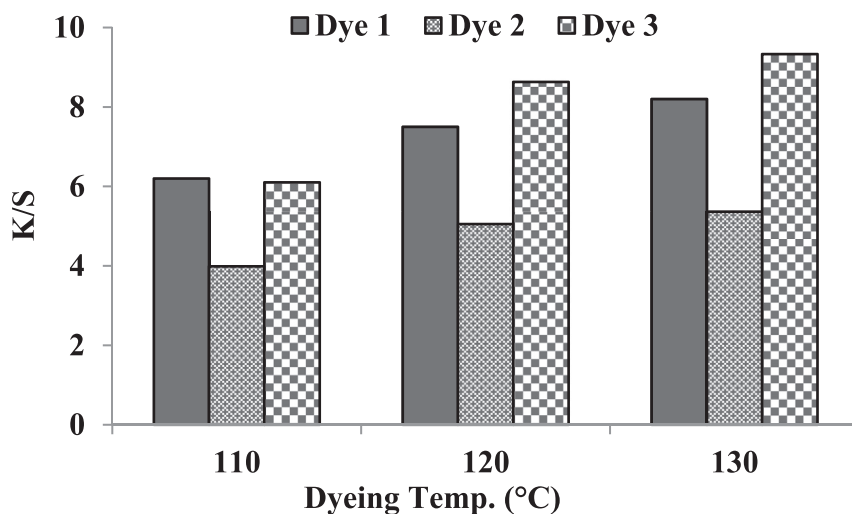


Fig. 8. Effect of dyeing temperature on the dye uptake (dye concentration 0.5% owf, dyeing time 30 min, and pH 4.5).

3.2.2. Fluorescence spectra

The emission spectra of the prepared dyes 1–3 was investigated in different environment at room temperature such as in solution, in polycrystalline form and on the surface of the dyed polyester fabrics. The emission maxima were summarized in Table 1 which showing only one sharp emission band for dyes 1, 2 at 546 and 548 respectively a shoulder vibronic band was observed for both dye 1 and 2 at 514 and 519 respectively, while in case of dye 3 a significant vibronic emission band appeared at 546 and 555 nm. The emission spectra for the prepared dyes on the surface of the dyed fabric and in powder form was showing a significant bathochromic shift in the range between 6–10 nm for the dyed fabrics and 26–37 nm in the case of powder as expected which ascribed to the rigid crystals and the inter/intra hydrogen bond as shown in Figs. 1, 2, and 3 [14, 19].

3.3. Dye application

The K/S values of the dyed polyester fabrics with different concentrations of dyes 1–3 (0.5–3% owf) were measured, and the results are given in Fig. 6. The obtained results showed that an increase of the dye concentration on dyeing polyester resulted in better K/S values. The

maximum K/S values of 9.63, 6.81 and 10.51 were obtained for dyes 1, 2 and 3, respectively at a high concentration of 3% owf. However, these values were not secured a significant increase compared to those obtained at low dye concentration of 0.5 % owf, possibly due to their high tinctorial strength with good dyeing properties.

For all dyes studied, the variation in dyeing pH does not provide a significant impact on dyeing behavior and each dye secured approximately the same K/S, over the range of acidic, neutral and alkaline pH, as shown in Fig. 7. Therefore, these dyes are likely to be considered as alkali stable disperse dyes. Also, it is observed from Fig. 8 that the K/S values of the polyester samples dyed with dyes 1–3 increase with increasing the dyeing temperature.

We can conclude that Br-phenyl based dye 2 exhibited lowest dye uptake values, in particular, compared to dye 1 as well as dye 3 due to the effect of the terminal Br atom. Moreover, dye 3 exhibited relatively high colour yield possibly due to the fact that its pyrrolinone coupling component contains electron rich heterocyclic thiophene substituent. This is in agreement with the results obtained by several authors [27, 28].

Table 2

Fastness properties of the dyed polyester fabrics using fluorescent disperse dyes 1–3.

Dye No	Rubbing fastness		Washing fastness*			Perspiration fastness*						Dry heat Fastness*				Light
						Acidic			Alkaline			180 °C		210 °C		
	Dry	Wet	Alt	SC	SP	Alt	SC	SP	Alt	SC	SP	SC	SP	SC	SP	
1	4-5	4-5	4-5	4-5	4-5	4-5	4-5	4-5	4-5	4-5	4-5	4	4	4	4	4
2	4-5	4-5	4-5	4-5	4	4-5	4-5	4	4-5	4-5	4	4-5	4	4-5	4	4-5
3	4-5	4-5	4	4-5	4-5	4-5	4-5	4-5	4-5	4-5	4-5	4-5	4	4-5	4	4-5

* Alt = alteration of color, SC = Staining on cotton, SP = Staining on polyester.

3.4. Fastness properties

The color fastness of the dyed polyester fabrics using dyes 1–3 are outlined in Table 2. All samples showed excellent fastness ratings to washing, rubbing perspiration and dry heat fastness. This may be due to the absence of anionic and ionizable groups such as OH, NH₂, COOH, etc. in the dye structure, which may affect the dye fastness to washing and perspiration in addition to the suitability of dye molecular, exhibiting very good fastness to dry heat. Light fastness of all dyes displayed approximately similar values with very good ratings because of the same pyrrolinone ester chromophoric system with no effective auxochromic substitutions.

4. Conclusion

A new hybrid of pure hydrazone fluorescent disperse dyes was prepared. The dyes are based on different derivatives of pyrrolinone esters bearing a coumarin moiety and fully characterized as mixture of *E/Z* isomer while the *Z* isomer was the most abundant isomer in the mixture. No significant solvatochromic effect was observed for the prepared dyes. These dyes displayed a significant fluorescence in the solid state. The dyeing application on polyester fabrics showed a high affinity to the fiber with bright colors due to its emission on the dyed fabrics with high color fastness properties.

Declarations

Author contribution statement

Tarek Aysha: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Mostafa Zain: Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Mohamed Arief: Conceived and designed the experiments; Wrote the paper.

Yehya Youssef: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

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Competing interest statement

The authors declare no conflict of interest.

Additional information

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