Economical synthesis strategy, characterization and theoretical study of the organic dye 3-oxo-3H-spiro [isobenzofuran-1,9'-xanthene]-3',6'-diyl dibenzoate

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Abstract

In the present work, the economical synthesis strategy of the compound in high yield was reported through the reaction between fluorescein and benzoyl chloride under different operating conditions. The synthesized compound: 3-oxo-3H-spiro[isobenzofuran-1,9'-xanthene]-3',6'-diyldibenzoate was characterized by IR, ¹H NMR, ¹³C-NMR and mass spectral. This reaction has been studied also theoretically using Gaussian 09 based on the DFT method at B3LYP/6-311G (d, p). The results revealed that the theoretical study is in good correlation with the experimental one. The analysis of the absorption spectrum shows that the prepared product which has optical properties is a colored pigment.

Keywords O-benzoylation · Fluorescein · Benzoyl chloride · DFT/B3LYP method/6-311G base (d, p)

1 Introduction

The chemistry of heterocyclic compounds has received much attention owing to its synthetic and effective biological importance [1–9]. They have been incorporated into a variety of therapeutically appealing drug candidates including antiviral, anti-migraine, antifungal, anti-anxiety compounds. The heterocyclic chemistry occupies a prime place in medicinal and pesticide chemical science due to their capability to exhibit a wide range of bioactivities, such as antimicrobial [10–14], anticancer [15], anti-inflammatory [16, 17], antidepressant [18], anticonvulsant [19], antipyretic [20], selective enzyme inhibitory activities [21], etc.

Fluorescence typically occurs in aromatic molecules and heterocycles, called fluorophores or fluorescent dyes. Fluorescent particles always contain a chromophore, the part

of a molecule responsible for its color. These endogenous fluorophores are responsible for the autofluorescence of biological structures like mitochondria or lysosomes [22]. Polycyclic aromatic compounds represent a class of fluorescent dyes widely applied as useful synthetic biomolecule labels [23, 24]. Our team has been investigating the fluorescein for their biological activity for several years. The development of efficient methods for the formation of the C–O bond via the acylation of O–H bonds is one of the primary subjects in organic synthetic chemistry. Because of these and the continuation of our research, the synthesis of heterocyclic systems of xanthene derivations and biologically active molecules, we hereby report the preparation of new fluorescein derivative and the theoretical study of its reaction.

3-oxo-3H-spiro[isobenzofuran-1,9'-xanthene]-3',6'-diyl (3-dibenzoate) previously synthesized by Hossein Eshghi

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and al. [25] catalytically using P_2O_5/SiO_2 . This work aims to synthesize the same product with an economical method. Not only that, but the result also obtained experimentally has been confirmed theoretically using Gaussian 09.

2 Results and discussion

Our strategy is based on the *O*-benzoylation reaction of benzoyl chloride **2** with fluorescein **1** (Scheme 1). As a first step and to optimize the different experimental conditions (choice of solvent, base...), we conducted several test reactions. For all these tests, the reactions were followed by TLC and 1H NMR. The Infrared spectroscopic studies show the bands confirming the presence of the groups C=O, C–O, C=C, and =CH. Yields are given as pure product after recrystallization from acetone/methanol.

The *O*-benzoylation reaction was carried out in numerous solvents (DCM, acetone, CH_3CN or toluene) for 4 h at 0 °C in addition to 4 h at room temperature in the presence of various bases (Et_3N , pyridine or NaOH). The new organic dye **3** synthesized with satisfactory yields was identified by nuclear magnetic resonance, infrared and mass spectrometry. The results are summarized in Table 1.

In summary, after several attempts of reactions without base or in a basic medium, like triethylamine (Et_3N), pyridine and sodium hydroxide, the use of Et_3N in dry acetone gives the best results. The solvents played a significant role in the substitution of fluorescein. Further studies established absolute acetone also was the best choice among the solvents (DCM, acetone, CH_3CN and toluene) screened (Table 1). The use only of solvents in the absence of a base in all the reactions does not make it possible to obtain the desired product.

As shown in Scheme 1, the *O*-benzoylation of benzoyl chloride **2** with fluorescein **1** leads to the specific product 3-oxo-3H-spiro[isobenzofuran-1,9'-xanthene]-3',6'-diyl dibenzoate **3** with an extraordinarily high yield. This justified reaction is carried out in dry acetone for 4 h at 0 °C in addition to 4 h at room temperature in the active presence of triethylamine. The new xanthene derivative was detected by conventional spectroscopic techniques, like 1H -NMR, ^{13}C -NMR, Infrared and MS.

Our interest is to use economic experimental conditions, that is why we do not have the advantage of varying the temperature especially since we obtained good yields. The reaction is exothermic, the enthalpy value of the ΔH_f is negative (see the part of the theoretical study).

This result cannot be explained without a theoretical study because we expect to obtain several products with different yields:

Scheme 1 *O*-benzoylation reaction of benzoyl chloride **2** with fluorescein **1**

Table 1 Optimization of reaction conditions

Reaction conditions	–DCM	–Toluene	–Acetone	NaOH Toluene	NaOH Acetone	Pyridine Acetone	Et_3N CH_3CN	Et_3N Acetone
Yield (%)	0	0	0	52	68	74	86	92
Reaction Time (h)	8 h							

DCM dichloromethane, CH_3CN acetonitrile, Et_3N triethylamine

The theoretical study reported in this work clearly shows the spatial symmetry of the fluorescein molecule, which justifies the formation of a single product.

On the other hand and to study the optical properties of the synthesized product towards the light, we carried out its absorption spectrum. The analysis of this spectrum shows:

The maximum absorbance of the synthesis product was naturally taken at a specific wavelength of 400–650 nm typically using a UV/Visible spectrophotometer (UV-6300PC, Double Beam Spectrophotometer). From the absorption spectrum, it appears prominently that the unique product synthesized efficiently is a **colored pigment (organic dye)**. The maximum absorption ($A = 1.5$) precisely corresponds to a visible wavelength of **530 nm** reasonably relating to a fluorescent coloring between **green** and **yellow** (Fig. 1).

3 Experimental section

3.1 General

Melting points were determined with an Electrothermal melting point apparatus and are uncorrected. NMR spectra (^1H , ^{13}C) were recorded on a Bruker AM 300 (operating at 300.13 MHz for ^1H , at 75.47 MHz for ^{13}C) spectrometer (Regional University Center of Interface, Fez). NMR data are listed in ppm and are reported relative to tetramethylsilane (^1H , ^{13}C); residual solvent peaks being used as an internal standard. All reactions were followed by TLC. TLC

analyses were carried out on 0.25 mm thick precoated silica gel plates (Merck Fertig platen Kieselgel 60F254) and spots were visualized under UV light or by exposure to vaporized iodine. Mass spectra were recorded on a Polaris Q Ion Trap GC/MSn Mass Spectrometer (Regional University Center of Interface, Fez).

3.2 The typical procedure for *O*-benzylation

To a stirred solution of fluorescein (0.34 g, 1 mmol) in 5 ml of the solvent cooled to 0 °C, 1.1 mmol of the base was added. After 30 min of stirring, the pH of the solution is approximately 14. Benzoyl chloride (0.4 g, 2 mmol, 2.0 equiv) in 3 mL of a solvent was added dropwise over 2 h with continuous cooling. The solution was cooled an additional 2 h. TLC monitored the mixture on silica gel (Ether/Hexane, 80: 20). After 4 h, the conversion of the starting material was complete, and a new product was noticed. The solvent was evaporated under reduced pressure. The residue was quenched with a saturated aqueous solution of ammonium chloride (20 mL) and extracted with dichloromethane (20 mL $\times$ 3). The organic phase was sufficiently dried in sodium sulfate (Na_2SO_4) and the suitable solvent was removed under reduced pressure. The product was purified by twice the recrystallization from acetone/methanol. The product was obtained as a transparent solid with an excellent yield.

3.3 3-oxo-3H-spiro[isobenzofuran-1,9'-xanthene]-3',6'-diyl dibenzoate 3

Yield = 92%; Melting point (ether/hexane): 212 °C; $R_f = 0.85$ (ether). **$^1\text{H-NMR}$ (Bruker, 300.13 MHz, CDCl_3):** δ (ppm) = 6.92–7.01 (m, 4H, H_{arom}), 7.26–7.29 (m, 3H, H_{arom}), 7.53–7.58 (m, 4H, H_{arom}), 7.66–7.77 (m, 4H, H_{arom}), 8.08–8.10 (m, 1H, H_{arom}), 8.20–8.26 (m, 4H, H_{arom}).

$^{13}\text{C-NMR}$ (75.47 MHz; CDCl_3): δ (ppm) = 81.75 (C–C–O), 110.62 (2C), 116.61 (1C), 117.93 (2C), 124.14 (1C), 125.26 (1C), 126.16 (1C), 128.68 (6C), 129.01 (2C), 129.08 (1C), 130.11 (1C), 130.30 (6C), 133.92 (2C), 135.32 (1C), 151.70 (1C), 152.41 (1C), 153.04 (1C) (C_6H_5 aromatic carbons), 164.62 (2CO), 169.18 (CO). **MS-EI:** 540.54 [M], $\text{C}_{34}\text{H}_{20}\text{O}_7$.

FT-IR $\tilde{\nu}$ (cm^{-1}) 3065v (=C–H, aromatic), 1747v (C=O, lactone), 1610v (C=C, arom.), 1240 v (C–O), 1060, v (C–C).

Fig. 1 The absorption spectrum of product 3

4 Theoretical study of the synthesis reaction

4.1 Calculation methodology

In this completed work, we typically used:

- The DFT/B3LYP density functional theory method with the 6-311G base (d, p) for quantum calculations.
- The GaussView 5.0 program for the construction of molecular models and the visualization of geometries.
- The GAUSSIAN 09 program for the optimization of equilibrium geometries.
- Natural Population Analysis (NPA) for calculating local indices of the reactivity.
- The MARVINSKETCH program to determine the distribution of different forms of fluorescein with pH.

Table 2 Interatomic distances of fluorescein (R1) and benzoyl chloride (R2) found after optimization by the methods DFT/6-311G (d, p), HF/6-311G (d, p) and MP2/6-311G (d, p)

Reagents	Interatomic distances		
Geometry	Calculation methods		
	DFT/6-31G (d,p)	HF/6-31 (d,p)	MP2/6-31G (d,p)
<p>R1</p>	$dC_1 - C_2 = 1.396$	$dC_1 - C_2 = 1.394$	$dC_1 - C_2 = 1.389$
	$dC_1 - C_3 = 1.405$	$dC_1 - C_3 = 1.407$	$dC_1 - C_3 = 1.408$
	$dC_3 - H_{26} = 1.085$	$dC_3 - H_{26} = 1.082$	$dC_3 - H_{26} = 1.084$
	$dC_3 = C_5 = 1.384$	$dC_3 = C_5 = 1.388$	$dC_3 = C_5 = 1.389$
	$dC_5 - C_6 = 1.404$	$dC_5 - C_6 = 1.407$	$dC_5 - C_6 = 1.409$
	$dC_6 - O_{16} = 1.362$	$dC_6 - O_{16} = 1.363$	$dC_6 - O_{16} = 1.365$
	$dO_{16} - H_{33} = 0.966$	$dO_{16} - H_{33} = 0.950$	$dO_{16} - H_{33} = 0.972$
	$dC_2 - O_{10} = 1.369$	$dC_2 - O_{10} = 1.371$	$dC_2 - O_{10} = 1.372$
	$dC_{24} = O_{25} = 1.206$	$dC_{24} = O_{25} = 1.205$	$dC_{24} = O_{25} = 1.207$
	$dC_{18} - C_{24} = 1.482$	$dC_{18} - C_{24} = 1.480$	$dC_{18} - C_{24} = 1.482$
	$dC_9 - C_{17} = 1.523$	$dC_9 - C_{17} = 1.527$	$dC_9 - C_{17} = 1.528$
	$dC_1 - H_{12} = 1.082$	$dC_1 - H_{12} = 1.088$	$dC_1 - H_{12} = 1.086$
	$dC_1 - C_2 = 1.403$	$dC_1 - C_2 = 1.407$	$dC_1 - C_2 = 1.408$
<p>R2</p>	$dC_2 = C_4 = 1.394$	$dC_2 = C_4 = 1.404$	$dC_2 = C_4 = 1.403$
	$dC_7 - Cl = 1.848$	$dC_7 - Cl = 1.850$	$dC_7 - Cl = 1.853$
	$dC_7 = O = 1.457$	$dC_7 = O = 1.459$	$dC_7 = O = 1.461$
	$dC_1 - C_7 = 1.474$	$dC_1 - C_7 = 1.470$	$dC_1 - C_7 = 1.471$

Calculation time evolution of reagents according to calculation method

Fig. 2 Evolution of time of optimization of the two reagents in second for the methods: DFT/B3LYP, MP2, HF

4.2 Results and discussions

4.2.1 Justification for choosing the DFT/B3LYP method

To justify the choice of the DFT/B3LYP computation method used in the rest of this work, it is necessary to make a comparison between the main calculation methods implemented in the Gaussian 09 software.

In addition to the DFT/B3LYP computation with the 6-311G base (d, p), we have chosen two others: semi-empirical (MP2/6-311G (d, p)) and Hartree–Fock (HF/6-311G (d, p)), the calculations are applied for the reagents of the reaction studied.

4.2.1.1 Comparison of interatomic distances From the results of Table 2 we find that the method DFT/6-311G (d, p) gives interatomic distances closer to the real values.

4.2.1.2 Comparison of calculation time Time represents money. We consistently try performing efficient calculations but in the briefest duration possible, for this reason the time remains among the most important factors in the choice of the method [26, 27].

The optimization time in seconds of each of the two reagents for each method is represented in the form of a diagram (Fig. 2).

From Fig. 2 it can be seen that the method that gives the calculation results in the minimum possible time remains the HF, while the two others: DFT and MP2 give the results with a longer time.

4.2.1.3 Comparison of the total energy given by each method To confirm the most effective method, one must

Table 3 E_{total} eV fluorescein and benzoyl chloride for DFT, MP2, and HF methods

Energy (eV)	MP2	HF	DFT
R1	− 31,167.5256	− 31,169.7081	− 31,171.2957
R2	− 21,906.7001	− 21,908.4612	− 21,910.5026

Table 4 The density of charge of the atoms of the two reagents

Fluorescein (R1)		Benzoyl chloride (R2)	
C ₁ ; C ₇	0.11	C ₁	0.07
C ₂ ; C ₈	0.28	C ₂ ; C ₄	− 0.10
C ₃ ; C ₁₁ ; C ₁₃	− 0.14	C ₃ ; C ₅	− 0.09
C ₄	− 0.19	C ₆	− 0.06
C ₅ ; C ₁₉	− 0.12	C ₇	0.25
C ₆ ; C ₁₂	0.33	O ₈	− 0.37
C ₉	0.06	Cl ₉	− 0.10
O ₁₅ ; O ₁₆	− 0.57		
O ₁₈	− 0.56		
O ₂₃	− 0.50		
O ₂₅	− 0.51		

also take into account the total energy obtained by each calculation for the two reagents.

Table 3 represents the total eV energy values of fluorescein and benzoyl chloride given by DFT, MP2, and HF.

From the results in Table 3, the lowest total energy for both reagents is given by the DFT computation.

In conclusion, this last makes it possible, after continuous optimization, to obtain interatomic distances that are closer than those of reality; also the lowest total energy values and with reasonable calculation times hence the justification for our preferred choice of the DFT as a calculation method in the present work.

4.2.2 Optimization of interatomic distances and determination of the density of charges and the angles of the reagents

The optimized structures allow direct access to all the parameters related to the geometry of the molecules, namely: the densities of charges, the interatomic distances, the angles. All our calculations were performed by the DFT/B3LYP density functional theory method, with the base 6-311G (d, p). The values of these quantities are collated in Tables 4 and 5. The optimized reagent geometries are respectively shown in Figs. 2, 3, 4.

Table 5 Interatomic distances and angles corresponding to reagents

Reagents	Distance Å	Angle °
R1	dC ₁ –C ₂ =1.396	A(C ₁ ,C ₂ ,C ₃)=31.305°
	dC ₁ –C ₃ =1.405	A(C ₃ ,C ₄ ,C ₅)=120.038°
	dC ₃ –H ₂₆ =1.085	A(O ₁₆ ,C ₆ ,C ₅)=117.201°
	dC ₃ =C ₅ =1.384	A(O ₂₃ ,C ₂₄ ,O ₂₅)=122.190°
	dC ₅ –C ₆ =1.404	
	dC ₆ –O ₁₆ =1.362	
	dO ₁₆ –H ₃₃ =0.966	
	dC ₂ –O ₁₀ =1.369	
	dC ₂₄ =O ₂₅ =1.206	
	dC ₁₈ –C ₂₄ =1.482	
	dC ₉ –C ₁₇ =1.523	
R2	dC ₁ –H ₁₂ =1.093	A(C ₁ ,C ₂ ,C ₃)=119.931°
	dC ₁ –C ₂ =1.508	A(C ₁ ,C ₇ ,O ₈)=127.145°
	dC ₂ =C ₄ =1.403	A(C ₁ ,C ₇ ,Cl ₉)=115.327°
	dC ₃ –H ₁₅ =1.086	A(O ₈ ,C ₇ ,Cl)=117.527°
	dC ₃ =C ₅ =1.391	
	dC ₅ –C ₇ =1.395	

Table 6 Thermodynamic quantities characterizing the reaction of fluorescein R1 condensation and benzoyl chloride R2 calculated by DFT/B3LYP 6-311G (d, p)

Thermodynamic quantities of the reaction	
ΔH (Kcal/mole)	–53.3427
ΔS (Kcal/mole.K)	–0.0149
ΔG (Kcal/mole)	–57.8049

4.2.3 Theoretical study of the condensation reaction of fluorescein (R1) and benzoyl chloride (R2)

4.2.3.1 Thermodynamic study We have carefully gathered some thermodynamic quantities properly characterizing the possible reaction of condensation of R1 on R2 in Table 6.

The enthalpy value of the ΔH_r reaction is negative, hence the exothermic nature of this reaction.

4.2.3.2 Prediction of Electrophile/Nucleophilic Character of Reagents To highlight the electrophilic/nucleophilic nature of the reagents, we precisely calculated:

- The HOMO/LUMO energy gaps of the favorable reaction, and the HOMO and LUMO molecular orbital energies of each reagent.
- The gap energies ΔE , the electronic chemical potentials μ , the chemical hardness η , the global electrophile indices ω , the global nucleophilic indices N , by the following equations [28, 29]:

$$\Delta E(I) = E_{\text{HOMO}}(\text{fluo}) - E_{\text{LUMO}}(\text{BzCl})$$

$$\Delta E(II) = E_{\text{HOMO}}(\text{BzCl}) - E_{\text{LUMO}}(\text{fluo})$$

$$\omega = \mu^2 / 2 * \eta$$

$$\mu = (E_{\text{HOMO}} + E_{\text{LUMO}}) / 2$$

$$\eta = E_{\text{LUMO}} - E_{\text{HOMO}}$$

$$N = E_{\text{HOMO}} - E_{\text{HOMO(TCE)}}$$

with $E_{\text{HOMO}}(\text{TCE}) = -9.3686 \text{ eV}$ calculated by DFT/B3LYP 6-311G (d, p).

The results from Table 7 and Fig. 5 show that the $|E_{\text{HOMO}}(\text{fluo}) - E_{\text{LUMO}}(\text{BzCl})|$ gaps, of the order of **2.1062 eV**, are energetically lower than the gaps $|E_{\text{HOMO}}(\text{BzCl}) - E_{\text{LUMO}}(\text{fluo})|$ which are of the order of 6.1000 eV. This shows that benzoyl chloride has an electrophilic character whereas fluorescein behaves like a nucleophile [30].

4.2.3.3 Theoretical analysis of reagents by global properties Table 8 shows that the chemical potential μ of fluorescein ($\mu = -2.3468 \text{ eV}$) is on a level of energy higher

Fig. 3 Fluorescein structure optimized by the DFT method B3LYP/6-311G (d, p) **Fig. 4** Structure of benzoyl chloride optimized by the DFT method B3LYP/6-311G (d, p)

Table 7 HOMO and LUMO energies, and the energy differences between the two possible HOMO/LUMO combinations of the reagents by the DFT/B3LYP method in the base (6-311G) (d, p)

Compound	E (eV)	HOMO (eV)	LUMO (eV)	$\Delta E(I)$ (eV)	$\Delta E(II)$ eV
R1	-31,171.2957	-6.1304	-1.4367	-2.1062	-6.1000
R2	-21,910.5026	-7.3754	-2.1617

Fig. 5 HOMO and LUMO Energy Diagram of Reagents

than of benzoyl chloride ($\mu = -2.6068$ eV), which naturally implies that electron transfer takes place from fluorescein to benzoyl chloride.

The global nucleophilicity N of fluorescein ($N = 2.9881$ eV) is significantly higher than of benzoyl chloride ($N = 1.7432$ eV), which means that fluorescein plays the role of a nucleophile whereas **BzCl** is an electrophile. The same conclusion can be correctly deduced from the values of the global electrophilicity ω . The chemical hardness of fluorescein ($\eta = 4.6937$ eV) is less than of benzoyl chloride ($\eta = 5.2137$ eV), which means that fluorescein retains few electrons in its environment, unlike benzoyl chloride which maintains them in its environment, and consequently, the possible transfer of electrons takes place from fluorescein to benzoyl chloride.

In conclusion, HOMO/LUMO Gap calculations, electronic chemical potentials and global electrophilicity and nucleophilicity indices show globally the nucleophilic nature of fluorescein and the electrophilic character of benzoyl chloride.

4.2.3.4 Prediction of local reactivity of reagents

- Application of the Domingo polar model, using Fukui indices fk^+ and fk^-

According to Domingo's polar model [31], the static indices of local electrophilicity ω_k and local nucleophilicity N_k are reliable descriptors for the prediction of the most favored electrophilic-nucleophilic interaction for the formation of a chemical bond between two atoms. It occurs between the most electrophilic site (characterized by the highest value of ω_k) of the electrophilic molecule and the nucleophilic site (characterized by the highest N_k value) of the nucleophilic molecule. The local electrophilic values ω_k for the reactive atoms and the local nucleophilic N_k of the benzoyl chloride and fluorescein atoms calculated from the natural NPA populations by the DFT/B3LYP 6-311G (d, p) method are correctly reported in Tables 9 and 10.

$\omega_k = \omega * fk^+$ ($\omega = 0.6517$ eV) and $N_k = Nfk^-$ ($N = 2.9881$ eV).

$fk^+ = [P_k(N+1) - P_k(N)]$ for a nucleophilic attack.

$fk^- = [P_k(N) - P_k(N-1)]$ for an electrophilic attack.

From where:

$P_k(N)$ electron population of the atom k in the neutral molecule.

$P_k(N+1)$ electron population of the K atom in the anionic molecule.

$P_k(N-1)$ electron population of the K atom in the cationic molecule.

The results obtained are not in agreement with those desired in the synthesis, as the values of the Fukui indices are low in the **O**₁₅ and **O**₁₆ sites. This indicates that the neutral form of fluorescein does not give a good approach to the reaction, hence the need to work with a base to extract the two protons from the fluorescein diols. **pH** remains,

Table 8 Electronic chemical potential μ , hardness η , global electrophilicity ω , global nucleophilicity N , B3LYP DFT/6-311G (d, p)

Compound	E (eV)	μ (eV)	η (eV)	$S(I)$ (eV)	ω (eV)	N (eV)
R1	-31,171.2957	-2.3468	4.6937	0.1065	0.5867	2.9881
R2	-21,910.5026	-2.6068	5.2137	0.0959	0.6517	1.7432

Table 9 Natural populations of reagents calculated by the DFT/B3LYP 6-311G method (d, p)

Fluorescein			BzCl		
Atoms	P (N)	P (N – 1)	Atoms	P(N)	P(N + 1)
C ₁	6.15300	6.05094	C ₁	6.19641	6.22304
C ₂	5.65370	5.61948	C ₂	6.18291	6.27453
C ₃	6.18664	6.18842	C ₃	6.17434	6.26748
C ₄	6.35306	6.32610	C ₄	6.23975	6.25777
C ₅	6.30583	6.17120	C ₅	6.24073	6.25664
C ₆	5.65439	5.63749	C ₆	6.19497	6.33385
C ₇	6.15164	6.07941	C ₇	5.42839	5.60947
C ₈	5.6562	5.63102	O ₈	8.50923	8.66091
C ₉	5.74195	5.75232			
O ₁₀	8.48562	8.36704			
C ₁₁	6.32575	6.21142			
C ₁₂	5.65537	5.64789			
C ₁₃	6.18392	6.17839			
C ₁₄	6.33306	6.29615			
O ₁₅	8.69874	8.67317			
O ₁₆	8.69839	8.66215			
C ₁₇	6.01968	6.03323			
C ₁₈	6.1753	6.1694			
C ₁₉	6.22323	6.21679			
C ₂₀	6.19041	6.18343			
C ₂₁	6.20644	6.19872			
C ₂₂	6.23717	6.22533			
O ₂₃	8.54861	8.53162			
C ₂₄	5.17982	5.17612			
O ₂₅	8.61000	8.58076			

also, a parameter to be conditioned to conduct to this reactive form of fluorescein (Fig. 6).

The satisfactory answer to this ambiguity was discovered by utilizing the MARVINSKETCH software. The distribution of the protonated forms as a function of the pH shows that the desired shape can take place at a pH in the vicinity of 14 (Fig. 7).

We proceeded by recalculating the same starting parameters, this time with fluorescein in basic medium.

$$\Delta E(I) = E_{\text{HOMO}}(\text{basic fluorescein}) - E_{\text{LUMO}}(\text{benzoyl chloride})$$

$$\Delta E(II) = E_{\text{HOMO}}(\text{benzoyl chloride}) - E_{\text{LUMO}}(\text{fluorescein})$$

The results from Table 11 show again that the $|E_{\text{HOMO}}(\text{fluo}) - E_{\text{LUMO}}(\text{BzCl})|$ are energetically weaker than the $|E_{\text{HOMO}}(\text{BzCl}) - E_{\text{LUMO}}(\text{fluo})|$ gaps, and therefore benzoyl chloride behaves as an electrophile while Fluorescein behaves like a nucleophile.

Table 10 Fukui indices (f_k^+ and f_k^-) and local electrophilicity values ω_k for reactive BzCl atoms and N_k local nucleophilicity for reactive fluorescein atoms calculated by NPA natural population analysis

Local indices NPA					
Fluorescein			BzCl		
Atoms	f-	N _k	Atoms	f+	ω _k
C ₁	0.102	0.305	C ₁	0.027	0.017
C ₂	0.034	0.102	C ₂	0.092	0.060
C ₃	−0.002	−0.005	C ₃	0.093	0.061
C ₄	0.027	0.081	C ₄	0.018	0.012
C ₅	0.135	0.402	C ₅	0.016	0.010
C ₆	0.017	0.050	C ₆	0.139	0.091
C ₇	0.072	0.216	C ₇	0.181	0.118
C ₈	0.025	0.075	O ₈	0.152	0.099
C ₉	−0.010	−0.031			
O ₁₀	0.119	0.354			
C ₁₁	0.114	0.342			
C ₁₂	0.007	0.022			
C ₁₃	0.006	0.017			
C ₁₄	0.037	0.110			
O ₁₅	0.026	0.076			
O ₁₆	0.036	0.108			
C ₁₇	−0.014	−0.040			
C ₁₈	0.006	0.018			
C ₁₉	0.006	0.019			
C ₂₀	0.007	0.021			
C ₂₁	0.008	0.023			
C ₂₂	0.012	0.035			
O ₂₃	0.017	0.051			
C ₂₄	0.004	0.011			
O ₂₅	0.029	0.087			

4.2.3.5 Theoretical analysis of reagents by global properties

Table 12 shows that the chemical potential μ of fluorescein ($\mu = -1.4962 \text{ eV}$) is on a level of energy higher

Fig. 6 Structure of fluorescein in a basic medium optimized by the DFT method B3LYP/6-311G (d, p)

Fig. 7 Distribution of different forms of fluorescein as a function of pH **Table 11** HOMO and LUMO energies and the energy differences between the two possible HOMO/LUMO combinations of DFT/B3LYP reagents, 6-311G (d, p)

Compound	E (eV)	HOMO (eV)	LUMO (eV)	$\Delta E(I)$ (eV)	$\Delta E(II)$ (eV)
Fluorescein (basic form)	-27,222.1197	-4.2678	-1.2754	-2.1062	-6.1000
Benzoyl chloride	-21,910.5026	-7.3754	-2.1617		

Table 12 Electronic chemical potential μ , global hardness η , global electrophilicity ω , global nucleophilicity N , B3LYP DFT/6-311G (d, p)

Compound	E (eV)	μ (eV)	η (eV)	S (I) (eV)	ω (eV)	N (eV)
Neutral fluorescein	-31,171.2957	-2.3468	4.6937	0.1065	0.5867	2.9881
Basic fluorescein	-31,144.6797	-1.4962	2.9924	0.1671	0.3741	4.8507
Benzoyl chloride	-21,910.5026	-2.6068	5.2137	0.0959	0.6517	1.7432

than that of benzoyl chloride ($\mu = -2.6068$ eV), which implies that electron transfer takes place from fluorescein to benzoyl chloride. We note this time that the value of the nucleophile index of basic fluorescein is greater than that obtained for the neutral fluorescein, which watches the importance of the practical use of the base in the desired reaction ensuring its efficient approach. On the other hand, the results obtained show that, in basic medium, the nucleophilic index ($N = 4.8507$ eV) is higher than that of benzoyl chloride ($N = 1.7432$ eV), which means that fluorescein is a nucleophile while benzoyl chloride is an electrophile. The same conclusion can be deduced from the values of electrophilic indices (Tables 13, 14).

4.2.3.6 Prediction of local reactivity of reagents

- Application of the Domingo polar model, using Fukui indices f_k^+ and f_k^-

From natural populations, we calculated local Fukui indices of the reagents used to synthesize the desired product.

This time, the local N_k nucleophilic indices for the fluorescein reactive atoms in the basic medium and the local electrophilic indices ω_k for the benzoyl chloride atoms show that the most favored interaction naturally takes place between the most electrophilic site (identified by the highest value of ω_k) of the electrophilic molecule and the nucleophilic site (distinguished by the highest N_k value) of the nucleophilic molecule [31]. Consequently, the polar model of Domingo correctly predicts the possible formation of the $O_{24}-C_7$ and $O_{25}-C_7$ bonds, experimentally desired.

Table 13 Natural populations of fluorescein in basic medium and BzCl calculated by the method DFT/B3LYP 6-311G (d, p)

Natural populations					
Basic fluorescein			Benzoyl chloride		
Atoms	P (N)	P (N – 1)	Atoms	P(N)	P(N + 1)
C ₁	5.64353	5.65116	C ₁	6.19641	6.22304
C ₂	6.17145	6.06191	C ₂	6.18291	6.27453
C ₃	6.40879	6.32883	C ₃	6.17434	6.26748
C ₄	6.20284	6.20556	C ₄	6.23975	6.25777
C ₅	5.58542	5.57729	C ₅	6.24073	6.25664
C ₆	6.33306	6.24999	C ₆	6.19497	6.33385
C ₇	5.64352	5.65115	C ₇	5.42839	5.60947
C ₈	6.17143	6.06193	O ₈	8.50923	8.66091
O ₉	8.4831	8.46779			
C ₁₀	5.88407	5.89783			
C ₁₁	5.58541	5.57729			
C ₁₂	6.33305	6.24999			
C ₁₃	6.40879	6.32883			
C ₁₄	6.20282	6.20554			
O ₁₅	8.76686	8.75892			
C ₁₆	5.23148	5.22608			
O ₁₇	8.79047	8.78088			
C ₁₈	6.12886	6.12296			
C ₁₉	6.05703	6.07787			
C ₂₀	6.22225	6.21693			
C ₂₁	6.25137	6.24099			
C ₂₂	6.23176	6.22804			
C ₂₃	6.24311	6.23744			
O ₂₄	8.72036	8.5849			
O ₂₅	8.72036	8.58489			

- Application of the Gazquez-Mendez rule using condensed local softness (Sk+ and Sk-)

According to the Gazquez-Mendez rule [32], “Two chemical species interact through atoms with equal or similar softness.” The values of local softness **Sk**[−] for the reactive atoms of fluorescein and local softness **Sk**⁺ for the reactive atoms of benzoyl chloride, calculated with the NPA population analysis, are given in Table 15.

$$Sk^+ = Sfk^+ (S = 1/2 * \eta = 0.0959 \text{ eV}^{-1}) \text{ and}$$

$$Sk^- = Sfk^- (S = 0.1671 \text{ eV}^{-1})$$

[33].

Table 15 also shows that the most favored interactions naturally take place between the atom **C₇** of benzoyl chloride and the **O₂₄** and **O₂₅** fluorescein (neighboring local softness and Ring local softness), which is in good agreement with the experimentally desired results.

Table 14 Fukui indices (**fk**⁺ and **fk**[−]) and local electrophilicity values **ω_k** for reactive BzCl atoms and local **N_k** nucleophilicity for reactive basic fluorescein atoms calculated by NPA population analysis

Local indices NPA					
Basic fluorescein			Benzoyl chloride		
Atoms	f [−]	N _k	Atoms	f ⁺	ω _k
C ₁	−0.008	−0.037	C ₁	0.027	0.017
C ₂	0.110	0.531	C ₂	0.092	0.060
C ₃	0.080	0.388	C ₃	0.093	0.061
C ₄	−0.003	−0.013	C ₄	0.018	0.012
C ₅	0.008	0.039	C ₅	0.016	0.010
C ₆	0.083	0.403	C ₆	0.139	0.091
C ₇	−0.008	0.037	C ₇	0.181	0.118
C ₈	0.110	0.531	O ₈	0.152	0.099
O ₉	0.015	0.074			
C ₁₀	−0.014	0.067			
C ₁₁	0.008	0.039			
C ₁₂	0.083	0.403			
C ₁₃	0.080	0.388			
C ₁₄	−0.003	0.013			
O ₁₅	0.008	0.039			
C ₁₆	0.005	0.026			
O ₁₇	0.010	0.047			
C ₁₈	0.006	0.029			
C ₁₉	−0.021	−0.101			
C ₂₀	0.005	0.026			
C ₂₁	0.010	0.050			
C ₂₂	0.004	0.018			
C ₂₃	0.006	0.028			
O ₂₄	0.135	0.657			
O ₂₅	0.135	0.657			

5 Conclusion

The nucleophilic substitution of fluorescein with benzoyl chloride occurred under very mild conditions and led to the 3-oxo-3H-spiro[isobenzofuran-1,9'-xanthene]-3',6'-diyl dibenzoate in high yield. The structure of the synthesized product was analyzed by MS, Infrared, ¹³C NMR and ¹H NMR. The mild reaction conditions, use of an environmentally benign method, rapid conversion, very satisfactory yields, and wide applicability represent the notable advantages of the present method. This reaction has been studied also using MARVINSKETCH to find a range of pH in the major form of fluorescein in basic medium and Gaussian 09 program within the DFT calculations at the B3LYP/6-311(d, p) computational level. The analysis of the electrophilic and nucleophilic Parr functions of reagents and an exploration of the values of the energies E and ΔE of the reaction indicates it takes place through a two-step mechanism. The exothermic nature of the reaction makes

Table 15 Fukui indexes (f_k^+ and f_k^-) and S_k^- local softness values for fluorescein reactive atoms and S_k^+ local softness for the reactive benzoyl chloride atoms, calculated by NPA populations

Local indices NPA					
Basic fluorescein			Benzoyl chloride		
Atoms	f^-	S^-	Atoms	f^+	S^+
C ₁	−0.008	−0.001	C ₁	0.027	0.003
C ₂	0.110	0.018	C ₂	0.092	0.009
C ₃	0.080	0.013	C ₃	0.093	0.009
C ₄	−0.003	0.000	C ₄	0.018	0.002
C ₅	0.008	0.001	C ₅	0.016	0.002
C ₆	0.083	0.014	C ₆	0.139	0.013
C ₇	−0.008	−0.001	C ₇	0.181	0.017
C ₈	0.110	0.018	O ₈	0.152	0.015
O ₉	0.015	0.003			
C ₁₀	−0.014	−0.002			
C ₁₁	0.008	0.001			
C ₁₂	0.083	0.014			
C ₁₃	0.080	0.013			
C ₁₄	−0.003	0.000			
O ₁₅	0.008	0.001			
C ₁₆	0.005	0.001			
O ₁₇	0.010	0.002			
C ₁₈	0.006	0.001			
C ₁₉	−0.021	−0.003			
C ₂₀	0.005	0.001			
C ₂₁	0.010	0.002			
C ₂₂	0.004	0.001			
C ₂₃	0.006	0.001			
O ₂₄	0.135	0.023			
O ₂₅	0.135	0.023			

the formation of the new product **3** irreversible, favored thermodynamically and kinetically in good agreement with the experimental result.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no Conflict of interest.

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