

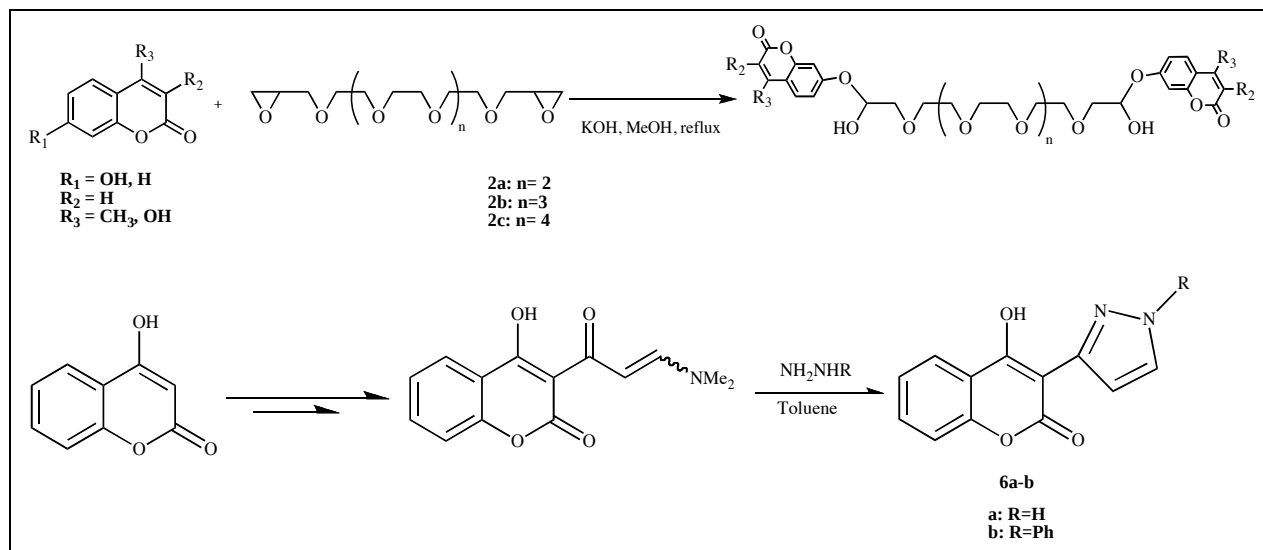
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The reactions between oligoethylene glycol diglycidyl ethers **2a-c** with both 7-hydroxy-4-methyl-2H-chromen-2-one and 4-hydroxy-2H-chromen-2-one lead to new hydroxy ethers **3** and **4** containing coumarin moieties in good yield. The synthesis of 3-(3-(dimethylamino)acryloyl)-4-hydroxy-2H-chromen-2-one **5** and new heterocyclic compounds 4-hydroxy-3-(1H-pyrazol-3-yl)-2H-chromen-2-one **6a**, 4-hydroxy-3-(1-phenylpyrazol-3-yl)-2H-chromen-2-one **6b** and 4-hydroxy-3-(isoxazol-3-yl)-2H-chromen-2-one **6c** is also described. All compounds were characterized by ¹H NMR, ¹³C{¹H} NMR, 2D-¹H-¹³C HMBC, 2D-¹H NOESY NMR, IR, and MS spectroscopy. Additionally, the antibacterial activity of the new products containing coumarin moiety was evaluated. This activity is clearly dependent on the chemical structure of compounds.

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INTRODUCTION

The 4-hydroxycoumarin-2-one derivatives represent, nowadays, an important group of organic compounds, which exhibit a broad biological activity spectrum [1–6] such as antibiotics [7,8], fungicides [9], anti-inflammatory [10], anticoagulant [11], and antitumor agents [12,13]. Additionally, these kinds of compounds are also extensively used as analytical reagents [14] and plant growth regulating modulators. Surprisingly their 7-hydroxy-2H-chromen-2-one analogues are less known [15].

The oligoethylene glycol diglycidyl ethers, which are prepared from epichlorohydrin and oligoethylene glycols, constitute a family of interesting compounds. They are used as cross-linking agents in polymer chemistry [16] and can be used as building blocks for synthesis of crown ether derivatives. In a recent patent [17], Ovchinnikov and Chernovianov reported the preparation of benzocrown

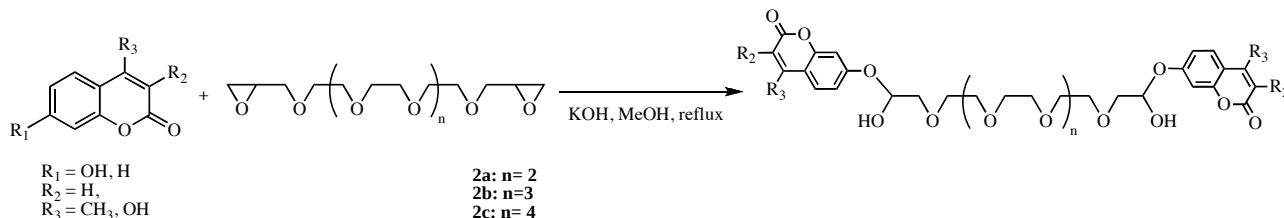
ether derivatives by the reaction of oligoethylene glycol diglycidyl ethers with catechol. T. Kikui *et al.* have also previously reported that a family of monoazacrown ethers containing two hydroxy groups can be obtained by reaction of glycidyl ethers with primary amines and ammonia in water and methanol [18]. It is important to emphasize that reactive crown ethers are potentially important key intermediate compounds for highly functionalized derivatives such as crown polymers [19,20], bis(crown-ethers) [21,22], lariat ethers [23–25], and synthetic ionophores [26, 27]. Nakatsuji *et al.* described the synthesis of novel crown ethers having two bromomethyl groups which showed that reactive groups in the crown ring provide new reactive macrocycles with new properties and therefore, they have new possible applications [28,29].

As a continuation of previous works in this area [30], we report herein the reactions between oligoethylene glycol diglycidyl ethers **2a-c** with both 7-hydroxy-4-

methyl-2*H*-chromen-2-one and 4-hydroxy-2*H*-chromen-2-one (Scheme 1). Additionally, we describe a simple and easy route to synthesize new heterocyclic compounds from 4-hydroxy-2*H*-chromen-2-one and their potential antibacterial activity have been investigated.

The proposed structure for the new 7,7'-(2,27-dihydroxy-4,7,10,13,16,19,22,25-octaaoctacosane-1,28-diyl)bis(oxy)]bis(4-methyl-2*H*-chromen-2-one) **3b** was determined by IR, ^1H , ^{13}C NMR, 2D- ^1H - ^{13}C HMBC, and 2D- ^1H NOESY spectroscopy. Its IR spectrum contain

Scheme 1

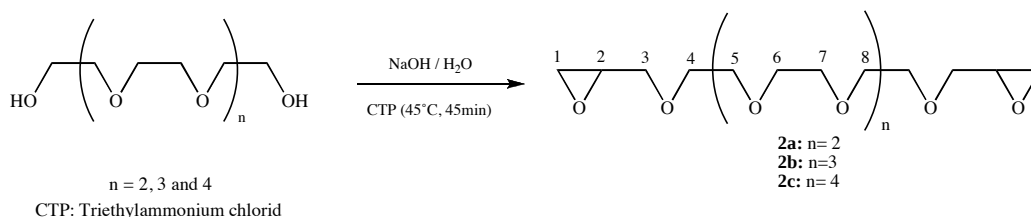


RESULTS AND DISCUSSION

Oligoethylene glycol diglycidyl ethers similar to **2a-c** have shown interesting properties in chemistry of polymers and are not widely known [31], particularly their ring opening reaction with coumarinic derivatives has not been studied. Oligoethylene glycol diglycidyl ethers **2a-c** were prepared in good yield (70–77%) by condensation of the proper oligoethylene glycol with epichlorohydrin in presence of sodium hydroxide using a catalyst for the phase transfer (Scheme 2).

bands at 1632 and 1712 cm^{-1} which are characteristic of the stretching vibrations of the coumarinic and ester $\text{C}=\text{O}$ bonds, while a broad OH stretching band comes at 3436 cm^{-1} . Its ^1H NMR spectrum is constituted by a reduced number of signals which are consistent with its symmetrical structure. The aromatic protons arose as a multiplet between 6.83 and 7.50 ppm. A singlet at 2.4 ppm was assigned to H_{12} protons, while the OH signal appeared at 2.88 ppm, which disappeared as the H was exchanged by D using D_2O . The ^{13}C $\{^1\text{H}\}$ NMR spectrum showed the signals for all expected carbon atoms (Experimental), being

Scheme 2



We attempted to establish the structural assignments of oligoethylene glycol diglycidyl ethers **2a-c** using ^1H , ^{13}C NMR, and MS spectra. The proposed molecular weight for compound **2a** was supported by mass spectroscopy. The ^1H NMR spectrum for compound **2a** is constituted by a signal related to H-1 at 2.56 that for H-2 at 2.74 and H-3 signal at 3.11 ppm. The NMR spectra signals for **2b** and **2c** are in agreement with their proposed structure.

Reaction in basic media of 2 equivalents of 7-hydroxy-4-methyl-2*H*-chromen-2-one with oligoethylene glycol diglycidyl ether **2b** afforded the expected coumarin hydroxy ether **3b** after 20 h in refluxing methanol (Scheme 3). The reaction was monitored by TLC that showed the exhaustion of 7-hydroxy-4-methyl-2*H*-chromen-2-one and the oligoethylene glycol diglycidyl ether **2b**.

the most significant signals, those for carbonyl C-2 (161.4), C-4 (155.0), C-12 (68.8), and C-13 (59.1 ppm). It is important to point out that the carbon signals were only possible to be assigned by its 2D- ^1H - ^{13}C HMBC spectra (Figure 1). The most important features are the clear correlation among the H-13 proton with carbon atoms C-12, C-14, and C-7. An important support for the proposed molecule assignment is the clear correlation among aromatic protons H-5, H-6, and H-8 and carbon C-7, which is bonded to the oxygen atom; consequently the C-13 is linked to C-12 and this last atom to C-7.

The proposed structure for **3b** was also in agreement with its 2D- ^1H NOESY spectrum. A clear cross peak between H-12 and H-14 and the hydroxylic proton suggests that these three H atoms are placed together on the same side of the average hydroxyl ether plane (Table 1).

Scheme 3

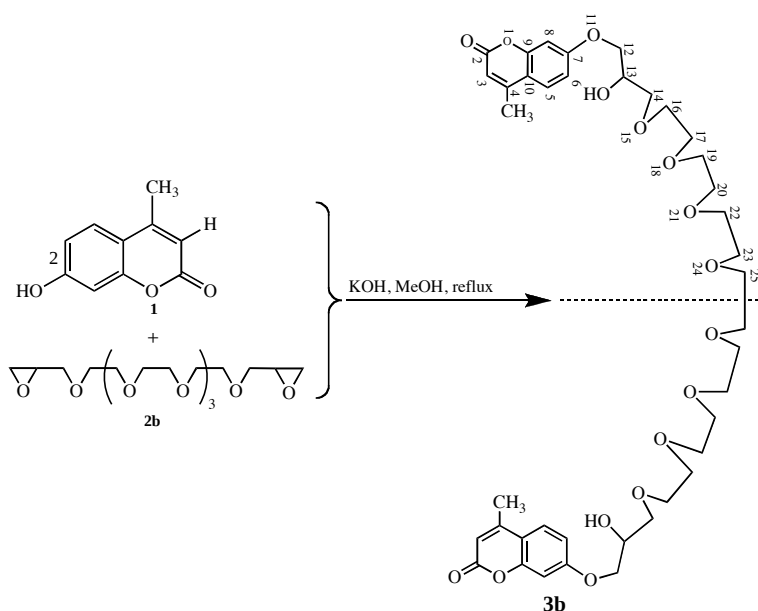


Table 1

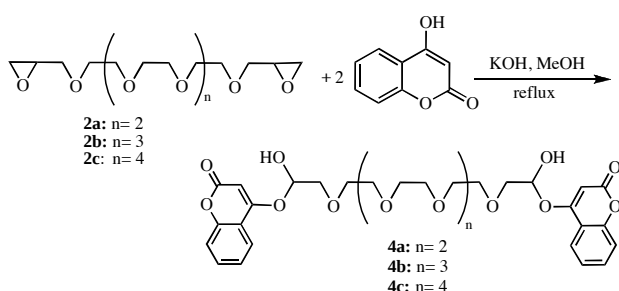
Correlations between HMBC and NOESY for compound **3b**.

Proton H-n	HMBC H-n-C-j	NOESY H-n-H-j
H-5	6, 7, 8	
H-6	6, 7, 8	
H-8	6, 7, 8	
H-13	12, 14	
H-12		OH
H-14		OH

Using similar conditions **3a** and **3c** were synthesized by reaction of 7-hydroxy-4-methyl-2H-chromen-2-one with oligoethylene glycol diglycidyl ethers **2a** and **2c**. Postulated structures for new coumarin hydroxy ethers **3a** and **3c** are in agreement with their IR, ^1H NMR, $^{13}\text{C}\{^1\text{H}\}$ NMR, IR, $2\text{D-}^1\text{H-}^{13}\text{C}$ HMBC, and $2\text{D-}^1\text{H}$ NOESY spectra.

4-Hydroxy-2H-chromen-2-one was treated with oligoethylene glycol diglycidyl ethers **2a-c** to give new coumarin hydroxy ethers **4a-c** in good yields by the same procedure that for obtaining compounds **3a-c** (Scheme 4).

Scheme 4

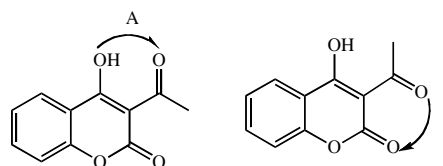


It should be noted that this new procedure to obtain coumarin hydroxy ethers constitutes a simple and convenient synthetic route to synthesize these kinds of compounds.

The 4-hydroxy-2H-chromen-2-one, which is substituted in the third position, is an excellent starting compound for an effective synthetic route for producing new heterocycles containing the coumarin moiety. These heterocyclic compounds are present in many synthetic and natural products such as drugs and pesticides [32,33]. These important applications have generated a considerable interest in this ring system and various 2,3- and 3,4-fused polycycles as well as open chain derivatives have been synthesized [34].

3-Acyl-4-hydroxy-2H-chromen-2-one can be a very good precursor for preparing fused ring systems through cyclization procedure **A** and **B** (Scheme 5).

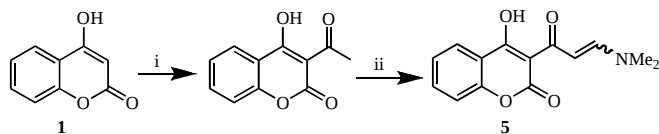
Scheme 5



These two possible cyclization pathways motivated us to attempt a convenient alternative preparation for the enaminone 3-(3-dimethylaminoacryloyl)-4-hydroxy-2H-chromen-2-one (**5**) that is expected to be more reactive than the 3-acyl-4-hydroxy-2H-chromen-2-one. The reaction of 4-hydroxy-2H-chromen-2-one with phos-

phorus oxychloride in the presence of acetic acid provided 3-acetyl-4-hydroxy-2*H*-chromen-2-one in high yield [35, 36], which using DMF–DMA in boiling toluene give 3-(3-(dimethylaminoacryloyl)-4-hydroxy-2*H*-chromen-2-one (5) (Scheme 6).

Scheme 6

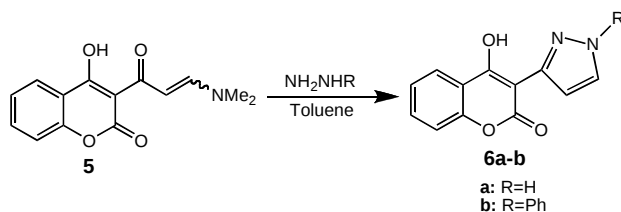


i: AcOH, POCl₃, 30 min, reflux; ii: DMF–DMA, toluene, reflux, 2 h

It should be noted that the dimethylamino group in enaminone 5 acts as a good leaving group by the reaction with binucleophilic compounds. The structure for 3-(3-(dimethylamino)acryloyl)-4-hydroxy-2*H*-chromen-2-one was assigned as the *E*-form by ¹H NMR spectrum. One of the main important findings was that the olefin proton coupling constant arose in the range of 13–16 Hz.

The reaction of 5 with hydrazine hydrate and phenylhydrazine in toluene affords respectively 4-hydroxy-3-(1*H*-pyrazol-3-yl)-2*H*-chromen-2-one (6a) and 4-hydroxy-3-(1-phenyl-1*H*-pyrazol-3-yl)-2*H*-chromen-2-one (6b) (Scheme 7).

Scheme 7



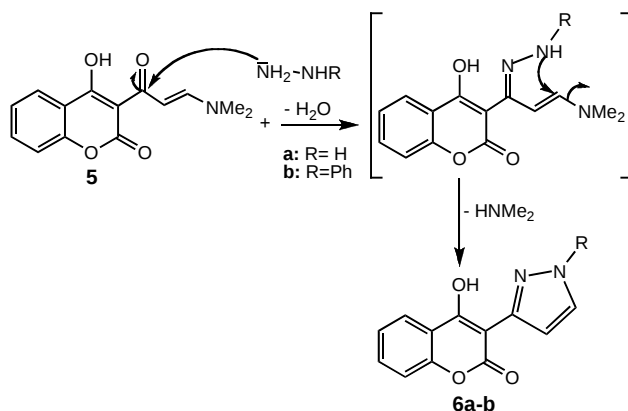
The synthesised compounds 6a–b were characterised by their ¹H, and ¹³C, and IR spectra as well as by elemental analysis.

There were characteristic bands in the IR spectra of compounds 6a for OH at 3331 cm⁻¹, and for C=O at 1702 cm⁻¹. In the ¹H NMR spectrum the signal related to the OH appears at δ13.48 ppm and exchanges with D₂O. The signal for the NH proton appears as broadened singlet at δ 12.51 ppm while the aromatic protons appeared as multiplets between 7.1–8.1 ppm.

In contrast, by refluxing in methanol 5 with hydroxylamine hydrochloride the 5-substituted isoxazole – 4-hydroxy-3-(isoxazol-3-yl)-2*H*-chromen-2-one (6c) was obtained in good yield. It is important to emphasize that it was not necessary to add any base for releasing hydroxylamine from its salt. Probably, the dimethylamine group formed through the reaction is stable enough to

favour this reaction. This evidence suggests that the probable process through these reactions occurred implicates an initial nucleophilic attack on the C=O bond, by the HN of amino group. The next step is a nucleophilic attack by the N-R amino atom on the C=C bond, which is accompanied by the elimination of a dimethylamine group generating the new coumarin derivative (Scheme 8) [37].

Scheme 8



Antibacterial activities. The new coumarin derivatives were screened *in vitro* for antimicrobial activities against Gram-positive bacteria *Staphylococcus aureus* (NCTC-7447), *Bacillus cereus* (ATCC-14579), and Gram-negative salmonella using the paper disk diffusion method for the antibiotic sensitivity technique [38]. The tested compounds were dissolved in DMF to obtain 1 mg/mL solution. The inhibition zones of microbial growth produced by different compounds were measured in millimetres after 48 h of incubation at 28°C. The results are summarized in Table 2.

Table 2

Antibacterial screening for compounds 3a, 4a, 5 and 6a

Compound	Concentration, mg/disk	Inhibition zone, diameter, mm
3a	1	20
	2	28
	4	28
4a	1	9
	2	15
	4	15
5	1	9
	2	12
	4	13
6a	1	13
	2	20
	4	20

The antibacterial activities for representative compounds 3a, 4a, 5, and 6a show that the activity against bacterial of these compounds is moderate, but

additionally clearly demonstrate that this kind of compounds could be effective antibacterial agents, their activity depending on their chemical composition.

CONCLUSION

We have synthesized new hydroxy ether compounds containing coumarin moieties by the reaction of oligoethylene glycol diglycidyl ethers **2a–c** with both 7-hydroxy-4-methyl-2H-chromen-2-one and 4-hydroxy-2H-chromen-2-one. All reactions provided products in good to excellent yields. Furthermore a synthetic route for a new family of heterocyclic compounds fused with 4-hydroxycoumarins-2-one moieties was described. The antibacterial activities for these compounds were evaluated. The moderate active antibacterial effects observed showed that this kind of compounds could be effective antibacterial agents. The synthesis of new heterocyclic fused compounds containing a coumarin moiety by the reaction of 3-(3-(dimethylamino)acryloyl)-4-hydroxy-2H-chromen-2-one with binucleophiles was observed for the first time. Works are in progress to synthesize new coumarin derivatives with antibacterial activity, which may be cheap useful pharmaceutical products.

EXPERIMENTAL

All reactions and manipulations were routinely performed. Reagents were obtained from Sigma-Aldrich-Fluka and used without purification. Mass spectra were obtained with a Hewlett-Packard 5880a spectrometer. In this case electron impact techniques were employed. Elemental analyses were performed using a Carlo Erba 1106 microanalyser at the University of Rennes 1 in France. Infrared spectra (λ in cm^{-1}) were recorded on an ATI Mattson Infinity series FT-IR spectrometer using potassium bromide pellets for solids and liquid films for oils. ^1H and ^{13}C NMR spectra were recorded on a Bruker AC300 spectrometer (300 and 75 MHz). The samples were dissolved in CDCl_3 (compounds **1–4**) and $\text{DMSO}-d_6$ (compounds **5–16**). The chemical shift values were referenced to tetramethylsilane and internal standard. Chemical shift values and IR data for all compounds are summarized in the experimental part and are in agreement with the proposed structures. Melting points were determined on a Büchi No510 apparatus and are uncorrected. Thin layer chromatography (TLC) was performed with silica gel plates HF254 (Merck), and the plates were viewed under UV-254 light. Silica gel (230–400 mesh) was used for chromatography separations.

3-Acetyl-4-hydroxycoumarin (1). To a solution of 4-hydroxy-2H-chromen-2-one (3 g, 1.86 mmol) in acetic acid (16 mL) was added phosphorus oxychloride (5.6 mL). The mixture was heated at reflux for 30 min. After cooling, the precipitate was collected and recrystallized from ethanol to give 3-acetyl-4-hydroxy-2H-chromen-2-one **1** as white needles. Yield 2.7 g (90%); mp 134–136°C. IR spectrum, $\delta \text{ cm}^{-1}$: 3185 (OH); 1705 (CO); 1700 (O–CO lactone). ^1H NMR spectrum (CDCl_3). δ ppm: 2.72 (3H, s, CH_3); 7.98 (1H, s, H-5); 7.95 (1H, dd, $^3J_{7,8}=8.35$, $^4J_{6,8}=1.2$, H-8); 7.1–7.4 (2H, m, H-6 + H-7);

17.69 (1H, s, OH). ^{13}C NMR spectrum (CDCl_3), δ ppm: 29.9 (CH_3); 178.5 (CO); 159.8 (C-4); 154.6 (C-2); 101.26 (C-3); 115.0–136.0 (C_{arom}). Mass spectrum, m/z (I, %): 204 $[\text{M}]^+$ (100); 189 (74); 161 (43). Anal: Calcd. For $\text{C}_{11}\text{H}_8\text{O}_4$: C, 64.71, H, 3.95. Found C, 64.92, H, 3.68.

Preparation of the oligoethylene glycol diglycidyl ethers 2,2'-(2,5,8,11,14,17-hexaoxaoctadecane-1,18-diyl)dioxirane (2a), 2,2'-(2,5,8,11,14,17,20,23-octaotetracosane-1,24-diyl)dioxirane (2b), and 2,2'-(2,5,8,11,14,17,20,23,26,29-decaoxatriacontane-1,30-diyl)dioxirane (2c) (General procedure). To a mixture of epichlorohydrin (27.0 g, 300 mmol), NaOH (12.0 g, 300 mmol), H_2O (1.2 g, 66 mmol), and triethylammonium chloride (0.33 g) (phase-transfer catalyst), was added oligoethylene glycol (1.75 g, 50 mmol) dropwise while stirring at 45°C. The reaction mixture was stirred for 40 min, diluted with water (10 mL), and extracted thoroughly with CH_2Cl_2 . The extract liquors were dried (magnesium sulphate) and evaporated under reduced pressure. The resulting residue was purified by distillation *in vacuo* to afford the pure oligoethylene glycol diglycidyl ethers **2a–c**.

2,2'-(2,5,8,11,14,17-Hexaoxaoctadecane-1,18-diyl)dioxirane (2a). This compound was obtained as yellow oil; Yield 1.75 g (70%). ^1H NMR spectrum (CDCl_3), δ ppm (J, Hz): 2.56 (2H, sys. ABX, $^1J_{\text{HH}} = 4.93$, $^2J_{\text{HH}} = 2.67$, H-1); 2.74 (1H, sys. ABX, $^1J_{\text{HH}} = 4.84$, $^2J_{\text{HH}} = 4.36$, H-2); 3.11 (2H, tdd, $^1J_{\text{HH}} = 1.13$, $^2J_{\text{HH}} = 2.87$, $^3J_{\text{HH}} = 6.42$, H-3); 3.38 (2H, 2dd, $^1J_{\text{HH}} = 5.98$, $^2J_{\text{HH}} = 13.2$, H-4); 3.63 (6H, m, H-6–8); 3.75 (2H, 2d, $^1J_{\text{HH}} = 2.91$, H-5). $^{13}\text{C}\{^1\text{H}\}$ NMR spectrum (CDCl_3), δ ppm: 44.11 (C-1); 50.71 (C-2); 71.88 (C-3), 70.50 (C-4), 70.61 (C-5–8). Mass spectrum, m/z (I, %): 350 $[\text{M}]^+$ (100).

2,2'-(2,5,8,11,14,17,20,23-Octaotetracosane-1,24-diyl)dioxirane (2b). This compound was obtained as yellow oil, Yield 1.87 g (75%). ^1H NMR spectrum (CDCl_3), δ ppm (J, Hz): 2.48 (2H, sys. AX, $^1J_{\text{AX}} = 4.55$, $^1J_{\text{BX}} = 3.14$, H-1); 2.66 (1H, sys. ABX, $^1J_{\text{AB}} = ^1J_{\text{BA}} = 4.97$, H-2); 3.02 (2H, m, H-3); 3.27 (2H, td, $^1J_{\text{HH}} = 11.79$, $^2J_{\text{HH}} = 5.93$, H-4); 3.54 (6H, m, H-7–10); 3.66 (2H, sys. AB, $^1J_{\text{AB}} = ^1J_{\text{BA}} = 7.65$, H-5); 3.92 (2H, sys. AB₂, $^1J_{\text{HH}} = 6.43$, H-6). $^{13}\text{C}\{^1\text{H}\}$ NMR spectrum (CDCl_3), δ ppm: 43.9 (C-1); 50.6 (C-2); 69.0 (C-3), 70.4 (C-4), 70.8 (C-5–10). Mass spectrum, m/z (I, %): 438 $[\text{M}]^+$ (100).

2,2'-(2,5,8,11,14,17,20,23,26,29-Decaoxatriacontane-1,30-diyl)dioxirane (2c). This compound was obtained as a white oil, Yield 1.92 g (77%). ^1H NMR spectrum (CDCl_3), δ ppm (J, Hz): 2.43 (2H, sys. AB, $^1J_{\text{AB}} = ^1J_{\text{BA}} = 5.05$, H-1); 2.62 (1H, sys. AB, $^1J_{\text{AB}} = ^1J_{\text{BA}} = 4.4$, H-2); 2.98 (2H, m, $^1J_{\text{HH}} = 3.05$, H-3); 3.21 (2H, sys. AB₂, $^1J_{\text{HH}} = 5.84$, H-4); 3.48 (6H, m, H-7–12); 3.61 (2H, sys. AB₂, $^1J_{\text{HH}} = 7.65$, H-5), 3.87 (2H, sys. AB, $^1J_{\text{AB}} = ^1J_{\text{BA}} = 5.13$, H-6). $^{13}\text{C}\{^1\text{H}\}$ NMR spectrum (CDCl_3): 43.79 (C-1); 50.46 (C-2); 68.96 (C-3), 70.32 (C-4), 71.75 (C-5–12). Mass spectrum, m/z (I, %): 526 $[\text{M}]^+$ (100).

Reaction of 7-hydroxy-4-methylcoumarin and 4-hydroxycoumarin with oligoethylene glycol diglycidyl ethers 2a–c. General synthesis for the preparation 3a–c and 4a–c. To a solution of KOH (0.1 g, 1.7 mmol) in methanol (10 mL) at 0°C was added 7-hydroxy-4-methyl-2H-chromen-2-one (0.3 g, 1.7 mmol) or 4-hydroxy-2H-chromen-2-one (0.3 g, 1.8 mmol) followed by the slowly addition of the oligoethylene glycol diglycidyl ether (**2a–c**) (0.4 g, 0.0011 mol (**2a**), 0.00091 mol (**2b**), 0.00076 mol) in 10 mL of MeOH. The reaction was then kept at reflux for overnight and the resulting coumarin hydroxy ether was purified by chromatography (silica gel; ethyl acetate–cyclohexane, 6:4).

7,7'-[(2,21-Dihydroxy-4,7,10,13,16,19-hexaoxadocosane-1,22-diyl)bis(oxy)]bis(4-methyl-2H-chromen-2-one) (3a). Yield 0.82 g (70%); mp 165–170°C. IR spectrum, δ cm⁻¹: 3430 (OH); 3009 (C-H), 1710 (C=O); 1632 (C=C), 1490 (C=C_{arom}), 1120 (C-O_{alcohol}). ¹H NMR spectrum (CDCl₃), δ ppm (J, Hz): 2.40 (3H, s, CH₃); 2.92 (1H, s, OH); 3.40–3.72 (8H, m, H-19, 20, 22); 3.92–4.35 (9H, m, H-12–14,16,17); 6.20 (1H, s, H-3); 6.85 (1H, s, H-8); 6.90 (1H, d, ¹J_{HH} = 8.57, H-6), 7.52 (1H, d, ¹J_{HH} = 8.75, H-5). ¹³C{¹H} NMR spectrum (CDCl₃), δ ppm: 18.7 (CH₃); 59.2 (C-13); 68.86 (C-12, 17, 19, 20, 22); 68.4 (C-14); 101.7 (C-8); 113.1 (C-6); 112.5 (C-3); 113.9 (C-10); 125.6 (C-5); 152.7 (C-9); 155.1 (C-4); 161.4 (C-7); 161.5 (C-2). Mass spectrum, *m/z* (I, %): 674 [M]⁺ (100). Found, %: C 60.60; H 6.40; O 33.30. C₃₄H₄₂O₁₄. Calculated, %: C 60.53; H 6.27; O 33.20.

7,7'-[(2,27-Dihydroxy-4,7,10,13,16,19,22,25-octaooxaococane-1,28-diyl)bis(oxy)]bis(4-methyl-2H-chromen-2-one) (3b). This compound was obtained as colourless needles, Yield 0.95 g (72%); mp 175–180°C. IR spectrum, δ cm⁻¹: 3436 (OH); 3009 (C-H), 1712 (C=O); 1632 (C=C); 1497 (C=C_{arom}), 1123 (C-O_{alcohol}). ¹H NMR spectrum (CDCl₃), δ ppm (J, Hz): 2.40 (3H, s, CH₃); 2.88 (1H, s, OH); 3.39–3.65 (10H, m, H-19 + H-20 + H-22 + H-23 + H-25); 3.96–4.23 (9H, m, H-12–14,16,17); 6.15 (1H, s, H-3); 6.83 (1H, s, H-8); 6.89 (1H, d, ¹J_{HH} = 8.57, H-6); 7.50 (1H, d, ¹J_{HH} = 8.75, H-5). ¹³C{¹H} NMR spectrum (CDCl₃), δ ppm: 18.65 (CH₃); 59.18 (C-13); 68.82 (C-12,17,19,20,22,23,25); 68.34 (C-14); 101.67 (C-8); 112.07 (C-6); 112.43 (C-3); 113.88 (C-10); 125.62 (C-5); 152.68 (C-9); 155.04 (C-4); 161.38 (C-7); 161.49 (C-2). Mass spectrum, *m/z* (I, %): 762 [M]⁺ (100). Found, %: C 60.1; H 6.80; O 33.70. C₃₈H₅₀O₁₆. Calculated, %: C 59.84; H 6.60; O 33.56.

7,7'-[(2,33-Dihydroxy-4,7,10,13,16,19,22,25,28,31-decaoxatetratriacontane-1,34-diyl)bis(oxy)]bis(4-methyl-2H-chromen-2-one) (3c). This compound was obtained as colorless prisms, Yield 1.12g (76%); mp 160–165°C. IR spectrum, δ cm⁻¹: 3440 (OH); 3071, 2896 (C-H), 1713 (C=O); 1612, 1386 (C=C_{arom}), 1147, 1068 (C-O_{alcohol}). ¹H NMR spectrum (CDCl₃), δ ppm (J, Hz): 2.37 (3H, s, CH₃); 2.78 (1H, s, OH); 3.36–3.78 (10H, m, H-19,20,22,23,25,26,28); 3.94–4.23 (m, 9-H, H-12–14,16,17); 6.11 (1H, s, H-3); 6.79 (1H, d, ¹J_{HH} = 1.94, H-8); 6.88 (1H, dd, ¹J_{HH} = 8.78, ²J_{HH} = 2.42, H-6); 7.47 (1H, d, ¹J_{HH} = 8.83, H-5). ¹³C{¹H} NMR spectrum (CDCl₃), δ ppm: 18.61 (CH₃); 68.77 (C-13); 69.39 (C-12); 69.48 (C-16,17,19,20,22,23,25,28); 70.80 (C-14); 101.62 (C-8); 112.01 (C-6); 112.35 (C-3); 113.82 (C-10); 125.58 (C-5); 152.61 (C-9); 155.01 (C-4); 161.28 (C-7); 161.48 (C-2). Mass spectrum, *m/z* (I, %): 886 [M]⁺ (100). Found, %: C 60.1; H 6.80; O 33.9. C₄₂H₅₈O₁₈. Calculated, %: C 59.29; H 6.86; O 33.85.

4,4'-[(2,21-Dihydroxy-4,7,10,13,16,19-hexaoxadocosane-1,22-diyl)bis(oxy)]bis(2H-chromen-2-one) (4a). This compound was obtained as colourless needles, Yield 0.74 g (65%); mp 130–135°C. IR spectrum, δ cm⁻¹: 3435 (OH); 2796 (C-H); 1705 (C=O); 1655 (OC=O); 1630 (C=C); 1108 (C-O). ¹H NMR spectrum (CDCl₃), δ ppm: 5.40 (1H, s, OH); 3.10–4.10 (15H, m, H-12–14,16,17,19,20,22); 4.62 (1H, s, H-3); 7.10–7.85 (4H, m, H-5–8). ¹³C{¹H} NMR spectrum (CDCl₃), δ ppm: 124.2 (C-10); 154.7 (C-9); 165.4 (C-2); 176.6 (C-4); 74.4 (C-14); 69.8 (C-12); 70.3 (C-13); 72.2 (C-16,17,19,20,22); 85.9 (C-3); 124.6 (C-5); 121.9 (C-6); 130.3 (C-7); 115.9 (C-8). Mass spectrum, *m/z* (I, %): 702 [M]⁺ (100). Found, %: C 61.60; H 6.70; O 21.60. C₃₆H₄₆O₁₄. Calculated, %: C 61.53; H 6.59; O 21.45.

4,4'-[(2,27-Dihydroxy-4,7,10,13,16,19,22,25-octaooxaococane-1,28-diyl)bis(oxy)]bis(2H-chromen-2-one) (4b). This

compound was obtained as colorless needles, Yield 0.858 g (75%); mp 145–147°C. IR spectrum, δ cm⁻¹: 3431 (OH); 2896 (C-H); 1703 (C=O); 1650 (OC=O); 1624 (C=C); 1106 (C-O); 1540 (C=C_{arom}). ¹H NMR spectrum (CDCl₃), δ ppm: 5.20 (1H, s, OH); 3.19–3.99 (23H, m, H-12–14,16,17,19,20,22,23,25); 4.66 (1H, s, H-3); 7.06–7.83 (4H, m, H-5 + H-6 + H-7 + H-8). ¹³C{¹H} NMR spectrum (CDCl₃), δ ppm: 123.95 (C-10); 154.89 (C-9); 165.49 (C-2); 175.50 (C-4); 74.45 (C-14); 69.91 (C-12); 70.25 (C-13); 72.89 (C-16,17,19,20,22,23,25); 85.97 (C-3); 124.76 (C-5); 121.92 (C-6); 130.21 (C-7); 115.96 (C-8). Mass spectrum, *m/z* (I, %): 790 [M]⁺ (100). Found, %: C 60.6; H 6.80; O 32.5. C₄₀H₅₄O₁₆. Calculated, %: C 60.75; H 6.83; O 32.41.

4,4'-[(2,33-Dihydroxy-4,7,10,13,16,19,22,25,28,31-decaoxatetratriacontane-1,34-diyl)bis(oxy)]bis(2H-chromen-2-one) (4c). This compound was obtained as a white solid; yield 0.80 g (70%); mp 150–160°C. IR spectrum, δ cm⁻¹: 3412 (OH); 2875 (C-H); 1722 (C=O); 1648 (OC=O); 1622 (C=C); 1534 (C=C_{arom}); 1108 (C-O). ¹H NMR spectrum (CDCl₃), δ ppm: 4.52 (1H, s, OH); 3.01–4.10 (23H, m, H-12–14,16,17,19,20,22,23,25,26,28); 5.56 (1H, s, H-3); 6.98–7.85 (4H, m, H-5–8). ¹³C{¹H} NMR spectrum (CDCl₃), δ ppm: 115.39 (C-10); 154.23 (C-9); 162.94 (C-2); 176.35 (C-4); 73.74 (C-14); 69.19 (C-12); 69.44 (C-13,16,17,19,20,22,23,25,26,28); 89.75 (C-3); 123.88 (C-5); 122.91 (C-6); 132.31 (C-7); 116.42 (C-8). Mass spectrum, *m/z* (I, %): 878 [M]⁺ (100). Found, %: C 60.2; H 6.80; O 32.7. C₄₄H₆₂O₁₈. Calculated, %: C 60.13; H 7.06; O 32.80.

3-(3-Dimethylaminoacryloyl)-4-hydroxy-2H-chromen-2-one (5). To a solution of the 3-acetyl-4-hydroxycoumarine-2-one (1) (2.04 g, 10 mmol) was added DMF–DMA (1.19 g, 10 mmol) in dry toluene (50 mL) and then the reaction mixture was refluxed for 2 h. The solvent was evaporated under vacuum and the residual material was triturated with petroleum ether (25 mL), filtered, evaporated under reduced pressure to provide the product as white needles. Yield 2.20 g (85%); mp 140–145°C. IR spectrum, δ cm⁻¹: 3439 (OH); 1700 (CO); 1619 (O–CO lactone). ¹H NMR spectrum (DMSO-D₆), δ ppm: 2.50 (6H, s, 2CH₃); 6.62 (1H, d, H_{ethyl}); 8.32 (1H, d, H_{ethyl}); 18.28 (OH); 7.29–7.95 (5H, m, H_{arom}). ¹³C{¹H} NMR spectrum (DMSO-D₆), δ ppm: 46.2 (CH₃); 182.9 (CO); 181.2 (C-4); 158.4 (C-2); 95.3 (C-3); 90.9 (CO–C); 160.9 (C=C–N); 118.8–153.7 (aromatic). Mass spectrum, *m/z* (I, %): 259 [M]⁺ (100).

The conversion of 3-(3-(dimethylaminoacryloyl)-4-hydroxy-2H-chromen-2-one (5) to adducts 6a-b. Hydrazine hydrate (0.048 g, 1.5 mmol) or phenylhydrazine (0.138 g, 1.5 mmol) was added to a solution of (3-dimethylaminoacryloyl)-4-hydroxy-2H-chromen-2-one (0.001 mol) in toluene (20 mL). The mixture was refluxed for 6 h. The formed precipitate was filtered and recrystallized from toluene.

4-Hydroxy-3-(1H-pyrazol-3-yl)-2H-chromen-2-one (6a). This compound was obtained as a yellow solid, Yield 0.273 g (80%); mp 120–130°C (CH₂Cl₂). IR spectrum, δ cm⁻¹: 1735–1755 (CO); 3331 (OH), 1702 (C=O ester). ¹H NMR spectrum (DMSO-D₆), δ ppm: 13.48 (1H, s, OH); 12.51 (1H, s, NH); 7.06–8.00 (5H, m, aromatic). ¹³C{¹H} NMR spectrum (DMSO-D₆), δ ppm: 160.5 (C-2); 164.6 (C-4); 94.9 (C-3); 146.8 (C=N); 116.6–132.9 (aromatic + C=N); 104.5 (C=C–N). Mass spectrum, *m/z* (I, %): 228 [M]⁺ (100). Found, %: C 63.2; H 3.4; O 21.1. N 12.5 Calculated, %: C 63.16; H 3.53; O 21.03. N 12.28;

4-Hydroxy-3-(1-phenyl-1H-pyrazol-3-yl)-2H-chromen-2-one (6b). This compound was obtained as a white solid, Yield

0.222 g (65%); mp 147–149°C (CH₂Cl₂). IR spectrum, δ cm⁻¹: 1725–1760 (CO); 3330 (OH), 1712 (C=O ester). ¹H NMR spectrum (DMSO-D₆), δ ppm: 13.48 (1H, s, OH); 7.20–8.10 (m, aromatic + CH₂). ¹³C {¹H} NMR spectrum (DMSO-D₆) δ ppm: 161.5 (C-2); 164.8 (C-4); 94.8 (C-3); 146.8 (C=N); 116.2–133.7 (aromatic + C=C–N); 104.6 (C=C–N). Mass spectrum, m/z (I , %): 304 [M]⁺ (100). Found, %: C 71.1; H 4.1; O 15.8; N 9.3. Calculated, %: C 71.05; H 3.97; O 15.77; N 9.21;

The conversion of 3-(3-(dimethylaminoacryloyl)-4-hydroxy-2H-chromen-2-one (5) to 4-hydroxy-3-(isoxazol-3-yl)-2H-chromen-2-one (6c). Hydroxylamine (0.048 g, 1.5 mmol) was added to a solution of (3-dimethylaminoacryloyl)-4-hydroxy-2H-chromen-2-one (0.001 mol) in methanol (20 mL). The mixture was refluxed for 3 h. The formed precipitate was filtered and recrystallized from ethanol.

This compound was obtained as a yellow solid, Yield 0.27 g (80%); mp 120–130°C. IR spectrum, δ cm⁻¹: 3430 (OH); 1731 (CO), 1613 (–O–CO–). ¹H NMR spectrum (DMSO-D₆), δ ppm: 10.36 (1H, s, OH); 6.79 (1H, d, CH₂); 7.35–8.20 (4H, m, aromatic). ¹³C {¹H} NMR spectrum (DMSO-D₆) δ ppm: 160.7 (C-2); 163.9 (C-4); 93.9 (C-3); 150.7 (C=N); 160.1 (C–O–N); 104.2 (C=C–O–N); 116.8–133.7 (aromatic). Mass spectrum, m/z (I , %): 229 [M]⁺ (100). Found, %: C 62.9; H 3.1; O 27.9; N 6.3. Calculated, %: C 62.89; H 3.08; O 27.92; N 6.11;

The experimental procedure for the antibacterial evaluation. The obtained products are dissolved in a suitable solvent, the determined quantities of this solution are put on the disc (example: 1 mg/disc, it is enough to dissolve 1 mg of the sample in 0.1 mL of CHCl₃), then the solvent is evaporated and the product remained. This prepared disc is called a treated disc. The second disc must be prepared: it is a disc of control (the solvent used can obtain active products, to ensure, the same volume has to be taken to dissolve the sample. The third disc must contain *gentamycin*, and the latter is for comparison. Then we put all three discs on the surface of gel containing bacteria, and then we close it at 37°C for 24 h, and we measure the diameters of the clear zones.

Antimicrobial reference standards.

Microorganisms/IZ ^{a,b}										
Sa	Se	MI	St	Ec	Pa	Fo	An	Al	Pe	
10 μ g	17	18	17	11	10	9	n.d	n.d	n.d	n.d

Microorganisms: Sa, *Staphylococcus aureus*; Se, *Staphylococcus epidermidis*; MI, *Micrococcus luteus*; St, *Salmonella typhimurium*; Ec, *Escherichia coli*; Pa, *Pseudomonas aeruginosa*; Fo, *Fusarium oxysporum*; An, *Aspergillus niger*; Al, *Alternaria* sp.; Pe, *Penicillium* sp. bIZ: Inhibition zone (mm).

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