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Efficient microwave-assisted synthesis, antibacterial activity and high fluorescence of 5 benzimidazolyl-2′-deoxyuridines

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ABSTRACT

A series of novel C-5 benzimidazolyl-2′-deoxyuridines was synthesized in good yields under solvent-free conditions and microwave irradiation from 5-formyl-2′-deoxyuridine and arylenediamine derivatives in the presence of $NaHSO_3$ as catalyst. Their absorption and fluorescence spectra were measured. They showed intense fluorescence around 400–500 nm with quantum yields between 0.3 and 0.5. All compounds studied in this work were screened for their antibacterial activities against a series of Gram positive and negative bacteria. The trifluoromethyl substituted benzimidazole derivatives showed some antibacterial activity.

1. Introduction

Structural modification of deoxyribonucleosides has been a prime target in medicinal chemistry. Introduction of a substituent to the heterocyclic ring may significantly influence their base-pairing abilities and selectivity for binding to targets. Many nucleoside analogues substituted at the 5-position of the pyrimidine, especially in the 2′-deoxyuridine series, show potent activity against viruses as diverse as herpes simplex virus (HSV-1 and HSV-2),[1] vaccinia virus (VV),[2] hepatitis B virus (HBV)[3] and varicella zoster virus (VZV)[4] with a potential to be activated by deoxynucleoside kinases in bacteria and thereby used as species–specific antibiotics for treatment of bacterial infections.[5]

The 5-position of pyrimidine nucleobases is an appropriate place to incorporate functionality as the site lies in the major groove of the duplex where appendages do not disrupt Watson–Crick base pairing.[6,7] On the other hand, fluorescent nucleoside analogues that are sensitive to their local environment have become powerful tools in biophysical studies of nucleic acid structure and dynamics. Fluorescent groups attached to 2′,3′-dideoxynucleotides have been key to automated DNA sequencing.[8]

Functionalized appendages may also increase the power and versatility of nucleic acids as receptors, ligands and catalysts. Linking natural nucleobases to fluorescent chromophores, typically via conjugating linkers (e.g. ethynyl), have been shown to produce fluorescent nucleoside analogues.[9–13]

The benzimidazole ring system is an important pharmacophore and has proven to be useful for the development of molecules with pharmaceutical properties. Benzimidazoles possess broad spectra of biological activities such as antiviral (anti-HIV), anticancer, antibacterial, antifungal and many other activities.[14,15] Based on these ideas, part of our research program is oriented towards the design, synthesis and implementation of new fluorescent nucleosides. They should only minimally disturb the structure of the natural nucleobases and display emission at long wavelengths (preferably in the visible range). In our project we conjugated the benzimidazole ring in 2-position to the uridine 5-position (Fig. 1) hoping to find a new class of fluorescent nucleosides with antibacterial and anti-HIV activities.

2. Results and discussion

Many heterocycles were introduced in C-5 position of 2′-deoxyuridine (furane, thiophene, imidazole, benzothiazole,

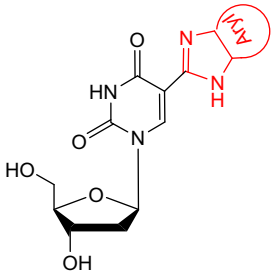

Figure 1.

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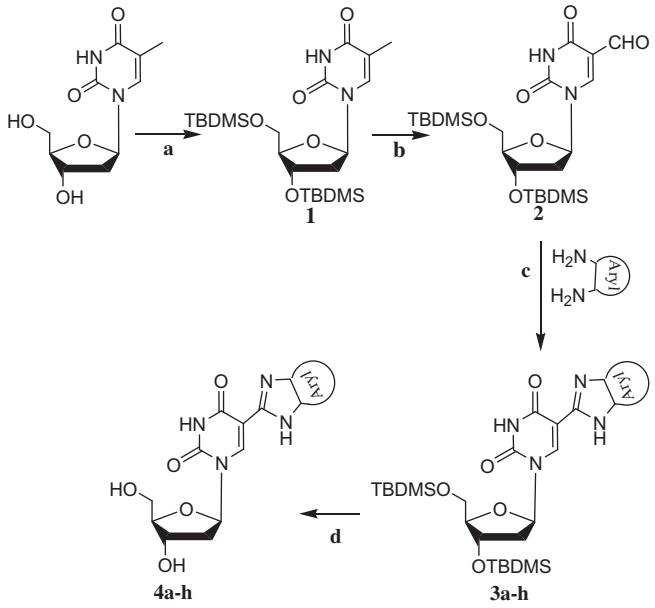
indole).[16–18] Cross-coupling reactions have facilitated the introduction of C-based substituents starting from 5-iododeoxyuridine. The Suzuki,[19] Stille[20] and Sonogahira[21] couplings have all been exploited successfully in this respect. To further broaden the chemical space not easily attainable by current cross coupling methods, the development of new synthetic methods will be very important to the chemical community. We report here the synthesis of new thymidine nucleoside analogues carrying benzimidazolyl group functionality appended at position C5. We have synthesized several analogues of 2′-deoxyuridine, investigated their potential applications as fluorescent probes and in addition tested the activity of these 5-benzimidazolyl-2′-deoxyuridines against Gram-positive and negative bacteria and HIV. Hence finding rapid means to synthesize benzimidazoles at the 5-position of uracil would be highly beneficial. Two approaches have been reported for the synthesis of benzimidazole and its derivatives. The first approach includes the coupling of *o*-phenylenediamines and carboxylic acids or various derivatives in the presence of strong acids by high temperature.[22] The second one is the condensation of *o*-diaminoaromatic compounds with aromatic aldehydes in a two-step procedure that includes an oxidative cyclodehydrogenation of 'aniline Schiff bases'. Various oxidative reagents, such as nitrobenzene,[23] benzoquinone,[24] sodium metabisulfite,[25] lead tetraacetate,[26] oxone,[27] tetracyanoethylene,[28] $I_2/KI/K_2CO_3/H_2O$[29] and even air,[30] are reported. Recently, sodium hydrogen sulfite ($NaHSO_3$)[31] has been successfully employed for a one-pot preparation of 2-aryl-benzimidazoles. To further explore the utility of simple one-pot, short reaction times, solvent-free and simple experimental conditions,[32] we decided to synthesise 5-benzimidazolyl-2′-deoxyuridine derivatives by the latter method.

The synthetic approach to 5-benzimidazolyl-2′-deoxyuridine involved first the preparation of 3′,5′-bis-*O*-(*tert*-butyldimethylsilyl)-thymidine **1** from the thymidine as shown in Scheme 1. The two hydroxyl groups in the deoxyribose of thymidine were protected with the *tert*-butyldimethylsilyl (TBDMS) group using *tert*-butyldimethylsilyl chloride (TBDMS-Cl) in DMF for 7 h to provide compound **1** in good yield.

Next, we investigated the oxidation of protected thymidine **1** to the corresponding acid. Molko and co-workers[33] reported the synthesis of 5-carboxy-2′-deoxyuridine by photosensitized oxidation using menadione in low yields and prolonged reaction times, (~16 h). Thus we followed the protocol of Gambari and co-workers[34] but failed to obtain the desired 5-carboxylate in a satisfactory yield. Next, we tried to introduce the formyl group to uracil. The protected nucleoside **1** was oxidized with 2 equiv of $K_2S_2O_8$, 0.38 equiv $CuSO_4 \times 5H_2O$ and 3.58 equivalents 2,6-lutidine in CH_3CN/H_2O at 65 °C for 2 h according to methodology developed by Matsuda group[35] and the resulting compound **2** was confirmed by ^1H, ^{13}C NMR and mass spectra.

Following this, the 5-benzimidazolyl-2′-deoxyuridine derivatives (Scheme 1) were prepared via condensation of 3′,5′-bis-*O*-(*tert*-butyldimethylsilyl)-5-(formyl)-2′-deoxyuridine **2** (1 equiv) and *o*-phenylenediamine derivatives (1.2 equiv) in the presence of catalytic amounts (0.4 equiv) $NaHSO_3$ under microwave irradiation without solvent in 2 min the desired nucleosides (**3a–h**) were observed as major products and obtained in good to excellent yields (67–90%). The final deprotection of TBDMS groups by tetra-*n*-butylammonium fluoride in tetrahydrofuran during 1 h at room temperature gave the corresponding nucleosides (**4a–h**) in high yields (Table 1).

The ^1H NMR spectra for compounds **3a** and **f** (see Supplementary data) were measured in $CDCl_3$. The benzimidazole NH appears at 11.7 ppm, the NH of uracil at 10.6 ppm for **3a**, the latter being broader. In the case of **3f** two signals appear for the benzimidazole at 12.0 and 11.3 ppm in a ratio of roughly 2:1. Theoretically one could expect 2 isomers for **3a** and 4 for **3f**. Assuming an intramolecular hydrogen bond, the possibilities are reduced to 1 and 2 isomers.



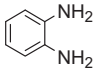
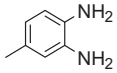
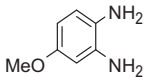
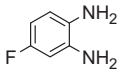
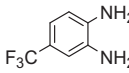
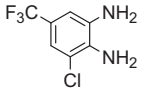
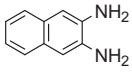
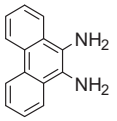
Scheme 1. Synthesis of benzimidazole attached 2′-deoxyuridines. Reagents and conditions: (a) TBDMSiCl, Imidazole, DMF, rt, 7 h; (b) $K_2S_2O_8$, $CuSO_4 \times 5H_2O$, 2,6-lutidine, H_2O/CH_3CN, 65 °C, 2 h. (c) $NaHSO_3$, MW, 250 Hz, 100 °C, 2 min; (d) TBAF, THF, rt, 1 h.

3. Spectral properties

3.1. Absorption spectra

The absorption spectra (JASCO UV–Vis) of 5-benzimidazolyl-2′-deoxyuridine derivatives (**4a–f**, Table 1) were taken in

Table 1
The synthesis of C-5 benzimidazolyl-2′-deoxyuridine under microwave irradiation

Entry	R	Product[a]	Yield[b] (%)	Product[a]	Yield[b] (%)
1		**3a**	90	**4a**	90
2		**3b**	72	**4b**	76
3		**3c**	86	**4c**	93
4		**3d**	82	**4d**	84
5		**3e**	67	**4e**	80
6		**3f**	70	**4f**	76
7		**3g**	84	**4g**	90
8		**3h**	85	**4h**	92

[a] All products were characterized by ^1H NMR, ^{13}C NMR, and mass spectrometry.
[b] Yields of isolated products.

dimethylsulfoxide as solvent at room temperature and the concentration of the solute was maintained at 44 μM. All absorption maxima are presented in Table 2 and spectra for all compounds are shown in Figure 2. 5-benzimidazolyl-2′-deoxyuridine exhibits a strong absorption around 330 nm and weaker bands between 270 and 290 nm and around 260 nm. The reason for the substantial bathochromic shift in the investigated compounds **4b** and **4c** (λ_{max} = 330–350.5 nm) relative to that of unsubstituted 5-(1*H*-benzimidazol-2-yl)-2′-deoxyuridine (**4a**) (λ_{max} = 326 nm) is the strong positive effect of the alkoxy and alkyl substituent, respectively. Electron withdrawing substituents show no or only small hypsochromic shifts in absortion maxima.

3.2. Emission spectra

Fluorescence emission spectra of compounds **4a–f** were recorded at 44 μM in dimethylsulfoxide as solvent (Fig. 3). The excitation of each molecule at their corresponding absorption band of each substituted benzimidazole shows single emission bands (Hitachi F-4500 fluorescence spectrometer) in the range of ~400 to ~500 nm which were assigned as emission from their locally excited states. Excitation and emission maxima and fluorescence quantum yields are reported in Table 2. The fluorescence quantum yields were determined according to the correlation of Crosby[36] using quinine sulfate (Φ = 0.55) in 0.1 N H_2SO_4 as standard.

5-benzimidazolyl-2′-deoxyuridine derivatives with electron-donating groups (alkyl and alkoxy) affects the position of the maxima of long-wavelength fluorescence which are shifted from λ_{max} = 442 nm, for **4a** (Table 2, entry 1) to λ_{max} = 501 nm for **4c** (Table 2, entry 3) with the same quantum yield (Φ = 0.29). Electron withdrawing substituents in **4d–f** again result in no shift in emission maxima compared to parent **4a** with the exception of **4e** that exhibits substantial hypsochromic shift to 404 nm with high quantum yield. The lower quantum yield and absence of emission maximum shift might be correlated with the halogene substituents in **4d** and **4f**

We further investigated the fluorescence of **4f** in aqueous solution at different pH values (Fig. 4). In acidic and neutral solution the fluorescence of **4f** is strong and there is no meaningful change in intensity. Above pH 11 the emission decreases significantly while maintaining emission wavelength.

Split NMR signals for unsymmetrical benzimidazoles as well as high fluorescence for all compounds might be explained by the formation of a hydrogen bridge between imidazole-NH and nucleobases carbonyle at C4 position. This arrangement would lead to a planar alignment and might explain high fluorescence for all compounds.[37] More profound explanation requires more detailed spectroscopic investigations to elucidate possible H-bond formation and formation of rotameres or tautomeres, respectively.

4. Biological results

The antibacterial activity was reported in terms of the minimum inhibitory concentration (MIC) values, which are defined as the lowest concentration of an antimicrobial that visibly inhibits the growth of the bacteria after an overnight incubation. The compounds (**4a–h**) were evaluated for their in vitro antibacterial activity against the following bacterial strains; *Staphylococcus aureus* (ATCC 13709 in vivo, ATCC 25923, oxford and MRSA in vivo), *Enterococcus faecalis* (ATCC 29212 VanS), *Enterococcus* faecium (Van A), *streptococcus pneumoniae* (VanA, ATCC49619, PenR and Blood effect), *Haemophilus influenzae* (ATCC 31517 MMSA), *Escherichia coli* (ATCC 25922) and *Pseudomonas aeruginosa* (ATCC 27853), using standard techniques and the minimum inhibitory concentration values (MICs).[38] The minimum inhibitory concentrations (MIC) of synthesized compounds against Gram positive and

Table 2
Spectroscopic properties of 5-benzimidazolyl-2′-deoxyuridine derivatives (**4a–f**) absorption and emission spectral data at room temperature in DMSO

Entry	Compounds	Solvent	λ_{abs} (nm)	λ_{em} (nm)	Quantum yield (Φ)%
1	**4a**	DMSO	326	442	0.29
2	**4b**	DMSO	330	468	0.24
3	**4c**	DMSO	336	501	0.29
4	**4d**	DMSO	326.5	445.5	0.10
5	**4e**	DMSO	327	404	0.58
6	**4f**	DMSO	324.5	446	0.22

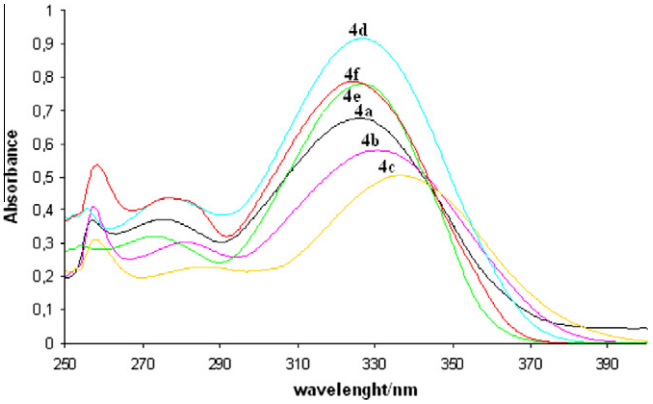


Figure 2. UV/Vis absorption spectra of compounds **4a–h** in DMSO (c = 44 μM).

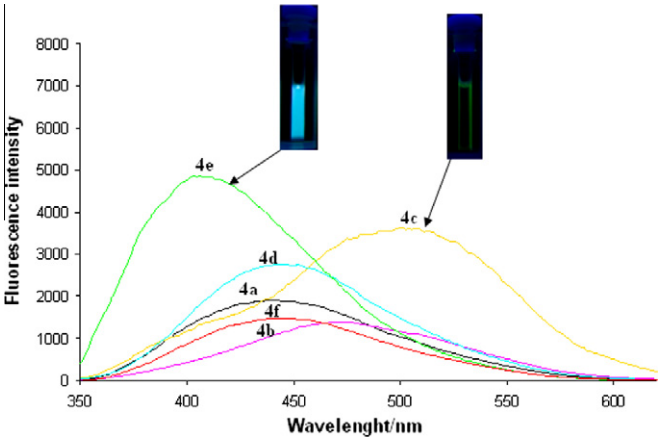


Figure 3. Fluorescence emission spectra of compounds **4a–h** in DMSO, excited at $\lambda_{abs\cdot max}$ (c = 44 μM).

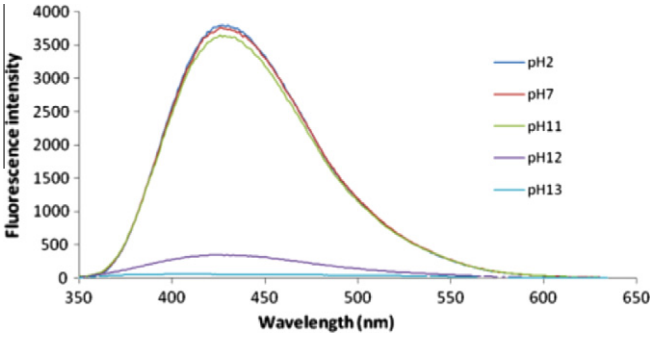


Figure 4. Fluorescence emission spectra of compound **4f** in aqueous solution at different pHs, excited at $\lambda_{abs\cdot max}$ (c = 44 μM).

Gram negative bacteria are summarized in Table 3. Ciprofloxacin and Linezolid were used as standard drugs for comparison.

As shown in Table 3 the 5-benzimidazolyl-2′-deoxyuridine derivatives showed effective activities against Gram-positive and Gram-negative bacteria. The compounds **4a**, **4b**, **4c**, **4d**, **4g** and **4h** showed antibacterial activity with a range of the MICs higher than 64 μg/ml and compounds **4e**, **4f** exhibit a good antibacterial activity, especially **4f** showed good activity against Gram-positive bacteria *S. aureus* (ATCC 13709 in vivo, ATCC 25923, oxford and MRSA in vivo) (2 μg/ml), *E. faecalis* (2 μg/ml), *E. faecium* (1 μg/ml) and *S. pneumoniae* (4–16 μg/ml), more potent than the tow reference drugs *S. aureus* (ATCC 13709 in vivo, ATCC 25923, oxford and MRSA in vivo) (2 μg/ml), *E. faecalis* (2 μg/ml), *E. faecium* (1 μg/ml) and *S. pneumoniae* (4–16 μg/ml), except in the presence of serum, but in the second test of this compound the activity could not be well reproduced.

The compounds were also evaluated for their antiviral activity.[39] None of the compounds exhibited specific antiviral activity against HIV, which means that they did not inhibit the replication (induction of viral cytopathogenicity).

5. Conclusions

New efficient synthetic routes were developed to prepare fluorescent compounds containing benzimidazoles in 5-position of pyrimidine nucleosides. The synthesis is based on the straightforward condensation of 5-formyl-2′-deoxyuridine and arylenediamine derivatives under a cooperative effect of microwave activation and $NaHSO_3$ catalysis. All 5-benzimidazolyl-2′-deoxyuridine derivatives were obtained in nearly quantitative yield after a short reaction time. We have investigated the absorption and fluorescence spectral properties of all compounds, most of them exhibit good to excellent fluorescence in the range of 400–500 nm with (Φ between 0.1 and 0.58). Antibacterial activities of these compounds are reported. Some of them showed potential antibacterial activity, none antiviral.

6. Experimental section

All chemicals were purchased from Alfa Aesar, Acros, Aldrich, Sigma or Fluka. Solvents were of laboratory grade. Thin-layer chromatography (TLC): aluminum sheets, silica gel 60 F_{254}, Column flash chromatography (FC). ^1H and ^{13}C NMR spectra were recorded in $CDCl_3$ and DMSO-d_6 on a Bruker AC 250 MHz and AC 400 MHz with TMS as an internal standard. Chemical shifts are given in ppm and spin–spin coupling constants, *J*, are given in Hz (s, singlet; d, doublet; t, triplet; m, multiplet and br, broad) (Bzm) Benzimidazole, Mass spectra were obtained using ESI/MS, MALDI-TOF. High-resolution (HRMS) mass spectrometry was performed on a MALDI Orbitrap XL (Thermo Fisher Scientific), equipped with a 337 nm Nitrogen-Laser. 4-Hydroxy alpha cyanocinnamic acid was used as matrix and the matrix peaks were used as lock masses for internal calibration.

6.1. Microwave irradiation experiments

All microwave irradiation experiments were carried out in a dedicated CEM-Discover mono-mode microwave apparatus, operating at a frequency of 2455 MHz with continuous irradiation power from 0 to 300 W ± 10%.

The electronic absorption spectra were obtained on Jasco V-650 spectrometer and fluorescence emission spectra were recorded on Hitachi F-4500 fluorescence spectrometer, in both cases using quartz cuvettes (1 cm). The fluorescence quantum yields were calculated by the comparative method relative to quinine sulfate ($\Phi = 0.55$) in 0.1 N H_2SO_4.

MICs were determined based on CLSI methodology[38] by a 2-fold broth dilution technique in Mueller Hinton (MH, pH 7.4 Biorad). For *S. pneumoniae* the medium was Brain Heart Infusion broth + 4% red blood cell extract. For *H. influenzae* the medium was HTM (Haemophilus Test Medium consisting of MH + 5 g/L yeast extract + hemin 15 mg/L + NAD 20 mg/L). Overnight cultures were diluted to obtain the final inoculum of 10^5 cfu/well. Incubation was 37 °C overnight in ambient air.

6.2. Sugar protection procedure

6.2.1. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-thymidine (1)

5 g (20.6 mmol) thymidine were added to 8.41 g (123.6 mmol) Imidazole and 9.31 g (61.8 mmol) TBDMS-Cl in 120 mL DMF at room temperature under argon atmosphere, the mixture was stirred for 7 h. The reaction was quenched with 8 mL MeOH, Diluted with 400 mL EtOAc. The solution was washed twice, each time with 200 mL H_2O, once with 200 mL saturated aq $NaHCO_3$ and once with 200 mL brine, dried (Na_2SO_4) and concentrated to afford a solid mass, which was then purified by column chromatography on silica gel; elution with EtOAc/hexane (1.5/8.5) afforded **1** in 96% yield. ^1H NMR (250 MHz, $CDCl_3$) δ 8.74 (br s, 1H, NH), 7.36 (s, 1H, H-6), 6.23 (dd, 1H, J = 7.8 and 5.8 Hz, $H_{1'}$), 4.29 (m, 1H, $H_{3'}$), 3.82 (dd, 1H, J = 4.7 and 2.2 Hz, $H_{4'}$), 3.70 (ddd, 1H, J = 28.5, 11.3 and 2.4 Hz, $H_{5'}$), 2.14 (ddd, 2H, J = 13.1, 5.8 and 2.5 Hz, $H_{2'a}$), 1.89 (m, 1H, $H_{2'b}$), 1.80 (s, 3H, CH_3), 0.82 (s, 9H, C–$(CH_3)_3$), 0.78 (s, 9H, C–$(CH_3)_3$), 0.00 (s, 6H, Si–$(CH_3)_2$), −0.03 (m, 6H, Si–$(CH_3)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 163.75 (C4), 150.29 (C2), 135.45 (C6), 110.82 (C5), 87.83 (C4′), 84.85 (C1′), 72.25 (C3′), 62.99 (C5′),

Table 3
Minimum inhibitory concentration (MIC) in μg/ml of medium

		Strains	Phenotype	Cipro	Liné	4a	4b	4c	4d	4e	4f*	4g	4h
1	*S. aureus*	Sa1	ATCC13709 in vivo	0.12	1	>64	>64	>64	>64	>64	2	>64	>64
2		Sa26	ATCC25923	0.25	1	>64	>64	>64	>64	>64	2	>64	>64
3		Sa26 + 10% serum human	Serum effect	0.25	1	>64	>64	>64	>64	>64	>64	>64	>64
4		Sa26 + 50% serum human	Serum effect	0.5	2	>64	>64	>64	>64	>64	>64	>64	>64
5		Sa4	Oxford	0.12	1	>64	>64	>64	>64	>64	2	>64	>64
6		Sa2	MRSA, in vivo	8	1	>64	>64	>64	>64	>64	2	>64	>64
7	*E. faecalis*	Ecalis1	ATCC29212 VanS	0.5	2	>64	>64	>64	>64	>64	2	>64	>64
8	*E. faecium*	Ecium1	VanA	16	0,5	>64	>64	>64	>64	64	1	>64	>64
9	*S. pneumoniae*	Pn1	ATCC49619	1	1	>64	>64	>64	>64	64	4	>64	>64
10		Pn9	PenR	0.5	0,5	>64	>64	>64	>64	32	4	>64	>64
11		Pn9 + 2.5%blood	Blood effect	0.5	0,25	>64	>64	>64	>64	64	16	>64	>64
12	*H. influenzae*	Hi3	ATCC 31517 MMSA	⩽0.03	16	>64	>64	>64	>64	4	>64	>64	>64
14	*E. coli*	Ec1	ATCC25922	⩽0.03	>32	>64	>64	>64	>64	>64	>64	>64	>64
16	*P. aeruginosa*	Pa1	ATCC 27853	0.25	>32	>64	>64	>64	>64	>64	>64	>64	>64

Cipro: Ciprofloxacin; Lin: Linezolid; *: second test showed results >64.

41.39 (C2′), 25.95 (C–CH_3), 25.76 (C–CH_3), 18.41 (C–CH_3), 18.02 (C–CH_3), 12.52 (CH_3), −4.62 (Si–CH_3), −4.82 (Si–CH_3), −5.35 (Si–CH_3), −5.44 (Si–CH_3). ESI-MS: *m*/*z* calcd for $C_{22}H_{42}N_2O_5Si_2$ [M+H] 471.26 found, 471.60.

6.3. Oxidative aldehyde formation

6.3.1. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-5-(formyl) -2′-deoxy-uridine (2)

6.0 g (12.75 mmol) **1** were dissolved in 200 mL of 1:1 (v/v) CH_3CN/H_2O and 6.89 g (25.5 mmol) K_2S_2O8, 1.17 g (4.72 mmol) $CuSO_4{\cdot}5H_2O$, and 5.28 mL (45.6 mmol) 2,6-lutidine were added. Heat the mixture with stirring at 65 °C. The reaction was monitored by TLC in 1:2 (v/v) EtOAc/hexane, twice (R_f = 0.50). The starting compound **1** disappeared after 2–3 h. A longer reaction time increases the amount of a more polar byproduct, so the reaction should be stopped before the complete disappearance of **1**. The reaction mixture was filtered off through a Celite pad and diluted with 500 mL EtOAc. The solution was washed three times with 300 mL of 10% aq EDTA, twice with 200 mL 0.5 N HCl, twice with 200 mL satd aq $NaHCO_3$ and once with 200 mL brine. The organic layers were dried over Na_2SO_4. Evaporation of solvent followed by flash chromatography with EtOAc/hexane (2/8) yielded **2** (50%). ^{1}H NMR (250 MHz, $CDCl_3$) δ 9.91 (s, 1H, CHO), 9.71 (br s, 1H, NH), 8.45 (s, 1H, H-6), 6.13 (dd, 1H, J = 7.5 and 5.8 Hz, $H_{1'}$), 4.31 (dd, 1H, J = 3.3 and 2.2 Hz, $H_{3'}$), 3.96 (d, 1H, J = 2.1 Hz, $H_{4'}$), 3.73 (ddd, 2H, J = 28.4, 11.4 and 2.6 Hz, $H_{5'}$), 2.35 (ddd, 1H, J = 13.1, 5.6 and 1.9 Hz, $H_{2'a}$), 1.94 (ddd, 1H, J = 13.2, 7.5 and 5.8 Hz, $H_{2'b}$), 0.79 (s, 9H, C–$(CH_3)_3$), 0.78 (s, 9H, C–$(CH_3)_3$), −0.01 (m, 12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 185.74 (CHO), 162.36 (C-4), 149.42 (C-2), 145.43 (C-6), 111.04 (C-5), 89.20 (C4′), 87.23 (C1′), 72.90 (C3′), 63.08 (C5′), 42.53 (C2′), 25.99 (C–CH_3), 25.78 (C–CH_3), 18.44 (Si–C), 18.04 (Si–C), −4.63 (Si–CH_3), −4.80 (Si–CH_3), −5.47 (Si–CH_3), −5.60 (Si–CH_3). ESI-MS: *m*/*z* calcd for $C_{22}H_{40}N_2O_6Si_2$ [M+H] 485.24 found 485.6

6.3.2. General procedure for the preparation of 3′, 5′-bis-*O* -(*tert*-butyldimethylsilyl)-5-(benzimidazol-2-yl)-2′-deoxy-uridines using $NaHSO_3$ under microwave irradiation (3a–h)

The mixture of 5-formyl-2′-deoxyuridine **2** (0.41 mmol), corresponding *o*-phenylenediamine (0.49 mmol) and sodium hydrogen sulfite (0.164 mmol) was irradiated in the microwave oven with 250 Hz for 2 min at 100 °C, the mixture was allowed to cool to room temperature and was extracted with ethyl acetate. The organic layer was dried over Na_2SO_4, and the solvent was evaporated under reduced pressure to give a crude product, which was purified by flash column chromatography on silica gel, using CH_2Cl_2–MeOH (98: 2) as eluent.

6.3.3. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-5-(1*H*-benzimidazol-2-yl)-2′-deoxyuridine (3a)

(90%) (250 MHz, $CDCl_3$) δ 11.70 (s, 1H, Bzm–NH), 10.60 (br s, 1H, NH), 8.88 (s, 1H, H-6), 7.54 (m, 1H, Bzm–H), 7.44 (m, 1H, Bzm–H), 7.09 (dd, 2H, J = 9.1 and 5.8 Hz, Bzm–H), 6.24 (dd, 1H, J = 7.7 and 5.8 Hz, $H_{1'}$), 4.34 (d, 1H, J = 4.9 Hz, $H_{3'}$), 3.94 (s,1H, $H_{4'}$), 3.75 (ddd, 2H, J = 13.9, 11.1 and 3.2 Hz, $H_{5'}$), 2.33 (dd, 1H, J = 12.6 and 5.3 Hz, $H_{2'a}$), 2.01 (ddd, 1H, J = 13.2, 7.6 and 5.8 Hz, $H_{2'b}$), 0.83 (s, 9H, C–$(CH_3)_3$), 0.73 (s, 9H, C–$(CH_3)_3$), 0.00 (m, 12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 162.48(C-4), 149.84(C-2), 145.50(Bzm–C), 142.95 (Bzm–C), 141.10(C-6), 134.23(Bzm–C), 122.39(Bzm–C), 122.07(Bzm–C), 118.61(Bzm–C), 111.67(Bzm–C), 104.73 (C-5), 88.97(C4′), 87.03(C1′), 73.23 (C3′), 63.30 (C5′), 42.09 (C2′), 26.07 (C–CH_3), 25.83 (C–CH_3), 18.46 (Si–C), 18.04 (Si–C), −4.62 (Si–CH_3), −4.75 (Si–CH_3), −5.25 (Si–CH_3), −5.56 (Si–CH_3). MALDI-TOF: calcd for $C_{28}H_{44}N_4O_5Si_2$ 572.29 found 572.84.

6.3.4. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-5-(6-methyl-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (3b)

(72%) The product is a mixture of two tautomers in the ratio 0.5:0.5. ^{1}H NMR (250 MHz, $CDCl_3$) δ 11.46 (s, 1H, NH), 10.52 (br s, 1H, NH), 8.85 (s, 1H, H-6), 7.31 (m, 2H, Bzm–H), 6.91 (t, 1H, J = 8.4 Hz, Bzm–H), 6.26 (m, 1H, $H_{1'}$), 4.37 (d, 1H, J = 5.2 Hz, $H_{3'}$), 3.96 (s, 1H, $H_{4'}$), 3.78 (dq, 2H, J = 11.3, 11.1 and 3.0 Hz, $H_{5'}$), 2.33 (m, 4H, Bzm–CH_3 and $H_{2'a}$), 2.07 (m,1H, $H_{2'b}$), 0.82 (m, 9H, C–$(CH_3)_3$), 0.75 (s, 9H, C–$(CH_3)_3$), 0.02 (m, 12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 162.60 (C-4), 149.77 (C-2), 145.26 (Bzm–C), 143.31 (Bzm–C), 140.82 (C-6), 134.25 (Bzm–C), 132.23 (Bzm–C), 123.91 (Bzm–C), 118.30 (Bzm–C), 111.25 (Bzm–C), 104.82 (C-5), 88.93 (C4′), 87.00 (C1′), 73.10 (C3′), 63.28 (C5′), 41.93 (C2′), 26.07 (C–CH_3), 25.84 (C–CH_3), 21.85 (Bzm–CH_3), 18.48 (Si–C), 18.06 (Si–C), −4.74 (Si–CH_3), −4.61 (Si–CH_3), −5.24 (Si–CH_3), −5.54 (Si–CH_3). MALDI-TOF: calcd for $C_{29}H_{46}N_4O_5Si_2$ 586.30, found 587.99.

6.3.5. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-5-(6-methoxy-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (3c)

(86%) The product is a mixture of two tautomers in the ratio 0.57:0.43 ^{1}H NMR (250 MHz, $CDCl_3$) δ 11.54 (s, 1H, Bzm–NH), 10.67 (br s, 1H, NH), 8.83 (s, 1H, H-6), 7.37 (dd, 1H, J = 24.6, 8.7 Hz, Bzm–H), 6.99 (dd, 1H, J = 25.9 and 1.9 Hz, Bzm–H), 6.74 (ddd, 1H, J = 8.3, 5.7 and 2.2 Hz, Bzm–H), 6.24 (m, 1H, $H_{1'}$), 4.36 (d, 1H, J = 5.27 Hz, $H_{3'}$), 3.95 (s, 1H, $H_{4'}$), 3.79 (m, 5H, OCH_3 and $H_{5'}$), 2.33 (m,1H, $H_{2'a}$), 2.05 (m,1H, $H_{2'b}$), 0.82 (m, 9H, C–$(CH_3)_3$), 0.75 (s, 9H, C–$(CH_3)_3$), 0.02 (m, 12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 162.54 (C-4), 156.25 (Bzm–C), 149.80 (C-2), 145.19 (Bzm–C), 140.48 (C-6), 137.55 (Bzm–C), 134.59 (Bzm–C), 128.89 (Bzm–C), 119.06 (Bzm–C), 111.83 (Bzm–C), 104.87 (C-5), 88.92 (C4′), 86.95 (C1′), 73.05 (C3′), 63.24 (C5′), 55.70 (Bzm–OCH_3), 41.97 (C2′), 26.07 (C–CH_3), 25.82 (C–CH_3), 18.46 (Si–C), 18.06 (Si–C), −4.62 (Si–CH_3), −4.77 (Si–CH_3), −5.27 (Si–CH_3), −5.54 (Si–CH_3). MALDI-TOF: calcd for $C_{29}H_{46}N_4O_6Si_2$ 602.30, found 603.73.

6.3.6. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-5-(6-fluoro-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (3d)

(82%) The product is a mixture of two tautomers in the ratio 0.6:0.4 ^{1}H NMR (250 MHz, $CDCl_3$) δ 11.74 (s, 1H, Bzm–NH), 10.74 (br s, 1H, NH), 8.81 (s, 1H, H-6), 7.32 (m,1H, Bzm–H), 7.10 (dd, 1H, J = 11.2 and 2.3 Hz, Bzm–H), 6.80 (dt, 1H, J = 10.7 and 2.0 Hz, Bzm–H), 6.27 (m,1H, $H_{1'}$), 4.34 (d, 1H, J = 4.0 Hz, $H_{3'}$), 3.98 (d, 1H, J = 5.4 Hz, $H_{4'}$), 3.77 (m, 2H, $H_{5'}$), 2.30 (dd, 1H, J = 12.8 and 5.2 Hz, $H_{2'a}$), 1.96 (dd, 1H, J = 13.25, 6.43 Hz, $H_{2'b}$), 0.84 (s, 9H, C–$(CH_3)_3$), 0.73 (s, 9H, C–$(CH_3)_3$), 0.02 (m, 12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 162.30 (C-4), 157.50 (Bzm–C), 149.93 (C-2), 146.66 (Bzm–C), 141.24 (C-6), 139.37 (Bzm–C), 134.24 (Bzm–C), 130.81 (Bzm–C), 119.07 (Bzm–C), 111.82 (Bzm–C), 104.56 (C-5), 89.10 (C4′), 87.02 (C1′), 73.36 (C3′), 63.28 (C5′), 42.36 (C2′), 26.07 (C–CH_3), 25.80 (C–CH_3), 18.45 (Si–C), 18.04 (Si–C), −4.68 (Si–CH_3), −4.82 (Si–CH_3), −5.28 (Si–CH_3), −5.65 (Si–CH_3). MALDI-TOF: calcd for $C_{28}H_{43}FN_4O_5Si_2$ 590.28, found 591.07.

6.3.7. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-5-(6-trifluoromethyl-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (3e)

(67%) ^{1}H NMR (400 MHz, $CDCl_3$) δ 12.01 (br s, 1H, Bzm–NH), 10.78 (br s, 1H, NH), 8.98 (s,1H, H-6), 7.72 (s,1H, Bzm–H), 7.49 (br s,1H, Bzm–H), 7.32 (br.s,1H, Bzm–H), 6.24 (m, 1H, $H_{1'}$), 4.36 (d, 1H, J = 5.0 Hz, $H_{3'}$), 3.98 (s, 1H, $H_{4'}$), 3.87 (m,1H, $H_{5'a}$), 3.74 (dd, 1H, J = 11.0 and 2.3 Hz, $H_{5'b}$), 2.31 (dd, 1H, J = 12.9 and 5.4 Hz, $H_{2'a}$), 2.02 (s, 1H, $H_{2'b}$),0.84 (s, 9H, C–$(CH_3)_3$), 0.73 (s, 9H, C–$(CH_3)_3$), 0.02 (m, 12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 162.27 (C-4), 149.94 (C-2), 147.01 (Bzm–C), 142.48 (Bzm–C and

C-6), 127.11 (Bzm–C), 125.00 (Bzm–C), 124.48 (Bzm–C), 122.79 (Bzm–C), 119.44 (Bzm–C and CF_3), 103.70 (C-5), 89.31 (C4′), 87.31 (C1′), 73.27 (C3′), 63.22 (C5′), 42.52 (C2′), 26.02 (C–CH_3), 25.76 (C–CH_3), 18.40 (Si–C), 18.01 (Si–C), −4.79 (2 × (Si–CH_3)), −5.30 (Si–CH_3), −5.66 (Si–CH_3). MALDI-TOF: calcd for $C_{29}H_{43}F_3N_4O_5Si_2$ 640.27, found 641.55.

6.3.8. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-5-(7-chloro-5-trifluoromethyl-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (3f)

(70%) The product is a mixture of two tautomers in the ratio 0.7:0.3 ^{1}H NMR (250 MHz, $CDCl_3$) δ 12.03 (s, 1H, Bzm–NH),11.30 (s, 1H, Bzm–NH), 10.80 (br s, 1H, NH), 9.56 (br s, 1H, NH), 9.00 (s, 1H, H-6), 8.94 (s, 1H, H-6), 7.70 (s, 1H, Bzm–H), 7.65 (s, 1H, Bzm–H), 7.36 (s,1H, Bzm–H), 7.34 (s, 1H, Bzm–H), 6.23 (dd, 1H, J = 11.9 and 5.8 Hz, $H_{1'}$), 4.40 (d, 1H, J = 5.2 Hz, $H_{3'}$), 4.01 (m, 1H, $H_{4'}$), 3.80 (m, 2H, $H_{5'}$), 2.41 (m,1H, $H_{2'a}$), 2.11 (m,1H, $H_{2'b}$), 0.81 (s, 9H, C–$(CH_3)_3$), 0.73 (m, 9H, C–$(CH_3)_3$), −0.00 (m, 12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 162.47 (C-4), 149.88 (C-2), 148.24 (Bzm–C), 142.81 (2 × Bzm–C), 142.34 (C-6), 134.35 (Bzm–C), 124.13 (Bzm–C), 119.30 (Bzm–C), 108.14 (Bzm–C), 103.39 (C-5), 89.35 (C4′), 88.08 (C1′), 73.04 (C3′), 63.17 (C5′), 42.32 (C2′), 25.91 (2 × (C–CH_3)), 18.41 (Si–C), 18.09 (Si–C), −4.78 (2 × (Si–CH_3)), −5.29 (Si–CH_3), −5.63 (Si–CH_3). MALDI-TOF: calcd for $C_{29}H_{42}ClF_3N_4O_5Si_2$ 674.23, found 675.07.

6.3.9. 3′,5′-Bis-*O*-(*tert*-butyldimethylsilyl)-5-(1*H*-naphtho[2,3-d]imidazol-2-yl)-2′-deoxyuridine (3g)

(84%) ^{1}H NMR (250 MHz, $CDCl_3$) δ 11.56 (s, 1H, Bzm–NH), 10.43 (s, 1H, NH), 9.01 (s, 1H, H-6), 8.01 (s,1H, Bzm–H), 7.84 (m, 2H, Bzm–H), 7.77 (m,1H, Bzm–H), 7.23 (m, 2H, Bzm–H), 6.25 (dd, 1H, J = 7.51 and 6.00 Hz, $H_{1'}$), 4.38 (d, 1H, J = 5.4 Hz, $H_{3'}$), 4.00 (d, 1H, J = 1.4 Hz, $H_{4'}$), 3.89 (dd, 1H J = 11.2 and 3.30 Hz, $H_{2'a}$), 3.74 (dd, 1H, J = 11.1 and 2.7 Hz, $H_{2'b}$), 2.41 (dd, 1H, J = 12.8 and 5.53 Hz, $H_{2'a}$), 2.08 (m,1H, $H_{2'b}$), 0.74 (s,9H, C–$(CH_3)_3$), 0.83 (s,9H, C–$(CH_3)_3$), 0.03 (m,12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 Hz, $CDCl_3$) δ 162.49 (C4), 149.73 (C2, Bzm–C), 143.34 (Bzm–C), 142.49 (C-6), 134.87 (Bzm–C), 130.57 (2 × (Bzm–C)), 128.27 (Bzm–C), 127.81 (Bzm–C), 123.66 (Bzm–C), 123.18 (Bzm–C), 115.06 (Bzm–C), 107.28 (Bzm–C), 104.10 (C-5), 89.31 (C4′), 87.50 (C1′), 73.23 (C3′), 63.30 (C5′), 42.42 (C2′), 25.98 (2 × (C–CH_3)), 18.49 (Si–C), 18.11 (Si–C), −4.59 (Si–CH_3), −4.72 (Si–CH_3), −5.21 (Si–CH_3), −5.55 (Si–CH_3). ESI-MS: m/z calcd for: calcd for $C_{32}H_{46}N_{45}Si_2$ [M+H] 623.30, found 623.80.

6.3.10. 3′,5′-Bis-*bis*-(*tert*-butyldimethylsilyl)-5-(1*H*-phenanthro[9,10-d]imidazol-2-yl)-2′-deoxyuridine (3h)

(85%) (250 MHz, $CDCl_3$) δ 11.78 (s, 1H, Bzm–NH), 10.13 (s, 1H, NH), 8.88 (s, 1H, H-6), 8.47 (m, 4H, Bzm–H), 7.45 (m, 4H, Bzm–H), 6.29 (t, 1H, J = 6.7 Hz, $H_{1'}$), 4.41 (m, 1H, $H_{3'}$), 4.00 (d, 1H, J = 1.9 Hz, $H_{4'}$), 3.81 (m, 2H, $H_{5'}$), 2.37 (dd, 1H, J = 12.8 and 5.7 Hz, $H_{2'a}$), 2.13 (m, 1H, $H_{2'b}$), 0.79 (s, 9H, C–$(CH_3)_3$), 0.71 (s, 9H, C–$(CH_3)_3$), −0.02 (m, 12H, (Si–$(CH_3)_2)_2$). ^{13}C NMR (250 MHz, $CDCl_3$) δ 162.34 (C-4), 149.43 (C-2), 142.76 (Bzm–C), 140.01 (C-6), 128.31 (2 × Bzm–C), 126.82 (4 × Bzm–C),125.26 (4 × Bzm–C), 123.43 (3 × Bzm–C), 122.42 (Bzm–C), 104.48 (C-5), 88.98 (C4′), 87.12 (C1′), 73.06 (C3′), 63.34 (C5′), 41.54 (C2′), 25.88 (2 × (C–CH_3)), 18.16 (Si–C), 18.41 (Si–C), −4.56 (Si–CH_3), −4.69 (Si–CH_3), −5.23 (Si–CH_3), −5.51 (Si–CH_3). MALDI-TOF: calcd for $C_{36}H_{48}N_4O_5Si_2$ 672.32, found 673.63.

6.3.11. General procedure for the deprotection of 3′, 5′-hydroxy groups (4a–h)

General procedure for preparing **4**: Compound **3** (0.1 mmol) was dissolved in approximately 15 mL of THF and added (0.25 mmol) TBAF/THF/H_2O the reaction was stirred at room temperature for 1 h. The solvent was removed under reduced pressure and the residue obtained was purified on silica gel column CH_2Cl_2 and MeOH (9/1).

6.3.12. 5-(1*H*-Benzimidazol-2-yl)-2′-deoxyuridine (4a)

(90%) ^{1}H NMR (250 MHz, DMSO-d_6) δ 12.17 (s, 1H, Bzm–NH), 11.91 (br s, 1H, NH), 8.80 (s, 1H, H-6), 7.57 (m,2H, Bzm–H), 7.13 (m, 2H, Bzm–H), 6.20 (t, 1H, J = 6.6 and 6.6 Hz, $H_{1'}$), 5.30 (d, 1H, J = 4.2 Hz, $O_{3'}$–H), 5.02 (t, 1H, J = 4.9 Hz, $O_{5'}$–H), 4.28 (m, 1H, $H_{3'}$), 3.86 (m, 1H, $H_{4'}$), 3.60 (t, 2H, J = 4.31 Hz, $H_{5'}$), 2.23 (m, 2H, $H_{2'}$). ^{13}C NMR (250 MHz, DMSO-d_6) δ 161.85 (C4), 149.47 (C2), 146.03 (Bzm–C), 140.90 (C6), 121.55 (4 × (Bzm–C)), 117.87 (Bzm–C), 112.17 (Bzm–C), 103.93 (C5), 87.79 (C4′), 85.27 (C1′), 70.48 (C3′), 61.34 (C5′), 40.02 (C2′). UV (DMSO) λ_{max} 326 nm (ε 15200). MALDI-TOF: calcd for $C_{16}H_{16}N_4O_5$ 344.11, found 344.10. HRMS (M+H) found 345.12002, calcd for $C_{16}H_{17}N_4O_5$ 345.11935.

6.3.13. 5-(6-Methyl-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (4b)

(76%) ^{1}H NMR (250 MHz, DMSO-d_6) δ 11.99 (s, 2H, Bzm–NH, NH), 8.69 (s, 1H, H-6), 7.37 (d, 1H, J = 7.9 Hz, Bzm–H), 7.29 (s, 1H, Bzm–H), 6.88 (m,1H, Bzm–H), 6.13 (t, 1H, J = 6.6 Hz, $H_{1'}$), 5.27 (br s, 1H, $O_{3'}$–H), 4.93 (br s, 1H, $O_{5'}$–H), 4.20 (d, 1H, J = 3.0 Hz, $H_{3'}$), 3.79 (dd, 1H, J = 6.9 and 3.8 Hz, $H_{4'}$), 3.53 (m, 2H, $H_{5'}$), 2.31 (s, 3H, Bzm–CH_3), 2.15 (m, 2H, $H_{2'}$). ^{13}C NMR (250 MHz, DMSO-d_6) δ 162.04 (C4), 149.62 (C2), 145.78 (Bzm–C), 140.56 (Bzm–C and C6), 130.62 (Bzm–C), 123.09 (2 × Bzm–C), 117.57 (Bzm–C), 111.80 (Bzm–C), 104.08 (C5), 87.79 (C4′), 85.24 (C1′), 70.51 (C3′), 61.39 (C5′), 57.54 (C3′), 40.03(C2′), 21.35 (Bzm–CH_3). UV (DMSO) λ_{max} 330 nm (ε 13200). ESI-MS: m/z calcd for $C_{17}H_{18}N_4O_5$ [M−H] 357.13, found 357.0. HRMS (M+H) found 375.13076 calcd for $C_{17}H_{19}N_4O_5$ 375.12991.

6.3.14. 5-(6-Methoxy-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (4c)

(93%) ^{1}H NMR (250 MHz, DMSO-d_6) δ 11.94 (s,1H, Bzm–NH), 11.81 (s,1H, NH), 8.65 (s, 1H, H-6), 7.35 (t, 1H, J = 7.8 Hz, Bzm–H), 7.01 (d, 1H, J = 16.16 Hz, Bzm–H), 6.67 (dd, 1H, J = 8.7 and 2.2 Hz, Bzm–H), 6.11 (t, 1H, J = 6.6 Hz, $H_{1'}$), 5.23 (d,1H, J = 4.0 Hz, $O_{3'}$–H), 4.95 (t, 1H, J = 4.8 Hz, $O_{5'}$–H), 4.18 (m, 1H, $H_{3'}$), 3.77 (d, 1H, J = 2.5 Hz, $H_{4'}$), 3.66 (s, 1H, Bzm–OCH_3), 3.52 (d, 2H, J = 4.0 Hz, $H_{5'}$), 2.13 (m, 2H, $H_{2'}$). ^{13}C NMR (250 MHz, DMSO-d_6) δ 161.85 (C4), 155.43 (Bzm–C), 149.48 (C4), 146.17 (Bzm–C), 145.16 (Bzm–C), 140.25 (C6), 136.97 (Bzm–C), 128.78 (Bzm–C), 118.32 (Bzm–C), 112.51(Bzm–C) 104.20 (C5), 100.43, 87.79 (C4′), 85.21 (C1′), 70.51 (C3′), 61.38 (C5′), 55.36 (Bzm–OCH_3), 40.03 (C2′). UV (DMSO) λ_{max} 336 nm (ε 11500). ESI/MS: m/z calcd for $C_{17}H_{18}N_4O_6$ [M−H] 373.12, found 373.1. HRMS (M+H) found 375.13076, calcd for $C_{17}H_{19}N_4O_6$ 375.12991

6.3.15. 5-(6-Fluoro-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (4d)

(84%) ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.29 (s, 1H, Bzm–NH), 11.95 (s,1H, NH), 8.83 (s, 1H, H-6), 7.58 (m,1H, Bzm–H), 7.36 (ddd, 1H, J = 16.5, 9.5, 2.1 Hz, Bzm–H), 7.00 (m, 1H, Bzm–H), 6.21 (t, 1H, J = 6.6 Hz, $H_{1'}$), 5.33 (d, 1H, J = 4.1 Hz, $O_{3'}$–H), 5.06 (t, 1H, J = 4.9 Hz, $O_{5'}$–H), 4.30 (m, 1H, $H_{3'}$), 3.89 (q, 1H, J = 3.9 Hz, $H_{4'}$), 3.62 (m, 2H, $H_{5'}$), 2.24 (dd, 2H, J = 8.2 and 3.69 Hz, $H_{2'}$). ^{13}C NMR (400 MHz, DMSO-d_6) δ 161.82 (C4),159.64 (Bzm–C), 149.45 (C2), 147.83 (Bzm–C), 141.27 (C6), 140.93 (Bzm–C), 130.99 (Bzm–C), 112.81(Bzm–C), 109.50 (Bzm–C), 109.75 (Bzm–C), 103.64 (C5), 87.82 (C4′), 85.34 (C1′), 70.46 (C3′), 61.31 (C5′), 39.96 (C2′). UV (DMSO) λ_{max} 326,5 nm (ε 20800). ESI-MS: m/z calcd for $C_{16}H_{15}FN_4O_5$ [M−H] 361.10, found 361.20. HRMS (M+H) found 363.11127, calcd for $C_{16}H_{16}FN_4O_5$ 363.10992.

6.3.16. 5-(6-Trifluoromethyl-1*H*-benzoimidazol-2-yl)-2′-deoxyuridine (4e)

(80%) ^{1}H NMR (250 MHz, DMSO-d_6) δ 12.58 (s, 1H, Bzm–NH), 12.02 (br s,1H, NH), 8.93 (s, 1H, H-6), 7.86 (m, 2H, Bzm–H), 7.45 (d,1H, J = 8.4 Hz, Bzm–H), 6.19 (t, 1H, J = 6.5 Hz, $H_{1'}$), 5.30 (br s, 1H,

$O_{3'}$–H), 5.03 (d, 1H, J = 1.1 Hz, $O_{5'}$–H), 4.28 (d, 1H, J = 2.8 Hz, $H_{3'}$), 3.88 (dd, 1H, J = 6.7 and 3.5 Hz, $H_{4'}$), 3.62 (d, 2H, J = 2.9 Hz, $H_{5'}$), 2.24 (m, 2H, $H_{2'}$). ^{13}C NMR (250 MHz, DMSO-d_6) δ 162.27 (C-4), 149.94 (C-2), 147.01 (Bzm–C), 142.48 (Bzm–C and C-6), 127.11 (Bzm–C), 125.00 (Bzm–C), 124.48 (Bzm–C), 122.79 (Bzm–C), 119.44 (Bzm–C and CF_3), 103.70 (C-5), 89.31 (C4′), 87.31 (C1′), 73.27 (C3′), 63.22 (C5′), 42.52 (C2′). UV (DMSO) λ_{max} 404 nm (ε 17700). ESI-MS: m/z calcd for $C_{17}H_{15}F_3N_4O_5$ [M−H] 411.10, found 411.6. HRMS (M+H), found 413.10773, calcd for $C_{17}H_{16}F_3N_4O_5$ 413.10673.

6.3.17. 5-(7-Chloro-5-trifluoromethyl-1*H*-benzimidazol-2-yl)-2′-deoxyuridine (4f)

(76%) ^{1}H NMR (250 MHz, DMSO-d$_6$) δ 12.85 (br s,1H, Bzm–NH), 12.13 (br s,1H, NH), 8.95 (s, 1H, H-6), 7.95 (s, 1H, Bzm–H), 7.59 (s, 1H, Bzm–H), 6.19 (t, 1H, J =6.51 Hz, $H_{1'}$), 5.34 (d, 1H, J = 4.20 Hz, $O_{3'}$–H), 5.01 (s, 1H, $O_{5'}$–H), 4.29 (d, 1H, J = 3.6 Hz, $H_{3'}$), 3.92 (d, 1H, J = 3.2 Hz, $H_{4'}$), 3.64 (t, 2H, J = 4.0 Hz, $H_{5'}$), 2.28 (dd, 1H, J = 7.9 and 3.6 Hz, $H_{2'}$). ^{13}C NMR (250 MHz, DMSO-d_6) δ 160.16 (C-4), 149.69 (C2), 148.89 (Bzm–C), 141.90 (Bzm–C), 141.53 (C-6), 134.99 (Bzm–C), 128.59 (Bzm–C), 122.36 (Bzm–C), 122.09 (Bzm–C), 118.10 (Bzm–C), 109.04 (Bzm–C), 102.01 (C5), 88.22 (C4′), 86.80 (C1′), 70.26 (C3′), 61.16 (C5′), 40.48 (C2′). UV (DMSO) λ_{max} 324,5 nm (ε 17800). ESI-MS: m/z calcd for $C_{17}H_{14}ClF_3N_4O_5$ [M−H] 445.06, found 445.6. HRMS (M+H) found 447.06844, calcd for $C_{17}H_{15}ClF_3N_4O_5$ 447.06776.

6.3.18. 5-(1*H*-Naphtho[2,3-d]imidazol-2-yl)-2′-deoxyuridine (4g)

(90%) ^{1}H NMR (250 MHz, DMSO-d_6) δ 12.27 (s,1H, Bzm–NH), 12.04 (s,1H, NH), 9.00 (s, 1H, H-6), 8.08 (d, 2H, J = 10.2 Hz, Bzm–H), 7.96 (t, 2H, J = 8.9 Hz, Bzm–H), 7.36 (m, 2H, Bzm–H), 6.23 (t, 1H, J = 6.5 Hz, $H_{1'}$), 5.36 (d, 1H, J = 4.18 Hz, $O_{3'}$–H), 5.11 (t, J = 4.9 Hz, $O_{5'}$–H), 4.32 (m, 1H, $H_{3'}$), 3.92 (d, 1H, J = 3.21 Hz, $H_{4'}$), 3.67 (t, 2H, J = 4.3 Hz, $H_{5'}$), 2.28 (m, 2H, $H_{2'}$). ^{13}C NMR (250 MHz, DMSO-d_6) δ 161.84 (C-4), 150.58 (Bzm–C), 149.49 (C-2), 143.02 (Bzm–C), 142.42 (C-6), 135.18 (Bzm–C), 129.79 (2 × Bzm–C), 127.93 (Bzm–C), 127.39 (Bzm–C), 123.40 (Bzm–C), 122.92 (Bzm–C), 113.98 (Bzm–C), 107.46 (Bzm–C), 103.45 (C5), 87.94 (C4′), 85.58 (C1′), 70.49 (C3′), 61.36 (C5′), 40.53 (C2′). ESI-MS: m/z calcd for $C_{20}H_{18}N_4O_5$ [M−H] 394.13, found 393.3. HRMS (M+H) found 395.13632, calcd for $C_{20}H_{19}N_4O_5$ 395.13500.

6.3.19. 5-(1*H*-Phenanthro[9,10-d]imidazol-2-yl)-2′-deoxyuridine (4h)

(92%) ^{1}H NMR DMSO-d_6 δ: 12.77 (s,1H, Bzm–NH), 11.90 (s,1H, NH), 8.81 (s, 1H, H-6), 8.75 (t, 2H, J = 7.4 Hz, Bzm–H), 8.58 (d, 1H, J = 7.3 Hz, Bzm–H), 8.50 (d, 1H, J = 6.9 Hz, Bzm–H), 7.59 (m, 4H, Bzm–H), 6.20 (t, 1H, J = 6.5 Hz, $H_{1'}$), 5.28 (d, 1H, J = 4.1 Hz, $O_{3'}$–H), 5.07 (t, 1H, J = 4.6 Hz, $O_{5'}$–H), 4.27 (m, 1H, $H_{3'}$), 3.84 (q, 1H, J = 3.3 Hz, $H_{4'}$), 3.61 (m, 2H, $H_{5'}$), 2.22 (t, 1H, J = 6.45 Hz, $H_{2'}$). ^{13}C NMR DMSO-d_6 δ:161.73 (C-4), 149.63 (C-2), 143.77 (Bzm–C), 140.45 (C-6), 127.44 (2 × Bzm–C), 126.95 (3 × Bzm–C), 126.68 (Bzm–C), 125.11 (3 × Bzm–C), 123.74 (3 × Bzm–C), 122.20 (4 × Bzm–C), 104.92 (C5), 87.84 (C4′), 85.27 (C1′), 70.47 (C3′), 61.28 (C5′), 40.49 (C2′).ESI-MS: m/z calcd for $C_{24}H_{20}N_4O_5$.[M−H] 443.14, found 443.3. HRMS (M+H) found 445.15130, calcd for $C_{24}H_{21}N_4O_5$ 445.15065.

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Supplementary data

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