CONJUGATES OF FERROCENE AND PURINE AND PURINE ISOSTERES: SYNTHESIS AND BIOLOGICAL EVALUATION

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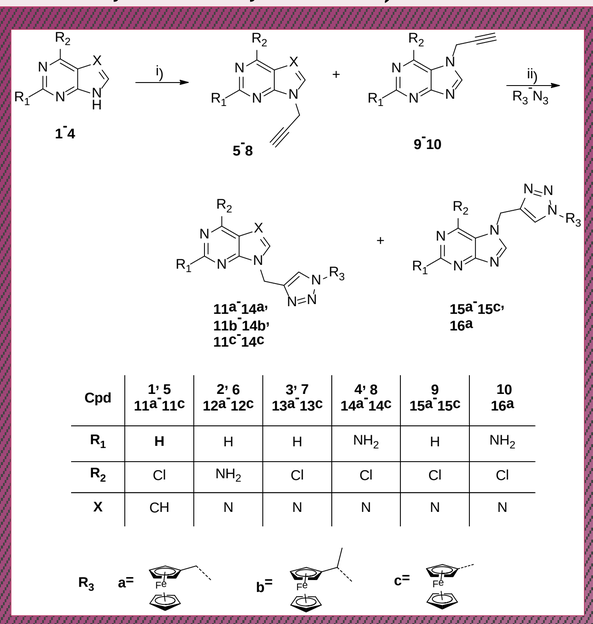
Introduction

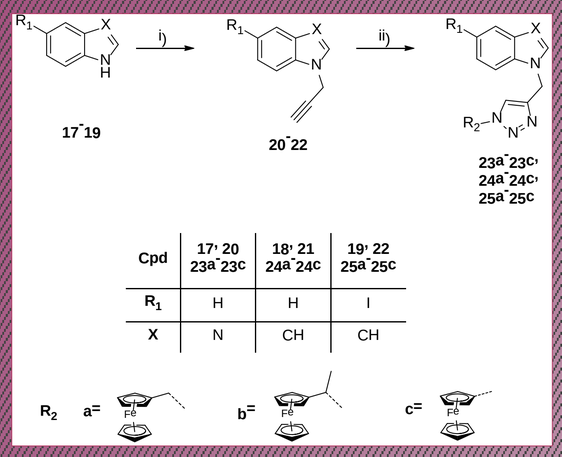
Cytotoxic nucleobase-derived compounds have gained a lot of importance in recent years for combating cancer of different types, usually in combination with other agents.[1,2] The interest in metal complexes of ferrocene-based ligands is due to their favorable physicochemical properties and reversible redox properties that enable ferrocene derivatives to be used as an excellent candidates for drug development.[3,4] Taking into consideration the biological relevance of nucleoside analogues of ferrocene, novel purine and purine isosteres containing a ferrocene N-1-substituted 1,2,3-triazole (**11a-11c, 12a-12c, 13a-13c, 14a-14c, 15a-15c, 16a, 23a-23c, 24a-24c, 25a-25c**) and a ferrocene 4-substituted 1,2,3-triazole moiety (**34a-34c, 35a-35c**) attached to variety of heterocyclic bases were prepared.



Cpd	1, 5 11a-11c	2, 6 12a-12c	3, 7 13a-13c	4, 8 14a-14c	9 15a-15c	10 16a
R_1	H	H	H	NH_2	H	NH_2
R_2	Cl	NH_2	Cl	Cl	Cl	Cl
X	CH	N	N	N	N	N

R_3 a= b= c=

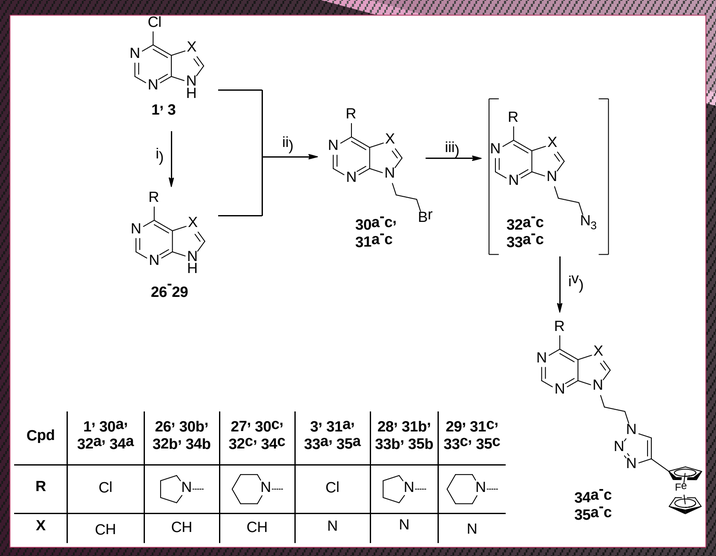
Scheme 1. Synthesis of novel purine and 7-deazapurine derivatives linked to C-4 of 1,2,3-triazolyl ferrocene. Reagents and conditions: (i) propargyl bromide, NaH, DMF, Ar atmosphere, 60 °C, 24 h; (ii) corresponding azide, $Cu(OAc)_2$, dry methanol, room temperature, 24 h.



Cpd	17, 20 23a-23c	18, 21 24a-24c	19, 22 25a-25c
R_1	H	H	I
X	N	CH	CH

R_2 a= b= c=

Scheme 2. Synthesis of novel benzimidazole and indole derivatives linked to C-4 of 1,2,3-triazolyl ferrocene. Reagents and conditions: (i) propargyl bromide, NaH, DMF, Ar atmosphere, 60 °C, 24 h; (ii) corresponding azide, $Cu(OAc)_2$, dry methanol, room temperature, 24 h.



Cpd	1, 30a, 32a, 34a	26, 30b, 32b, 34b	27, 30c, 32c, 34c	3, 31a, 33a, 35a	28, 31b, 33b, 35b	29, 31c, 33c, 35c
R	Cl	N	N	Cl	N	N
X	CH	CH	CH	N	N	N

Scheme 3. Synthesis of novel purine and 7-deazapurine derivatives linked to N-1 of 1,2,3-triazolyl ferrocene. Reagents and conditions: (i) heterocyclic base, corresponding amine, KOH, H_2O, MW: 100 °C, 400 W; (ii) heterocyclic base, NaH, 1,2-dibromoethane, dry DMF, room temperature, 24 h; (iii) *N*-propargylated heterocyclic base, NaN_3, acetone, 60 °C, 12 h; (iv) corresponding terminal azide, $Cu(OAc)_2$, ethynylferrocene, dry methanol, room temperature, 12 h.

Table 1. In vitro growth inhibitory effects of synthesized compounds (**11a–11c, 12a–12c, 13a–13c, 14a–14c, 15a–15c, 16a, 23a–23c, 24a–24c, 25a–25c, 34a-34c, 35a-35c)** on selected tumor cell lines and their measured kinetic solubility range and chrom logD

Compd	IC_{50} (μM) SW620	CFPAC-1	HepG2	HeLa	HFF	Kinetic solubility (μM)	Chrom logD
11a	31.58 ±0.84	32.91 ±2.46	41.96 ±5.93	38.25 ±2.30	8.57 ±1.95	<1	5.62
11b	31.96 ±0.34	31.26 ±4.51	34.52 ±1.36	31.38 ±1.04	8.88 ±3.46	3-10	5.94
11c	**9.07 ±1.21**	35.03 ±5.44	31.94 ±4.32	27.74 ±3.23	5.89 ±1.88	3-10	5.62
12a	>100	>100	>100	>100	56.63 ±15.60	30-100	2.85
12b	74.95 ±8.93	70.15 ±6.50	68.60 ±9.15	76.40 ±3.41	18.44 ±2.64	>100	3.09
12c	>100	>100	>100	>100	54.50 ±4.96	3-10	2.64
13a	**14.38 ±3.02**	22.83 ±4.31	19.89 ±5.12	25.20 ±4.31	3.98 ±0.17	>100	4.27
13b	36.56 ±3.42	43.83 ±10.94	38.77 ±10.11	40.34 ±4.52	6.79 ±1.06	30-100	4.64
13c	51.78 ±1.90	54.67 ±5.13	54.66 ±3.21	46.78 ±6.72	6.77 ±2.78	>100	4.19
14a	60.23 ±24.36	49.47 ±5.03	60.13 ±5.73	71.43 ±16.06	>100	30-100	3.62
14b	98.74 ±19.71	67.36 9.12	77.10 ±5.92	>100	11.53 ±5.09	30-100	3.90
14c	81.33 ±1.23	90.50 ±1.83	>100	>100	24.55 ±4.45	<1	3.50
15a	27.21 ±5.30	34.39 ±0.43	36.01 ±1.81	28.62 ±8.28	13.80 ±4.17	>100	3.82
15b	**15.50 ±3.24**	28.59 ±2.68	37.11 ±1.84	28.97 ±0.09	0.88 ±1.06	>100	4.15
15c	45.47 ±4.52	72.25 ±13.70	81.71 ±17.05	69.25 ±25.47	29.59 ±4.28	>100	3.68
16a	>100	>100	>100	>100	5.30 ±5.36	<1	3.37
23a	96.98 ±13.55	64.57 ±12.18	59.90 ±10.94	>100	23.83 ±3.32	10-30	4.56
23b	>100	68.03 ±13.67	83.6 0±8.61	89.63 ±13.65	31.80 ±9.81	30-100	4.84
23c	>100	>100	>100	>100	>100	30-100	4.45
24a	61.99 ±8.79	78.05 ±6.61	53.55 ±5.05	68.72 ±2.91	16.72 ±8.61	3-10	>6.57
24b	>100	87.63 ±1.79	>100	89.64 ±7.64	>100	3-10	>6.57
24c	>100	84.54 ±15.76	90.87 ±11.51	88.12 ±1.18	>100	3-10	>6.57
25a	>100	>100	>100	78.52 ±9.36	>100	1-3	>6.57
25b	>100	>100	>100	>100	>100	1-3	>6.57
25c	>100	>100	71.79 ±3.44	79.44 ±1.82	4.75 ±1.70	1-3	>6.57
34a	>100	>100	>100	>100	>100	10-30	4.92
34b	>100	>100	>100	>100	>100	10-30	4.02
34c	57.65 ±6.55	45.78 ±9.11	57.61 ±13.80	71.12 ±15.89	20.16 ±1.54	10-30	5.49
35a	78.18 ±5.69	65.28 ±6.23	48.09 ±7.85	63.00 ±6.06	6.00 ±2.99	10-30	3.70
35b	>100	>100	>100	>100	>100	30-100	3.58
35c	>100	>100	>100	>100	6.83 ±2.17	10-30	5.02

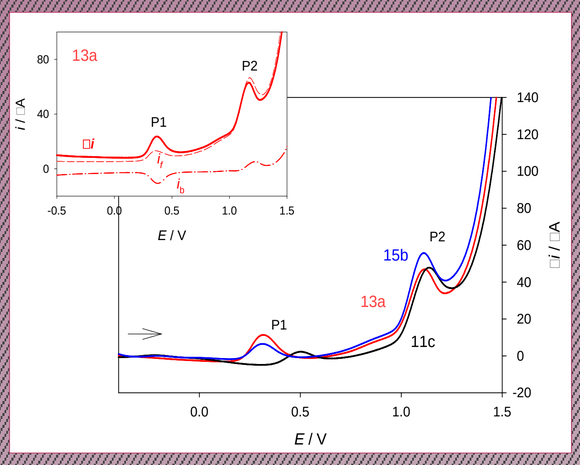


Figure 1. Square-wave voltammograms for the oxidation of **11c, 13a** and **15b** in 0.5 mol/L $NaClO_4$ at pH 9.

Table 2. The permeability (Papp) values of 11a-c, 13a, 15a-b (with and without P-gp inhibitor) and stability values of 11a-c, 13a, 15a-b in liver microsomes of various species

Compd	P_{app} (AB) (x10^{-6} cm/s)	P_{app} (BA) (x10^{-6} cm/s)	Efflux ratio (BA/AB)	P_{app} (AB) (x10^{-6} cm/s)	P_{app} (BA) (x10^{-6} cm/s)	Efflux ratio (BA/AB)	*in vivo* CL_h (% LBF)	
	without P-gp inhibitor			with P-gp inhibitor			Mouse	Human
11a	8.4	18.0	2.2	29.4	21.1	0.7	98	90
11b	11.8	17.4	1.5	19.7	20.5	1.1	100	97
11c	5.3	14.9	2.9	15.5	16.5	1.1	99	95
13a	8.3	23.8	2.9	29.8	25.3	0.9	98	65
15a	4.8	33.5	7.0	16.9	13.7	0.8	97	58
15b	3.2	33.2	10.7	11.7	12.9	1.1	99	79

Biological evaluation

Among all tested compounds, derivatives **11a–11c, 13a–13c** and **15a–15c** exhibited the best inhibitory effects, particularly on colorectal adenocarcinoma (SW620) cells (Table 1). *N*-9 and *N*-7 isomers of 6-chloropurine **13a** and **15a** containing ferrocenylmethylene unit show high solubility, good permeability and moderate stability. Although **15b** showed good physicochemical properties, this compound, together with **11a–11c**, is metabolically unstable (Table 2). Voltammetric analysis showed that the oxidation of ferrocenyl group in **11c** requires higher potential compared to oxidation in **13a** and **15b**, that may contribute to higher potential antioxidant properties of **13a** and **15b** containing ferrocenylmethylene moiety than that of **11c** (Fig. 1).

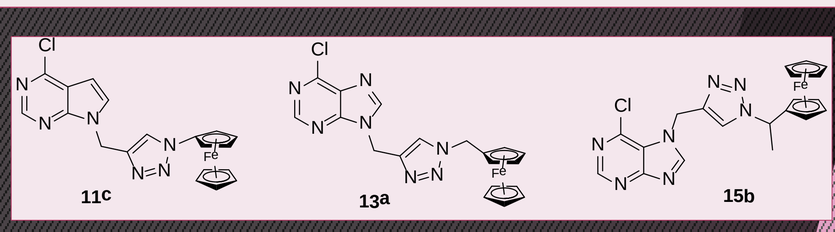


Figure 2. Structures of most potent derivatives **11c, 13a** and **15b**

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