Epigenetic drug discovery at the academia

CRC 992 Medical Epigenetics

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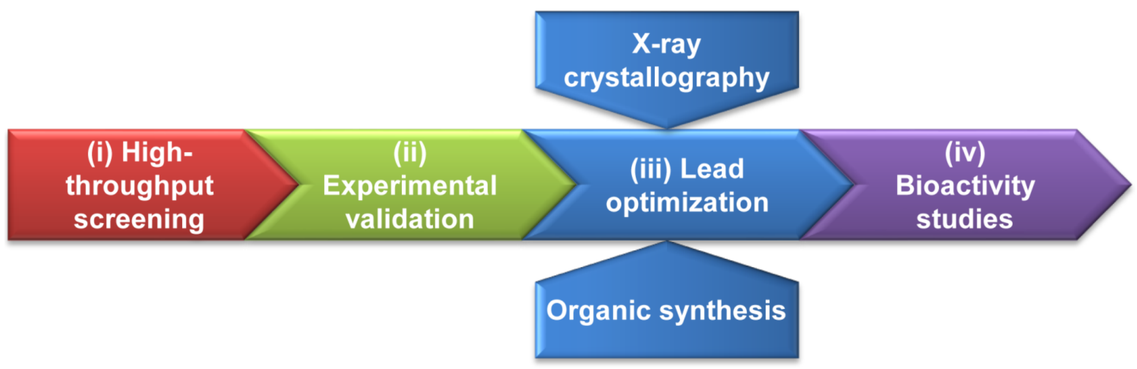
General overview

Bromodomains are epigenetic mark 'readers' that specifically recognize ε-*N*-acetylated lysine residues. Their potential as **therapeutic targets** has attracted much attention due to their implication as regulators of disease-relevant gene expression. BET is the most studied bromodomain subfamily so far, and it has been characterized as a key determinant in several types of cancer, particularly **leukemia** [1,2]. We have performed a structure-based virtual screening and identified 4-acyl pyrroles as a novel class of bromodomain inhibitors [3].

Conclusions

4-acyl pyrrole derivatives represent a novel class of potent BET bromodomain inhibitors. We show that rational design allows for the modulation of human bromodomains beyond the BET subfamily.
The established collaborative platform presented herein, built within the CRC 992 Medical Epigenetics, is an **efficient academic resource** for the development of inhibitors of disease-relevant epigenetic targets. As a result, we are currently expanding the scope of our work to other less studied epigenetic regulators.

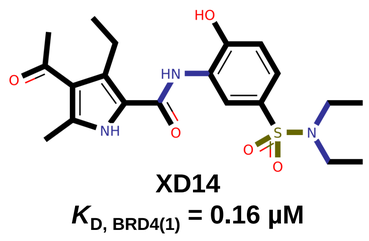
Academic drug discovery pipeline



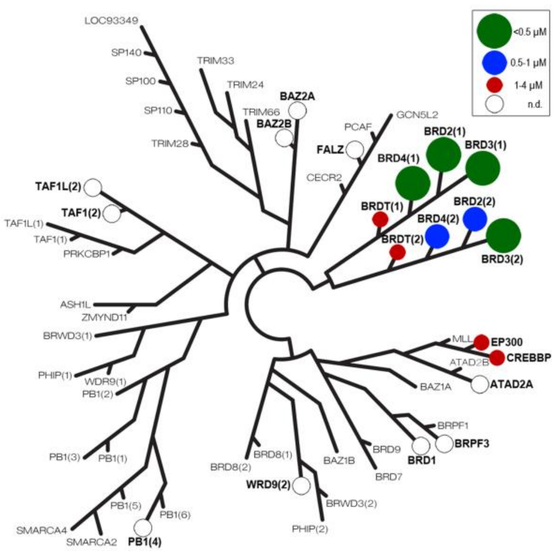
(i) Few tens of promising candidates are selected from millions of chemicals using efficient *in silico* techniques.
(ii) Their binding affinity is evaluated experimentally by ITC, α-screen, ThermoFluor, etc.
(iii) Validated lead compounds are subjected to further structure-based optimization.
(iv) The biological activity of the hits is explored in *in vitro* cellular assays.

Discovery of XD14

We successfully applied the presented academic pipeline and identified the potent BET bromodomain inhibitor **XD14**, which features a novel 4-acyl pyrrole core.



XD14
$K_{D,\ BRD4(1)}$ = 0.16 µM



Bromodomain **selectivity profiling** shows that XD14 inhibits the BET bromodomain family, as well as the bromodomains of CREBBP and EP300.

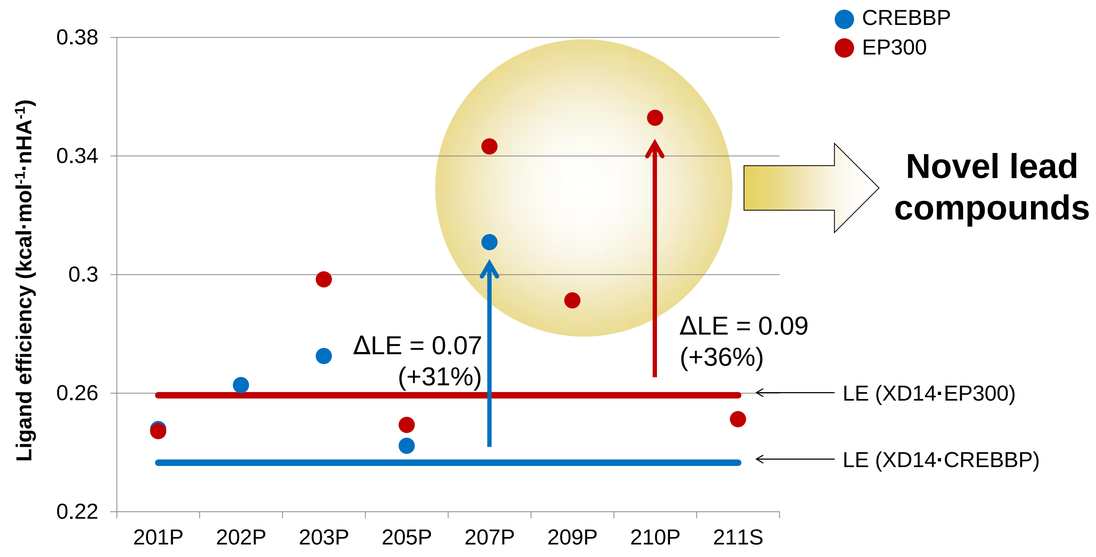
XD14 exhibits potent, specific inhibition of cell proliferation of leukemic cell lines HL60 and SR. The compound is not toxic neither in cellular nor animal models.

	Cell line	GI (%)
Breast cancer	T-47D	31.0
	HS 578T	18.0
	BT-549	15.3
	MCF7	15.1
	MDA-MB-468	7.7
	MDA-MB-231/ATCC	2.4
Colon cancer	KM12	24.3
	HCT-116	10.9
	SW-620	7.3
	HCT-15	4.7
	HT29	1.2
	COLO 205	-4.7
Leukemia	HL-60(TB)	65.0
	SR	64.2
	K-562	42.0
	MOLT-4	34.8
	CCRF-CEM	23.0
	RPMI-8226	21.8
Melanoma	UACC-62	23.5
	M14	15.2
	SK-MEL-5	12.9
	UACC-257	10.3
	MDA-MB-435	9.6
	SK-MEL-2	9.1
	SK-MEL-28	7.2
	MALME-3M	4.7
Prostate cancer	PC-3	17.8
	DU-145	2.6

	Cell line	GI (%)
CNS cancer	SNB-75	29.4
	SF-295	10.0
	U251	6.6
	SNB-19	1.3
	SF-268	0.2
Non-small cell lung cancer	NCI-H522	42.4
	NCI-H322M	25.4
	HOP-92	15.8
	NCI-H226	9.7
	NCI-H23	8.9
	HOP-62	5.7
	A549/ATCC	4.1
	NCI-H460	3.2
Ovarian cancer	IGROV1	27.3
	OVCAR-4	14.8
	OVCAR-3	11.2
	OVCAR-8	6.7
	SK-OV-3	6.4
	NCI/ADR-RES	3.2
	OVCAR-5	-6.7
Renal cancer	UO-31	31.3
	786-0	15.6
	A498	14.5
	SN12C	9.4
	CAKI-1	6.1
	ACHN	3.6
	TK-10	-0.7
	RXF 393	-1.0

Design of CREBBP and EP300 inhibitors

We have used the **4-acyl pyrrole** scaffold to rationally design compounds with improved affinity towards **CREBBP** and **EP300** compared to XD14:



[1] R.K. Prinjha, J. Witherington, K. Lee, "Place your BETs: the therapeutic potential of BET bromodomains", *Trends Pharmacol. Sci.*, 2012, 33(3), 146-53.

[2] X. Lucas, S. Günther, "Targeting the BET family for the treatment of leukemia", *Epigenomics* (in press).

[3] X. Lucas, D. Wohlwend, M. Hügle, K. Schmidtkunz, S. Gerhardt, R. Schüle, M. Jung, O. Einsle, S. Günther, "4-acyl pyrroles: mimicking acetylated lysines in histone code reading", *Angew. Chem. Int. Ed. Engl.*, 2013, 52(52), 14055-9.

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