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amines: Biological and Clinical Perspectives

Interactions Between New Polyamine Analogs and Bovine Serum Amine Oxidase

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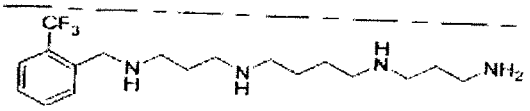
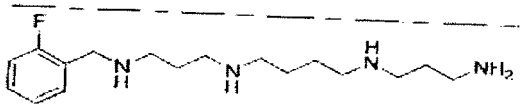
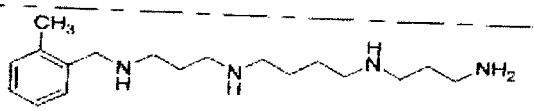
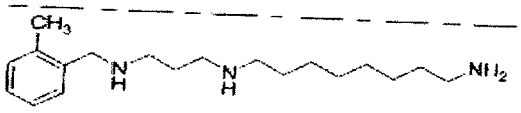
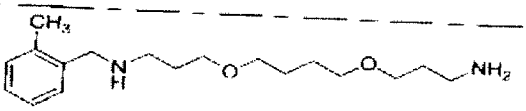
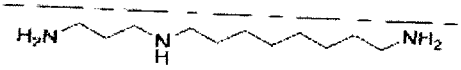
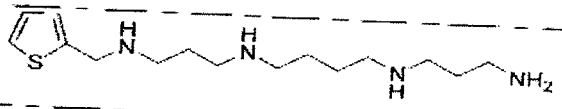
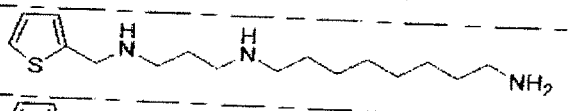
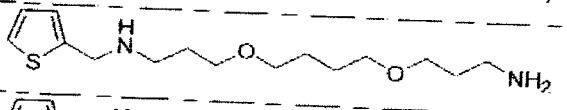
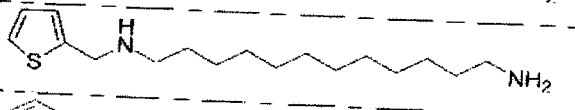
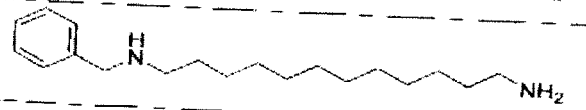
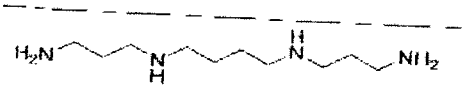
Natural polyamines putrescine, spermidine and spermine are ubiquitous polycationic compounds present in significant amounts in nearly every prokaryotic and eukaryotic cell type. Spermidine and spermine primarily exist in aqueous solution at pH 7.4 as fully protonated polycations (1). Such ubiquitous chemical entities play an important role in cell growth and proliferation, in the synthesis of proteins and nucleic acids, in both normal and cancer cells.

In order to develop new amino oxidase (AO) spermine-based ligands, several spermine analogs were synthesized with the aim to improve their enzymatic oxidative deamination. Kinetic observations were carried out in buffer phosphate 0.01M, at pH 7.4-7.6 (1,2), in order to perform cytotoxicity studies on cancer cells using BSAO (bovine serum amine oxidase) enzyme in the presence of polyamine analogs. BSAO catalyzes, in the presence of O_2, the oxidative deamination of spermine, spermidine and their analogs, providing the formation of the products: H_2O_2 and aldehyde(s). The kinetic assays were performed using the new synthesized compounds in the range between 0.05-1 mM in the presence of BSAO. As shown in Table 1 the addition of thiophene group on terminal amines and substitution of the inner nitrogen atoms of spermine with oxygen atoms, allows obtaining kinetic parameter values approximately similar to those of spermine (Table 1).

The improvement of the kinetic parameters at pH 7.6, respect to pH 7.4 ones, is probably due to a better interaction between amine-oxidase with polyamine analogs. Such as interaction leads to the formation of the Schiff base (1,2). Using our expertise on AutoDock program (3,4), structure based (SB) studies were performed o assess, both the binding mode of the reported analogs and the role of the involved residues; furthermore, using he AutoDock side-chain flexibility feature the topaquinone (TPQ) cofactor behaviour was analysed in depth to ave an overview of the enzymatic process and determine what chemical characteristics should be considered for rational drug design. Knowledge of these requirements could be useful to predict, moreover, the binding rientation of new highly potential BSAO substrates, details will be reported. As future perspective, these

Table 1. Kinetic parameters of some polyamine analogs determined in buffer phosphate 0.01 M at pH 7.6

	Polyamines analogs	Vmax µM/s	K_m (µM)	k_c (s^{-1})
	BD 32	0.47	17	0,61
	BD 33	0.40	38	0,52
	BD 9	0,13	7,2	0,25
	EB 20	0.33	97	0,04
	EB 22	0.36	3,78	0,47
	EB 23	0.24	10	0,31
	BD 28	0.29	6	0.59
	EB 27	0.36	8,37	0,46
	EB 26	0.36	7	0,46
	AN 224	0.15	5	0.33
	BZA Diado	0,13	14	0.29
	Physiological polyamines	Vmax µM/s	K_m (µM)	K_C (s^{-1})
	Spm	0.65	3.15	1,5

molecules will be assayed alone or in combination with BSAO on several cancer cells, with the aim to evalu their cytotoxic effects that could be taken into consideration as new approach in anti-cancer therapy.

References:

1. S. Saccoccio, A.Minarini, A.Milelli, V.Tumiatti, G. Tempera, N.Viceconte, R.Stevanato and E.Agostinelli (2010) Oxidative deamination of new polyamine analogs by bovine serum amine oxidase: a kinetic study. 2nd International Conference on the Role of Polyamines and their Analogs in Cancer and other Diseases Tivoli (Rome, Italy) December 1-6.

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