Studies on the interaction of mycotoxins and macrocycles by molecular modelling

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ABSTRACT

The goal of our work is to develope a selective sensor to detect the gene wrecking and carcinogenic mycotoxin molecules [1,2]. Hence our research group applies functionalized gold nanoparticles and thin films to measure these analytes. In this investigation modified thiolated macrocycle molecules (mainly cyclodextrins) were applied to functionalise the gold surfaces on the nanoparticles.

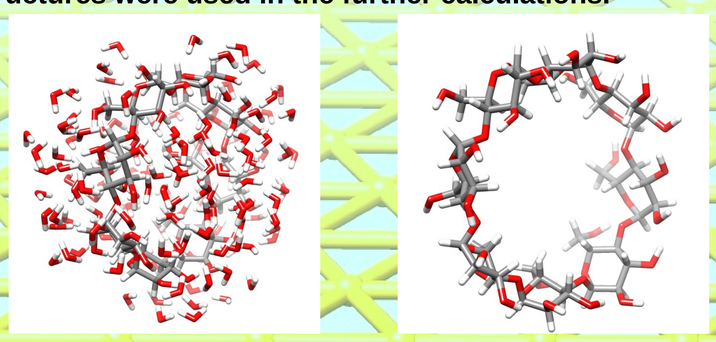
The cyclodextrins containes α-D-glucopyranoside unites, which connect to a ring with 1,4 glycosidic bonds. The best known three cyclodextrins are the α-, β- and γ-cyclodextrin, these form six, seven and eight glucopyranose molecules, respectively. In the inner cavity, there are hydrogen atoms and the oxygens of the 1,4 glycosidic bonds. The hydroxyl groups are placed on the flange of the hoop. So, the inner cavity of the cyclodextins is hydrophobic, and the outer surface is hydrophilic. This property makes cyclodextrin molecules able to form inclusion complexes with hydrophobic molecules like the aflatoxins. The hydrophobic character and the hydrogen donor hydroxyl groups warrants the relatively strong binding. The modification of cyclodextrins with the suitable chemical groups gives the selectivity of the sensors.

To choose of the appropriate modified cyclodextrin molecules and predict the binding affinities to mycotoxins, we investigated the complexes with molecular modelling.

To improve our fundamental understanding of the nanomaterials functionalized with macrocycle molecules, we use the tools of the molecular docking, molecular mechanics (MM) and semiempirical quantum chemistry methods.

Keywords: sensors, mycotoxins, macrocycles, docking

To prepare the cyclodextrin structures, we optimized them with PM6-DH+/COSMO method, added an r = 12 Å water sphere. The cyclodextrin coordinates were fixed and the sphere was pre optimized with the SP4 force filed (VegaZZ). Finally the cyclodextrin in the sphere was optimized with the PM6-DH+ method. These cyclodextrin structures were used in the further calculations.



The gamma cyclodextrin molecule.

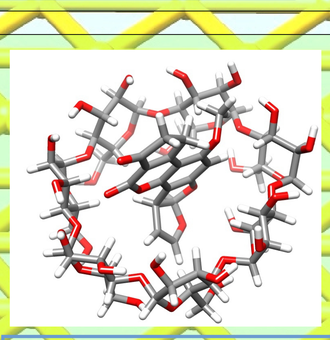
No.	Produced by:	Occurrence:	Group:	Toxin name:
1	Aspergillus species of fungi, such as A. flavus and A. Parasiticus	cotton, peanuts, spices, pistachios, maize	Aflatoxins	Aflatoxin B1
2				Aflatoxin B2
3				Aflatoxin G1
4				Aflatoxin G2
5				Aflatoxin M1
6				Aflatoxin M2
7	metabolite			Aflatoxin Q1
8				Aflatoxin P1
9				Aflatoxin D1
10	Penicillium and Aspergillus species	beverages: beer, wine	Ochratoxins	Ochratoxin A
11				Ochratoxin B
12				Ochratoxin C
13				Ochratoxin TA
14	Over a dozen species of Penicillium and several species of Aspergillus	wheat, rice, corn, barley, oats, rye, and food colored with Monascus pigment	Citrinin	Citrinin
15	Ergot or ergot fungi refers to a group of fungi of the genus Claviceps.	infecting the grain of developing cereals such as wheat and maize	Ergot Alkaloids	ergotamine
16				ergocornine
17				ergocristine
18				ergocryptine
19				agroclavine
20				ergometrine
21	Aspergillus and Penicillium	rotten fruits (mainly in apple)	Patulin	Patulin
22	Over 50 species of Fusarium	infecting the grain of developing cereals such as wheat and maize	Fusarium toxins / fumonisins	Fumonisin B1
23				Fumonisin B2
24			Fusarium toxins / A Trichothecene	T-2 toxin
25				HT-2 toxin
26				diacetoxyscirpenol (DAS)
27			Fusarium toxins / B Trichothecene	deoxynivalenol (DON)
28				nivalenol
29				15-acetyldeoxynivalenol
30				3-acetyldeoxynivalenol
31			Fusarium toxins / Zearalenone	Zearalenone

The mycotoxin molecules used during the calculations, and the docked molecules below.

We used random conformation search (SP4 force field, VegaZZ), and the best conformations were optimized with the new PM7 semiempirical method implemented in the MOPAC2012 software, using the COSMO implicit solvatation modell.

On the right huge table we summarized te results of the AutoDock VINA virtual screening. We used Gasteiger charges.

These quantities are relative scores from the AutoDock VINA score. The colour indicates the strength of the binding. (yellow: above -20.0 ; light red: between -20.0 and -25.0; red: between -25.0 and -30.0 and white: below -30.0) Molecules in the white cells binds strongest.



The complex of the beta-cyclodextrin and Aflatoxin B1 molecule. The aflatoxins binds in this way, the methoxy group do not moves to the cavity.

No.	Toxin name:	Alpha cyclodextrin		Beta cyclodextrin		Gamma cyclodextrin	
		AutoDock Vina / {PM6-DH+/explicit water}	AutoDock Vina / {PM6-DH+/COSMO}	AutoDock Vina / {PM6-DH+/explicit water}	AutoDock Vina / {PM6-DH+/COSMO}	AutoDock Vina / {PM6-DH+/explicit water}	AutoDock Vina / {PM6-DH+/COSMO}
1	Aflatoxin B1	-23.0	-23.4	-29.3	-28.5	-26.4	-25.9
2	Aflatoxin B2	-23.4	-23.4	-29.3	-28.5	-26.4	-25.9
3	Aflatoxin G1	-23.8	-23.4	-28.5	-28.0	-27.6	-26.8
4	Aflatoxin G2	-24.3	-23.8	-29.7	-28.0	-27.6	-25.5
5	Aflatoxin M1	-24.3	-24.3	-29.3	-28.0	-27.6	-26.4
6	Aflatoxin M2	-24.3	-23.8	-29.3	-28.0	-27.6	-26.4
7	Aflatoxin Q1	-23.8	-23.4	-28.5	-27.2	-27.6	-25.1
8	Aflatoxin P1	-23.4	-23.4	-30.1	-28.9	-27.6	-26.8
9	Aflatoxin D1	-24.3	-20.9	-28.0	-26.8	-24.7	-23.4
10	Ochratoxin A	-22.6	-21.3	-26.4	-25.9	-25.1	-23.8
11	Ochratoxin B	-22.6	-21.3	-27.6	-25.9	-24.3	-23.8
12	Ochratoxin C	-20.1	-18.8	-25.1	-23.4	-23.8	-23.8
13	Ochratoxin TA	-23.8	-20.9	-26.8	-25.1	-25.9	-24.7
14	Citrinin	-20.9	-21.3	-24.7	-20.9	-23.0	-22.2
15	ergotamine	-24.7	-23.4	-29.7	-28.5	-31.8	-30.5
16	ergocornine	-24.7	-23.4	-26.8	-25.5	-28.9	-30.1
17	ergocristine	-25.1	-24.3	-27.2	-27.2	-28.0	-28.0
18	ergocryptine	-22.6	-22.2	-26.8	-26.8	-29.7	-28.0
19	agroclavine	-20.5	-18.8	-23.8	-20.9	-24.3	-22.6
20	ergometrine	-21.3	-20.9	-25.5	-22.6	-25.1	-23.4
21	Patulin	-23.0	-20.5	-21.3	-19.7	-16.7	-16.3
22	Fumonisin B1	-16.7	-16.7	-18.8	-17.2	-18.0	-15.9
23	Fumonisin B2	-16.3	-16.3	-18.4	-18.8	-18.4	-17.6
24	T-2 toxin	-20.1	-20.5	-23.8	-22.6	-25.5	-22.6
25	HT-2 toxin	-20.5	-20.9	-24.3	-26.4	-25.9	-23.4
26	diacetoxyscirpenol (DAS)	-21.8	-21.3	-26.4	-23.4	-25.9	-24.7
27	deoxynivalenol (DON)	-23.8	-22.6	-30.1	-27.6	-26.4	-25.1
28	nivalenol	-23.8	-24.3	-30.1	-28.0	-28.5	-25.5
29	15-acetyldeoxynivalenol	-24.3	-22.6	-27.6	-27.2	-27.2	-25.5
30	3-acetyldeoxynivalenol	-21.3	-21.3	-25.9	-25.5	-27.2	-25.9
31	Zearalenone	-24.7	-22.6	-30.1	-28.5	-26.4	-25.9

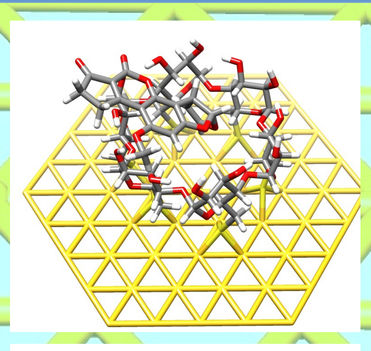
The best docked poses were optimized with the semiempirical PM6-DH+, and PM6-DH2 dispersion and hydrogen bond interaction corrigated methods, with the COSMO implicit solvation model. The energies shows the size selective property of CDs.

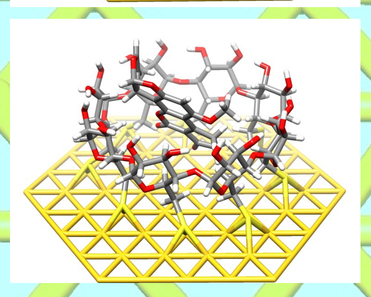
			PM6-DH2, COSMO	PM6-DH+, COSMO	PM6-DH2	PM6-DH
		aflatoxin	ΔH^{o}_{b} / kJ/mol	ΔH^{o}_{b} / kJ/mol	ΔH ratio	ΔH ratio
ACD	1	Aflatoxin B1	-97.12	-94.62	1	1
	2	Aflatoxin B2	-104.01	-86.41	1	1
	3	Aflatoxin G1	-105.28	-103.36	1	1
	4	Aflatoxin G2	-90.78	-104.75	1	1
	5	Aflatoxin M1	-119.89	-125.43	1	1
	6	Aflatoxin M2	-111.14	-106.29	1	1
	7	Aflatoxin Q1	-103.96	-104.57	1	1
	8	Aflatoxin P1	-100.17	-99.56	1	1
BCD	1	Aflatoxin B1	-121.11	-103.67	1.25	1.10
	2	Aflatoxin B2	-125.01	-127.83	1.20	1.48
	3	Aflatoxin G1	-121.68	-83.67	1.16	0.81
	4	Aflatoxin G2	-121.36	-121.98	1.34	1.16
	5	Aflatoxin M1	-137.94	-154.89	1.15	1.23
	6	Aflatoxin M2	-135.01	-122.76	1.21	1.15
	7	Aflatoxin Q1	-113.36	-102.82	1.09	0.98
	8	Aflatoxin P1	-121.99	-104.75	1.22	1.05
GCD	1	Aflatoxin B1	-70.05	-60.17	0.72	0.64
	2	Aflatoxin B2	-70.93	-59.81	0.68	0.69
	3	Aflatoxin G1	-105.38	-66.60	1.00	0.64
	4	Aflatoxin G2	-106.95	-52.01	1.18	0.50
	5	Aflatoxin M1	-100.98	-104.70	0.84	0.83
	6	Aflatoxin M2	-124.71	-101.23	1.12	0.95
	7	Aflatoxin Q1	-134.57	-113.13	1.29	1.08
	8	Aflatoxin P1	-103.92	-111.28	1.04	1.12

From the ratio of the standard binding enthalpy between the ACD, BCD and GCD, it is evident that the BCD binds the strongest the aflatoxin molecules. It is good agreement with the docking experiments, and with the experimental measurements.

We investigated the binding of the thiolated cyclodextrins on gold surface. The gold surface model was a gold (111) lattice plane (one layer gold atoms). The cyclodextrins were positioned on the center of the plane and optimized with PM6-DH+ semiempirical method. Then the aflatoxin molecules were docked to the optimized strucrures with the UCSF DOCK and AutoDock VINA softwares. The PM6 charges were used with UCSF DOCK. In the case of autodock the smaller diameter site was closed with a plane of carbon atoms.

			cyclodextrine on gold	cyclodextrine on gold	cyclodextrine on gold
		toxin	Continous Score	Hawkins GB/SA Score	Autodock VINA Score
ACD	1	Aflatoxin B1	-22.8	-14.89	-22.18
	2	Aflatoxin B2	-23.25	-15.18	-22.59
	3	Aflatoxin G1	-23.38	-16.07	-22.59
	4	Aflatoxin G2	-23.6	-16.36	-23.01
	5	Aflatoxin M1	-24.46	-14.41	-23.43
	6	Aflatoxin M2	-25.29	-15.67	-22.59
	7	Aflatoxin Q1	-23.84	-14.74	-23.01
	8	Aflatoxin P1	-22.57	-13.78	-22.59
BCD	1	Aflatoxin B1	-28.04	-18.53	-30.54
	2	Aflatoxin B2	-28.92	-20.08	-30.12
	3	Aflatoxin G1	-28.63	-18.55	-31.38
	4	Aflatoxin G2	-29.75	-20.88	-30.54
	5	Aflatoxin M1	-30.28	-18.46	-30.54
	6	Aflatoxin M2	-30.98	-18.53	-30.54
	7	Aflatoxin Q1	-27.41	-18.21	-29.71
	8	Aflatoxin P1	-27.63	-17.87	-30.54
GCD	1	Aflatoxin B1	-30.53	-23.02	-28.87
	2	Aflatoxin B2	-29.69	-20.89	-28.87
	3	Aflatoxin G1	-28.58	-20.04	-29.29
	4	Aflatoxin G2	-31.12	-21.94	-28.87
	5	Aflatoxin M1	-30.38	-20.89	-29.71
	6	Aflatoxin M2	-32.75	-22.02	-29.29
	7	Aflatoxin Q1	-28.82	-19.92	-28.45
	8	Aflatoxin P1	-30.66	-21.14	-29.29





On the left side pictures (ACD and GCD on gold model, with Aflatoxin B1 molecule) can be seen, that the Aflatoxin molecules are too large to bind to the alpha-cycldextrin cavity. In the gamma-cyclodextrin, the ligand is too far from the wall of te cavity, so the binding is weak. The beta-cyclodextrine has the optimal size to bind Aflatoxin molecules. The AutoDock VINA scores are show this too in the table above. We need more shopisticated caltulations to explain the differences between the binding of Aflatoxin molecules to modified gold surfaces.

CONCLUSION

- **We docked 31 different dangerous mycotoxin molecules to the alpha, beta, gamma cyclodextrines.**
- **We calculated the standard binding entalphy of the cyclodextrin – aflatoxin complexes.**
- **We found a good agreement between the AutoDock VINA scores and the calculated standard binding enthalpies.**
- **We started to investigate the aflatoxin adsorption, and the mycotoxin - cyclodextrin interaction with semiempirical quantum chemistry and molecular docking.**
- **These calculations are the early attempt, to understand these systems.**
- **To get better result we need to use MD (REMD) and DFT methods, to investigate the conformation space of modified cyclodextrines and the thiol-gold interactions.**

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