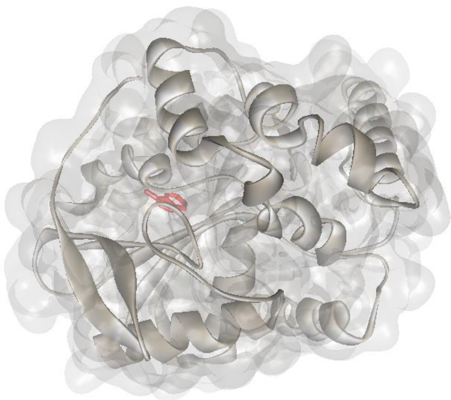
Task 3

23.11.2012

Mutant identification

Your first real task in biotechnological laboratories BTP (Biotechnology for purification) is to distinguish between two mutant variants X1 and X2 of a protein which is used for the decomposition of toxic substances in drinking water. Accidentally, labels of reaction reservoirs with enzymes were confused. You have found out that both types of enzyme contain one tryptophan. Additionally, you know that tryptophan in case of mutant X1 is placed much closer to the surface and therefore it is in contact with surrounding solution more than in the case of the second variant of protein X2.

X1



X2

The addressing of this task is important for the supply of drinking water to residents in an area affected by drought. You can use your knowledge of fluorescence quenching for the problem solution. You remember that the protein with tryptophan close to the surface can be determined from the dependence of fluorescence intensity on the concentration of the quencher. For quenching fluorophore, the basic Stern-Volmer equation can be applied:

$$\frac{F_0}{F} = 1 + K_{SV}[Q]$$

where F_0 is fluorescence intensity in the absence of quencher, F is the fluorescence intensity in the presence of quencher, K_{SV} is Stern-Volmer constant and [Q] is the concentration of the quencher.
You have carried out measurements of fluorescence intensity of proteins taken from reservoirs A and B. The fluorescence intensity was measured in the absence of the quencher. Then you measured fluorescence drop after gradual addition of quencher (acrylamide). Your obtained values of fluorescence intensity are in the table below.
Plot the dependence of relative fluorescence intensity decrease on the acrylamide concentration in the form of Stern-Volmer graph and answer the following questions:

1. **Is acrylamide dynamic or static quencher?**
2. **What are the constants K_{SV} corresponding to each mutant variants of the enzyme?**
3. **Determine in which reservoir the enzyme X1 is located.**

Please send me your answers together with Stern-Volmer plot for A and B via email.
Correct answer = 0.5 point

		reservoir	Acrylamide concentration [M]					
			0	**0.1**	**0.2**	**0.3**	**0.4**	**0.5**
1	Bencúrová, Petra	**A**	944	911	891	870	853	834
		B	944	794	697	621	560	510
2	Dabravolski, Siarhei	**A**	977	943	922	901	883	864
		B	977	822	722	643	580	528
3	Dubec, Vít	**A**	940	908	887	867	850	831
		B	940	791	694	619	558	508
4	Dudová, Zdenka	**A**	951	918	898	877	860	841
		B	951	800	703	626	565	514
5	Dvořák, Jan	**A**	960	927	907	886	868	849
		B	960	808	709	632	570	519
6	Fabišik, Matej	**A**	986	952	931	910	891	872
		B	986	830	729	649	585	533
7	Fedorko, Jan	**A**	938	906	886	865	848	829
		B	938	790	693	617	557	507
8	Fialová, Martina	**A**	957	924	903	882	865	846
		B	957	805	707	630	568	517
9	Holek, Michal	**A**	986	952	931	910	891	872
		B	986	830	729	649	585	533
10	Kočka, Martin	**A**	975	942	921	899	881	862
		B	975	821	720	642	579	527
11	Míka, Matěj	**A**	981	947	926	905	886	867
		B	981	825	724	646	582	530
12	Obacz, Joanna Agnieszka	**A**	957	924	903	882	865	846
		B	957	805	707	630	568	517
13	Partyka, Jan	**A**	936	904	884	864	846	828
		B	936	788	692	616	556	506
14	Přikrylová, Terézia	**A**	994	959	938	917	898	878
		B	994	836	734	654	590	537
15	Rájecký, Michal	**A**	984	950	929	908	890	870
		B	984	829	727	648	584	532
16	Reichman, Pavel	**A**	962	929	908	887	870	851
		B	962	810	711	633	571	520
17	Sochorová, Jana	**A**	964	931	910	889	871	852
		B	964	811	712	635	572	521
18	Škubník, Karel	**A**	953	920	900	879	861	842
		B	953	802	704	627	566	515
19	Tylichová, Zuzana	**A**	970	936	915	894	876	857
		B	970	816	716	638	575	524