A quick comparison of blocking effectiveness of SophoMer™ F10 and BSA

Introduction

Two simple systems were used for comparison of the blocking properties of **SophoMer™ F10** and Bovine serum albumin (BSA), which are independent on the variant of immunoassay arrangement. Both utilize a standard 96w immunoassay plates and as a source of signal the Horseradish Peroxidase (HRP) together with Tetramethylbenzidine (TMB). The first approach uses the HRP alone (test A), for the second one a commercial conjugate of HRP with a rabbit polyclonal antibody (test B) is used.

Material

Elisa 96w plates (Greiner, # 705071), HRP (Pierce, # 31490), HRP conjugated Rabbit IgG x beta-lactoglobuline (Elisa Development ltd), TMB (Diarect AG, # TMBW-1000), BSA (Protease free, IVD grade), **SophoMer™ F10**, other chemicals and equipment from common suppliers.

Test A protocol

- Clean 96w polystyrene microwells were used
- Dosing of 200µL HRP 5mg/L in phosphate buffer containing either **SophoMer™ F10** or BSA at three concentrations, 0.1, 0.5 and 2 g/L respectively (each concentration in quadruplets)
- Incubation 16h at RT, no shaking
- Aspiration, washing 4x 350 µL with washing solution containing NaCl and Tween 20
- Dosing 200 mL of TMB solution
- 10 min incubation
- Dosing 50 µL 2M HCL
- OD measurement at 450 nm.

Test B protocol

- Clean 96w polystyrene microwells were used
- Dosing of 200µL HRP conjugated Rabbit IgG x beta-lactoglobuline diluted 1:500 in phosphate buffer containing either **SophoMer™ F10** or BSA at three concentrations, 0.2, 1.0 and 5.0 g/L respectively (each concentration in quadruplets)
- Incubation 120 minutes at RT, no shaking
- Aspiration, washing 4x 350 µL with washing solution containing NaCl and Tween 20
- Dosing 200 µL of TMB solution
- 10 min incubation
- Dosing 50 µL 2M HCL
- OD measurement at 450 nm

Results and discussion

Results of measurement from both protocols including the variation coefficients are presented in the following tables.

Tab 1. Results from protocol A experiments

	BSA / g/L			SophoMer™ F10 / g/L		
	0.1	**0.5**	**2**	**0.1**	**0.5**	**2**
	OD 450					
1	3.023	1.240	0.265	0.581	0.123	0.068
2	3.154	1.145	0.247	0.487	0.119	0.068
3	3.338	1.235	0.470	0.584	0.115	0.069
4	3.198	1.183	0.273	0.562	0.121	0.069
mean	**3.178**	**1.200**	**0.314**	**0.554**	**0.119**	**0.069**
***C.V.* %**	**4.09**	**3.77**	**33.44**	**8.17**	**2.92**	**0.680**

Tab 2. Results from protocol B experiments

	BSA / g/L			SophoMer™ F10 / g/L		
	0.2	**1**	**5**	**0.2**	**1**	**5**
	OD 450					
1	2.024	0.358	0.059	0.224	0.080	0.042
2	1.867	0.363	0.071	0.184	0.068	0.040
3	2.074	0.367	0.069	0.202	0.075	0.041
4	2.099	0.421	0.084	0.218	0.073	0.043
mean	**2.016**	**0.377**	**0.077**	**0.207**	**0.074**	**0.041**
***C.V.* %**	**5.16**	**7.85**	**20.7**	**8.61**	**6.93**	**2.28**

As in the system there is no other source of the signal than the one from non-specific adsorption of HRP or conjugate on the wells, it can be presumed that the lower signal the better prevention of passive sorption of both materials on the previously non treated wells. The signal reduction achieved by using **SophoMer™ F10** is much more significant than the reduction achieved by using the same concentration of BSA. The concentration of **SophoMer™ F10** needed to reach the comparable effect is at least five times lower when compared to BSA and the signal noise level has lower C.V.

Conclusion

In both systems **SophoMer™ F10** showed better blocking performance then classic BSA blocker.