

Test on DNA adsorption to plastic materials - 30 September 2006

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Background

A contamination of plastics with chemicals as well as the loss of samples on the tube walls are two important defects which can have great impact on the quality of biological sample preparation. It has been observed that DNA can bind to polypropylene, and that the interaction of DNA with tube walls induces a change of conformation, which can go as far as complete denaturation. The choice of a product with low binding adsorption quality is therefore of crucial importance. In the following study REMP products (MTP384, STBR96-300, STBR384, STBR-900*) have been tested against three competitor products with regard to DNA adsorption.

Method

A DNA fragment of 120 base pairs is radioactively end-labeled, following standard protocols, using polynucleotide kinase and [γ -³²P]-ATP, and purified by preparative polyacrylamide gel electrophoresis, electroelution, and ethanol precipitation.

The labeled DNA fragment (~0.1 ng per sample, representing ~10 000 cpm) is incubated for 16 hr at 37°C in plastic tubes, either in 10 mM Tris-HCl pH 7.6, 1 mM EDTA, 2.5 M NaCl (high salt conditions) or in 10 mM Tris-HCl pH 7.6, 1 mM EDTA, 100 mM NaCl (medium salt conditions).

After incubation, the liquid is carefully removed and transferred to a second tube. The radioactivity present in both tubes, containing respectively the bound and the unbound fraction, is counted in a scintillation counter, and the percentage of DNA bound to the tube relative to the total DNA input is calculated.

To ensure that the results are statistically significant, from 12 to 34 samples are prepared for each kind of material. For each material the histograms shown represent the number of samples with a given percentage of DNA binding, by intervals of 5% .

Materials tested

- MTP384 : since individual tubes cannot be extracted from these plates, the DNA bound was redissolved in a solution containing a detergent (1% Triton X-100) and transferred to a tube for counting.
- STBR96-300
- STBR384
- STBR96-900*
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* test still ongoing – expected results similar to STBR96-300

Controls

- Competitor 1: standard low-binding tubes, known from previous tests to show extremely low DNA adsorption
- Competitor 2: (product a) : polypropylene tubes widely used in molecular biology laboratories
- Competitor 2: (product b): recently introduced tube advertised as very low DNA binding
- Competitor 3: low-priced polypropylene tubes

Results

Tests were performed at two different ionic conditions.

High salt concentration : 2.5 M NaCl. These are very stringent conditions, as the interactions of DNA with plastic seem to rely mostly on hydrophobic interactions and are strongly stimulated by high ionic strength.

STBR384 and STBR96-300 show extremely little DNA binding, and are comparable Competitor 1 or to Competitor 2 (Product b). The very slight differences between all these tubes may not be significant in terms of DNA adsorption, but may rather reflect the differences in the geometries of the bottoms of the tubes, as the presence of sharp angles makes it more difficult to remove absolutely all the liquid.

MTP384 present a moderate DNA binding, with most of the samples averaging 20% while a few samples present a strong binding

Summary : under high salt conditions, STBR96-300 and STBR384 are as good as Competitor 1 tubes that we use as reference, (which are equivalent to Competitor 2 (Product b)). MTP384 are better than average.

Medium salt concentration : 0.1 M NaCl, i.e. salt conditions that are nearly physiological.

Under such conditions differences are less striking, as DNA adsorption never reaches very high values. However, here again STBR96-300 and STBR384 are excellent and equivalent to Competitor 1 or Competitor 2 (Product b). MTP384 shows some DNA adsorption, to an extent that does not seem to be significantly different from what is observed for Competitor 2 (Product a) tubes.

General conclusion : excellent quality of STBR96-300 and STBR384

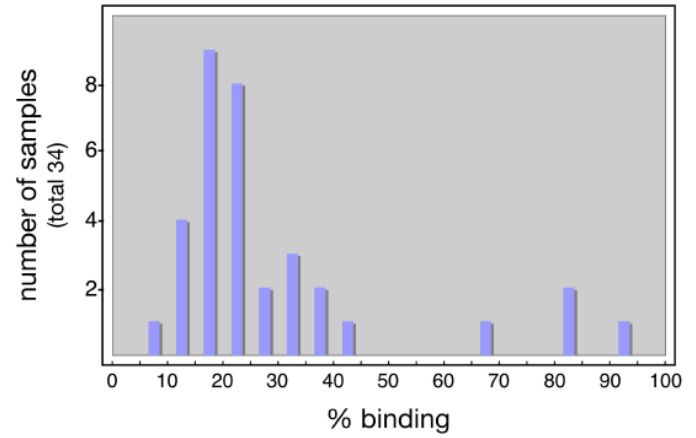
	Competitor 1	REMP STBR 96-300	REMP STBR 384	REMP MTP 384	Competitor 2 Product a	Competitor 2 Product b	Competitor 3
2,5 M NaCl							
Average	3,00 %	5,01 %	3,47 %	29,45 %	71,83 %	3,92 %	77,93 %
Standard deviation	1,14 %	1,62 %	1,27 %	20,86 %	15,27 %	0,40 %	12,74 %
0,1 M NaCl							
Average	1,13 %	3,17 %	1,93 %	11,57 %	7,86 %	3,00 %	31,99 %
Standard deviation	0,30 %	1,69 %	0,39 %	5,37 %	6,44 %	1,39 %	29,82 %

Table 1

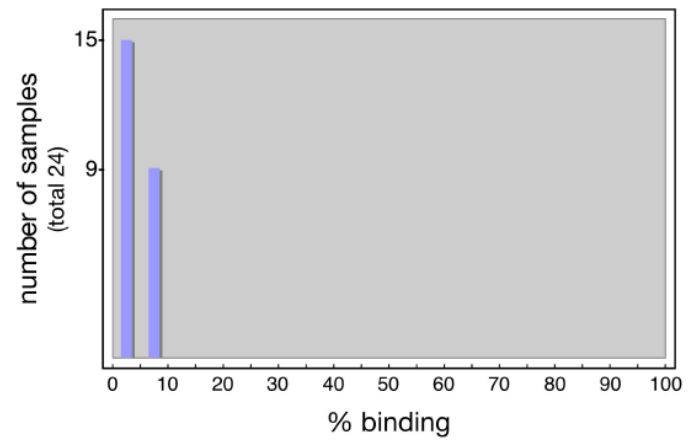
Average and standard deviations of DNA adsorption values (% of input DNA bound), as a function of the material studied.

Incubation
in
2.5 M NaCl

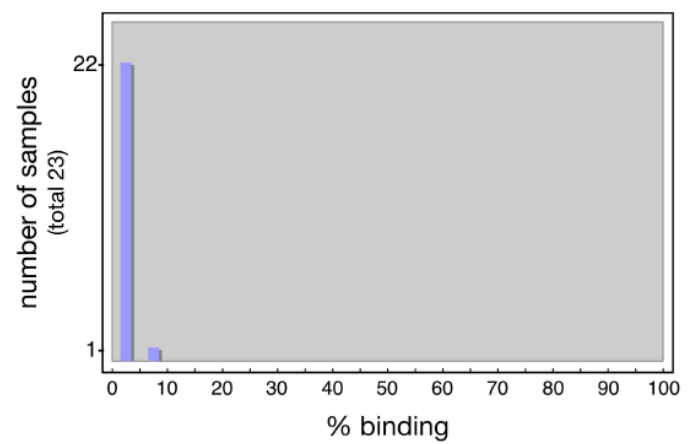
**REMP
MTP384**



**REMP
STBR96-300**

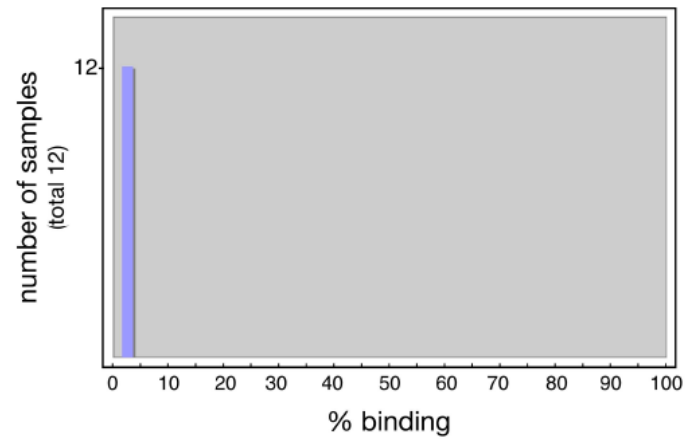


**REMP
STBR384**

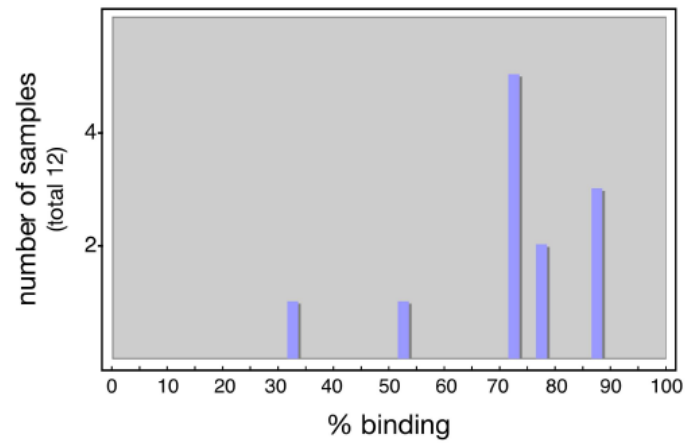


Incubation
in
2.5 M NaCl

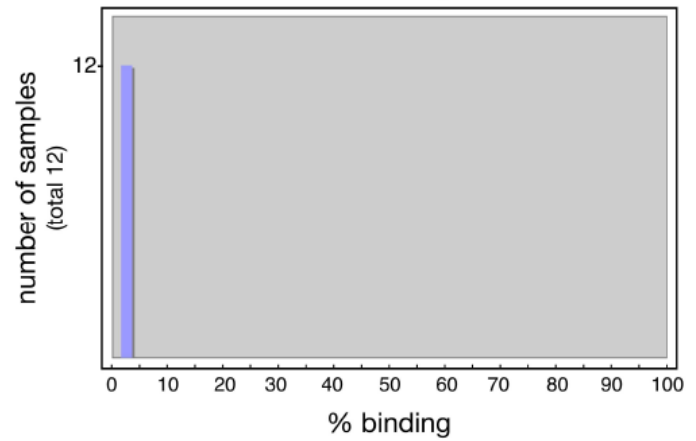
Competitor 1



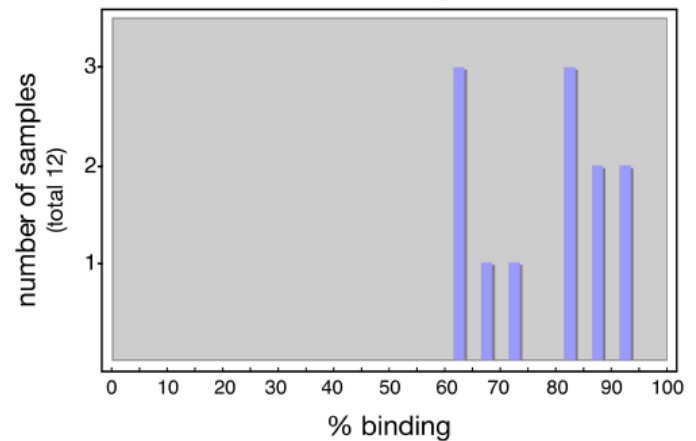
**Competitor 2
Product a**



**Competitor 2
Product b**

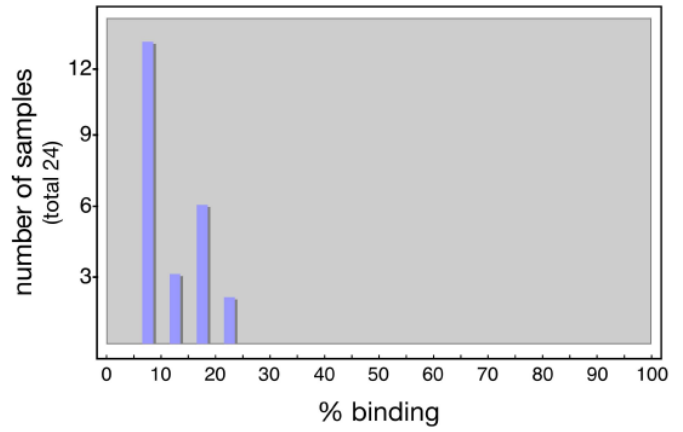


Competitor 3

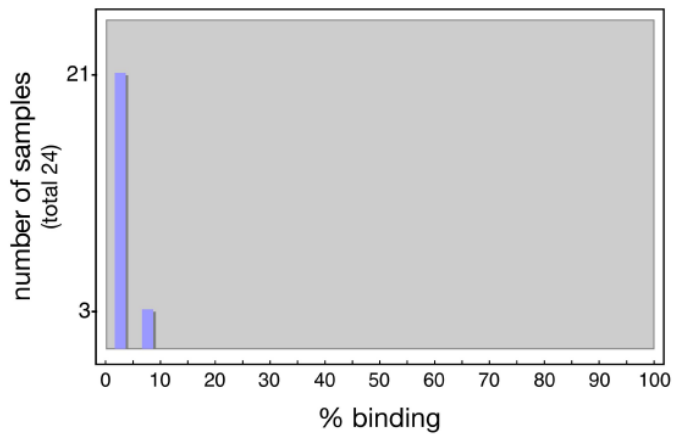


Incubation
in
0.1 M NaCl

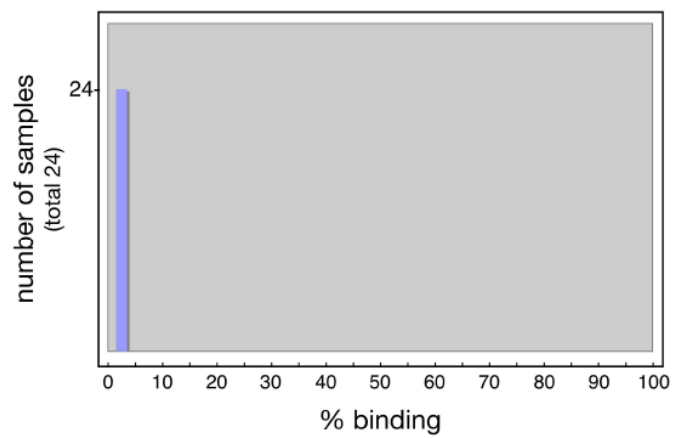
**REMP
MTP384**



**REMP
STBR96-300**

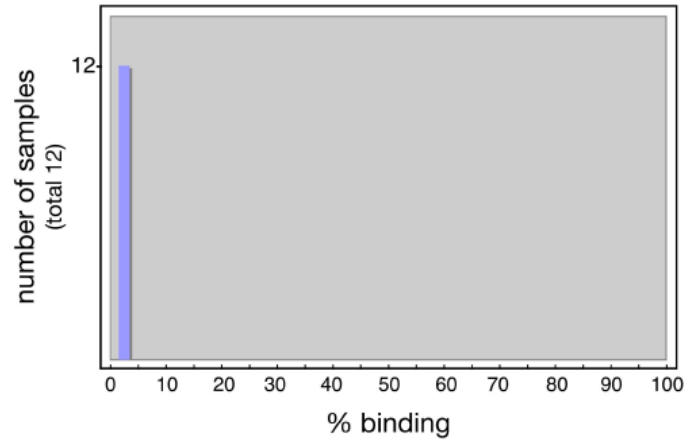


**REMP
STBR384**

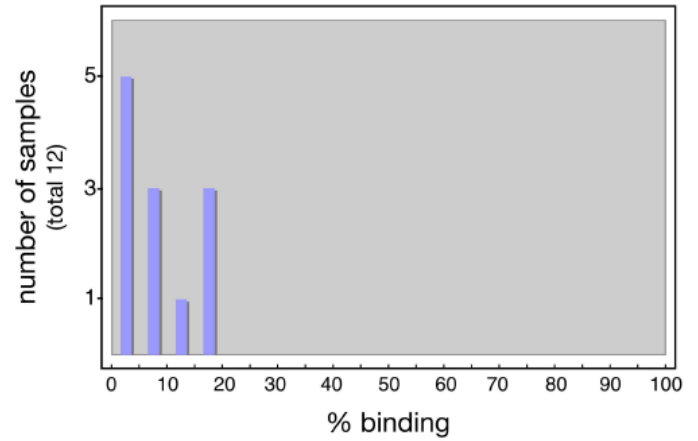


Incubation
in
0.1 M NaCl

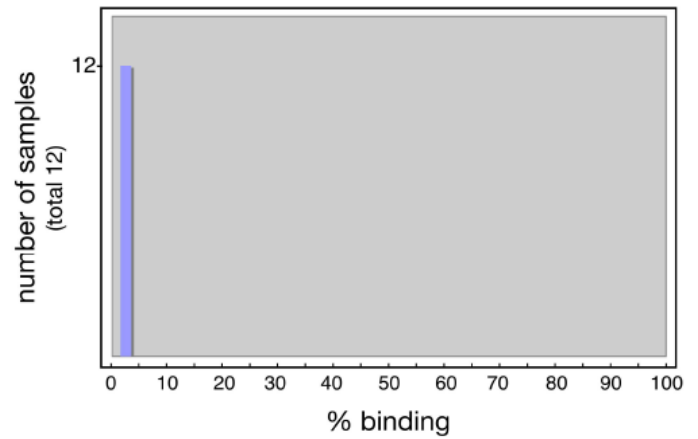
Competitor 1



**Competitor 2
Product a**



**Competitor 2
Product b**



Competitor 3

