

# Supplementary informations

## Functional characterization of a monoclonal antibody epitope using a lambda phage display-deep sequencing platform

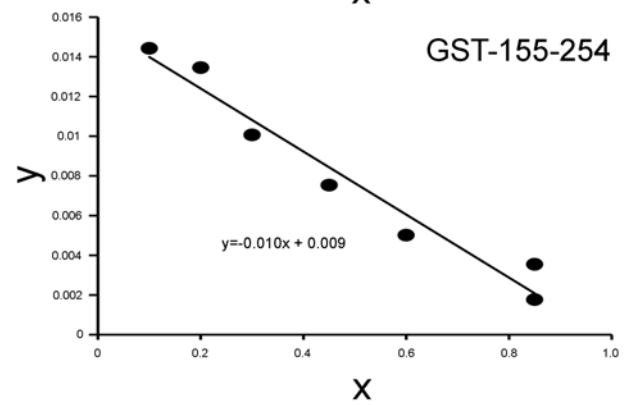
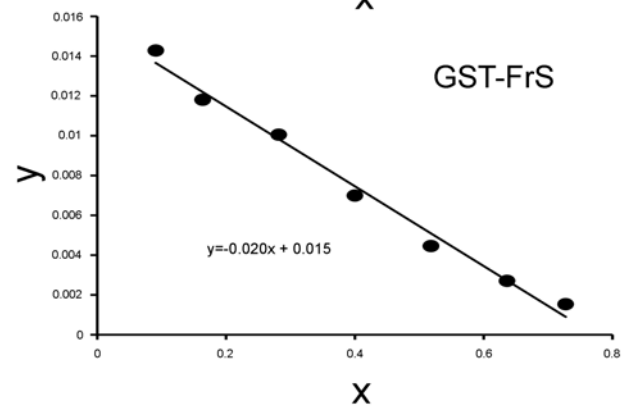
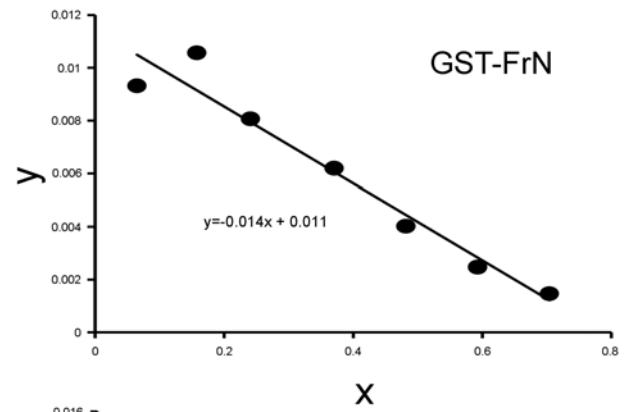
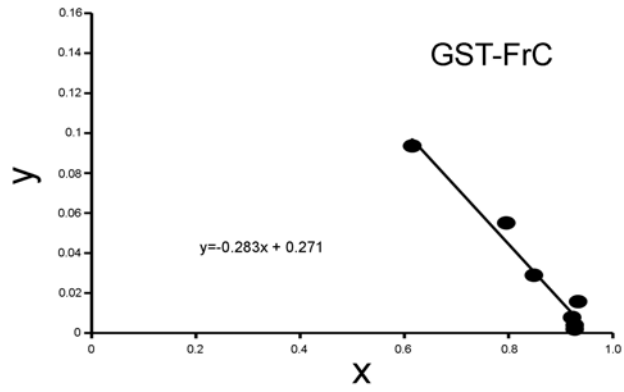
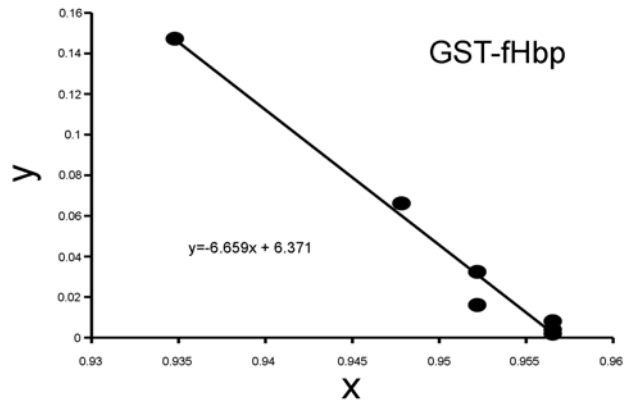
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**Supplementary note 1**

The probability that a gene fragment is cloned in its natural ("authentic") frame in the amino-terminal region of the D capsid protein is calculated as  $1/2 \times 1/3 \times 1/3 = 1/18$ , where the first value (1/2) is the probability that the insertion occurs in the forward direction, and the second and third values (1/3) correspond to the probability that the recombinant fragment is in frame at the 5' and 3' ends, respectively.

Fig. S1



**Supplementary Figure 1.** Determination of  $K_D$  values for mAb 12C1 of the fHbp fragments identified in the present study, as reported in Table 1. The panels show, for each antigen, data plotted on x and y axes, according to the method of Friguet *et al.*<sup>1</sup>, as detailed in the Materials and Methods section.  $x=A_0-A/Ax$ , where  $A_0$  is the measured ELISA absorbance of the sample containing the mAb alone, and  $A$  is the absorbance for a given concentration of free antigen in solution.  $y$  is proportional to the fraction of bound antibody in solution, divided by the concentration of free antigen in solution in the mixture:

$$y = \frac{\frac{A_0 - A}{A_0}}{C_{FN} - C_{Ab} \left( \frac{A_0 - A}{A_0} \right)}$$

The experimental data points are fitted to straight line whose slope is equal to  $-1/K$  and whose y-axis intercept point is  $1/KD$ . Shown are representative plots from 4 independent experiments producing similar results.

- 1 Friguet, B., Chaffotte, A. F., Djavadi-Ohanian, L. & Goldberg, M. E. Measurements of the true affinity constant in solution of antigen-antibody complexes by enzyme-linked immunosorbent assay. *Journal of immunological methods* **77**, 305-319 (1985).