

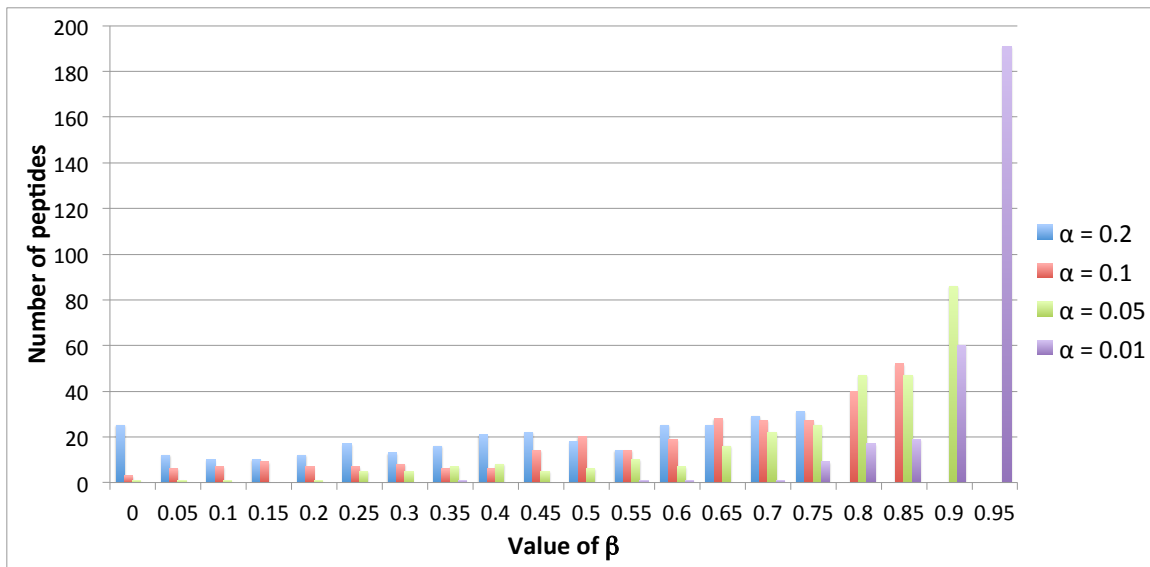
## The dependence of false negative probabilities (values of $\beta$ ) on $\alpha$ .

As described in the main paper, t-tests were used to compare the mean intensity of the treatment with the mean intensity of the control for each unique peptide on the array. We chose 0.2 as our threshold for rejecting the null hypothesis (i.e. that the means of the treatment and control are equal) for a given peptide. Peptides for which we rejected the null hypothesis were later used for pathway analysis.

We show here that, while 0.2 may seem like a liberal threshold, it is necessary to avoid high false negative probabilities (values of  $\beta$ ) for the majority of the peptides. While we show this only for one comparison (between the proximal compartment of Animal 1 and the control compartment of Animal 1), the same trends hold for the other comparisons performed in this study.

For each peptide in the aforementioned comparison, the value of  $\beta$  was calculated using the R package *pwr* for four different values: 0.2 (our chosen  $p$ -value threshold), 0.1, 0.05, and 0.01. The distribution of  $\beta$  values for each of these values of  $\alpha$  is given in Figure S1.

Figure S1 shows that using smaller values of  $\alpha$  would result in very large values of  $\beta$  for a significant proportion of peptides. For instance, if  $\alpha = 0.05$  is used, 227 out of the 300 peptides on the array have  $\beta \geq 0.7$  whereas this is true of only 60 peptides when  $\alpha = 0.2$ . As such, using a high value of  $\alpha$  is necessary to avoid large numbers of false negatives.



**Figure S1: Distribution of values of  $\beta$  for four different values of  $\alpha$  for the comparison between the proximal compartment and the control compartment of Animal 1.** The values on the  $x$  axis represent a range of values of  $\beta$ ; for example, “0” represents values of  $\beta$  in the range  $[0,0.05)$ . The  $y$  axis represents the number of peptides having values of  $\beta$  falling within that range.