**Supporting Information**

**Fisher’s exact test:**

Rectal and vaginal challenges were compared by Fisher’s exact test using GraphPad Prism 6. No statistically significant association (i.e. the data does not support rejection of the null hypothesis) between protection and the type of the challenge was observed for any of the antibody dose groups with p values >0.05 (p=0.44 for 10 mg/kg treatment groups, p=1 for 2 mg/kg and 0.4 mg/kg treatment groups). It should be noted that Fisher’s exact test can only falsify a null-hypothesis of “similar efficacy”.

A power analysis, as implemented in R statistical package http://rpackages.ianhowson.com/cran/statmod/man/power.html, suggested that ~15 macaques per group (about 100 macaques for the entire experiment) would be needed to provide statistically significant estimate of the differences between the protection probabilities for different challenge routes assuming it is similar to that seen in the original experiment (0.1).

**Logistic regression analysis**

Rectal and vaginal challenges were compared by **Logistic regression analysis** using SAS, version 9.2. This analysis gives an estimated odds ratio (OR) of 2.25 for the effect of route of challenge with a corresponding confidence interval of [0.30 ; 16.28]. In line with the wide confidence interval the p value is 0.4308. This analysis therefore provides no evidence of a clear difference in antibody protection by both challenge routes but it can also not be concluded that there is no difference. The logistic regression analysis did show a very significant effect of dose of antibody with a p value of 0.0026.

Nevertheless, the confidence interval of the OR can still give us some information. An 16-fold difference in protection (e.g. 5% for rectal and 80% for vaginal), similar to the possible difference in transmission rates, would correspond to an OR of 76; an 8-fold difference in protection (e.g. 8% for rectal and 80% for vaginal), would correspond to an OR of 36, whereas a 4-fold difference in protection (e.g. 20% for rectal and 80% for vaginal), would correspond to an OR of 16. This suggests that it is unlikely that the difference between rectal and vaginal protection is greater than an order of magnitude (~4-8 fold). If a more precise approximation is needed than a follow-up study should be designed that is sufficiently powered to demonstrate a difference ranging from a smaller than ~4-fold difference, up to no difference. The feasibility of this however can be questioned. To estimate how large an experiment should be in order to prove that no significant difference exists, we performed a thought experiment. If we e.g. require an OR proven within [0.5; 2], corresponding e.g. to one route providing 80% protection and the other 90% or v.v., then the experiment would require about 30 times more animals.