SUPPLEMENTAL MATERIAL

Jarjour et al., http://www.jem.org/cgi/content/full/jem.20132409/DC1

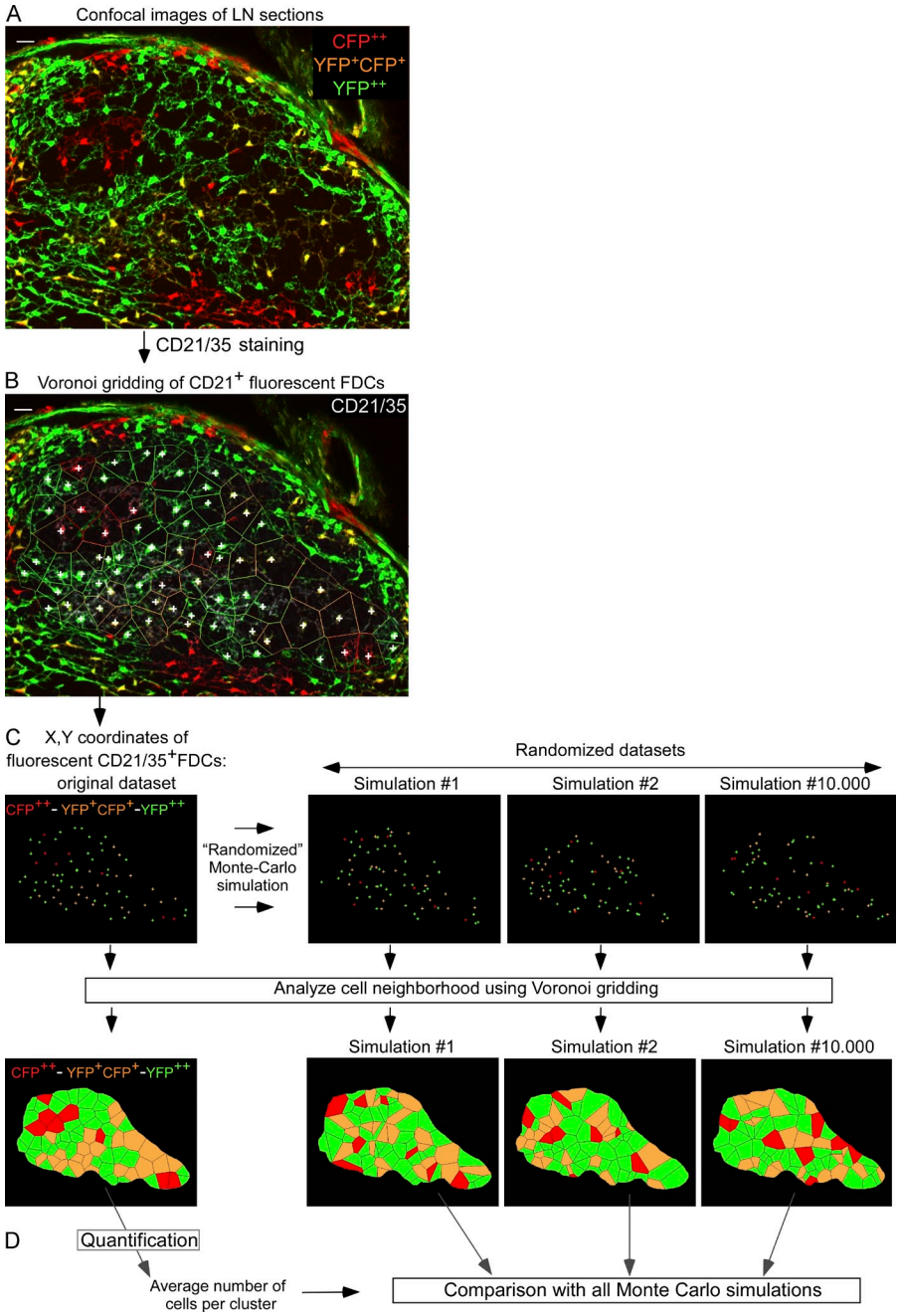


Figure S1. Quantification of the FDC clustering index and significance evaluation using Voronoi gridding and Monte Carlo simulations. Please refer to Materials and methods for further details. Quantification of FDC clustering index in the LN follicle of a Wnt-1Cre Ubow$^{+/+}$ mouse is taken as an example. Cells of interest (CFP^{++}, YFP^{++}, and CFP^{+}/YFP^{+} CD21/35^{+} FDCs) were first manually labeled within microscopic images (A). Automatic computation of the corresponding Voronoi diagrams (B) was then combined with Monte Carlo simulations (C) to obtain a quantitative statistical measure of the spatial distribution (i.e., cluster formation or diffuse cell spreading) of the FDCs (D). Bars, 25 µm.

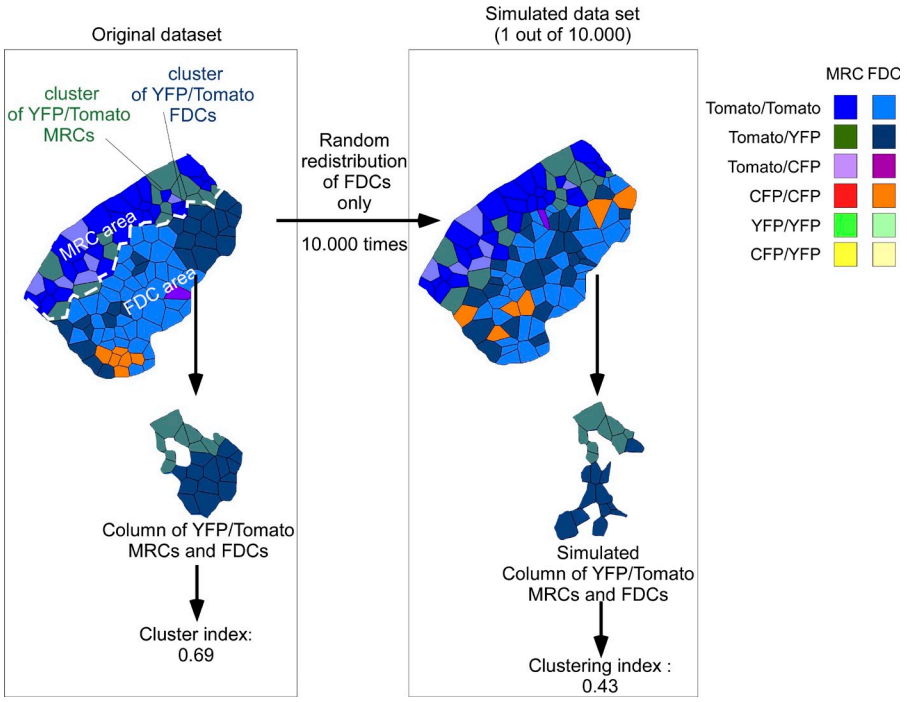


Figure S2. Flow chart explaining the calculation of the FDC/MRC clustering index shown in Fig. 3 E. Please refer to Materials and methods for further details.

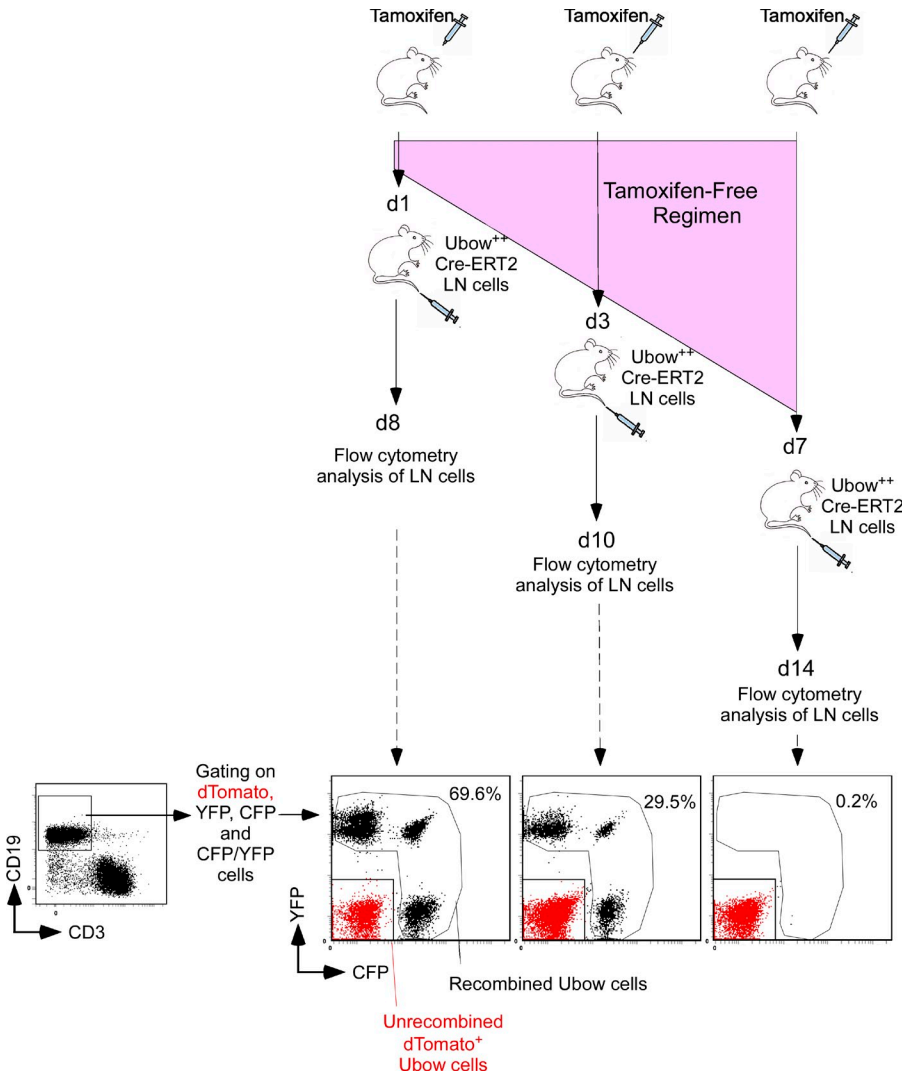
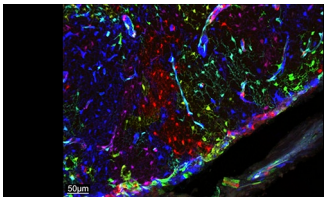


Figure S3. Tamoxifen remains active several days in vivo. WT mice were treated with tamoxifen (3 mg administered for 3 consecutive days i.p). This initial phase of treatment was followed by a "chase" period of 1, 3, and 7 d in which mice were maintained in a tamoxifen-free regimen. To measure the ability of the residual tamoxifen to trigger Cre-mediated Ubow recombination, total LN cells (10^7) isolated from a Ubow^{++} CreRT2 mouse were adoptively transferred i.v. in each of these mice. 1 wk later, mice were killed and the colors of the adoptively transferred cells analyzed by flow cytometry. If active tamoxifen was still present in any of these tamoxifen-treated mice, the adoptively transferred Ubow cells should have undergone recombination and switched from Tomato to CFP or YFP expression during this period. On the contrary, a lack of recombination in these cells would indicate a lack of detectable residual tamoxifen activity. Data on CD19$^+$ B cells indicate that tamoxifen activity was no longer present 7 d after the end of the tamoxifen treatment (same results for CD3$^+$ T cells). For this reason, all the tamoxifen-treated mice used in this study were injected with CFA/OVA 7 d after the last tamoxifen injection, thus avoiding any late recombination event.



Video 1. Supporting video of Fig. 3 D in which one can observe multiple columns composed of RANK-L$^+$ MRCs and CD21/35$^+$ FDCs sharing a similar color. This movie was created with a 50-µM-large Z stack dataset of confocal images acquired with a 20× objective.