

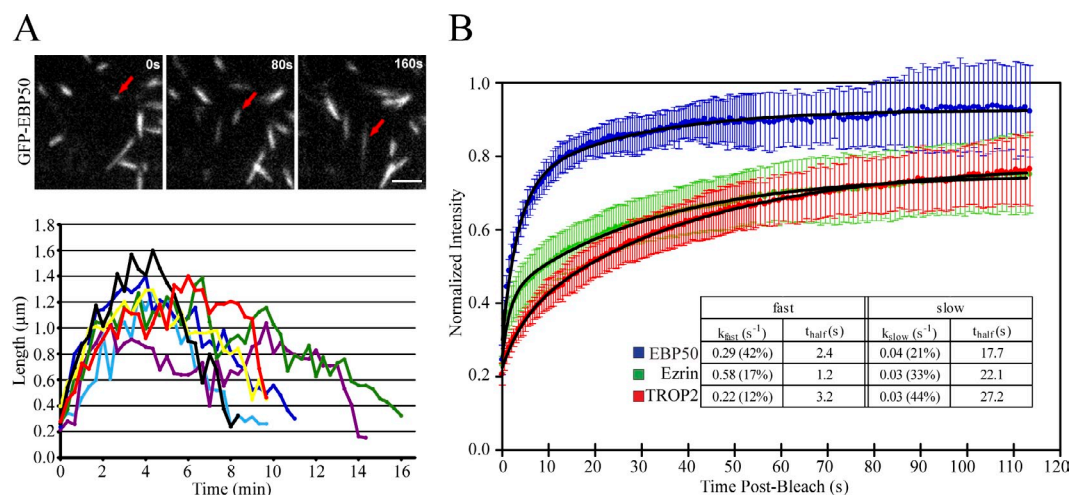
Garbett and Bretscher, <http://www.jcb.org/cgi/content/full/jcb.201204008/DC1>

Figure S1. **Microvilli and their components are dynamic.** (A, top) Time points from JEG-3 cells expressing GFP-EBP50 to observe the microvillar lifecycle. Red arrows show a microvillus form and grow to its mature length. Bar, 2 μm . (bottom) Length measurements of individual microvilli over time show similar initial growth rates but variable lifetimes. (B) Photobleaching recovery curves of GFP-tagged EBP50 ($n = 13$), ezrin ($n = 19$), and TROP2 ($n = 16$). Error bars show SD, and black lines represent fitted curves with corresponding rates shown in the table.

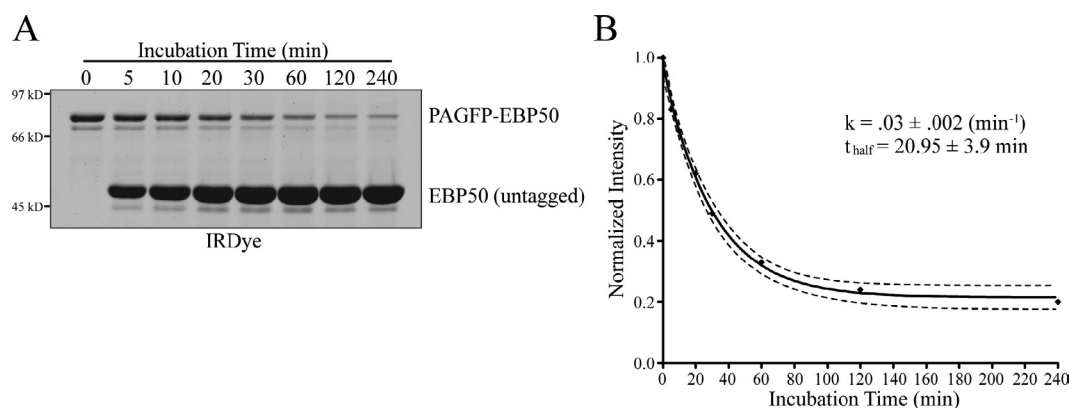


Figure S2. **EBP50 associates with ezrin stably in vitro.** (A) Limiting amounts of PAGFP-EBP50 prebound to ezrin FERM beads were competed off with untagged-EBP50 for the times indicated. Gel was stained for total protein with IRDye. (B) Normalized amount of the remaining bound PAGFP-EBP50 from the representative experiment in A was fit to a single exponential decay curve shown as solid line. Dotted lines indicate the 95% confidence interval. Rate and half-life are indicated (\pm SD).

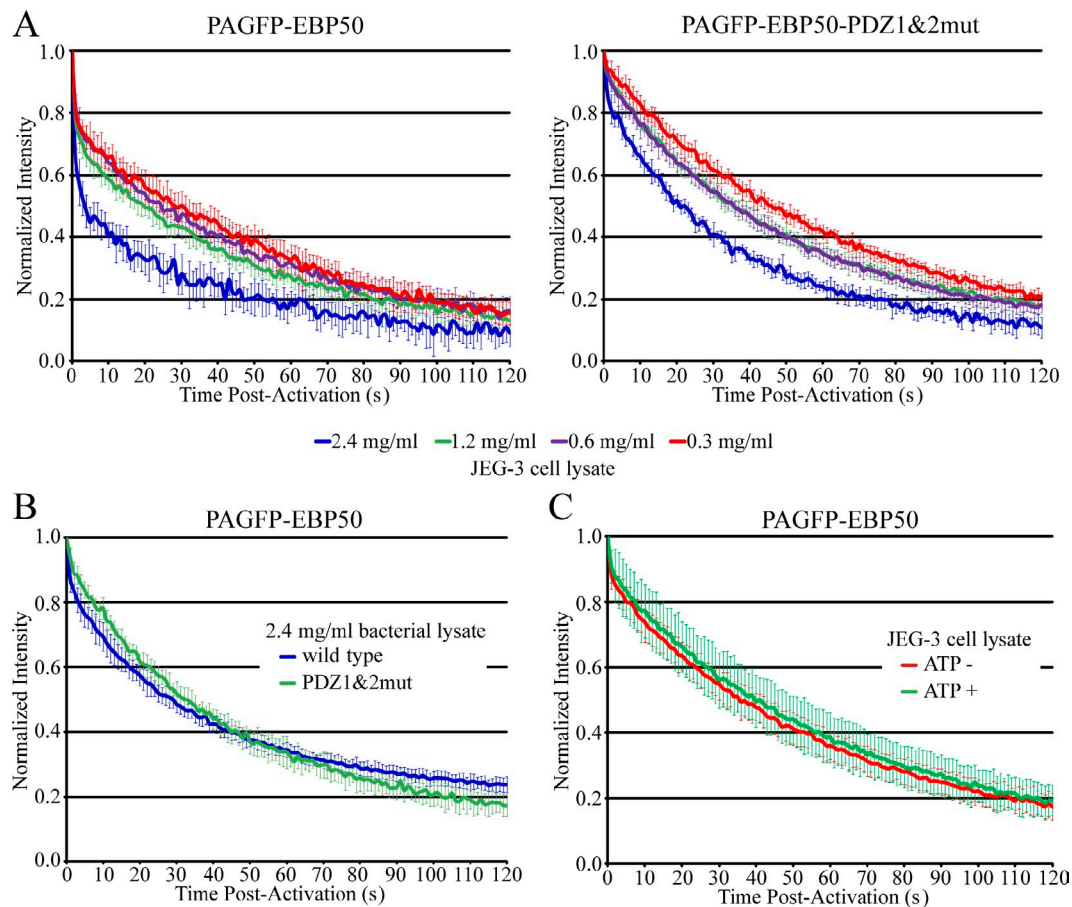


Figure S3. **Lysate effects on EBP50 dissociation from ezrin.** (A) Increasing concentrations of JEG-3 cell lysate were added to the in vitro photoactivation assay. Graphs represent the photoactivation decay curves for PAGFP-tagged EBP50 wild type (left) and PDZ1&2 mutant (mut; right) with increasing amounts of lysate. $n \geq 6$ for all curves. (B) Graph with addition of bacterial lysate to the in vitro photoactivation assay using either PAGFP-tagged wild type ($n = 9$) or PDZ1&2 mutant ($n = 8$). (C) Graph with addition of 2.4 mg/ml JEG-3 cell lysate depleted of ATP (ATP-; $n = 18$) or with ATP regeneration system (ATP+; $n = 17$) using PAGFP-tagged wild-type EBP50. All error bars show SD.