Supplementary information
(consisting of 4 figures) to the report entitled:

Dendritic spine dynamics in synaptogenesis after repeated LTP inductions:
Dependence on pre-existing spine density.

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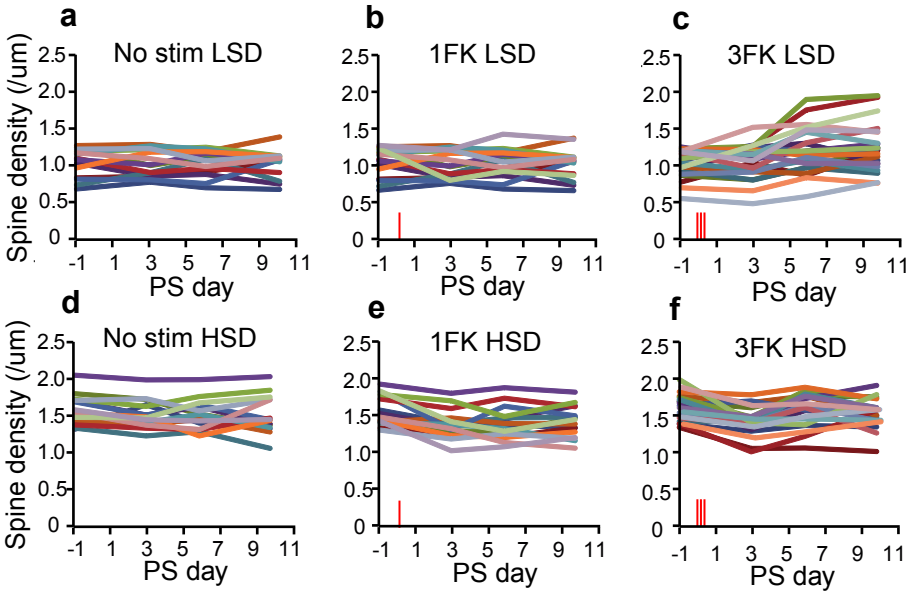
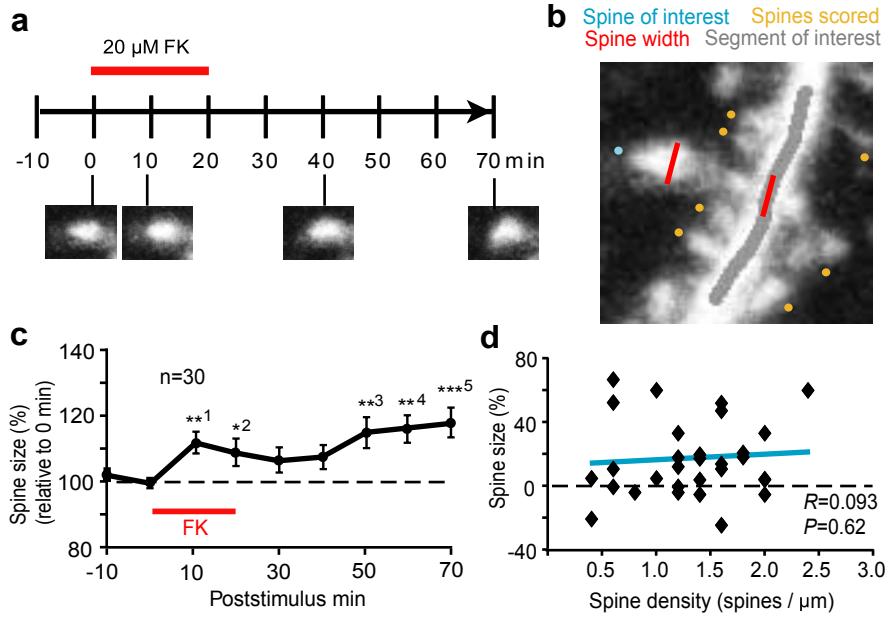


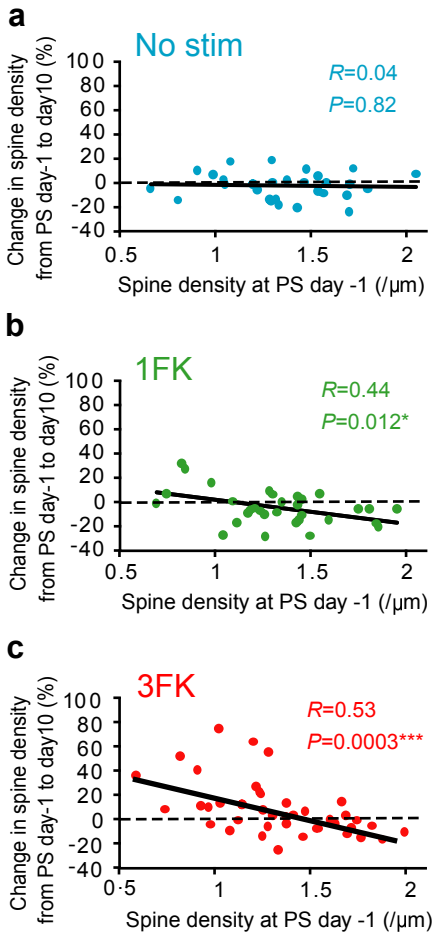
Supplementary Figure S1
Dendritic spine dynamics as seen on an individual segment basis.

(a-c) Dynamics in dendritic segments having low pre-existing spine densities (≦1.3 spines/um) as classified by stimulus conditions.
(d-f) Dynamics in dendritic segments having high pre-existing spine densities (>1.3 spines/um) as classified by stimulus conditions. The dynamics as seen on an average basis is shown in Text Fig. 4. In each panel lines shown in different colors represent individual segments. This figure demonstrates that the spine number increase occurs preferentially in the dendritic segments having low pre-existing spine density (c).



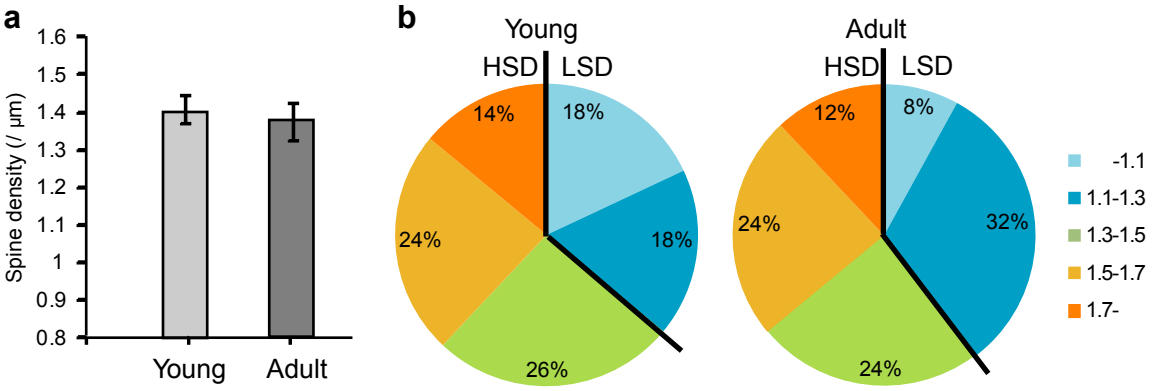
Supplementary Figure S2
Correlation between spine size enlargement as an index of LTP induction and spine residence.

(a) Timeline of experimental procedures. Inset images depict the fate of a representative spine.
(b) A sample image showing definitions. Spines (demarcated by orange dots) are counted along the gray line (5 um) drawn superimposed on the dendritic shaft. Maximal width (not volume) of the half peak fluorescence measured in parallel to the dendritic shaft is shown in red.
(c) Spine enlargement following the passage of time. Statistic comparison was made by paired t-test for each time point as compared with that at 0 min. The number of spines examined is 30. P values are 0.0011 for **1, 0.036 for *2, 0.0018 for **3, 0.0013 for **4, and 4.1 x 10-4 for ***5.
(d) Magnitude of spine enlargement as a function of pre-existing spine density. Blue line indicates the regression line, showing no correlation. Statistics was made by linear regression analysis. R is regression coefficient and P is its reliablity (Spearman's test).



Supplementary Figure S3
Correlation between spine density change and pre-existing spine density.

Generation and retraction are indicated as plus and minus values, respectively. Values were obtained by comparing images at PS day -1 and PS day 10 in samples of No stim (a), 1FK (b), and 3FK (c).Each dot represents one dendritic segment. The numbers of segments examined are 30, 32, and 42 for No stim, 1FK, and 3FK specimens, respectively. In each panel, thick line indicates the regression line. R is regression coefficient and P is its reliablity (Spearman's test).



Supplementary Figure S4
Spine density in the apical dendrite of CA1 pyramidal neurons in mouse brain in vivo.

(a) Mean dendritic densities of young (3 weeks) and adult (3 months) mice. The number of animals and dendritic segments examined were 3 and 50, respectively, for both young and adult mice. Statistic comparison was made by paired t-test but no significant difference was detected.
(b) Classification of dendritic segments having low and high spine densities. A relative decrease of segments having <1.1 spines/um and a relative increase of the segments having 1.1-1.3 spines/um may be correlated with the animals' accumulation of experience.