Real-time PCR

Total RNA from human B cells was extracted with RNAPURE kit (RP1202; BioTeke) and reverse-transcribed with a PrimeScript[™] RT reagent Kit (RR037A; Takara). The resulting cDNA was analyzed for the expression of various genes with SsoAdvancedTM SYBR® Green supermix (Bio-Rad) on a CFX96 Touch Real-Time System (Bio-Rad) and the appropriate primers for 'test genes'. *cd* 19 5' primer: cccaccaggagattcttcaa, *cd* 19 3' primer : gggcacatacaggctttgtt. fcγr II b 5' primer : actcatccaagcctgtgacc, fcγr II b 3'primer: attgtgttctcagccccaac. *btk* 5' primer : gcaccagtctccacaagtga, *btk* 3' primer : tcctgcccatttttatctcg. Fig.S1

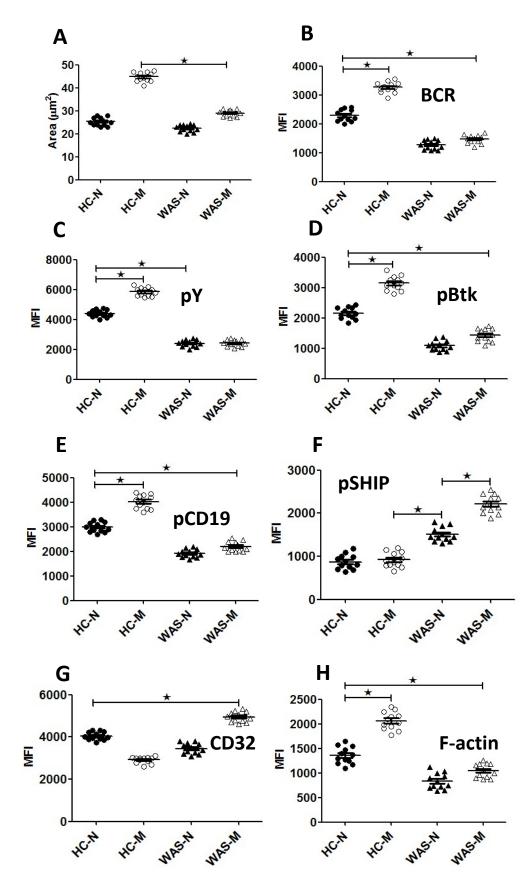


Fig S1. A combined data of TIRFM analysis from 12 patients.

IRM and TIRFM analysis of PBMCs (P1-12) that were incubated with membrane-tethered Fab'-anti-Ig. The quantification of B-cell contact area (A) at 5 min from the average of HCs and WAS (P1-12) B cells .The quantification of MFI of BCR (B) at 5 min, pY (C) at 3 min, pBtk (D) at 5 min, pCD19 (E) at 5 min, pSHIP (F) at 5 min, CD32 (G) at 5 min and F-actin (H) at 3 min in the B-cell contact zone from the average of HCs and WAS

(P1-12) B cells and each dot represents one patient. TIRFM analysis is from >300 individual cells of three individual experiments. *p<0.01, compared to WAS naïve or memory B cells.