Supporting Information

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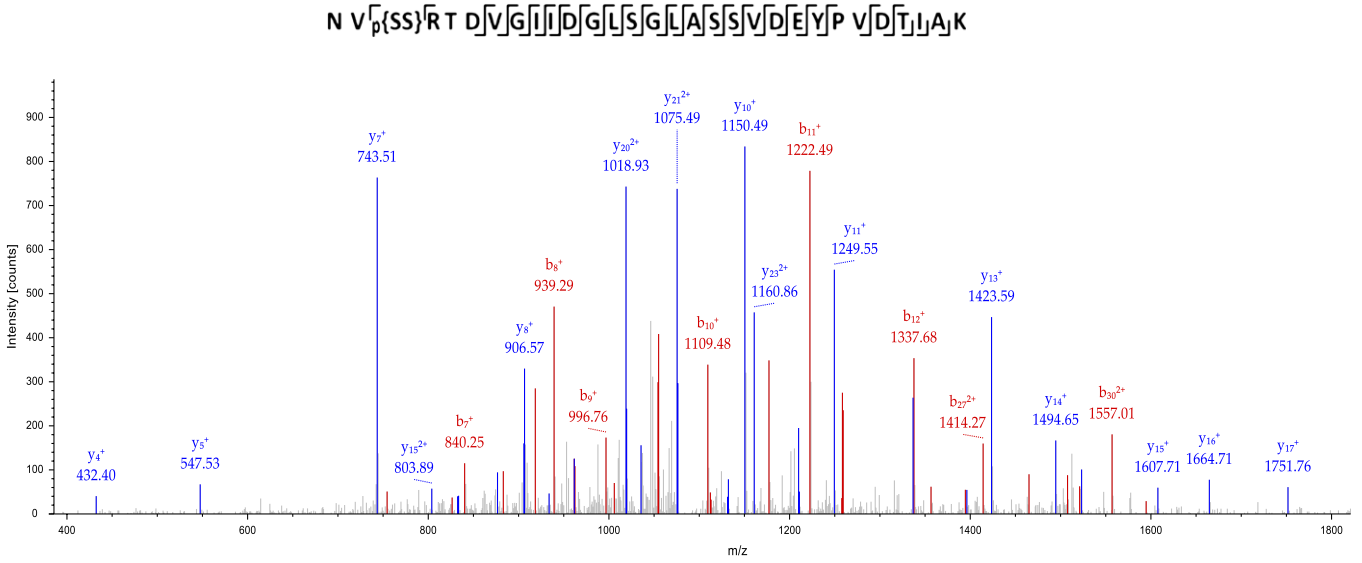

Fig. S1. Identification of the AMP-activated protein kinase (AMPK) phosphorylation site in recombination-activating gene 1 protein (RAG1). Collision-induced dissociation (CID) spectrum confirmed that a RAG1 peptide containing S528 was singly phosphorylated. The fragment ion masses localized the phosphorylation site to either S527 or S528. The peptide fragments N-terminal to the fragmentation sites (b) are shown in blue and the peptide fragments C-terminal to the fragmentation sites (y) are shown in red.

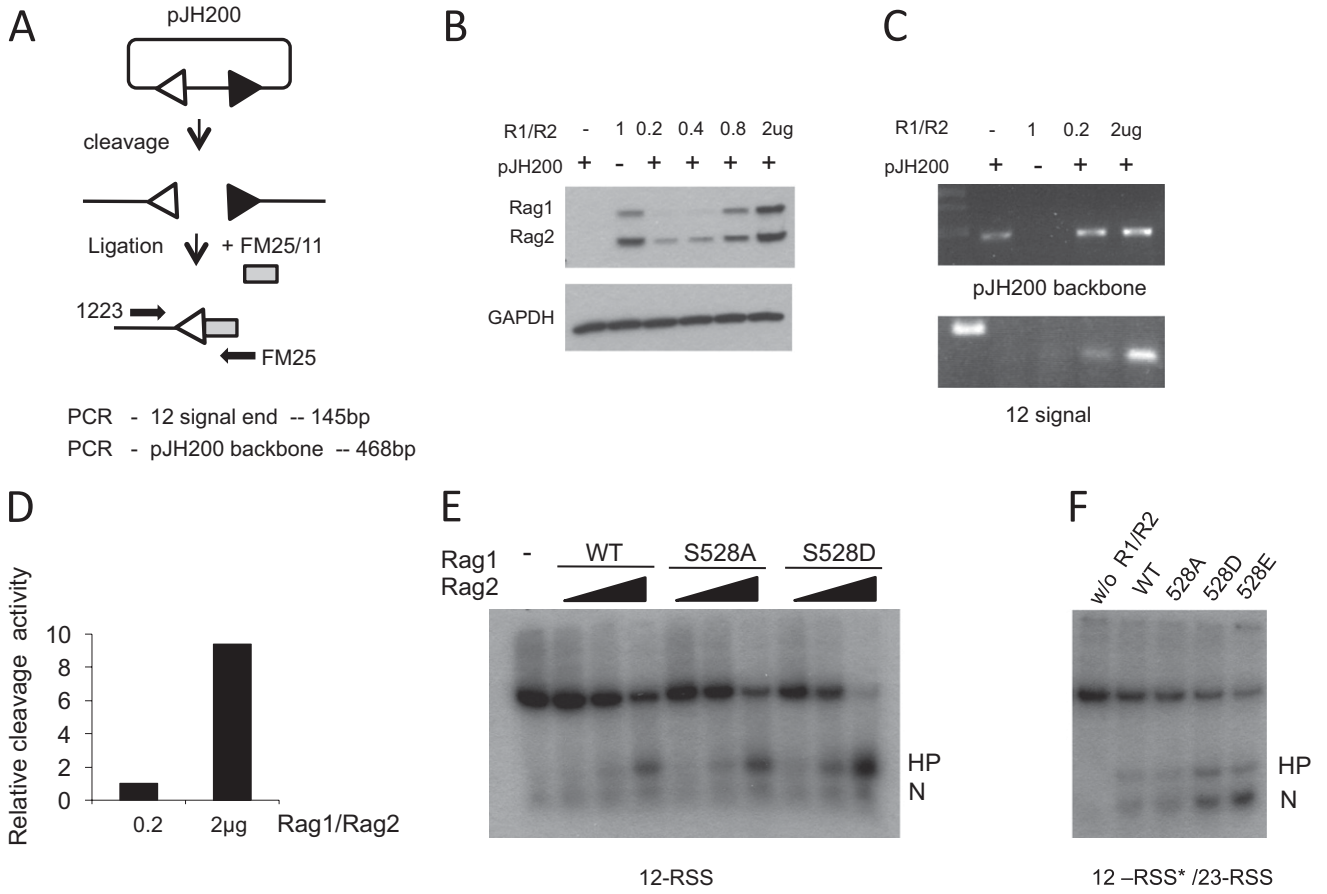

Fig. S2. Extrachromosomal cleavage of pJH200 (1) recombination substrate was detected by ligation-mediated PCR (LM-PCR), using different expression levels of WT RAG1/RAG2 to establish an approximately linear range. (*A*) Diagram of the extrachromosomal cleavage assay is shown. (*B*) 293T cells were transfected with different amounts of RAG1/RAG2(R1/R2) while maintaining a constant amount of transfected pJH200. RAG1/RAG2 expression was evaluated by Western blot. GAPDH was used as a loading control. (*C*) DNA cleavage in 293T cells transfected with 0.2 or 2 μg of RAG1/RAG2 was compared. Cleavage was detected by LM-PCR of 12-recombination signal sequence (RSS). (*D*) The cleavage detected in *C* was quantified. (*E*) Various amounts of WT and S528 mutants of RAG1 were incubated with RAG2 and with radiolabeled 12-RSS oligonucleotide and $MnCl_2$ in an in vitro cleavage reaction. The nick (N) and hairpin (HP) cleavage products were analyzed by TBE-urea gel. (*F*) HP and N formation from a coupled cleavage reaction contained HMGB1, $MgCl_2$, RAG1 (WT and mutants), and WT RAG2, with radiolabeled 12-RSS and unlabeled 23-RSS.

1. Hesse JE, Lieber MR, Gellert M, Mizuuchi K (1987) Extrachromosomal DNA substrates in pre-B cells undergo inversion or deletion at immunoglobulin V-(D)-J joining signals. *Cell* 49(6): 775–783.