Supplemental Data

Supplemental Experimental Procedures

Constructions of deletion mutants- The region II of BAEBL was cloned from the pBSV-Fc-BAEBL-8His into the *Bam*HI/*Eco*RI site of pBluescript II KS (+) plasmid. The resultant plasmid, designated pBluescript-BAEBL, was used as a template for the following PCR. The 5' fragment and 3' fragment of BAEBL were amplified by PCR using the primers shown in Table S1, ligated with *Nhe*I or *Pst*I, and cloned into the *Bam*HI/*Eco*RI site of pBSV-Fc-8His vector. The resultant plasmids were designated pBSV-Fc-BAEBL-ΔGAG3-8His, pBSV-Fc-BAEBL-ΔGAG4-8His, and pBSV-Fc-BAEBL-ΔGAG5-8His, respectively. To generate pBSV-Fc-BAEBL-ΔGAG1-8His, QuickChange II Site-Directed Mutagenesis Kit (Stratagene, Cedar Creek, TX) and primers shown in Table S1 was used according to the manufacturers' instructions. The BAEBL containing the desired nucleotide change was cloned into the *Bam*HI/*Eco*RI site of pBSV-Fc-8His.

Supplemental Tables

Table S1. The list of primers used to generate the expression vectors for motif-deleted BAEBLs.

Expression plasmid		Direction	Sequence [a]
pBSV-Fc-BAEBL-ΔGAG1-8His		Forward	5'-GGATCCCAATATACGTTTATACAGgcAgcTACTgcTTTGTTTGCTTGTGG-3'
		Reverse	5'-CCACAAGCAAACAAAgcAGTAgcTgcCTGTATAAACGTATATTGGGATCC-3'
pBSV-Fc-BAEBL-ΔGAG3-8His	5' fragment	Forward	5'-GTAAAACGACGGCCAG-3'
		Reverse [b]	5'-TTTgCTagcTgcCATAAATGATGTATATGAAGAACATG-3'
	3' fragment	Forward [b]	5'-AAAgctAGcgcAACACAAATGGAGGTTTTG-3'
		Reverse	5'-CAGGAAACAGCTATGAC-3'
pBSV-Fc-BAEBL-ΔGAG4-8His	5' fragment	Forward	5'-GTAAAACGACGGCCAG-3'
		Reverse [c]	5'-ACCtgcAgcTAAAAGATATGTACGTCCAAG-3'
	3' fragment	Forward [c]	5'-ACcTgcaGGTgcTGAGGAAGATTATAAGGAAC-3'
		Reverse	5'-CAGGAAACAGCTATGAC-3'
pBSV-Fc-BAEBL-ΔGAG5-8His	5' fragment	Forward	5'-GTAAAACGACGGCCAG-3'
		Reverse [c]	5'-CTCTgcagcTTTCTCACAAAAATCGTCTC-3'
	3' fragment	Forward [c]	5'-AActgcAGAGgcAATATATTCATTTGAGTCATTTAAGG-3'
		Reverse	5'-CAGGAAACAGCTATGAC-3'

[a] The mutated nucleotides are in lower case. [b] *Nhe*I site is underlined. [c] *Pst*I site is underlined.

Table S2. *Kd* of the binding of motif-deleted BAEBL with HS.

Protein name	*Kd* (M) [a]	p [b]
BAEBL/Fc	$8.18 \pm 2.54 \times 10^{-9}$	
BAEBL ΔGAG1/Fc	$6.94 \pm 2.57 \times 10^{-9}$	0.749
BAEBL ΔGAG3/Fc	$4.25 \pm 0.15 \times 10^{-9}$	0.198
BAEBL ΔGAG4/Fc	$8.17 \pm 1.97 \times 10^{-9}$	0.997
BAEBL ΔGAG5/Fc	$1.08 \pm 0.84 \times 10^{-8}$	0.391

[a] Data are shown as mean ± standard error of the mean.

[b] Differences between the value of mutant BAEBL and that of wild-type BAEBL were assessed by *t*-test.