Supplementary Fig. S1 (Kasper)

a  
CBP wild type allele

Targeting Construct

Targeted Allele

Targeted Allele after Cre mediated recombination in ES cells

Nhel digest w/ Int. probe

~10 kb  
~6.5 kb

Southern

PCR (primers p3 and p4)

exon

loxp site

PCR primer

342-393 AA deletion (with introduced Nhel site)

PCR on RT cDNA

~9 kb  
~4 kb

Southern

p300 ΔCH1

PCR on RT cDNA

b  
p300 wild type allele

Targeting Construct

Targeted Allele

Targeted Allele after Flp mediated recombination in ES cells

Nhel digest w/ Int. probe
Supplementary Fig. S1. Deletion mutations were introduced into the CH1 domains of CBP (a) and p300 (b) in ES cells by homologous recombination. The NeoTK drug selection cassette was excised in ES cells by transient expression of Cre (a) or Flp (b) recombinase. Restriction sites are indicated: C, Clal; H, HindIII; K, KpnI; N, Nhel; X, XbaI. Homologous targeting confirmed by Southern blot (EcoRV digest for CBP, EcoRI digest for p300) using external probes (indicated) and PCR (primers p1 and p2). Screening after NeoTK cassette excision by PCR (primers p3 and p4) and Southern blot using Nhel digest and internal probes (indicated). A diptheria toxin gene (DTA) was included in the p300 construct. Correct splicing of the RNA was determined by PCR using primers in exons 3 and 5 of CBP or p300 and RTcDNA from CBPΔCH1/ΔCH1 (a) or p300ΔCH1/ΔCH1 (b) MEFs and confirmed by sequencing the products. PCR on RTcDNA using primers in exons 2 and 12 for CBP and exons 3 and 13 for p300 also gave single products of the correct size in MEFs homozygous for the ΔCH1 mutant alleles (data not shown).