Supplementary Data S1

Methods and Materials

Generation of BAC transgenic mice

The BAC recombination was done using previous proved system [1]. Briefly, the BAC clone was first recombined to remove the two *loxP* elements in the pBACe3.6 vector sequence. Next, the CreER^{T2} targeting sequence was incorporated into the second exon of the *dcx* BAC DNA, silencing the *dcx* open reading frame. The *neo* selection marker was then removed by arabinose-induced *FLP-FRT* mediated recombination. All recombinants were screened by PCR and checked by restriction-enzyme digestion mapping on a pulse-field gel. Recombinant BAC DNA was extracted using the SDS-NaOH method, and purified by CsCl gradient centrifugation or by sepharose 4B-CL chromatography.

BAC DNA linearized by digestion with *Not*I was microinjected into the pronuclei of C57B6L fertilized eggs. Founders were screened using the following primers: forward, 5'-GGG TAT TCC CTG GAG GCT GT-3'; reverse, 5'-TTC TTG CGA ACC TCA TCA CT-3'. For the genotyping of CreER^{T2}, a second pair of primers was used: forward, 5'-ATT TGC CTG CAT TAC CGG TCG-3'; reverse, 5'-CAG CAT TGC TGT CAC TTG GTC-3'. For expression pattern screening, the founder was crossed to wild-type C57/B6L mice, and then each offspring was crossed to reporter mice, and their descendants were examined independently. Descendants with the predicted expression pattern were back-crossed to wild-type C57/B6L mice and the purified line was expanded.

LacZ staining

Embryonic slices with 30 μm thickness were mounted on gelatin coated slides. The slides were incubated in 0.5 mg/ml X-gal in LacZ reaction solution at 37 for 2-12 h. The slides were then washed with PBS solution three times for 10 min. Finally, the slides were mounted in 80% glycerol PBS medium. Images were taken using a Nikon

fluorescence microscope with a CCD camera.

Supplementary reference

1. Lee EC, Yu D, Martinez de Velasco J, *et al.* A highly efficient *Escherichia coli*-based chromosome engineering system adapted for recombinogenic targeting and subcloning of BAC DNA. *Genomics* 2001; **73**:56-65.