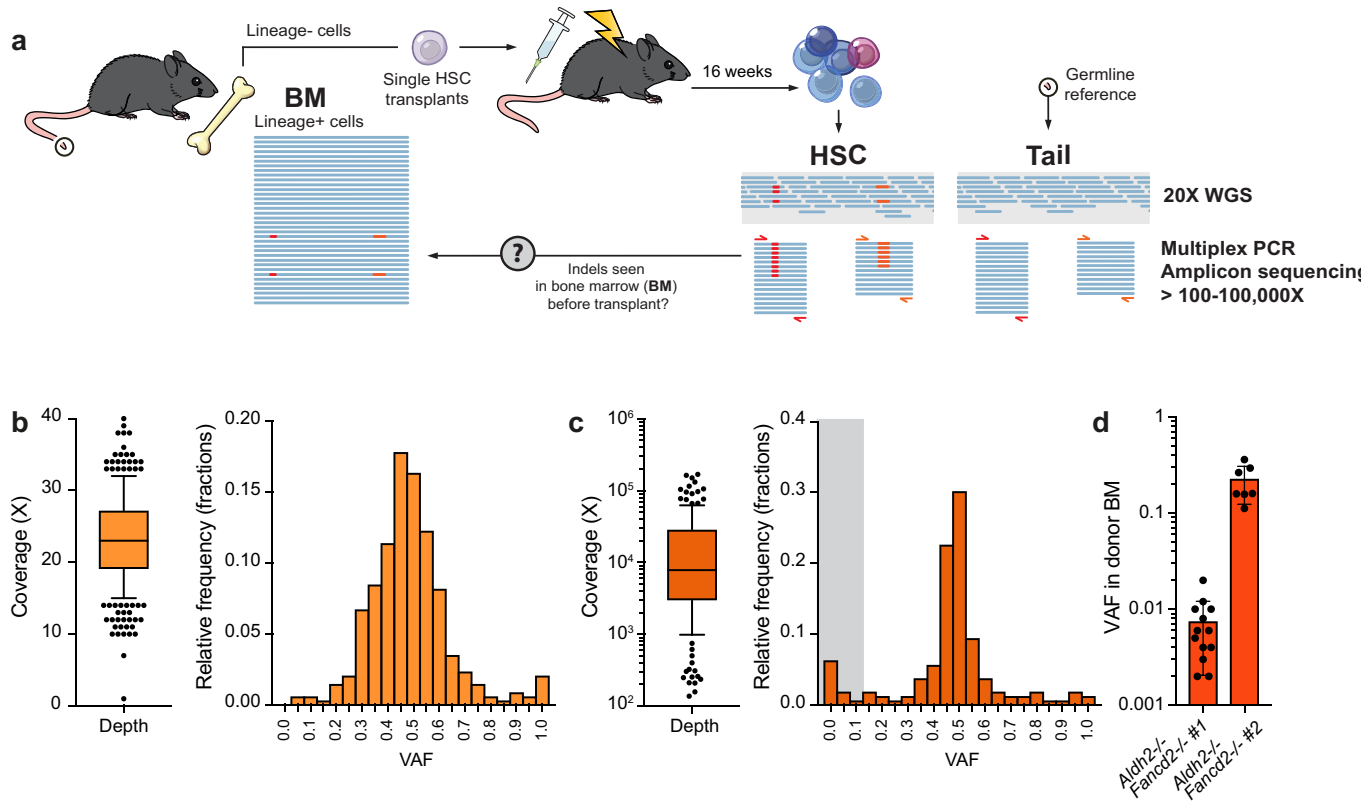

Extended Data Figure 9 | Validation of indels by targeted deep sequencing. a, Scheme depicting the generation of HSC clones by transplantation of single stem cells, subsequent whole-genome sequencing and validation of indel calls by amplicon deep sequencing. On the basis of the indel location from 20× whole-genome sequencing, we designed multiplex PCRs and deep sequenced the PCR products to higher coverage (100–100,000×) to confirm that the calls were not sequencing artefacts. In addition, we attempted to detect indels in DNA samples of bone-marrow cells from the mice that provided the transplanted HSCs. **b**, Coverage depth and VAF of the filtered set of indel calls from whole-genome sequencing ($n = 342$ indels; box plot shows the mean, box edges represent the first and third quartiles, whiskers extend over 10–90% of data). **c**, Coverage depth and VAF of the indel calls from deep sequencing validation ($n = 159$ locations; box plot shows the mean, box edges represent the first and third quartiles, whiskers extend over 10–90% of data). One hundred and fifty-nine locations had coverage greater than 100× and were used for the analysis. We could validate the presence of 91.2% of the initial calls; 14/159 (8.8%) calls had VAF <0.1 and were deemed false positives (indicated by grey shading). Note that the VAF distribution is centred tightly around 0.5, confirming the clonal nature of most indels. **d**, We used targeted deep sequencing to look for indel calls in bone-marrow samples from the mice that provided the transplanted HSCs. In most cases, the calls were below the detection limit of the assay (VAF <0.0001). However, we could detect indels from two *Aldh2*$^{-/-}$*Fancd2*$^{-/-}$ HSCs, indicative of 'clonal haematopoiesis' in these mice (accounting for 0.7 and 21.4% of blood production, respectively). Data shown as mean and s.e.m.; $n = 13$ and 7 indels.