

Fig. S1. Schematic representation of the typical targeted allele configurations used in this study. (A) *Dusp3* was targeted using a promoter-driven selectable marker. (B) *Ap4e1* was targeted using a promoterless selectable marker. Both were studied as a reporter tagged knockin allele (tm1aWtsi) and as a reporter tagged deletion allele (tm1bWtsi). Tm1bWtsi alleles were derived from the parental tm1aWtsi allele via *Cre/loxP* mediated excision.

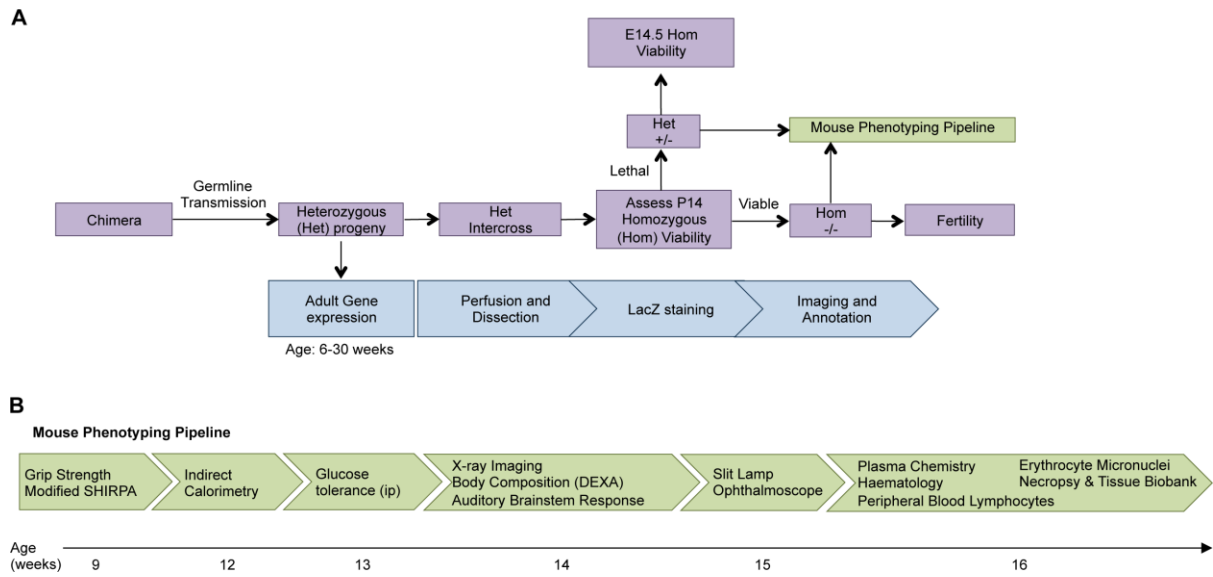


Fig. S2. Overview of the typical workflow each knockout mouse line progressed through including (A) adult *lacZ* reporter gene expression profiling and (B) adult clinical phenotyping.

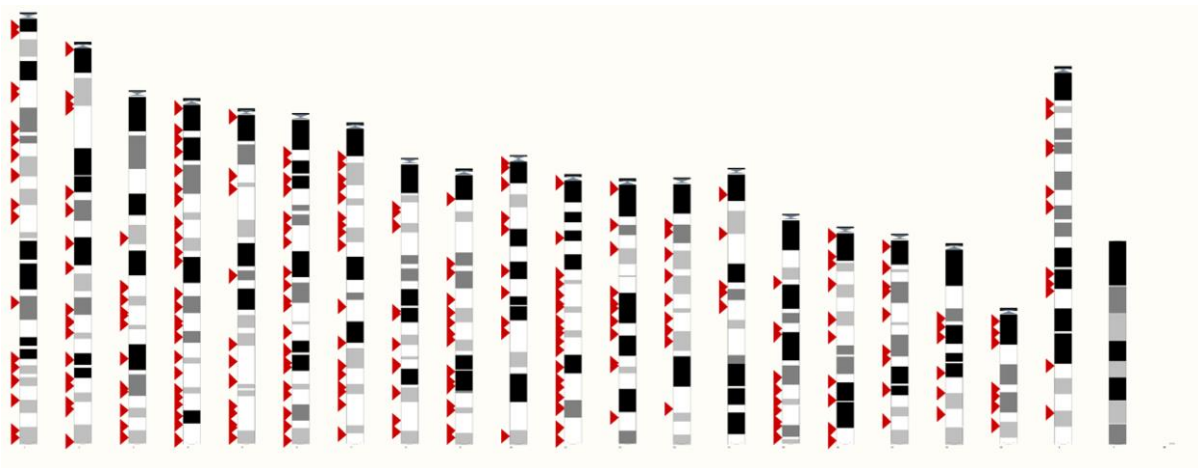


Fig. S3. Illustrative karyotype demonstrating the genomic distribution of the 424 genes studied (red arrowhead) across all chromosomes except Y.

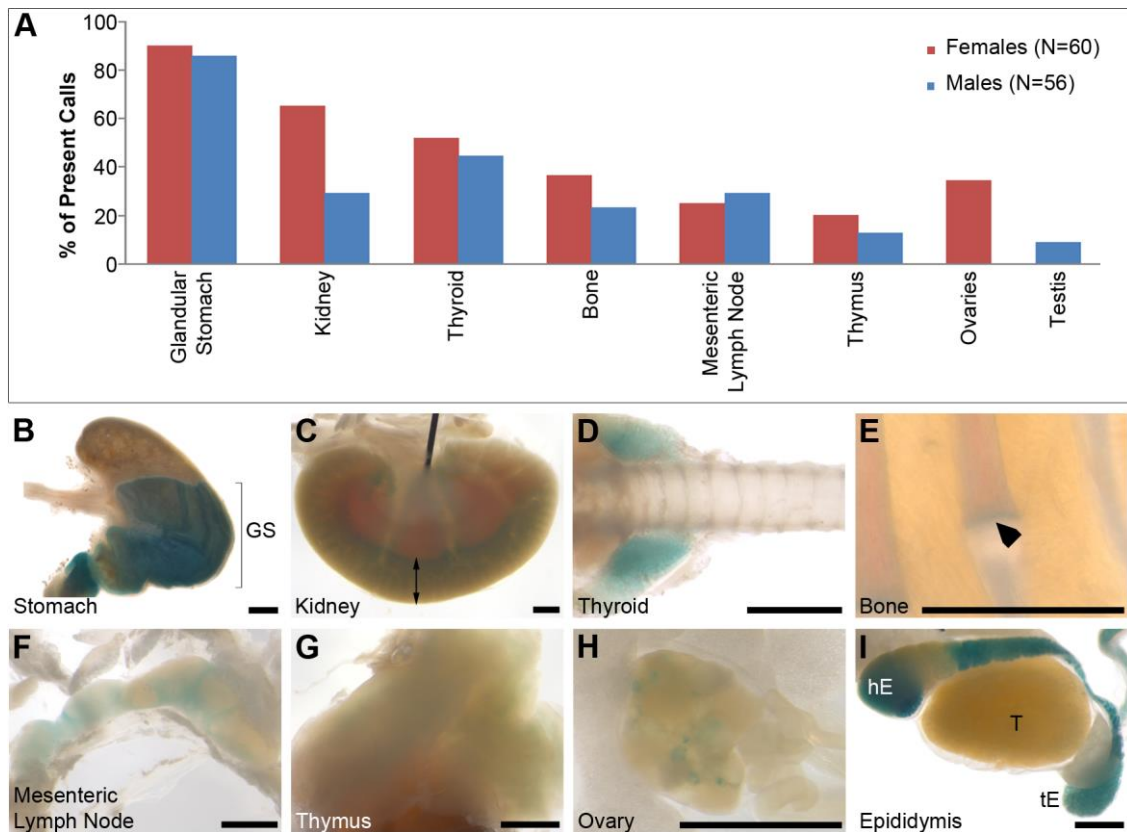


Fig. S4. Incidence, location and worst case examples of background interference observed in wild type animals (n = 116). To monitor background interference, wild type control animals were included each time mutant mice were processed. Glandular stomach (GS), testis (T), head of the epididymis (hE), tail of the epididymis (tE), kidney cortex (double arrow), bone (black arrowhead).

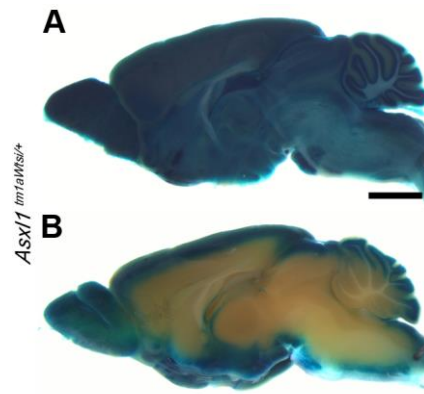


Fig. S5. Comparison of stain penetration of the brain in *Asx1^{tm1aWtsi/+}* animals using (A) a thick midline section or (B) the whole brain demonstrated improved penetration using thick midline sections.

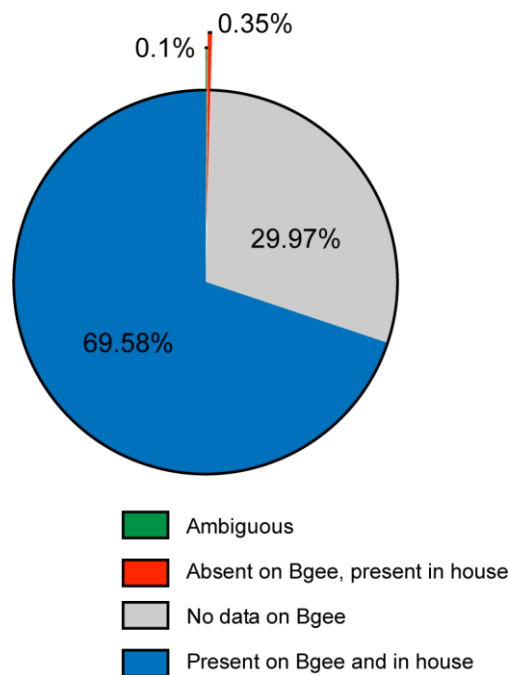


Fig. S6. Comparison of 100% consistent in-house calls of *present* with Bgee data revealed 69.58% of calls agreed (2,802 calls of in-house *present*, Bgee *present*), 0.35% disagreed (14 calls of in-house *present*, Bgee *absent*), 0.1% were ambiguous (4 calls of in-house *present*, Bgee *ambiguous*) and 29.97% represented novel data (1,207 calls of in-house *present*, Bgee *no data*).

Table. S1. Table presenting summary expression profiles for each of the 449 alleles studied. This encompassed 424 unique genes, 25 of which were studied in the tm1aWtsi and tm1bWtsi allelic configuration (highlighted in pink in column A). For each tissue a call of *present*, *not detected*, *ambiguous* or *no data* was assigned. Calls appended with + (e.g. Present+) are derived from a single mouse. Calls appended with ++ (e.g. Present++) are inferred using a defined set of rules (see Materials and methods section). All other calls were derived from multiple mice and were fully concordant within our dataset. Further annotation of each mouse line and gene is included. Most are self-explanatory. (Column BN) Homozygous viability at post-natal day 14 is presented as lethal, subviable or viable.

See separate excel spreadsheet

[Click here to Download Table S1](#)

Tab. S2. For each gene, a standard set of 39 tissues were assessed in the 424 unique genes studied (16,536 possible outcomes). For each structure a call of *present*, *not detected*, *ambiguous* or *no data* was assigned. The breakdown of these outcomes is presented across all data and split based on the confidence of each call. The majority of calls were derived from multiple mice and were fully concordant (100% concordance). The remaining calls were inferred using a defined set of rules (see Materials and methods section) or derived from a single mouse.

Expression analysis outcome	All Data	100% concordance	Rule based inference	N=1
Present	5404	3567	776	1061
Not detected	10295	8548	454	1293
Ambiguous	488	250	61	177
No Data	349	349	-	-

Tab. S3. For each gene, the total number of *lacZ* reporter positive tissues was calculated and grouped into one of five categories (0-3, 4-10, 11-20, 21-30 and 31-39 positive structures). Within each category, the number of genes for which homozygous mice were lethal, subviable and viable at P14 is presented. These data complement Fig. 5.

Number <i>lacZ</i> positive tissues	Number Lethal	Number Subviable	Number Viable	Total	% Viable	<i>P</i> value from Fisher's exact test comparison with 0-3 tissues
0-3	20	10	88	118	74.6	-
4-10	20	6	72	98	73.5	0.8771
11-20	21	9	48	78	61.5	0.0588
21-30	23	9	30	62	48.4	0.0009
31-39	22	9	14	45	31.1	<0.0001