***eLife’s* transparent reporting form**

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**Sample-size estimation**

* You should state whether an appropriate sample size was computed when the study was being designed
* You should state the statistical method of sample size computation and any required assumptions
* If no explicit power analysis was used, you should describe how you decided what sample (replicate) size (number) to use

Please outline where this information can be found within the submission (e.g., sections or figure legends), or explain why this information doesn’t apply to your submission:

No statistical methods or power analyses were used to predetermine sample sizes, but our sample sizes are similar to those reported previously (Susuki et al., 2013). Data distribution was assumed to be normal. This information can be found in Figure legends 1,2,3,4 and MATERIALS AND METHODS-Statistical analysis section.

**Replicates**

* You should report how often each experiment was performed
* You should include a definition of biological versus technical replication
* The data obtained should be provided and sufficient information should be provided to indicate the number of independent biological and/or technical replicates
* If you encountered any outliers, you should describe how these were handled
* Criteria for exclusion/inclusion of data should be clearly stated
* High-throughput sequence data should be uploaded before submission, with a private link for reviewers provided (these are available from both GEO and ArrayExpress)

Please outline where this information can be found within the submission (e.g., sections or figure legends), or explain why this information doesn’t apply to your submission:

Biological replicates are indicated as the number of animals used for each experiment; technical replicates include the number of nodes, neurons, and AIS counted and are also indicated in figure legends. No data points were excluded in this manuscript. This information can be found in Figure legends 1,2,3,4

**Statistical reporting**

* Statistical analysis methods should be described and justified
* Raw data should be presented in figures whenever informative to do so (typically when N per group is less than 10)
* For each experiment, you should identify the statistical tests used, exact values of N, definitions of center, methods of multiple test correction, and dispersion and precision measures (e.g., mean, median, SD, SEM, confidence intervals; and, for the major substantive results, a measure of effect size (e.g., Pearson's r, Cohen's d)
* Report exact p-values wherever possible alongside the summary statistics and 95% confidence intervals. These should be reported for all key questions and not only when the p-value is less than 0.05.

Please outline where this information can be found within the submission (e.g., sections or figure legends), or explain why this information doesn’t apply to your submission:

Unpaired, two-tailed Student’s t-test was performed for statistical analysis unless otherwise indicated. Individual data points were plotted in figures with N per group less than 10. P-values, exact values of N and precision measures are provided in source data. This information can be found in Figure legends 1,2,3,4 and MATERIALS AND METHODS-Statistical analysis section.

(For large datasets, or papers with a very large number of statistical tests, you may upload a single table file with tests, Ns, etc., with reference to sections in the manuscript.)

**Group allocation**

* Indicate how samples were allocated into experimental groups (in the case of clinical studies, please specify allocation to treatment method); if randomization was used, please also state if restricted randomization was applied
* Indicate if masking was used during group allocation, data collection and/or data analysis

Please outline where this information can be found within the submission (e.g., sections or figure legends), or explain why this information doesn’t apply to your submission:

Samples were allocated into experimental groups based on genotypes. Data were collected and processed randomly and analyzed using GraphPad Prism and Microsoft Excel. Experimenters were blinded to genotype in the following experiments: all behavioral experiments comparing *SptbF/F* and *Nes-cre; SptbF/F* mice, all behavioral experiments comparing *Sptbn4F/F* and *Nes-cre; Sptbn4F/F* mice, all analyses of AnkG spacing at AIS, all analyses of AnkG, Nav channel and GFP fluorescence intensity, and all analyses of EEG recordings. Experimenters were not blinded to genotype in the behavioral tests comparing *SptbF/F*; *Sptbn4F/F* and *Nes-cre; SptbF/F*; *Sptbn4F/F* mice due to obvious motor impairments. No data points were excluded. This information can be found in Figure legends 1,2,3,4 and MATERIALS AND METHODS-Statistical analysis section.

**Additional data files (“source data”)**

* We encourage you to upload relevant additional data files, such as numerical data that are represented as a graph in a figure, or as a summary table
* Where provided, these should be in the most useful format, and they can be uploaded as “Source data” files linked to a main figure or table
* Include model definition files including the full list of parameters used
* Include code used for data analysis (e.g., R, MatLab)
* Avoid stating that data files are “available upon request”

Please indicate the figures or tables for which source data files have been provided:

This information can be found in a ‘source data’ excel file.