**Purpose**: to understand temporal dynamics of PNN expression across experience.

* Do dynamics vary in specific regions over time / with repeated exposure to experience under our learning paradigm? Do these dynamics differ between WT and Het (in adolescence and/or adulthood)??
  + Here, I’ve chosen to start with 12-week-old adults (all sectioned with uniform thickness, stained simultaneously, and imaged with identical parameters).
  + This analysis will help to determine if this is worth pursuing long-term (more replicates, across ages).

**Notes**

* This cohort consists of 6 conditions (outlined below). Comparisons should be made *between conditions for each genotype* and *between genotypes for each condition*.
  + All 6 are mapped, and I have completed the 3 WT conditions thus far. I will have the Het conditions completed in 7-10 days.
* This cohort was mapped using the Allen Brain Institute Common Coordinate Framework (Allen CCF) in an ImageJ plugin, ABBA (Aligning Big Brains & Atlases), which provides significantly more adaptability in our image processing than using static maps. New parameters are listed in the data spreadsheet, and explanations / details for each parameter are provided below:
  + ***Rostral-Caudal Spatial Locations***
    - Old Method: The location of each tissue section along the rostral-caudal axis were previously identified by “map number.”
    - New Method: Using the Allen CCF, tissues are now identified by “z-position” along the rostral-caudal axis (unit = mm).
      * The principle is the same but using the Allen CCF in ABBA allows spatial adjustments of each tissue position in 10.0 µm increments.
  + ***XY Orientation (symmetrical vs. asymmetrical)***
    - Old Method: Images were split by hemisphere and mapped individually to account for asymmetry in tissue sections.
    - New Method: ABBA can adjust the Allen CCF automatically in both X- and Y-dimensions, allowing for a full tissue to be appropriately mapped for quantification.
      * Examples of positive and negative rotations in both dimensions are provided in **Figure 1**.
  + ***Subregional Specificity***
    - Old Atlas: Static maps from the Paxino’s mouse brain atlas show subregions of a specific size which appear and disappear at specific map numbers. Borders were adjusted during ROI creation to omit background.
    - New Atlas: ABBA adjust the atlas with extremely high specificity to each tissue section. Depending on tissue orientation / location, subregions can (***a***.) be quite small (smallest area of 3 completed conditions so far was 0.0005 mm2), and (***b***.) vary between left and right hemispheres.
* I have lumped most of the visual cortex subregions into a single region (VIS) and all auditory subregions into a single region (AUD).

**Conditions (n = 6)**

* **NW**: Naïve WT, no surrogate experience
* **SW P0-P1**: Surrogate WT with 2 consecutive days of experience
* **SW P0-P5**: Surrogate WT with 6 consecutive days of experience
* **NH**: Naïve Het, no surrogate experience
* **SH P0-P1**: Surrogate Het with 2 consecutive days of experience
* **SH P0-P5**: Surrogate Het with 6 consecutive days of experience

**Subregions for Comparison (n = 23)**

* **ACAd** (Anterior Cingulate Area – Dorsal Part)
* **ACAv** (Anterior Cingulate Area – Ventral Part)
* **AUD** (Auditory Cortex)
* **GU** (Gustatory Area)
* **MOp** (Primary Motor Area)
* **MOs** (Secondary Motor Area)
* **RSPagl** (Retrosplenial Area – Agranular Part)
* **RSPd** (Retrosplenial Area – Dorsal Part)
* **RSPv** (Retrosplenial Area – Dorsal Part)
* **SSp-bfd** (Primary Somatosensory Cortex – Barrel Field Subregion)
* **SSp-ll** (Primary Somatosensory Cortex – Lower Limb Subregion)
* **SSp-m** (Primary Somatosensory Cortex – Mouth Subregion)
* **SSp-n** (Primary Somatosensory Cortex – Nose Subregion)
* **SSp-tr** (Primary Somatosensory Cortex – Trunk Subregion)
* **SSp-ul** (Primary Somatosensory Cortex – Upper Limb Subregion)
* **SSp-un** (Primary Somatosensory Cortex – Undefined Subregion)
* SSs (Secondary Somatosensory Cortex)
* **TEa** (Temporal Association Area)
* **VIS** (Visual Cortex – All Areas)
* **VISa** (Visual Cortex – Anterior Area)
* **VISC** (Visceral Area)
* **VISp** (Primary Visual Cortex)
* **VISrl** (Visual Cortex – Rostrolateral Area)

Diagram

Description automatically generated

**Figure 1**. Examples of positive and negative rotations in the X- and Y-dimensions. Adjustment of the x-axis orientation facilitates better alignment of tissue edges while also changing the area of each subregion overall. Adjustments to the y-axis orientation assist in accounting for asymmetry.