**Transcriptomic Mapping of the 5-HT Receptor Landscape**

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**Supplemental Information**

# Supplementary Figures

**Figure S1. Htrs transcription and hierarchical clustering visualization.**

(A) Top: Linear regression model fit between prevalence (y axis) and amount of intracellular expression (x axis) for each Htrs. Bottom: violin plots showing distribution of intracellular amount of expression for each Htrs. (B) Heatmaps showing relationship between neurotransmitter, class and neighborhoods. (C) Bar plots representing the size of each group when cells are categorized by neurotransmitter (top), neighborhood (middle) and class (botttom).

**Figure S2. UMAP distribution of each Htr**

UMAP-associated neurotransmitter, class and dissection region metadata (first row) and UMAP distribution of each Htr.

**Figure S3. Htrs features across neighborhoods and classes**

(A) Htrs prevalence across neighborhoods (left). Confusion matrix related to the decoding of neighboroods classification from Htrs transcription (middle). SHAP values matrix associated to the neighborhood decoding (right). (B) Confusion matrix related to the decoding of classes classification from Htrs transcription (left). SHAP values matrix associated to the class decoding (right).

**Figure S4. Correlation and co-localization patterns across neighborhoods**

(A) Correlation matrices between pairs of Htrs in each neighborhood. (B) co-localization matrices of Htrs transcription in each neighborhood.

**Figure S5. Htr1d transcription**

(A) On the left, Htr1d prevalence across neighborhoods with squared Pearson correlation coefficient (R²) between RNA-seq and MERFISH dataset. On the right, amount of Htr1d RNA detected using RNA-seq (top) and MERFISH (bottom). (B) Amount of co-localization with each Htrs by cells expressing Htr1d RNA (left). Number of Htrs RNA detected in cells expressing Htr1d RNA (right). (C) Prevalence of Htr1d RNA across all classes of cells in RNA-seq and MERFISH dataset. Inset represents the linear regression between the two datasets. On the right, absolute number of cells expressing Htr1d RNA by class ranked in descending order (top ten). (D) Prevalence of Htr1d RNA across divisions (left) and structures (right). Inset represents the proportion of cells expressing Htr1d RNA that belongs to enriched clusters. (E) Top: Prevalence of cells from enriched clusters across the antero-posterior axis, identified in the scRNA-seq dataset and cross-referenced in the MERFISH dataset. Bottom: average amount of RNA expression found in enriched clusters cross-referenced in the MERFISH dataset. (F) Expression of Htr1d RNA detected by MERFISH in 4 representative slices. Border color represents the position on the antero-posterior axis.

**Supplementary Figure S6. Htr5a transcription**

(A) On the left, Htr5a prevalence across neighborhoods with squared Pearson correlation coefficient (R²) between RNA-seq and MERFISH dataset. On the right, amount of Htr5a RNA detected using RNA-seq (top) and MERFISH (bottom). (B) Amount of co-localization with each Htrs by cells expressing Htr5a RNA (left). Number of Htrs RNA detected in cells expressing Htr5a RNA (right). (C) Prevalence of Htr5a RNA across all classes of cells in RNA-seq and MERFISH dataset. Inset represents the linear regression between the two datasets. On the right, absolute number of cells expressing Htr5a RNA by class ranked in descending order (top ten). (D) Prevalence of Htr5a RNA across divisions (left) and structures (right). Inset represents the proportion of cells expressing Htr5a RNA that belongs to enriched clusters. (E) Prevalence of cells from enriched clusters across the antero-posterior axis, identified in the scRNA-seq dataset and cross-referenced in the MERFISH dataset.

**Figure S7. Htr5b transcription**

(A) On the left, Htr5b prevalence across neighborhoods with squared Pearson correlation coefficient (R²) between RNA-seq and MERFISH dataset. On the right, amount of Htr5b RNA detected using RNA-seq (top) and MERFISH (bottom). (B) Amount of co-localization with each Htrs by cells expressing Htr5b RNA (left). Number of Htrs RNA detected in cells expressing Htr5b RNA (right). (C) Prevalence of Htr5b RNA across all classes of cells in RNA-seq and MERFISH dataset. Inset represents the linear regression between the two datasets. On the right, absolute number of cells expressing Htr5b RNA by class ranked in descending order (top ten). (D) Prevalence of Htr5b RNA across divisions (left) and structures (right). Inset represents the proportion of cells expressing Htr5b RNA that belongs to enriched clusters. (E) Prevalence of cells from enriched clusters across the antero-posterior axis, identified in the scRNA-seq dataset and cross-referenced in the MERFISH dataset.

**Supplementary Figure S8. Htr2b transcription**

(A) On the left, Htr2b prevalence across neighborhoods with squared Pearson correlation coefficient (R²) between RNA-seq and MERFISH dataset. On the right, amount of Htr2b RNA detected using RNA-seq (top) and MERFISH (bottom). (B) Amount of co-localization with each Htrs by cells expressing Htr2b RNA (left). Number of Htrs RNA detected in cells expressing Htr2b RNA (right). (C) Prevalence of Htr2b RNA across all classes of cells in RNA-seq and MERFISH dataset. Inset represents the linear regression between the two datasets. On the right, absolute number of cells expressing Htr2b RNA by class ranked in descending order (top ten).

**Figure S9. Htr6 transcription**

(A) On the left, Htr6 prevalence across neighborhoods with squared Pearson correlation coefficient (R²) between RNA-seq and MERFISH dataset. On the right, amount of Htr6 RNA detected using RNA-seq (top) and MERFISH (bottom). (B) Amount of co-localization with each Htrs by cells expressing Htr6 RNA (left). Number of Htrs RNA detected in cells expressing Htr6 RNA (right). (C) Prevalence of Htr6 RNA across all classes of cells in RNA-seq and MERFISH dataset. Inset represents the linear regression between the two datasets. On the right, absolute number of cells expressing Htr6 RNA by class ranked in descending order (top ten).

**Figure S10. Htr3a transcription**

(A) On the left, Htr3a prevalence across neighborhoods with squared Pearson correlation coefficient (R²) between RNA-seq and MERFISH dataset. On the right, amount of Htr3a RNA detected using RNA-seq (top) and MERFISH (bottom). (B) Amount of co-localization with each Htrs by cells expressing Htr3a RNA (left). Number of Htrs RNA detected in cells expressing Htr3a RNA (right). (C) Prevalence of Htr3a RNA across all classes of cells in RNA-seq and MERFISH dataset. Inset represents the linear regression between the two datasets. On the right, absolute number of cells expressing Htr3a RNA by class ranked in descending order (top ten). (D) Prevalence of Htr3a RNA across divisions (left) and structures (right). Inset represents the proportion of cells expressing Htr3a RNA that belongs to enriched clusters. (E) Top: Prevalence of cells from enriched clusters across the antero-posterior axis, identified in the scRNA-seq dataset and cross-referenced in the MERFISH dataset. Bottom: average amount of RNA expression found in enriched clusters cross-referenced in the MERFISH dataset. (F) Expression of Htr3a RNA detected by MERFISH in 4 representative slices. Border color represents the position on the antero-posterior axis.

**Figure S11. Htr3b transcription**

(A) On the left, Htr3b prevalence across neighborhoods with squared Pearson correlation coefficient (R²) between RNA-seq and MERFISH dataset. On the right, amount of Htr3b RNA detected using RNA-seq (top) and MERFISH (bottom). (B) Amount of co-localization with each Htrs by cells expressing Htr3b RNA (left). Number of Htrs RNA detected in cells expressing Htr3b RNA (right). (C) Prevalence of Htr3b RNA across all classes of cells in RNA-seq and MERFISH dataset. Inset represents the linear regression between the two datasets. On the right, absolute number of cells expressing Htr3b RNA by class ranked in descending order (top ten).

**Table 1. Neighborhoods descriptions**

Description of cells belonging to each neighborhood group.

**Table 2. Region acronyms**

Allen Mouse Brain Common Coordinate Framework (CCF) acronyms.

**Table 3. Htrs defining features**

Table listing the defining feature of each Htr trascription pattern.