

Choline Acetyltransferase expression in the central nervous system of the pond snail *Lymnaea stagnalis*



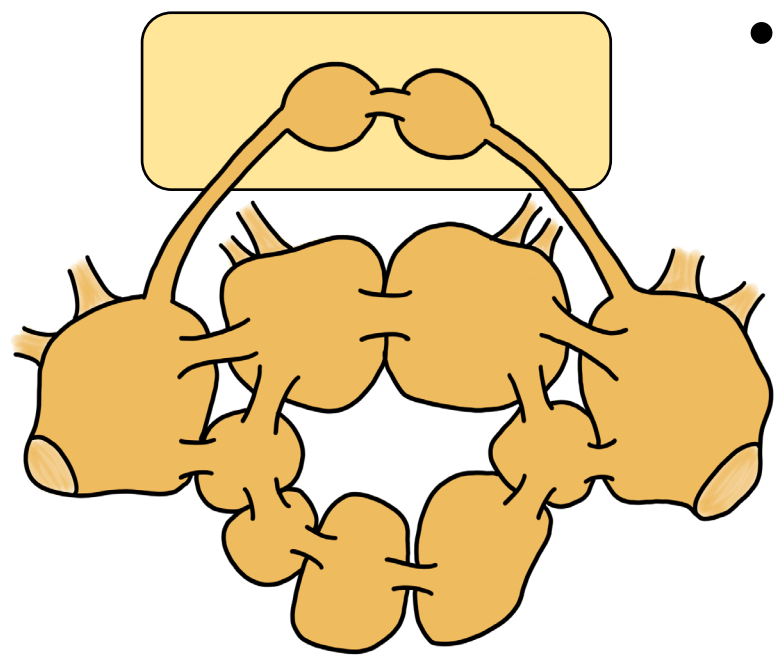
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Introduction

Gastropod Neuroanatomy:

- Gastropods have been key models in understanding how nervous systems control behavior
 - Neuroanatomy underlies function
- Structure ↔ Function
- L. stagnalis* Nervous System
- Central brain
 - Focus: Buccal ganglia



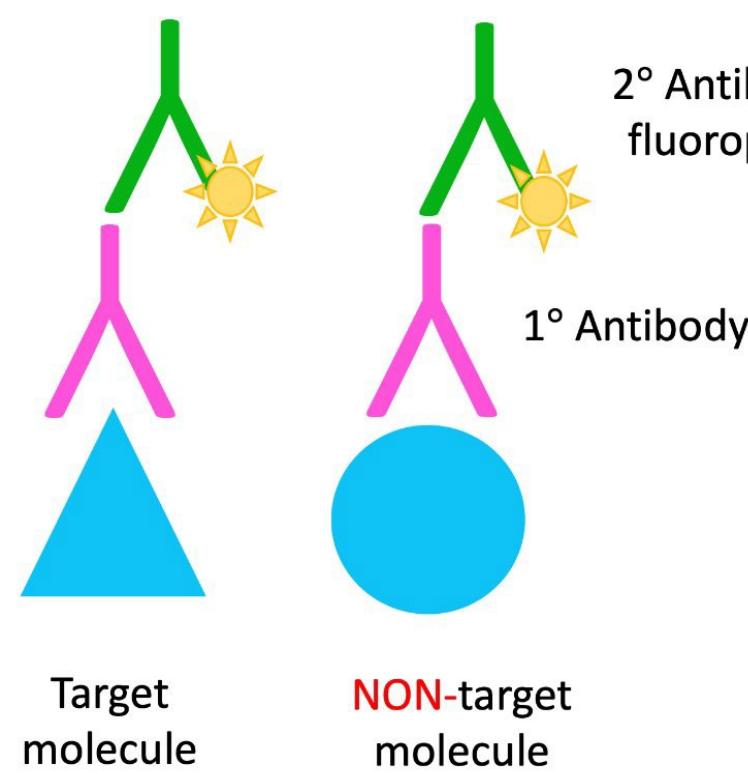
Labelling Comparison:

- Traditional = IHC
- Immunohistochemistry**
- Based on protein structure (using an antibody)
 - Visualization of protein expression
- New = HCR
- in situ Hybridization Chain Reaction**
- Based on gene sequences
 - Visualization of mRNA expression

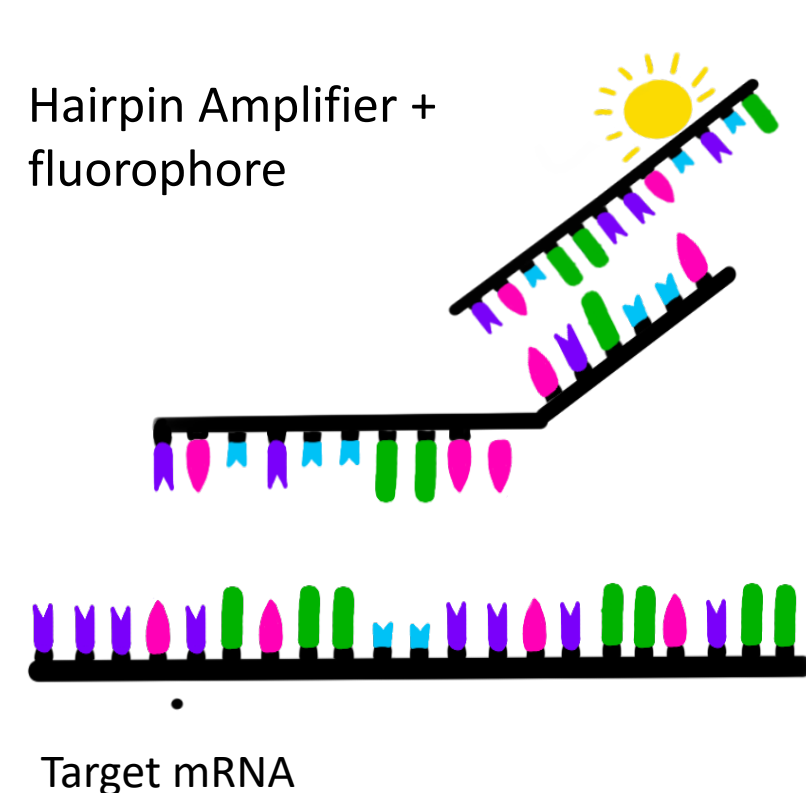
Target:

- Choline acetyltransferase (ChAT)
- Enzyme
 - Synthesizes acetylcholine (a neurotransmitter)
- 4 methods compared:
3 IHC antibodies vs HCR

IHC



HCR

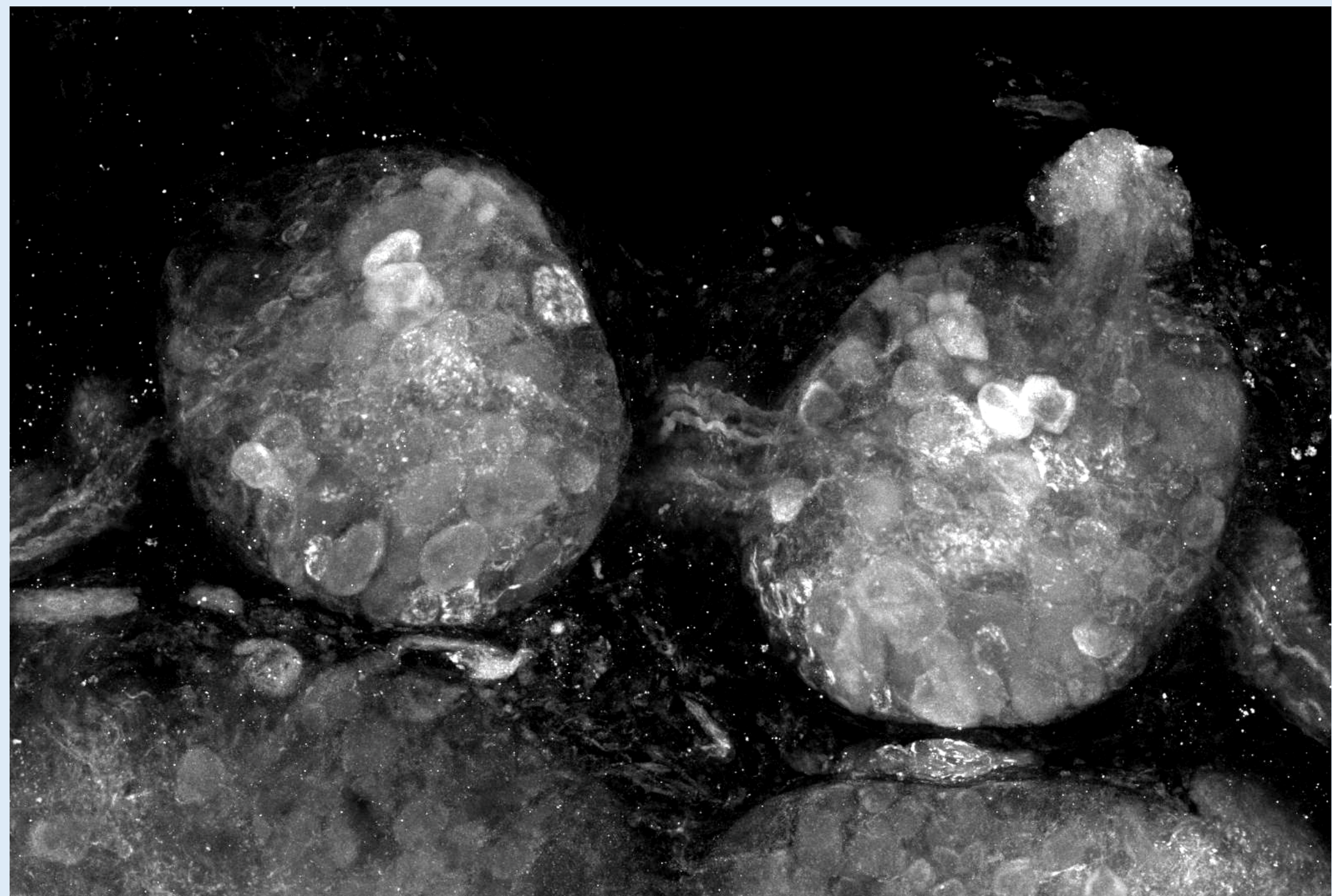


Hypothesis: Labelling in all methods will produce similar expression

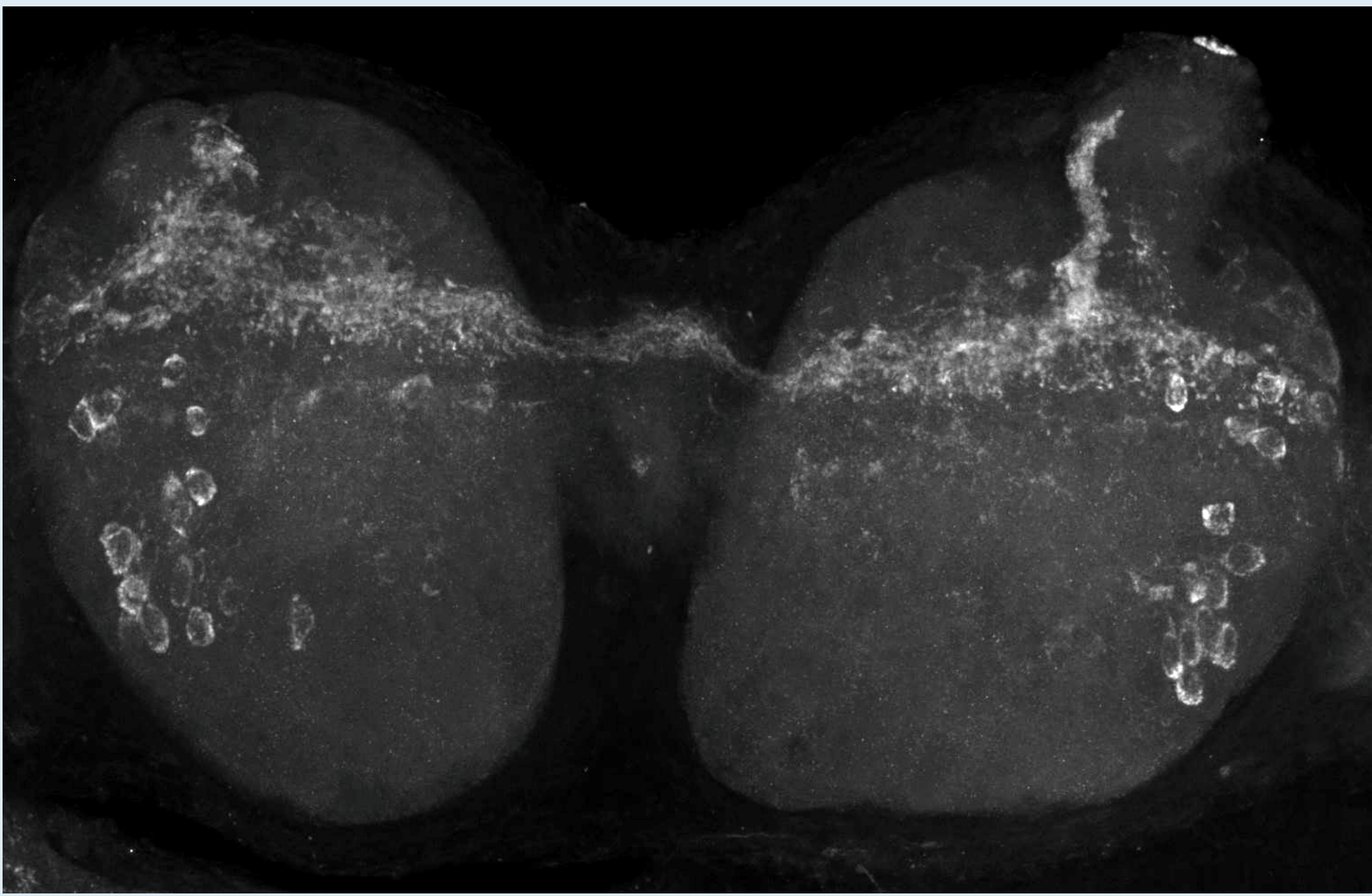
Results

Central Nervous System: Buccal Ganglia

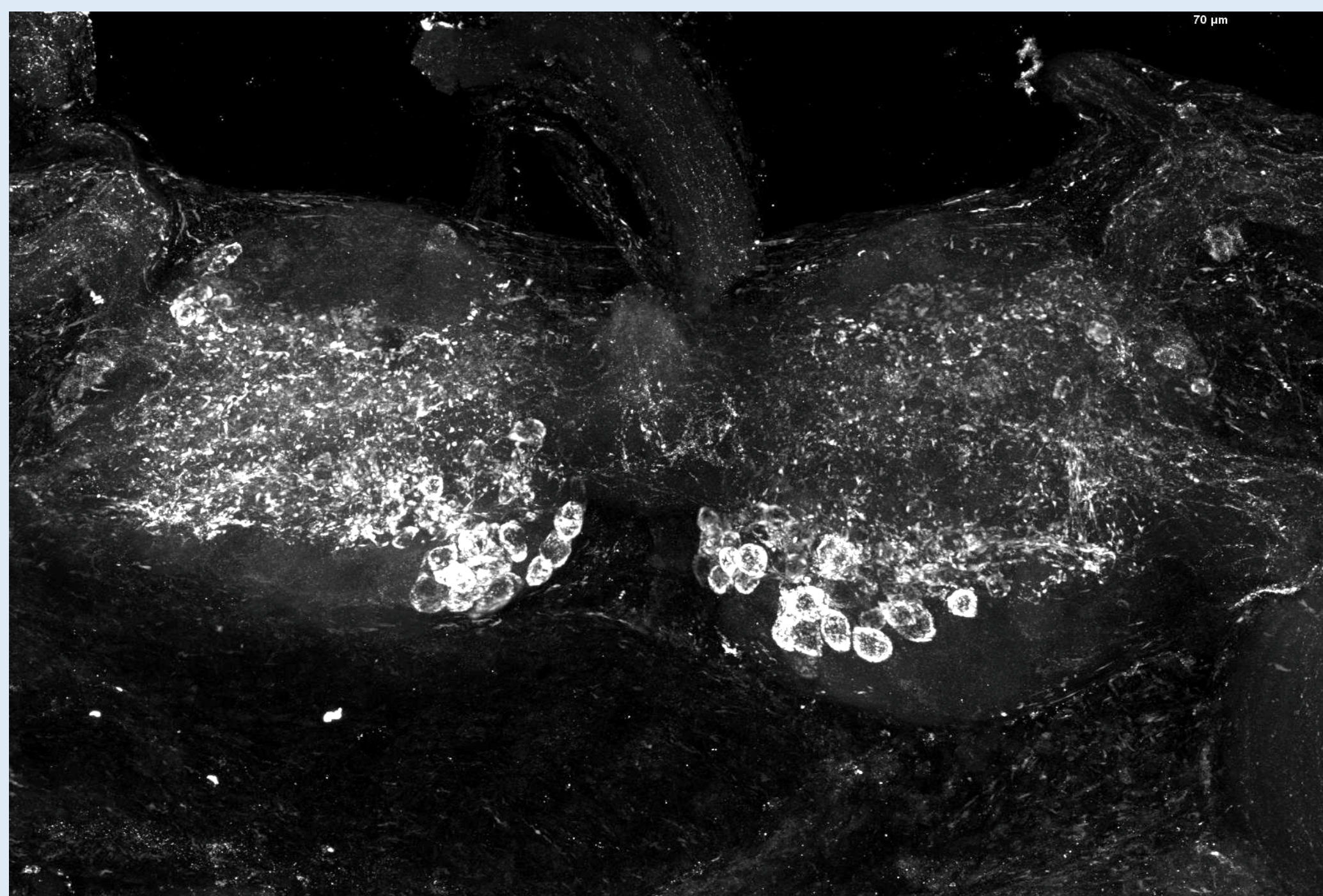
IHC



ChAT1

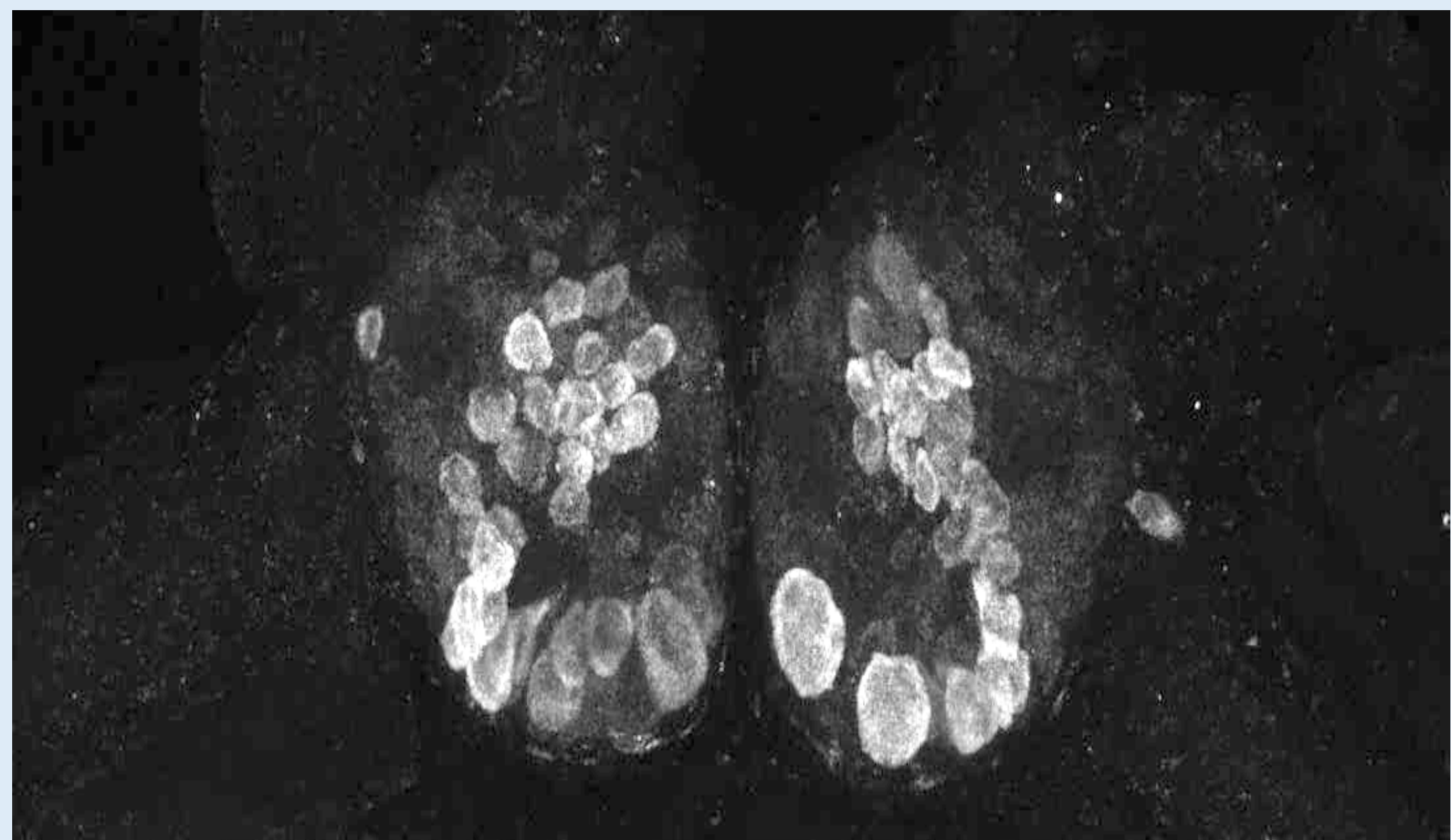


ChAT2



ChATCB

HCR



Cell Analysis

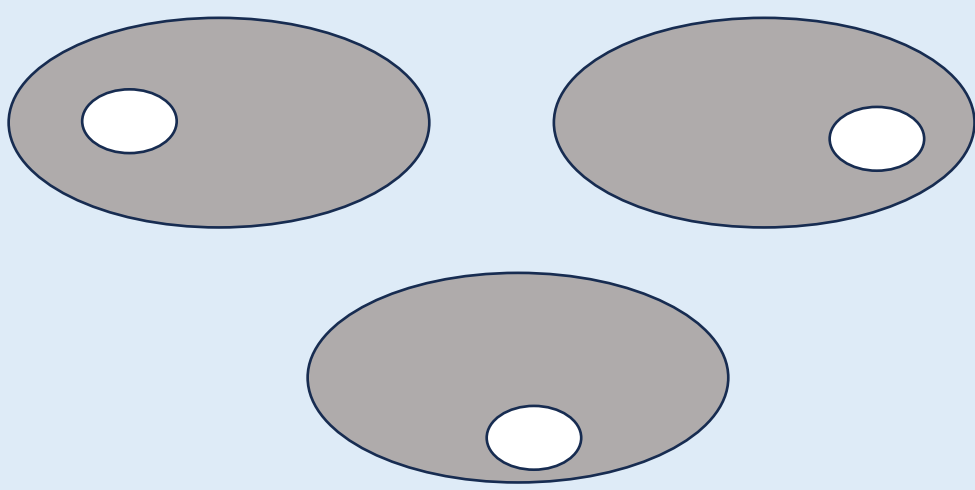
i. Cell shape



ii. Cell size



iii. Cell location



Cell Characteristics

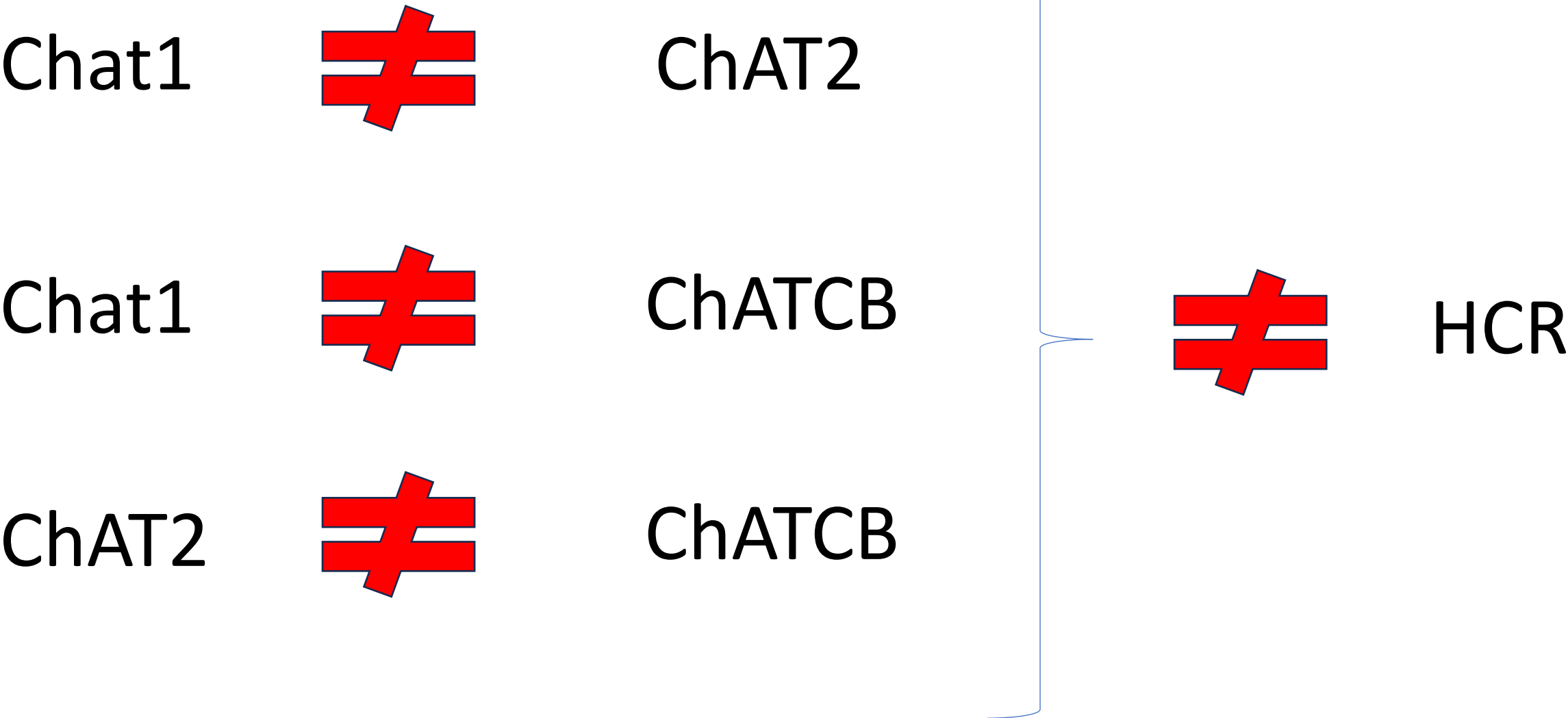
ChAT1 → Circular, large, medial

ChAT2 → Oblong, small, lateral

ChATCB → Oblong, small, posterior/medial

HCR → Circular, large, distributed

Overall IHC vs HCR Label Comparison



Similarities:

- ChAT 1 and HCR center clusters
- ChAT1 and ChATCB small posterior/medial clusters

Conclusions

Probable explanation: Labelling of different versions

Possible explanations:

- One or more not labelling ChAT
- Labelling different versions of ChAT
 - Splice variants
 - Post translational modifications
 - Isoforms

Conclusion: Hypothesis was not supported

Acknowledgements

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Lymnaea stagnalis



Future Directions

- Determine specific labeling of antibodies
- Explore ChAT in the *Lymnaea* genome