Introduction:
Understanding the basis of brain function requires knowledge of cortical operations over wide spatial scales, and quantitative analysis of brain activity in well-defined brain regions. Matching an anatomical atlas to brain functional data requires substantial labor and expertise. We have developed an automated machine learning-based registration and segmentation approach for quantitative analysis of mouse mesoscale cortical images. A deep learning model identifies nine cortical landmarks using only a single raw fluorescent image. Another fully convolutional network was adapted to delimit brain boundaries. This anatomical alignment approach was extended by adding three functional alignment approaches that use sensory maps or spatial-temporal activity motifs. We present this methodology as MesoNet, a robust and user-friendly analysis pipeline using pre-trained models to segment brain regions as defined in the Allen Mouse Brain Atlas. This Python-based toolbox can also be combined with existing methods to facilitate high-throughput data analysis.

Method:
Set up of wide-field calcium imaging and definition of landmarks.

Performance of brain-to-atlas transformation for clustering cortical activity motifs.

Method & results:
Performance of automated landmark estimation.

Testing the brain-to-atlas approach across different lines of fluorescent protein mice.

Conclusion:
We developed atlas-to-brain and brain-to-atlas alignment approaches using anatomical landmarks and brain boundaries. We also extend our pipeline to make use of functional sensory maps and spontaneous cortical activity motifs. We included a GUI in order to improve ease of use for non-specialists, enabling the use of our toolbox in a variety of projects and conditions.

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