## Linking physiology and morphology in two types of honeybee projection neurons.

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It is widely acknowledged that a neuron's function is reflected in both, its morphology and its electrophysiological properties. However it is very difficult to conclude from either of these two characteristics to the other one. Here we use hierarchical clustering and machine learning to explore differences between two types of projection neurons (PNs) in the honey bee antennal lobe (AL). These morphologically similar neuron families leave the AL via different tracts, the lateral (I-APT PNs) and the medial Antenno Protocerebral Tract (m-APT PNs). The existence of these two separated pathways suggests functional differences between its neurons. But is this assumption supported by systematic differences in electrophysiological properties? And if so, which properties are most helpful to separate m-APT PNs from I-APT PNs?

We analyzed data from 122 extracellularly recorded AL units, which, based on electrode placement were unambiguously identified as belonging to the m- or the I-APT [1]. For each unit well established measures of electrophysiological response activity (features) were estimated. To find the set of features which most efficiently describes the difference between units from I- and m-tract, we performed hierarchical clustering (Euclidian distances, Wards linkage) based on the principal components (PCs) of every possible combination of properties. We validated the resulting set of features by sorting neurons using a support vector machine (SVM) as an alternative approach.

We find that significant between-group differences exist for individual features. Alone however, none of these features suffices to classify I-APT and m-APT units. Clustering by means of electrophysiological properties separates I-APT units and m-APT units significantly above chance level (Matthews correlation coefficient 0.47, chance level 0.19). The features which contributed most to the separation of units from different tracts were CV2 (a measure of spike time irregularity), Fano-factor (spike count variability), spontaneous firing rate and a unit's lifetime sparseness (odor tuning width). Sorting units with a support vector machine (SVM)approach performed superior to hierarchical clustering (Matthews correlation coefficient 0.65). Measures which contributed to the SVM-model were again, CV2, Fano-factor and measures of firing rate.

Our results indicate that electrophysiological properties of units from the I-APT and the m-APT show subtle differences in characteristic electrophysiological properties. What has been described often a times phenomenologicaly, are differences in odor specificity, firing frequency and sometimes rate profiles [2,3,4]. These phenomena are well captured in those properties we identified as best separators: life time sparseness (odor specificity), CV2 (spike time irregularity), Fano-factor and rate measures (frequency and profile). We conclude that hierarchical clustering is a useful tool to identify properties of electrophysiological activity, which efficiently describe differences between morphologically distinct PNs. SVMs offers a superior method to predict PN morphology from electrophysiology.

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