Automatic High Throughput Measurement of Feeding Behavior in Drosophila

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Introduction

Feeding is an essential component of an animal's behavior and better understanding of the principles governing food intake will provide future insights into the interaction between physiology and behavior. Fruit flies (*Drosophila melanogaster)* have emerged as a powerful model to study the molecular and neuronal mechanisms underlying feeding behavior, but it remains challenging to quantify feeding in flies, due to their small size and the minute quantities of food they ingest. Existing methods are a potential limiting factor in the study of feeding in *Drosophila* because of their poor temporal resolution, reliance on manual annotation and inability to access the behavior of individual animals. Here, we have developed and validated an automated, high resolution behavioral monitoring system called flyPAD (fly Proboscis and Activity Detector), that uses capacitance–based measurements to detect the physical interaction of individual *Drosophila* with food.

Methods

The device uses ultralow current capacitance to digital converters to sample capacitance across 2 electrodes at 100 Hz and is capable of measuring feeding behavior of individually housed flies. To allow for high throughput behavioral analysis our system can record the behavior of up to 32 individual flies. We have furthermore developed an algorithm to automatically analyze the capacitance signal to extract behavioral features based on the shape of the capacitance trace. Our algorithm detects individual proboscis extension events ("sips") as well as periods of activity on the food ("activity bouts"). In order to validate our approach we have performed simultaneous capacitance and high resolution video recording.

Results

A comparison of the ethograms generated by manual annotation with the results of our automated method confirmed the accuracy of our approach. The number of "sips" detected by our algorithm was strongly and significantly correlated with the number of proboscis contacts with the food (Rho=0.874, $p<<0.0001$). The algorithm detected 92.5% of the sips tabulated via manual scoring, while missing 7.5% and generating 7.5% false sips. We have found that feeding from a non-liquid food induces a pattern of highly stereotyped rhythmic proboscis extensions and retractions that is suggestive of an underlying central pattern generator controlling the feeding motor program. By adapting the luciferase bioluminescent techniques to measure the intake in single flies we measured that 1 "sip" detected by the flyPAD corresponds on average to an intake of 1 nl of food. The analysis of ingestion dynamics and the microstructure of meals allowed us to dissect the behavioral elements mediating the homeostatic response of the fly to starvation and satiation. These results uncover several similarities with rodents and humans, highlighting a potential conservation of strategies that regulate food intake across phyla.

Conclusions

Our work opens up new avenues for studying the neuronal basis of feeding behavior and food intake at different time scales in an automated and high-throughput manner. The ability to subdivide homeostatic changes in feeding into specific changes in feeding strategies opens up new opportunities to explore how molecular mechanisms,

neuronal circuits and metabolic processes interact to control food intake and nutrient homeostasis to optimize life history traits such as reproduction and aging.