



## Startle Responses and Behavioural State Switching

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Under sudden and uncertain threat, animals can trigger a switch into prolonged defensive behavioural and physiological states. Startle responses—rapid, short-latency motor reactions like flinches, jumps, or pauses—are widespread across phyla. We hypothesise that startles play a role in behavioural state switching, interrupting ongoing behaviours and facilitating the onset of sustained defensive responses.

We first addressed how to classify startle responses. Rather than focusing on a single behaviour, we tracked thousands of flies responding to looming stimuli and observed diverse, short-latency, transient responses. To determine the startle response time window, we trained a supervised neural network to identify whether a loom had occurred or not solely based on body part movement. By shifting time windows, we identified a startle behavioural signature in a time window centred on loom offset. Next we used an interpretable classifier (JAABA) to calculate hundreds of features from body part coordinates, learn thresholds that partition the data and weight their contribution to the classification. We aimed to identify startle subtypes through clustering, however the weights of features relating to velocity dominated. Jumps and pauses clustered separately, potentially reflecting different startle magnitudes.

Startle responses show modulation by prior behavioural state. To study threat-potential, flies were exposed to looming or control stimuli, then tested with blackout stimuli, which elicit jumps and pauses but not sustained responses. Loomed flies jumped more to blackouts and in both groups jumping flies were invariantly stationary beforehand. Our next aim is to manipulate prior velocity in closed loop or optogenetically to understand how it shapes startle. As enhanced startle reactivity is a hallmark of anxiety disorders, *Drosophila* may offer a powerful, high-throughput platform for future investigation.

**startle, defensive behaviour, behavioural state, interpretable classifiers**