

## **Why sniff fast? High-frequency sniffing, odor discriminations, and receptor neuron activation in the behaving rat**

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Detection of olfactory stimuli by olfactory receptor neurons (ORNs) depends on sniffing, a complex behavior that controls odorant flow into the nasal cavity. In rodents, sniff frequency varies from ~1.5 - 12 Hz. Several studies have shown that freely-moving animals increase sniff frequency to >4 Hz around the time of odor sampling, implying that high-frequency ('fast') sniffing is important for odor discrimination. However, we recently reported that, in a head-fixed odor discrimination paradigm that removes confounds from locomotion, rats can accurately perform odor discriminations without engaging in fast sniffing (Verhagen et al. 2007, *Nat Neurosci* 10:631). Here, we further explore the importance of sniffing patterns in odor discrimination using the head-fixed paradigm, while also imaging ORN input to the olfactory bulb (OB) during task performance. We used a two-odor, lick/no-lick discrimination task and continuously measured sniffing in Long-Evans female rats. Head-fixed rats spent the majority of their time sniffing at low frequencies (~2.5 Hz), similar to sniff frequencies in resting, unrestrained rats. Most rats showed only slight (~1 - 2 Hz) increases in frequency around the time of odor presentation, even though discrimination accuracy was high (>85%). Some rats, however (3/11), often exhibited brief bouts of fast (5 - 8 Hz) sniffing during the first 500 ms of odor onset. These bouts consisted of 3 - 4 inhalations which were smaller in amplitude and shorter in duration than most other sniffs. Imaging ORN responses evoked during these fast-sniff bouts revealed that the early sniffs in the bout typically failed to evoke any ORN responses, and that the bout ended as soon as ORNs were activated. For all rats, sniff frequency did not increase significantly even when we increased task difficulty by asking animals to discriminate between enantiomer pairs or discriminate similar binary odor mixtures, nor did they increase while learning new odor discriminations. Reducing odor intensity or increasing mixture similarity to the point where discriminations failed also did not elicit high-frequency sniffing. These results differ from those of earlier studies in freely-moving rats performing a nose-poke into an odor port, and suggest that fast sniffing is not necessary, or even important, for odor discrimination. Instead, fast sniffing may be a general strategy used in scanning or searching for changes in the olfactory environment.