The circadian clock in the mammalian olfactory system: mechanisms of entrainment and functional aspects

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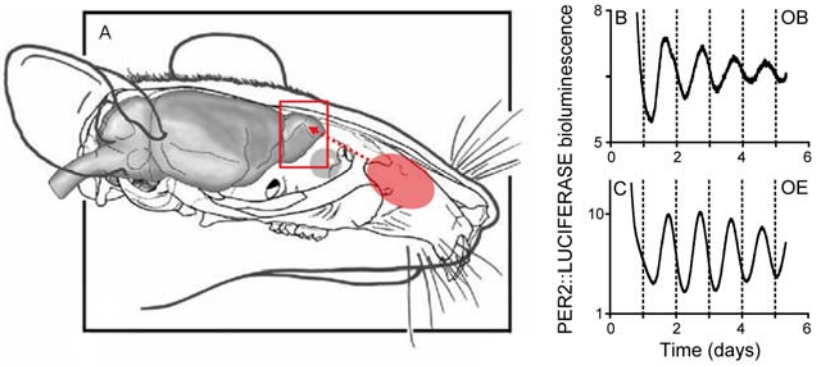
Overview:
In mammals, the suprachiasmatic nuclei (SCN) of the anterior hypothalamus control a multitude of daily (circadian) physiological and behavioral processes, for example, the sleep-wake cycle or the periodical release of hormones. Although circadian oscillations have been described in many mammalian peripheral tissues, as for example in liver cells and fibroblasts, the function of these local clocks is still unknown. There is abundant evidence for the majority of these local clocks being driven by the SCN. In this respect, it is remarkable that we recently found SCN-independent oscillations in the olfactory bulbs (OB) of the live rat. The function of this OB clock is still unknown, however, previous studies indicate that the OB does not only impact the circadian system, but also influences the photoperiodic regulation of reproduction in rodents.



A) Lateral view of a mouse head: the olfactory epithelium (OE, red area) of the nasal cavity projects directly to the OB (red rectangle) (modified after Lennington *et al*., Reprod Biol Endocrinol, 2003). B, C: Explants of mice expressing luciferase under the control of the clock gene *Per2* (PER2::LUC knockin mice) display bioluminescence in the presence of luciferin. *Per2*-driven bioluminescence (counts/5min x 10^4) oscillates in the OB (B) and in the OE (C) for at least 4 days.

Rationale:
The goal of this project is to investigate the effects of daily recurring odor and light signals on the synchronization behavior (entrainment) and clock gene expression in mice. In particular, we focus on the interplay between different components of the olfactory system (olfactory epithelium (OE), OB) and the SCN.
A central question of my research is whether the olfactory clock can be entrained by periodic odor stimulation in the absence of the SCN. Subsequently, we will study the effects of specific olfactory stimuli and common olfactory neurotransmitters on single cells of the OB and OE.
Ultimately, we strive to establish a functional link between olfactory synchronization of the OB clock and the regulation of reproduction.

We study these questions using the following methods:

- Individual wheel-running registrations in order to detect circadian behavior while applying light and odor stimuli
- real-time bioluminescence assays with tissue cultures of PER2::LUC knockin mice
- immunohistochemistry with frozen sections
- qPCR and Western blot
- single cell low-light bioluminescence imaging of tissue and dispersal cultures

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Curriculum Vitae

2006 - present **Project leader** in the Chronobiology group of Prof. Achim Kramer, Universitätsklinikum Charité, Berlin, Germany.

The circadian clock in the mammalian olfactory system: mechanisms of entrainment and functional aspects.

Uncovering the molecular mechanisms regulating rhythmic olfactory sensitivity in mammals

2003 – 2006 DFG-funded research grant and **postdoc** with Prof Erik Herzog, Washington University in St. Louis, USA.
Circadian clocks in the mammalian brain: Are the suprachiasmatic nuclei required for sustained oscillations in other brain areas?

1997 - 2002 **Doctoral thesis** at the Max-Planck-Research Centre for Ornithology, Andechs/ Ludwig-Maximillians-University, Munich, Germany.
Molecular and neurochemical characterization of the hypothalamic circadian oscillator in the house sparrow (*Passer domesticus*)

Selected Publications

Abraham, U.; Westermark, P.; Granada, A.;.Heine, M.; Kramer, A and Herzel, H. (2010) Coupling governs entrainment range of circadian clocks. *Mol. Syst. Biol.* **6**:438

Keller, M.; Mazuch, J.; Abraham, U.; Eom, G.; Herzog. E.D.; Volk, H.-D.; Kramer, A.; and Maier, B. (2009) A circadian clock in macrophages controls inflammatory immune responses. *Proc Natl Acad Sci U S A* **106**(50):21407-12.

Gross, S.; Abraham, U.; Herzog, E.D.; Piwnica-Worms, D.R. (2006) Continuous delivery of D-luciferin by implanted micro-osmotic pumps allows true real-time bioluminescence imaging of luciferase activity *in vivo. Molecular Imaging* **6**(2), 121-130.

Abraham, U.; Prior, J.D.; Granados-Fuentes, D.; Piwnica-Worms, D.R. and Herzog, E.D. (2005) Independent circadian oscillations of *Period1* in specific brain areas *in vivo* and *in vitro. J. Neurosci*. **25**(38), 8620-8626.

Granados-Fuentes, D.; Prolo, L. M.; Abraham, U. and Herzog, E.D. (2004) The suprachiasmatic nucleus entrains, but does not sustain, circadian rhythmicity in the olfactory bulb. *J. Neurosci.* **24**(3), 615-619.

Abraham, U.; Albrecht, U. and Brandstätter, R. (2003) Hypothalamic circadian organization in birds. II. Clock gene expression. *Chronobiol. Int.*, **20**(4), 657-669.

Abraham, U.; Albrecht, U.; Gwinner, E. and Brandstätter, R. (2002) Spatial and temporal variation of *passerPer2* gene expression in two distinct cell groups of the suprachiasmatic hypothalamus in the house sparrow (*Passer domesticus*). *Eur. J. Neurosci.*, **16**, 429-436.