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Modulations of olfactory responses by GAP junctions uncoupling in the OM.

Mikaël LE BOURHIS, Bertrand BRYCHE, Denise GREBERT, Edith PAJOT, Patrice CONGAR and Nicolas MEUNIER.

Glial cells not only act as supporting cells, they are clearly involved in nervous communication. Modulation of odorant detection takes place as early as the peripheral level, on the olfactory sensory neurons. Those neurons are surrounded by glial like cells called **sustentacular cells (SCs)**. These cells maintain both the structural and ionic integrity of the olfactory epithelium. They are electrically coupled through **GAP junctions** (Vogalis *et al.*, 2005).

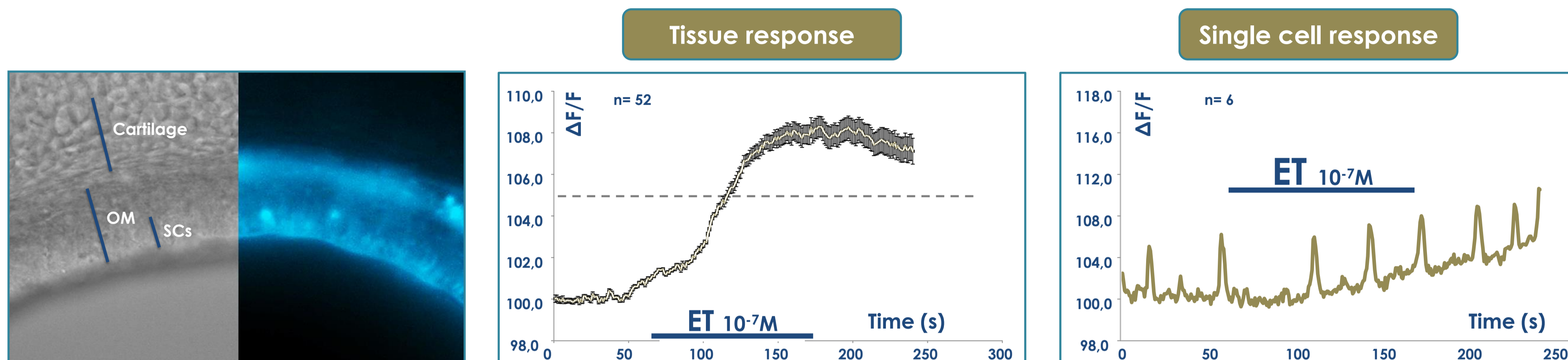
Using **endothelin (ET)** and **carbenoxolone (CBX)** to uncouple GAP junctions (Blomstrand *et al.*, 2004), the aim of this study is to test the involvement of SC's GAP junctions in olfactory responses.

Introduction

Results

I - ET activates SCs ex vivo

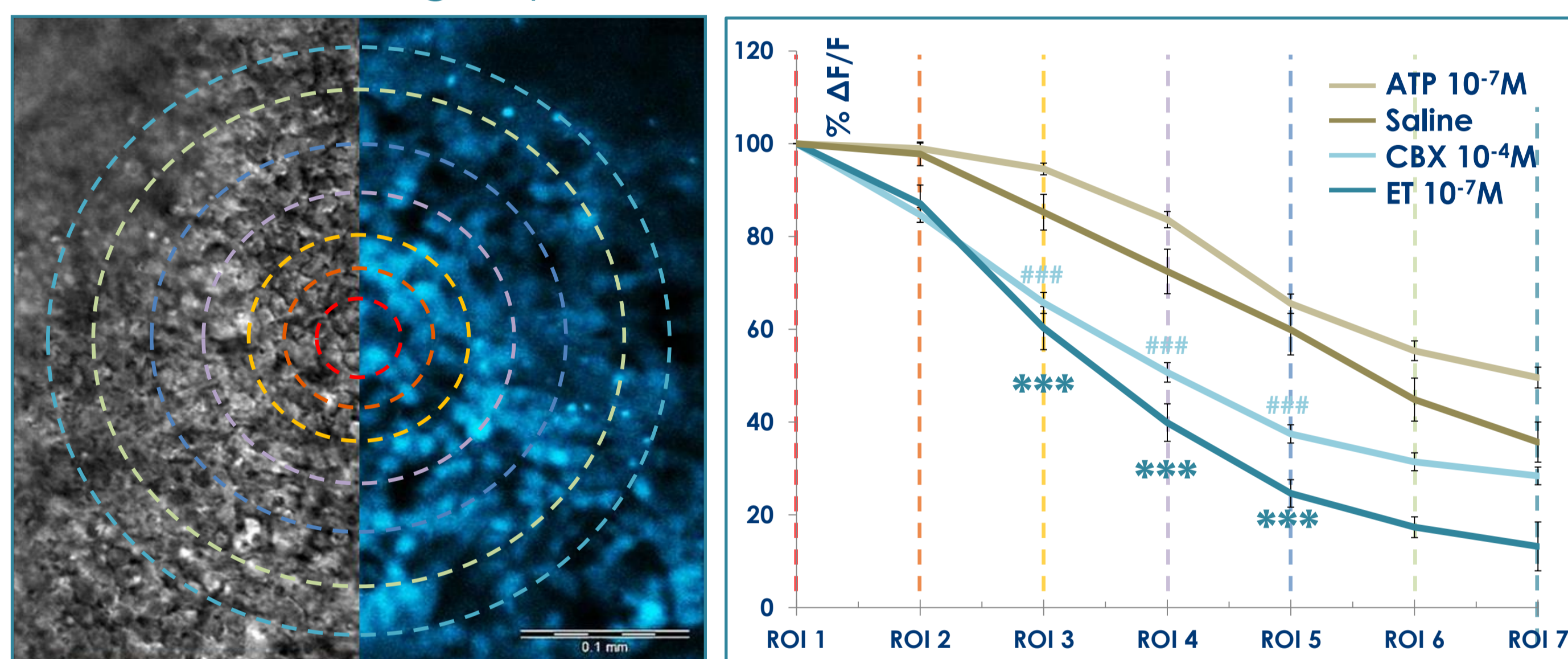
Method: Olfactory Mucosa (OM) slices 200µm (Wistar P0-P4), loaded with Oregon Green (488nm), Calcium Imaging.



I - ET evokes both tonic and phasic calcium responses in the SCs.

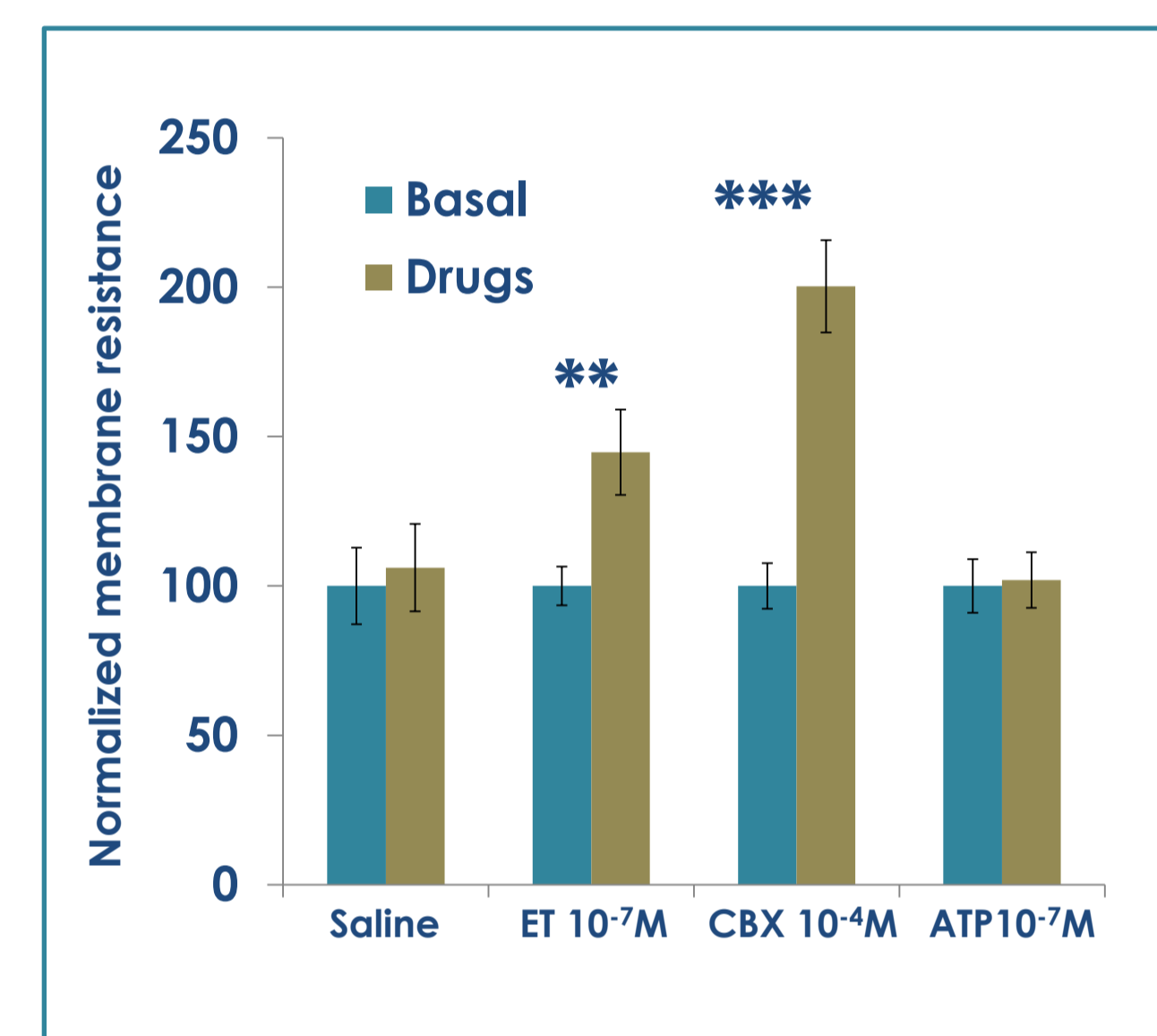
II - Plasticity of GAP junctions ex vivo and in vitro.

Method: Olfactory Mucosa explant (Wistar P7), loaded with Oregon Green, Electric stimulation and Calcium Imaging. n > 20 for each group.



CBX and ET decrease spreading of the calcium wave.

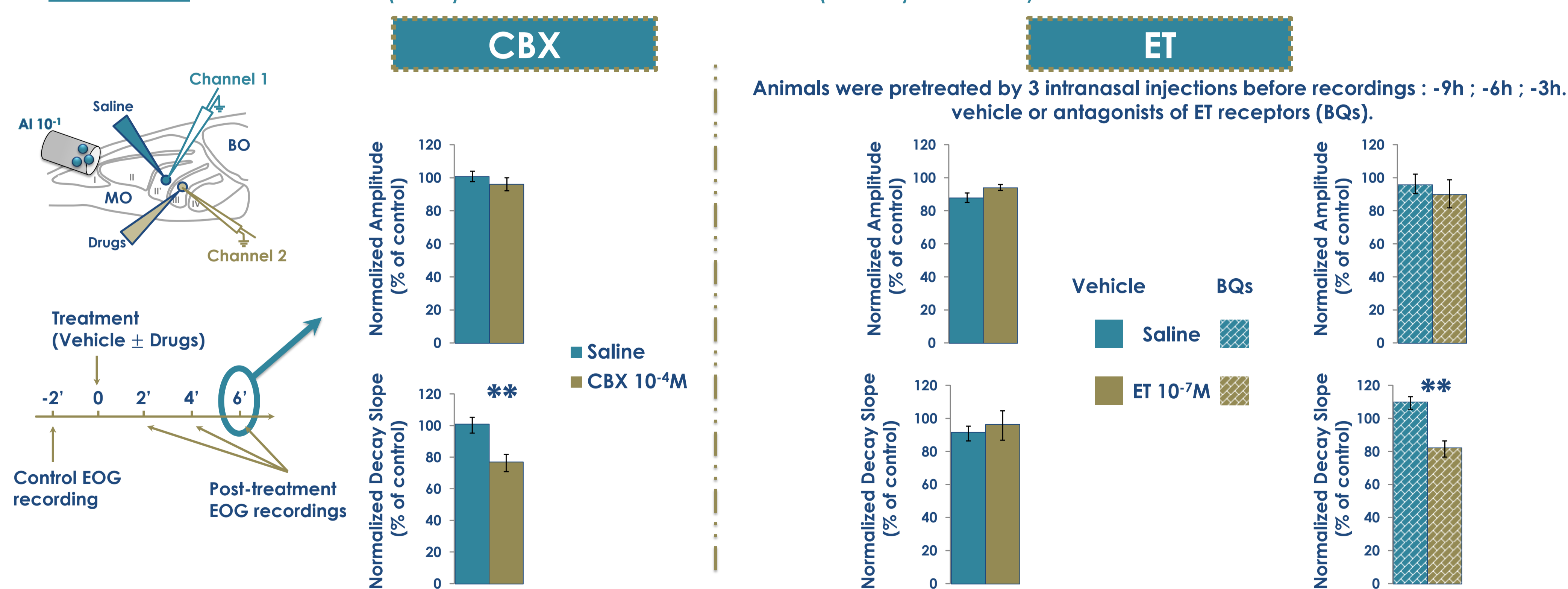
Method: Olfactory Mucosa Primary Culture, Whole cell Voltage Clamp. n > 15 for each group.



CBX and ET increase membrane resistance of SCs.

III - ET and CBX modify olfactory responses.

Method: Hemi-Head (P30), Electro-OlfactoGram (EOG), Isoamyl Acetate 1:10 in mineral oil, n = 6 in each group.



Repolarization kinetics of the olfactory response, but not amplitude, are affected by GAP junctions uncoupling.

III - GAP junctions of SCs are involved in olfactory responses.

Conclusion

SCs respond *in vitro* (Gouadon *et al.*, 2010) and *ex vivo* to ET. This peptide acts as an autocrine and paracrine factor. ET induces **uncoupling of SC's GAP junctions** both *in vitro* and *ex vivo*, like the classically used GAP junction uncoupling agent CBX. **Such modulations affect olfactory responses.** Both ET and CBX treatments increase EOG signal repolarization kinetics. **Our results are consistent with an implication of glial-like cells in the peripheral modulation of the olfactory information.** This has to be confirmed using patch clamp on the knobs of olfactory sensory neurons.

- Blomstrand F *et al.*, Endothelins regulate astrocyte gap junctions in rat hippocampal slices. *Eur J Neurosci.* (2004)
- Gouadon *et al.*, Endothelin evokes distinct calcium transients in neuronal and non-neuronal cells of rat olfactory mucosa primary cultures. *Neuroscience.* (2010)
- Fivos Vogalis *et al.*, Electrical Coupling in Sustentacular Cells of the Mouse Olfactory Epithelium. *J. Neurophysiol.* (2005)

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