**Identification of an orphan GPCR, GPR151, as a potential actor in reproduction and metabolic function**

Jacquinet, Charlotte1 ; Franssen, Delphine1; Dupuis, Nadine2; Lopez Rodriguez, David1; Gerard, Arlette1; Hanson, Julien2; Parent, Anne-Simone1

1GIGA Neurosciences, Neuroendocrinology Unit, University of Liège, Belgium
2GIGA, Laboratory of Molecular Pharmacology, University of Liège, Belgium

Reproductive health has been deteriorating worldwide, as evidenced by the increasing prevalence of puberty and fertility disorders. The mechanisms involved in this phenomenon need to be better understood in order to find better treatment for reproductive disorders. The gonadotropin-releasing hormone (GnRH) neurons in the hypothalamus govern the hypothalamic–pituitary–gonadal axis which is responsible for all aspects of the reproductive function.

We previously showed that early postnatal exposure to bisphenol A (BPA), a ubiquitous endocrine disruptor, disrupts neuroendocrine sexual maturation in female rats. Perinatal exposure to 25 ng/kg/d induced delayed maturation of GnRH secretion, whereas exposure to 5 mg/kg/d induced early maturation of GnRH secretion1. Using RNA sequencing, several hypothalamic genes were identified to be affected by both BPA doses. GPR151 was the most affected gene. GPR151 mRNA levels increased in the hypothalamus after exposure to the low dose of BPA and decreased after exposure to the high dose. GPR151 is an orphan GPCR belonging to the rhodopsin family. This receptor has been identified in the fibres of cholinergic neurons of the habenula projecting to the interpeduncular nucleus in mice. In this area, GPR151 is expressed in the presynaptic membrane and associated with synaptic vesicles2. A phylogenetic study based on the sequence and structure of GPCRs indicates that GPR151 has homology with galanin and kisspeptin receptors3, two factors known to regulate GnRH secretion and play a role in the onset of puberty and reproduction. GPR151 is also expressed in the liver and plays a role in gluconeogenesis4.

We identified GPR151 in the median eminence in the proximity of GnRH fibres, in male and in female rats at PND 80. Using β-galactosidase labeling in GPR151 KO mice in which the *GPR151* gene has been replaced by the Lac Z coding sequence, we observed cell bodies in the preoptic area, where GnRH cell bodies are located. Moreover, GPR151 mRNA expression in female rat hypothalamus increases throughout pubertal development with significant differences between PND 15 (1.00 ± 0.51) versus PND 25 (3.94 ± 0.49; p < 0.01) and PND 80 (5.32 ± 0.80; p < 0.001). To understand the role of GPR151 in GnRH neurons, we overexpressed GPR151 in GnV3 cells, a GnRH immortalized cells line, and observed an increase in GnRH release in the medium compared to cells transfected with the vector alone. Because β-arrestin is responsible for GPR151 internalization after its activation, we used a β-arrestin recruitment assay to analyze the capacity of kisspeptin and galanin to activate the receptor. Neither kisspeptin nor galanin induced the recruitment of β-arrestin but this assay allowed to show that GPR151 appears to have a constitutive activity. Based on GPR151 role in glucose metabolism4, we measured GPR151 mRNA expression in the mediobasal hypothalamus of rat exposed to endocrine disruptor with or without high-fat diet. GPR151 mRNA expression was higher in rat fed with high-fat diet compared to standard diet (p<0,006, ANOVA 2). Our data indicates that GPR151 could be involved in the hypothalamic regulation of GnRH release and/or energy balance.

1. Franssen, D. *et al.* Delayed neuroendocrine sexual maturation in female rats after a very low dose of bisphenol a through altered gabaergic neurotransmission and opposing effects of a high dose. *Endocrinology* **157**, 1740–1750 (2016).

2. Antolin-Fontes, B. *et al.* The habenular G-protein–coupled receptor 151 regulates synaptic plasticity and nicotine intake. *Proc. Natl. Acad. Sci. U. S. A.* **117**, 5502–5509 (2020).

3. Kakarala, K. K. & Jamil, K. Sequence-structure based phylogeny of GPCR Class A Rhodopsin receptors. *Mol. Phylogenet. Evol.* **74**, 66–96 (2014).

4. Bielczyk-Maczynska, E. *et al.* G protein-coupled receptor 151 regulates glucose metabolism and hepatic gluconeogenesis. *bioRxiv* 2022.05.25.493489 (2022) doi:10.1101/2022.05.25.493489.