

CB1-5-HT2A heteromers in schizophrenia patients: human studies in pro-neurons of the olfactory epithelium

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Introduction

Despite multiple clinical and pre-clinical studies investigating schizophrenia, the neurobiological basis of this disease is still unknown. The dysregulation of the serotonergic system, in particular, the 5-HT_{2A} receptor and the endocannabinoid system have been postulated as possible causes of schizophrenia.

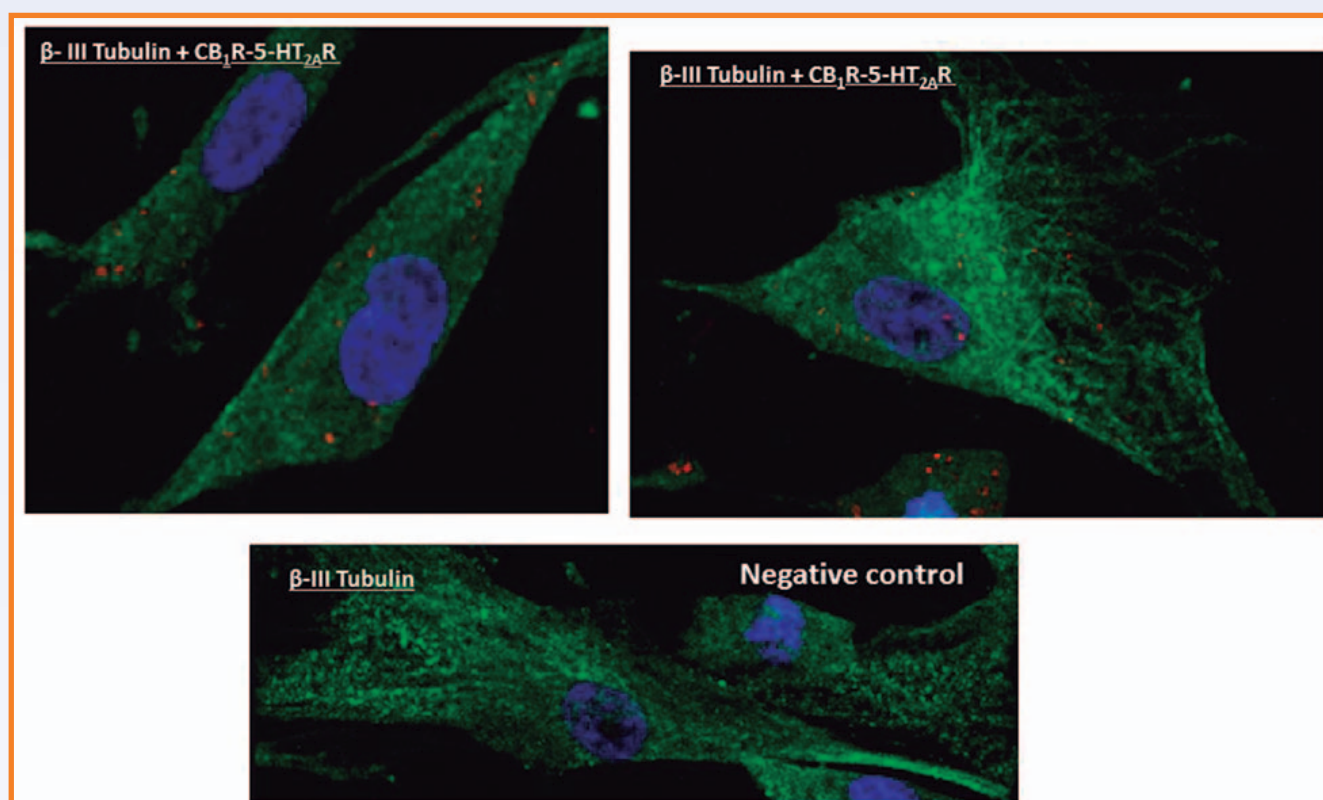
Methods

We recruited a group of 9 healthy volunteers, and 7 patients diagnosed with schizophrenia, which were treated with atypical antipsychotics, were clinically stable and had an illness duration range from 1 up to 15 years. The patients were diagnosed with schizophrenia from the medical record and confirmed by the Structured Clinical Interview for DSM Disorders. The expression of CB1-5-HT_{2A} receptor heteromers in primary cultures of pro-neurons from the olfactory epithelium was quantified using proximity ligation assays and confocal microscopy. The patients and healthy controls were matched by age and gender.

Results

- Olfactory epithelium pro-neurons were viable and expressed the neuronal marker, III- β tubulin. We also established the presence and the functionality of CB1-5-HT_{2A} receptor heteromers in these cells using the proximity ligation and cAMP activity assays, respectively (Figure 1).

Figure 1. Immunofluorescent staining with β -III tubulin and CB1-5-HT_{2A} heteromers by PLA healthy controls

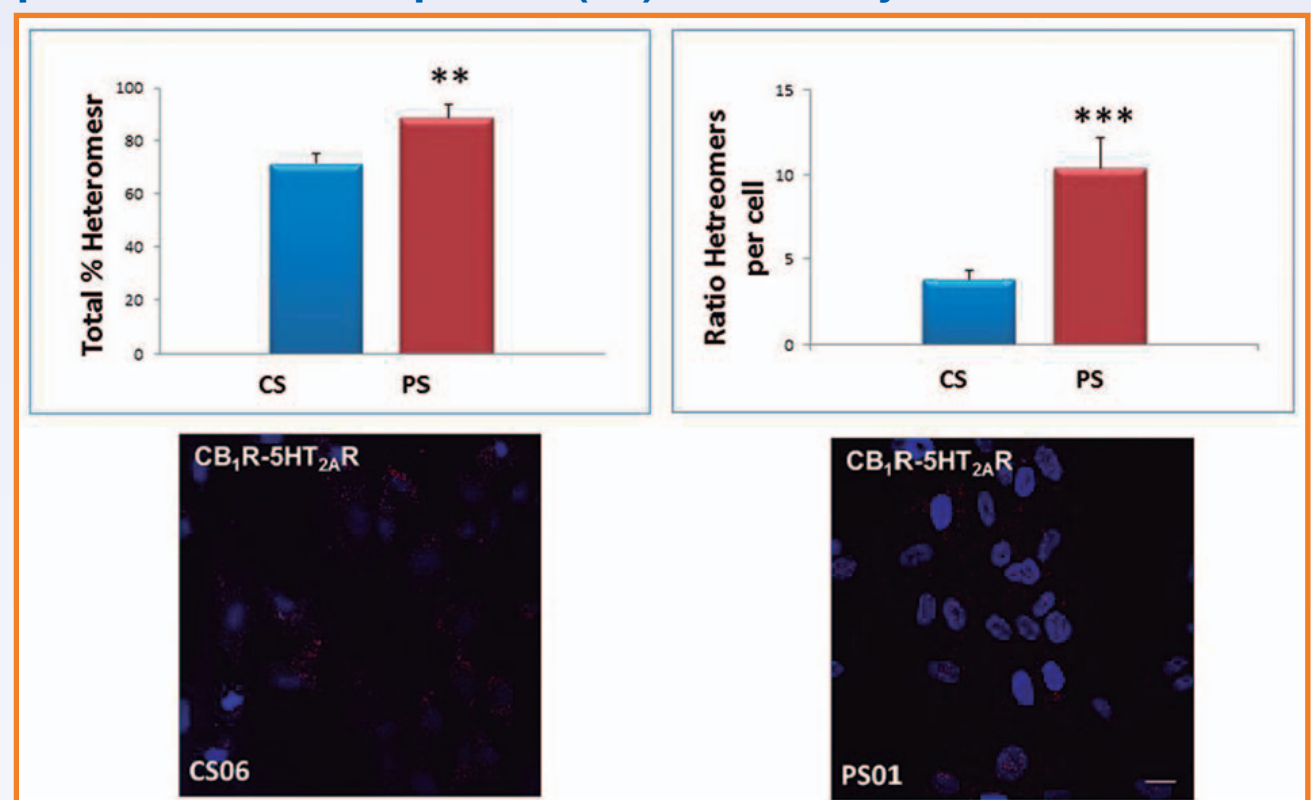


Objectives

The aim of this study is to evaluate the expression of CB1-5-HT_{2A} receptor heteromers in primary cultures of pro-neurons from the olfactory epithelium in schizophrenia patients and control subjects.

- Heteromer expression was significantly increased in schizophrenia patients with respect to controls (Figure 2).

Figure 2. CB1-5-HT_{2A} heteromers in control subjects (CS), and patients with schizophrenia (PS) detected by in situ PLAs



Conclusions

- This highly innovative methodology will allow the non-invasive, low-cost study of new biomarkers for schizophrenia in a model closely related to the central nervous system.

References

- Benítez-King, G., Riquelme, A., Ortiz-López, L., Berlanga, C., Rodríguez-Verdugo, M. S., Romo, F., ... Domínguez-Alonso, A. (2011). A non-invasive method to isolate the neuronal lineage from the nasal epithelium from schizophrenic and bipolar diseases. *Journal of Neuroscience Methods*, 201(1), 35–45. <http://doi.org/10.1016/j.jneumeth.2011.07.009>
- Benítez-King, G., Valdés-Tovar, M., Trueta, C., Galván-Arrieta, T., Argueta, J., Alarcón, S., ... Solís-Chagoyán, H. (2016). The microtubular cytoskeleton of olfactory neurons derived from patients with schizophrenia or with bipolar disorder: Implications for biomarker characterization, neuronal physiology and pharmacological screening. *Molecular and Cellular Neuroscience*, 73(101), 84–95. <http://doi.org/10.1016/j.mcn.2016.01.013>
- Viñals, X., Moreno, E., Lanfumey, L., Cordoní, A., Pastor, A., de La Torre, R., ... Robledo, P. (2015). Cognitive Impairment Induced by Delta9-tetrahydrocannabinol Occurs through Heteromers between Cannabinoid CB1 and Serotonin 5-HT_{2A} Receptors. *PLoS Biology*, 13(7), e1002194. <http://doi.org/10.1371/journal.pbio.1002194>

***Acknowledgements:** This work was supported by grants from DIUE de la Generalitat de Catalunya (2014SGR 680), Instituto de Salud Carlos III, (PI14/00210.) and (PI10/01708) FIS-FEDER Funds. LG is supported by the Instituto de Salud Carlos III through a “Río Hortega” Spanish government research grant (CM14/00111).