

Abstract

Novel Interactions between Mas and Angiotensin Receptors and Their Functionality Modulatory Role for the Brain RAS[†]

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Abstract: The renin–angiotensin system (RAS) not only plays an important role in controlling blood pressure, but also participates in almost every process to maintain homeostasis in mammals. The occurrence of RAS in the basal ganglia suggests that the system may be targeted to improve the therapy of neurodegenerative diseases. We found heteromers formed by Mas and angiotensin receptors, and addressed their functionality in neurons and microglia. Novel interactions formed by MAS/AT₁R and MAS/AT₂R were discovered by using resonance energy transfer techniques. In the heterologous system, we showed that the three receptors—MasR, AT₁R, and AT₂R—can interact to form heterotrimers (Figure S1). The functionality of individual and interacting receptors was assayed by measuring levels of the second messengers cAMP and Ca²⁺ in transfected human embryonic kidney cells (HEK-293T) (Figure S2) and primary cultures of striatal cells. Expression (Figure S3) and functionality (Figure S4) were assayed in parallel in primary cultures of microglia treated or not treated with lipopolysaccharide and interferon- γ (IFN- γ) to simulate neuroinflammation conditions. The proximity ligation assay (PLA) was used to assess heteromer expression in parkinsonian and dyskinetic conditions (Figure S4). In all cases, agonist-induced signaling was reduced upon coactivation, and in some cases just by coexpression. In addition, the blockade of signaling of two receptors in a complex by the action of a given (selective) receptor antagonist (cross-antagonism) was often observed. The negative modulation of calcium mobilization (mediated by AT₁R activation), the multiplicity of possibilities on RAS affecting the MAPK pathway, and the disbalanced expression of heteromers in dyskinesia yield new insights into the operation of the RAS system, how it becomes unbalanced, and how a disbalanced RAS system can be rebalanced. Furthermore, RAS components in activated microglia warrant attention in drug development approaches to address neurodegeneration.

Keywords: Parkinson; GPCR; RAS; Mas; AT₁; AT₂; angiotensin

Supplementary Materials: The following are available online at <https://www.mdpi.com/article/10.3390/IECBS2021-10656/s1>: Figure S1: Interaction between angiotensin (AT₁ and AT₂) and Mas receptors in a heterologous expression system, Figure S2: Functionality of AT₁Mas and AT₂Mas Hets in a heterologous expression system; Figure S3: Expression of AT₁Mas and AT₂Mas Hets in the

striatum of parkinsonian and dyskinetic rats; Figure S4: Functionality of AT1Mas and AT2Mas Hets in activated microglia.

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