Chronic sodium bromide relieves autistic-like deficits in the Oprm1 mouse model of autism and modulates the activity of serotonin and dopamine receptors in vitro

**Introduction:**

Autism spectrum disorders (ASD) are complex neurodevelopmental diseases whose diagnosis lies on the detection of impaired social skills together with restricted and repetitive behavior and interests (DSM-5). Although the etiology of ASD remains mostly unknown, impaired excitation/inhibition ratio appears as a common mechanistic feature. Bromide ion is known to reduce hyperexcitability, possibly by competing with chloride ions at channels and transporters and may thus have therapeutic potential in ASD. We evaluated the therapeutic potential of bromide ion in the Oprm1+/− mouse model of ASD and the molecular mechanisms involved in bromide treatment, notably effects on GPCRs.

**Chronic bromide treatment relieves behavioral deficits in Oprm1+/− mice:**

Behavioral deficits in Oprm1+/− mice: Impact of chronic bromide treatment in behavioral deficits of Oprm1+/− mice, in the two core symptoms of TSA: Social abilities (upper panel) and stereotypes (bottom left panel), and on Anxiety (bottom right panel), a common concomitant symptom. Statistics: All: genotype x treatment effect, except for 3-chamber test: two-way ANOVA followed by Newman–Keuls post hoc test. 3-chamber test: four-way ANOVA with one repeated measure: stimulus—mouse versus toy followed by Newman–Keuls post hoc test. *P <0.05; **P<0.01; ***P<0.001 vs. 0 mM X. Data are presented as mean ± SEM.

**Chronic bromide treatment relieves behavioral deficits in Oprm1+/− mice in a dose-dependent manner**

RT-qPCR in Oprm1 mouse line under NaBr treatment:

Gene expression fold changes (A) and hierarchical clustering (B) in Oprm1 mouse line under chronic treatment of 250 mg/kg of NaBr. Clusters groups (left to right): Glutamate family; GABA family; GPCRs and Excitability family. Statistics: Red: *P<0.05; **P<0.01; ***P<0.001 vs. Oprm1+/− Saline. Blue: #P<0.05; ##P<0.01 Oprm1+/− Br vs. Sal. Figures in brackets indicate the fold changes between Oprm1+/− Br and Sal. Data are presented as mean ± SEM.

**Chronic bromide treatment in Oprm1 knockout mice: transcriptomics**

Bromide impacts the expression of GABA_A subunits involved in E/I balance, Arc involved in neuronal plasticity and Grm2, Htr7 (Nacc) and Grm4 (Nacc) in signalization.

**Bromide and chloride ion effects on 5-HT and dopamine receptor pharmacology**

BRET-based cAMP production assay

(A) Representative Dose-Response Curves (N=3) of 5-HT1R with different Locke’s buffer modified concentrations of CI− and Br−. A clear shift in EC50 is observed between low and high concentration of either CI− or Br−, with a stronger effect of Br−. (B) For each GPCR, agonist used is under brackets and ΔpEC50 is calculated against pEC50 at 0mM X. As D2R is G-coupled (AMP inhibition), experiments are done with 10 μM of Forskolin (activation of adenylate cyclase). Statistics: 1-way ANOVA with Dunnett’s posttest value of *P<0.05; **P<0.01; ***P<0.001 vs. 0 mM X. Data shown represent the means ± SEM of at least 3 experiments.

Bromide and chloride ion behave as Positive Allosteric Modulators on D1R, 5-HT6R and 5-HT7R, but not D2R and 5HT4R

**Conclusion:**

The beneficial effects of bromide administration in a genetic murine model of ASD and its impact on both gene expression and GPCR pharmacology predicts high translational potential in patients with autism, despite high heterogeneity in etiology and symptoms. The next step will be to test the effects of bromide on mGlur4 and mGlur2 in vitro, keeping in mind that CI− is already known as a good Positive Allosteric Modulator (PAM) of mGlur4, but not such a good one for mGlur2.