

## Species-specific differences in cerebellar cannabinoid 1 (CB<sub>1</sub>) receptor function

**Introduction.** We have recently identified species-specific effects of cannabis extracts which suggested differential CB<sub>1</sub> expression and functional receptor activation by  $\Delta^9$ -tetrahydrocannabinol (THC) [1]. Here, we extend these data on CB<sub>1</sub> receptor function in cerebellar membranes from different species.

**Methods.** [<sup>3</sup>H]-SR141671A (a CB<sub>1</sub> receptor antagonist) saturation binding and THC (a CB<sub>1</sub> receptor partial agonist)-stimulated [<sup>35</sup>S]-GTP $\gamma$ S binding assays were performed in cerebellar membrane preparations from mouse, rat, chicken, dog and human tissue. Assays were conducted in triplicate and 5 separate assays performed in each case. Analyses of saturation binding data were conducted by non-linear regression and fitted to a one-binding site model to determine maximal number of binding sites B<sub>max</sub> and the equilibrium dissociation constant K<sub>D</sub>. GTP $\gamma$ S binding data were analysed using a sigmoidal concentration-response model to determine EC<sub>50</sub> and maximum response (E<sub>max</sub>). Statistical significance was determined using an ANOVA followed by a Tukey's post hoc test on raw data.

**Results.** In saturation binding studies, a significant reduction in B<sub>max</sub> was seen in human (P<0.05 vs mouse and rat) and dog (P<0.05 vs mouse) cerebella membranes (Table 1); there were no significant changes in K<sub>D</sub> between species. THC-stimulated GTP $\gamma$ S binding showed significant differences in E<sub>max</sub> elicited by CB<sub>1</sub> receptor activation (Table 1) with a rank order of chicken = rat = dog > mouse = human (P<0.05 for all members of each group) was seen; there were no significant changes in EC<sub>50</sub> between species.

**Table 1. Cerebellar CB<sub>1</sub> receptor binding data for different species**

	Saturation binding		GTP $\gamma$ S binding	
	B <sub>max</sub> (pmol mg <sup>-1</sup> )	K <sub>D</sub> (nM)	EC <sub>50</sub> (nM)	E <sub>max</sub> (%)
Chicken (n= 5)	1.44 ± 0.2	1.57 ± 0.7	107 ± 10	33.9 ± 1.8
Rat (n=5)	1.80 ± 0.4	1.06 ± 0.1	92 ± 30	33.6 ± 2.5
Mouse (n=5)	2.40 ± 0.4	2.30 ± 0.6	138 ± 49	12.0 ± 1.8 <sup>ψ</sup>
Dog (n=5)	0.80 ± 0.2 <sup>*</sup>	0.54 ± 0.2	170 ± 59	27.4 ± 1.8
Human (n=5)	0.46 ± 0.1 <sup>*δ</sup>	2.07 ± 0.3	25 ± 9.9	11.3 ± 2.4 <sup>ψ</sup>

\* p<0.05 vs mouse; <sup>δ</sup> p<0.05 vs rat; <sup>ψ</sup> p<0.05 vs each of chicken, rat and dog

## Conclusions

We identify significant species-selective differences in CB<sub>1</sub> expression and functional receptor activation. Overall, human had a lower CB<sub>1</sub> receptor activity profile which confirm that THC effects in animal tissue models may be poorly predicted of those on human CB<sub>1</sub> receptor-mediated processes.

## References

[1] Whalley BJ, Lin H, Bell L, Hill T, Patel A, Gray RA, Roberts CE, Devinsky O, Bazet M, Williams CM, Stephens, GJ (2018) Species-specific susceptibility to cannabis-induced convulsions. Br J Pharmacol Epub ahead of print Feb 19.