Preclinical Imaging of Adolescent Cannabidiol on Brain Structure and Functional Connectivity

Colon-Perez, L., Pompilus, M. (UF, Psychiatry), Carney, P.R. (UNC Chapel Hill), and Febo, M. (UF, Psychiatry)

Cannabiol as a potential treatment for adolescent behavioral disorders

Several lines of evidence support the use of cannabidiol (CBD) as a broad acting medication to alleviate autism spectrum disorder (ASD) comorbidities. CBD has been reported to alleviate psychosis and anxiety, facilitate REM sleep, and suppress seizure activity, which are all outcomes that may benefit children with an ASD. A major goal of this R03 grant is to characterize the *in vivo* neural mechanisms of action of cannabidiol (CBD) in unaffected and in ASD brain. Our main hypothesis is that administering CBD during adolescence normalizes structural and functional deficits in an animal model of ASD. To this end, we devised two specific aims, one of which is close to completion (Aim 1) and the other is underway. This proposal uses the rat valproic acid (VPA) model of ASD to explore the main hypothesis. Using the VPA rat model, we determined the effect of chronic CBD treatment during adolescence on functional connectivity, as measured by resting state fMRI (Aim 1). In Aim 2, which is now in progress, we determine the effects of CBD on structural white matter connectivity and tissue microstructural integrity measured by diffusion MRI (dMRI) (Aim 2).

Methods

Pregnant rats arrived at gestational day 10 (GD10) and were randomly assigned to saline controls or VPA treated dams. On GD 12.5, rats were administered either saline or VPA (600mg/kg at 2.5ml/kg). Dams were then left undisturbed, singly housed with food and water ad lib and nesting material. On postnatal day 1 (PD1), pups were assessed for body weights, sex ratios, anogenital distances and for VPA related developmental defects. Offspring were then randomly assigned postnatally to either vehicle treated or CBD treated groups (CBD at 100 mg/kg in 1:1:18 ethanol:cremaphore:saline vehicle; volume at 1ml/kg subcutaneous, SC). CBD or vehicle was administered from PD32 to PD47. In an initial pilot experiment we encountered difficulties scanning adolescent mice (PD32) using our fMRI anesthesia protocol. Thus, we conducted the study using a cross sectional design (Imaging on PD48 and behavioral testing PD49-52). At PD48 there were no issues preventing us from collecting data from physiologically stable animals. Rats were imaged at 4.7 Tesla under 0.2 mg/kg/h dexmedetomidine, SC and 0.5% isoflurane in 70%N_2/30%O_2. Data were processed using pipelines established and published by our group. We carried out network analyses on the multiple groups (n=10/group with 50% females): Saline-Vehicle, Saline-CBD, VPA-Vehicle, VPA-CBD. Images were processed as indicated by our recent work(1-3).

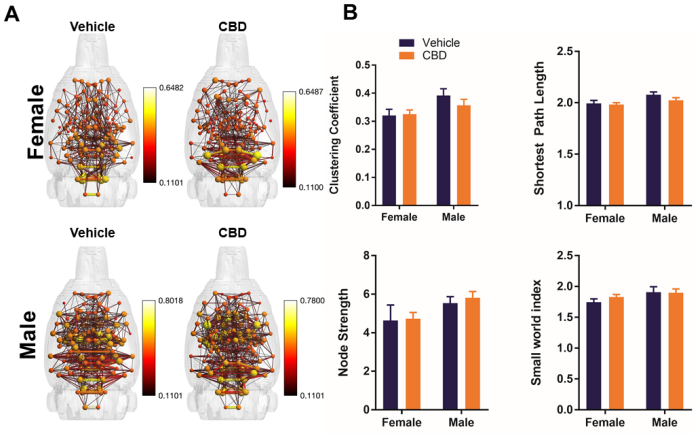

Fig. 1 CBD administration (100 mg/kg, SC) over the course of early adolescence had no effect on functional connectivity in male and female rats. A) 3D rat brain connectomic maps comparing node strength and z score values of male and female rats. B) Results for functional connectomic metrics showing no effect.

Results and Discussion

As indicated in Figure 1 (above), we failed to observe any statistically significant effects of CBD treatment during adolescence (PD32-47) on functional connectivity. We used a robust analysis approach involving network (connectomic) metrics from the field of graph theory. This allows use to focus the analysis on correlation values above a significance threshold (in this case z>0.3) and it also allows us to determine not only changes in connectivity strength but also the arrangement of neuronal interactions in the brain (network organization). In spite of this robust approach there was no effect in male or female rats, regardless of whether they were prenatally exposed to VPA or saline (VPA data not shown). In support of these negative findings, we also failed to observe behavioral changes with CBD treatment.

Conclusions

At this time, we are processing diffusion MRI data sets, which we collected using a multi shell sequence in order to determine the effect of CBD or VPA (or sex) on neurite orientation dispersion and density, as well as free water. These studies are currently under way.

Acknowledgements

The National High Magnetic Field Laboratory is supported by the National Science Foundation through NSF/DMR-1157490/1644779 and the State of Florida. Supported also by NIH/NIDA grant R03DA042971 to Marcelo Febo.

References

[1] Thompson, M.F., *et al*., J Psychopharmacol, **32**, 332-43 (2018)
[2] Nephew, B. C., *et al*., J Affect Disord, **229**, 213-23-16 (2018)
[3] Colon-Perez, L. M., *et al*., Neuropharmacol, 137, 178-93 (2018)