IN VITRO AND IN VIVO PROFILE OF CYB003: A NOVEL, DEUTERATED PSILOCYBIN ANALOG FOR THE POTENTIAL TREATMENT OF MAJOR DEPRESSIVE DISORDER

Michael Palfreyman, Joan Krakowsky, Michael Morgan, Clinton Canal, Pradip Pathare, Ken Avery, Amy Reichelt, Tina Mueller, Joshua Hartsel, Alex Nivorozhkin, Amir Inamdar, Geoffrey Varty

Cybin Inc., 5600-100 King St West, Toronto, ON M5X 1C9, Canada

NYSE:CYBN
NEO:CYBN

Abstract

Background: Phase 2 clinical studies have demonstrated that short-term administration of psilocybin has beneficial effects on Major Depressive Disorder (MDD) and Treatment-Resistant Depression (TRD). While psilocybin is efficacious and inherently safe, there is significant variability between patients in their psychedelic and side-effect experiences. This variability is likely due to psilocybin acting as a pro-drug that requires de-phosphorylation to the psychoactive metabolite, psilocin. CYB003 is a novel deuterated analog of psilocybin that may offer benefits over psilocybin. The aim of these studies was to compare the in vitro pharmacology and in vivo activity of CYB003 to psilocin and conduct Investigational New Drug (IND)-enabling studies to allow the initiation of clinical studies.

Methods: Pharmacological profiles of CYB003 and psilocin were compared using serotonin (5-HT) receptor binding and functional assays to evaluate potency, efficacy, and selectivity at serotonin receptors; both compounds were also screened for activity at a panel of over 100 proteins. Additionally, both compounds were evaluated in the mouse head twitch response (HTR) assay (a behavioral model of psychedelic-like effects). ADME and pharmacokinetic (PK) studies were also completed followed by dose-range finding studies in rodent and non-rodent species. Finally, IND-enabling studies conducted to Good Laboratory Practice (GLP) standards were completed; specifically, repeat dose toxicology studies, and cardiovascular, respiratory and CNS safety pharmacology assessments.

Results: The pharmacological and selectivity profile of CYB003 was similar to that seen with psilocin. CYB003 binds and activates the 5-HT_{2A} receptor (5-HT_{2A} Ki: CYB003 37 nM; psilocin 31 nM) and induces the HTR in mice [No. of HTR (0-30 min): CYB003 8.9 ± 1.3; psilocin 12.6 ± 1]. ADME studies indicated little metabolism in vitro and PK/PD studies showed a good correlation between plasma levels and in vivo activity. Safety pharmacology study findings were consistent with activation of 5-HT receptors, reflecting findings seen with other psychedelic compounds, including psilocybin. GLP-toxicological studies established safe and potentially effective doses, and will support multiple dose clinical studies. Collectively, these data permitted the initiation of Phase 1/2a clinical studies ***www.clinicaltrials.gov ID: NCT05385783: A study of a Psilocybin Analog (CYB003) in Participants with Major Depressive Disorder***. The Phase 1/2a protocol in participants with MDD is now on-going.

Conclusion: Our findings confirm that CYB003 has the appropriate pharmacology and safety profile to proceed to clinical trials in patients with MDD. These data further support the potential of CYB003 to have superior properties to psilocybin and is likely to offer considerable clinical benefits.

Overview

Cybin's CYB003 program aims to develop a novel Second-Generation psychedelic compound based on psilocybin that will provide therapeutic mental health benefits with improved bioavailability and reduced duration of action.

By leveraging Cybin's cutting-edge medicinal chemistry expertise, we have successfully generated CYB003, a proprietary deuterated psilocybin analog.

CYB003 may harness the therapeutic benefits of psilocybin while potentially reducing the duration of the clinical visit and diminishing side-effects associated with psilocybin.

CYB003 is designed to achieve less variability in plasma levels, faster onset of action, shorter duration of effect and potentially better tolerability versus oral psilocybin, for an overall better outcome for patients.

Here, we present the in vitro and in vivo profile of CYB003, and findings from safety pharmacology/toxicology studies, that along with pharmacokinetic data (see accompanying poster), were used to predict safe and efficacious human doses for an on-going Phase 1/2a trial in Major Depressive Disorder (MDD).

Introduction

Psychedelic drugs, such as psilocybin, have clinical potential for the treatment of psychiatric conditions including anxiety (Nutt 2019), depression (Carhart-Harris 2021, Goodwin 2022), OCD (Moreno 2006), cancer-related depression and anxiety (Griffiths 2016; Ross 2016), and substance use disorders (Nielson 2018, Noorani 2018).

Notably, psilocybin was recently granted '*breakthrough therapy*' designation by the US Food and Drug Administration (FDA) for MDD (Staines 2019), and treatment-resistant depression (TRD, Compass Pathways 2018) – recognizing the potential for substantial outcome improvements over current treatments (FDA 2018).

Psilocybin is a prodrug, which is dephosphorylated to produce its active form, psilocin, that acts as an agonist at a variety of serotonin (5-hydroxytryptamine [5-HT]) receptors, including the 5-HT_{2A} receptor (Nichols, 2004).

Following dephosphorylation, the psychoactive effects of psilocybin occur within 1 hr after dosing, peak around 2-3 hr, and diminish by 6 hr (Griffiths 2018). Based on this time course, in the clinical trial setting, protocols mandate observation until approximately 8 hr after dosing.

While psilocybin administration in a therapeutic setting represents a potential revolution in mental health therapy, the delayed onset of effects, patient-to-patient variability, and potential adverse reactions such as nausea, may present some development challenges.

At Cybin, we have focused on developing a deuterated psilocybin analog, CYB003, with faster onset, reduced variability and a shorter duration of action in order to retain the therapeutic effects of psilocybin but improve patient and clinical experience.

Aims and Hypothesis

- Compare the in vitro pharmacology and in vivo (behavior and pharmacokinetics) effects of CYB003 to both psilocybin and psilocin.
- Determine if CYB003 offers any potential improvements over psilocybin that could translate to better outcomes in the patient.
- Determine if CYB003 is safe and well-tolerated in both a rodent and a non-rodent species.

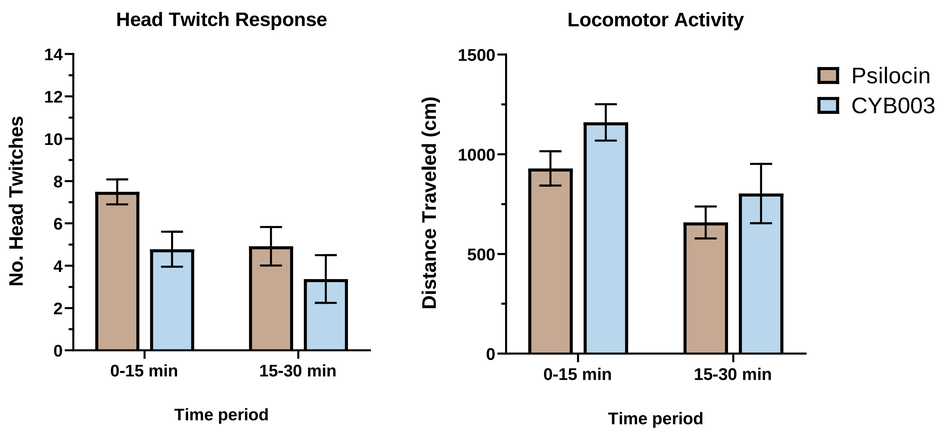
Results

Serotonin Receptor & Off-Target Profile

Serotonergic Receptors	% Inhibition @ 10 µM (Ki, nM)	
	CYB003	Psilocin
Human 5-HT_{1A}	96	96
Rat 5-HT_{1B}	80	82
Human 5-HT_{2A}	100 (37)	99 (31)
Human 5-HT_{2B}	100 (26)	100 (21)
Human 5-HT_{2C}	98 (92)	98 (95)
Human 5-HT_{5A}	99	99
Human 5-HT_{6}	98	98
Human 5-HT_{7}	98	102

Secondary Targets	% Inhibition @ 10µM	
	CYB003	Psilocin
Human Adrenergic α_{2A}	70	63
Rat L-Type Ca^{2+} channel	54	56
Human Histamine H_1	75	78
Human DA transporter	58	32
Human NE transporter	59	41
Human $Na_V1.5$ channel	6.6	6.6
Human $Ca_V1.2$ channel	No Inhibition	No Inhibition
hERG channel	25.6	43.5

Mouse Head Twitch Response (HTR) & Locomotor Activity (LMA)



- CYB003 produced a HTR in mice - indicative of in vivo 5-HT2A receptor engagement - that was not significantly different from psilocin.
- Similarly, CYB003's effects on LMA where indistinguishable from psilocin.

Conclusions

- CYB003 exhibits a psilocin-like serotonin receptor binding profile, as well as similar off-target activity.
- CYB003 induces head twitch responses and hyperactivity in mice and the effects are similar to psilocin.
- Safety pharmacology and toxicology studies demonstrated that CYB003 is well-tolerated in the rat (50-200 mg/kg, PO) and dog (0.5 mg/kg, PO).
- In conclusion, CYB003 mirrors the in vitro binding profile and in vivo activity of psilocin and has been preclinically demonstrated to be a safe and well-tolerated orally active molecule.

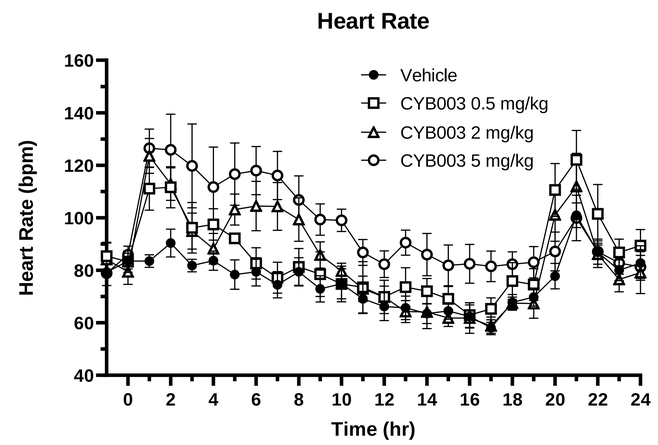
Further Investigations

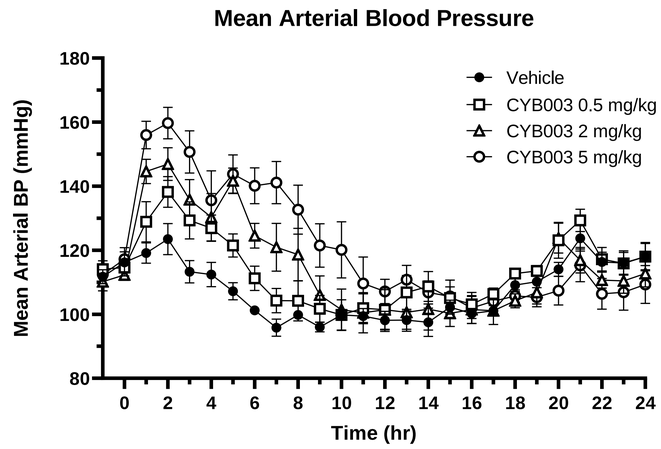
- CYB003 has progressed into US-based clinical studies for the treatment of MDD. **ClinicalTrials.gov Identifier: NCT05385783**

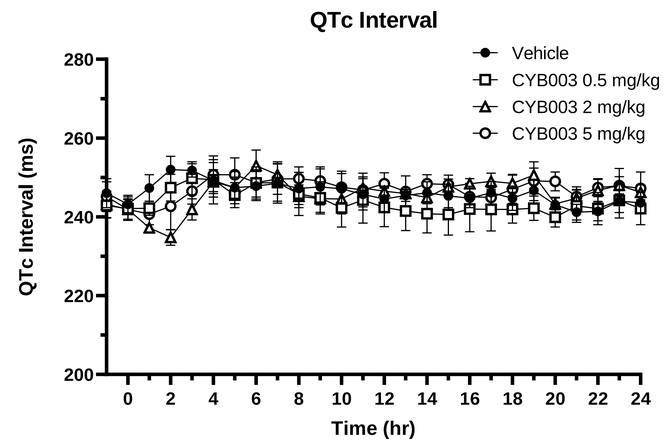
Cybin Inc @CybinInc cybininc

Results

Cardiovascular Effects in the Telemeterized Dog







- CYB003 produced a dose-dependent increase in both blood pressure and heart rate. The duration of these effects also appeared to be dose-related.
- No effect on QTc Interval.

References

Carhart-Harris, R. et al. (2021) N Engl J Med, 384, 1402–1411.

COMPASS Pathways. (2018). COMPASS Pathways receives FDA Breakthrough Therapy designation for psilocybin therapy for treatment resistant depression: Compass Pathways.

FDA. (2018). Breakthrough Therapy FDA. https://www.fda.gov/patients/fast-track-breakthrough-therapy-accelerated-approval-priority-review/breakthrough-therapy.

Goodwin, G. M. et al. (2022) N Engl J Med, 387, 1637-1648.

Griffiths, R. R. et al. (2016) J Psychopharmacol, 30, 1181–1197.

Griffiths, R. R. et al. (2018) J Psychopharmacol, 32(1), 49-69.

Nichols D.E. (2004) Pharmacol Ther. 101(2):131-181.

Nielson, E. M. et al. (2018) Front Pharmacol, 9(132), 1–13.

Noorani, T. et al. (2018) J Psychopharmacol, 32(7), 756–769.

Nutt, D. (2019) Dialogues Clin Neurosci, 21(2), 139 147.

Ross, S. et al. (2016) J Psychopharmacol, 30(12), 1165–1180.

Staines, R. (2019) In: Pharmaphorum. https://pharmaphorum.com/news/fda-tags-psilocybin-drug-as-clinical-depression-breakthrough-therapy/.