Pharmacokinetic Evaluation of Subcutaneously Administered ZYN001 in Male Sprague-Dawley Rats

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Poster # 8

Background

- The antinociceptive and antihyperalgesic properties of delta-9-tetrahydrocannabinol (THC), a cannabinoid and the primary psychoactive component in Cannabis sativa,[1] have been extensively reported[2,3]
- These findings have prompted the development of orally administered THC-containing medications, which have been associated with psychotropic effects, such as altered perception, change in mood, and euphoria[2,4]
- In some patients, these side effects may limit the therapeutic use of THC[4]
- ZYN001 is:
  - A synthetic pro-drug of THC
  - Formulated for delivery via a transdermal patch
  - Not derived or extracted from botanicals
- The pro-drug technology facilitates the transport of THC, which is naturally hydrophobic, across the stratum corneum and into the systemic circulation
- Chemically, ZYN001 is the D-(-)-glyceric acid ester of THC
- Unlike THC, ZYN001 can be absorbed into the skin transdermally
- After crossing the stratum corneum, ZYN001 is hydrolyzed back to THC and glyceric acid by esterases in the skin (Figure 1)
- The transdermal patch is a non-invasive, non-oral dosage form that may be able to achieve sustained, consistent plasma levels of THC while avoiding the common psychoactive adverse events associated with high plasma levels of THC

Objective

The objective of this study was to evaluate the in vivo pharmacokinetics of ZYN001 — specifically to confirm in vivo that ZYN001 is hydrolyzed to THC.

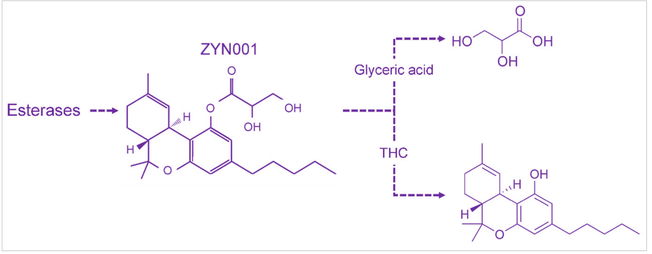
Methods

- A total of 3 male experimentally naïve Sprague-Dawley rats were included in this study
- During the acclimation period, animals were observed daily with respect to general health and any signs of disease

Methods *cont.*

- They were housed individually in cages and maintained under controlled conditions before testing and during the testing period
- Rats were given a single subcutaneous bolus injection of ZYN001 at a dose of 1 mg/kg
- Dose volumes were individualized by body weight
- Plasma samples were analyzed by liquid chromatography-tandem mass spectrometry

Figure 1. Hydrolysis of ZYN001 into glyceric acid and THC



Methods *cont.*

- Plasma concentrations of ZYN001; THC; its main active metabolite, 11-hydroxy-delta-9-tetrahydrocannabinol (THC-OH)[5]; and its main inactive metabolite, 11-nor-delta-9-tetrahydrocannabinol-9-carboxylic acid (THC-COOH),[5] were measured
- Blood samples were obtained at baseline and at 0.08, 0.25, 1, 3, 6.67, 24.11, 30.1, 47.75, 54, and 74.75 hours postdose
- Immediately after the samples were harvested, plasma was separated, and 50 µL of plasma was extracted with solvent to precipitate the proteins
- Samples were centrifuged (10,000 g x 3 minutes), supernatant was removed, and the samples were evaporated to dryness under nitrogen gas
- Samples were reconstituted with acetonitrile and analyzed

Results

- ZYN001 rapidly converted to THC within 0.08 hours (≤4.8 minutes)
- THC plasma concentration ranged from 159.6 at 0.08 hours postdose to 93.3 ng/mL at 74.75 hours postdose
- Plasma concentrations of ZYN001 ranged from 26.4 ng/mL at 0.08 hours postdose to 17.4 ng/mL at 74.75 hours postdose
- Concentration of metabolites were consistent and low (<5 ng/ml) at each timepoint postdose
  - Concentrations of THC-OH:
    - Were 1.4 ng/mL at 0.08 hours postdose
    - Decreased in roughly linear fashion to 0.1 ng/mL at 74.75 hours postdose
  - For THC-COOH, concentrations were 4.1 ng/mL at 0.08 hours postdose and 1.9 ng/mL at 74.75 hours postdose
- The plasma concentrations versus time for ZYN001, THC, THC-OH, and THC-COOH are summarized in Table 1 and illustrated in Figure 2

Figure 2. Plasma concentration vs time in rats after 1 mg/kg subcutaneous administration of ZYN001

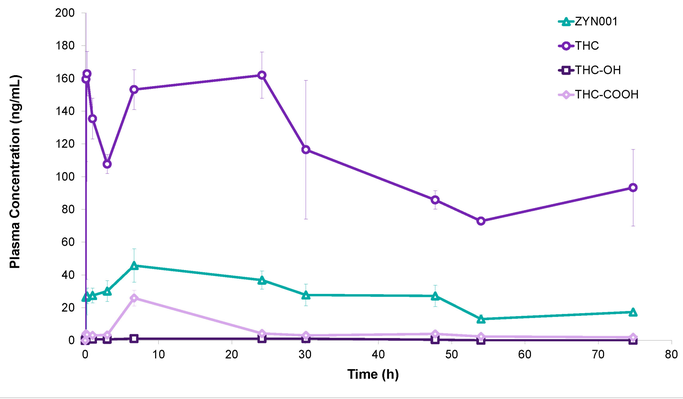


Table 1. Plasma concentrations of ZYN001 in Sprague-Dawley rats (n = 3)

Hours postdose	0	0.08	0.25	1	3	6.67	24.11	30.1	47.75	54	74.75
ZYN001	0	26.4	27.2	27.4	30.1	45.8	36.9	27.8	27.2	13.0	17.4
THC	0	159.6	162.9	135.4	107.7	153.2	162.0	116.5	85.8	72.9	93.3
THC-OH	0	1.4	1.0	0.8	0.7	1.1	1.1	1.1	0.5	0.1	0.1
THC-COOH	0	4.1	3.6	3.1	3.4	25.9	4.3	3.1	4.0	2.4	1.9

Conclusions

- **Rat subcutaneous dosing proved to be an excellent tool for observing chemical characteristics of a THC prodrug delivered just beneath the skin's surface**
- **ZYN001 was rapidly hydrolyzed to THC within 5 minutes of subcutaneous dosing**
- **Low levels of THC's main metabolites, THC-OH and THC-COOH, were observed over the course of the study period**
- **Since THC-OH is a potent psychoactive metabolite that crosses the blood-brain barrier more easily than THC, low levels in plasma may reduce the likelihood that patients will experience treatment-emergent psychotropic effects**
- **Based on the results of this study, ZYN001, given in the form of a transdermal delivery system, should rapidly hydrolyze to THC, bypass first pass metabolism to THC's main metabolites and thereby reduce treatment-emergent psychotropic effects in patients**

References

1. Gaoni YM, Mechoulam R. *J Am Chem Soc*. 1964;86:1646–1647.; 2. Fine PG, Rosenfeld MJ. *Rambam Maimonides Med J*. 2013;4(e0022):1–15.; 3. Aggarwal SK. *Clin J Pain*. 2013;29:162–171.; 4. Skrabek RQ et al. *J Pain*. 2008;9:164–173.; 5. Burstein SH. *Bioorg Med Chem*. 2014;22:2830–2843.

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