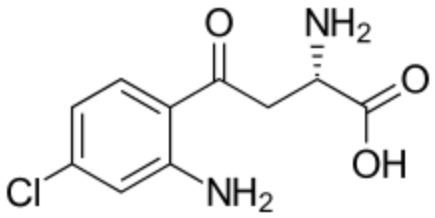


L4-Chloro-kynurenine

PETER LASCARIDES, DO

Treasurer, Eastern Pain Association
Assistant Clinical Professor of PM&R
NYIT College of Osteopathic Medicine, Glen Head, NY
Pain Medicine Physician
Pain Institute of Long Island, Riverhead, NY

Disclosures

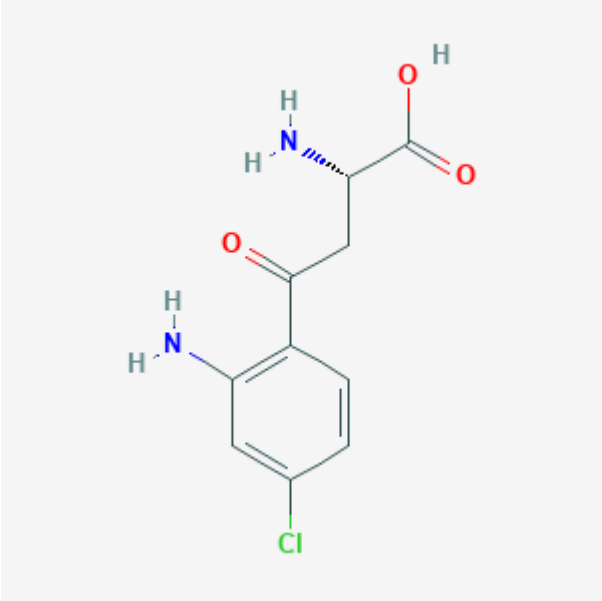
- None

L4-Chlorokynurenine - Outline

- Background
- Pharmacology
- Clinical Studies
- Outcomes
- Future Use

L4-Chlorokynurenine

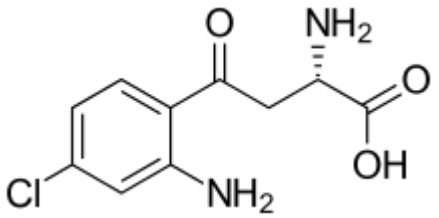
- L4-Chlorokynurenine (L4-Cl-KYN) is a drug candidate under studies for potential treatment of major depressive disorder, and pain.



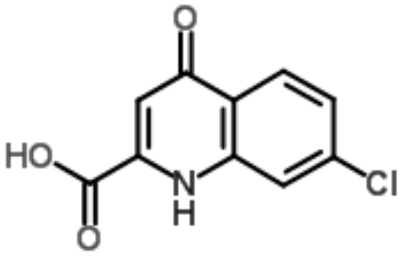
L4-Chlorokynurenine

- It is an orally active small molecule prodrug of 7-chlorokynurenic acid (7-Cl-KYNA)

- 7-Cl-KYNA is a potent and highly selective noncompetitive antagonist of the Glycine-B site of the NMDA receptor



L4-Cl-KYN



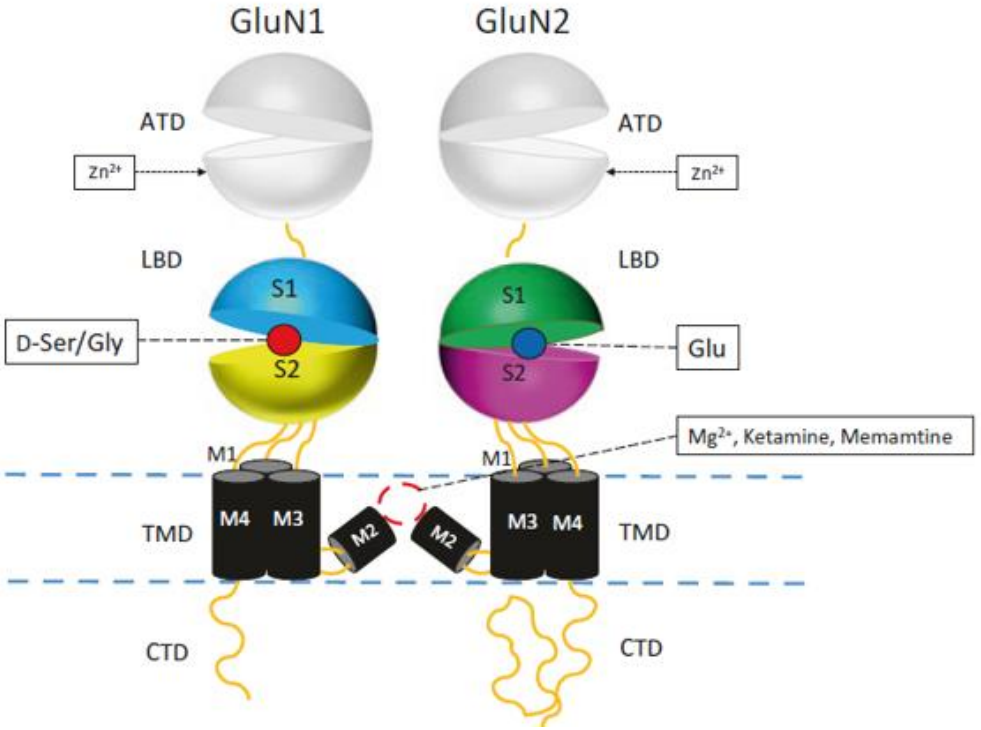
7-Cl-KYNA

NMDA Receptors

- Play a role in the development of neuropathic pain, which is associated with a variety of causes including:
  - Infectious Diseases
    Zoster, HIV, Leprosy
  - Central/ Peripheral injuries to the Nervous System
    - Nerve/ Spinal Cord Damage, Phantom limb pain, MS
  - Systemic and Autoimmune diseases
    - Sarcoidosis, DM
  - Toxic Agents
    EtoH, Chemotherapeutics
  - Inherited conditions
    - CMT, Fabry's Disease
  - Complex Regional Pain Syndrome

NMDA Receptors

- Activation requires binding of agonist (glutamate) and coagonist (glycine or D-serine), and release from voltage dependent Mg2+ that blocks receptor.



Mori,Hisashi. Chapteer 1: Overview of the NMDA Receptor. In: Hashimoto, Kenji et. Al. The NMDA Receptors, Springer Publishing, Switzerland 2017: 7

NMDA Receptors

- Prolonged nociceptive stimulation activates and upregulates NMDA receptors, causing amplified conduction of pain signals to central sites (Central Sensitization).
- Clinically, this may present as spontaneous pain, hyperalgesia and allodynia. [1]

NMDA Receptor Antagonists

- In theory, blocking NMDA receptors should disrupt the pain process, reducing symptoms
- The positive effects of targeting this receptor however, seem to come with a price

NMDA Receptor Antagonists

- The most potent and best studied drug is Ketamine, however its clinical use is limited due to anesthetic, cognitive, and psychotomimetic side effects[2]

NMDA Receptor Antagonists

- Other NMDAR inhibitors include:
  - Magnesium
  - Memantine
  - Amantadine
  - Riluzole
  - Dextrometromorphan
- Unfortunately have weaker affinities for the receptor minimizing both their efficacy.[3]

NMDA Receptor

- When compared with classic NMDAR antagonists, Glycine- B Receptor antagonists seem to have a better safety profile, and do not cause the adverse side effects that are associated with “classic” NMDAR antagonists
- Why aren’t they in use?

L4-Chlorokynurenine

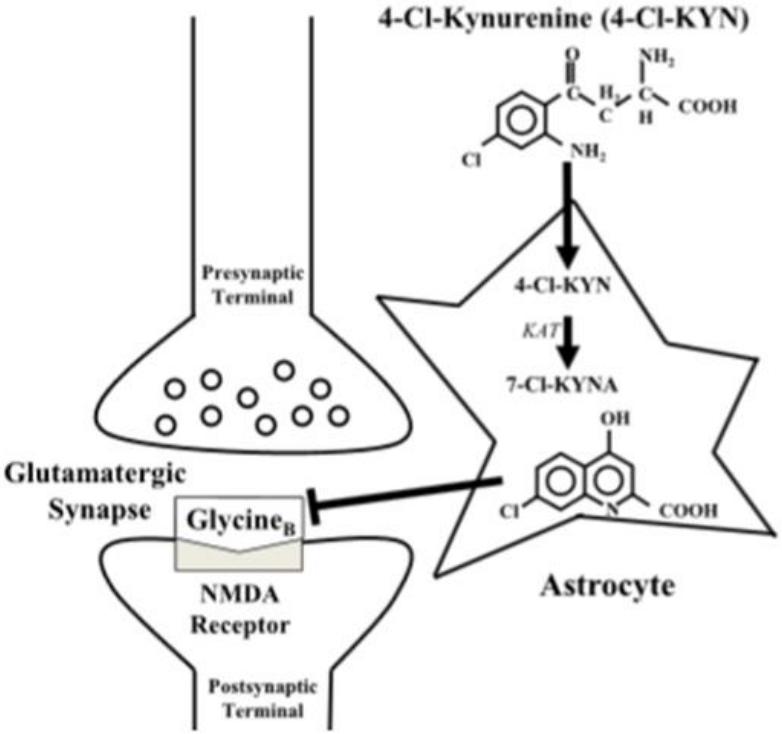
- 7-Cl-KYNA has been widely used as a pharmacological probe to study the biology of the NMDA receptor, documented in studies as early at 1988 [4]

L4-Chlorokynurenine

- Unfortunately the therapeutic use of 7-Cl-KYNA is limited by its poor penetration of the blood-brain barrier[5]
- As a result L4-Cl-KYN (AV-101), has been developed for use in humans, it is currently being studied in a Phase II clinical trials

L4-Chlorokynurenine

- Penetrates the blood–brain barrier via the large neutral amino acid transporter
- Accumulated in astrocytes where it is then converted to 7-Cl-KYNA by kynurenine aminotransferase (KAT) [6,7]



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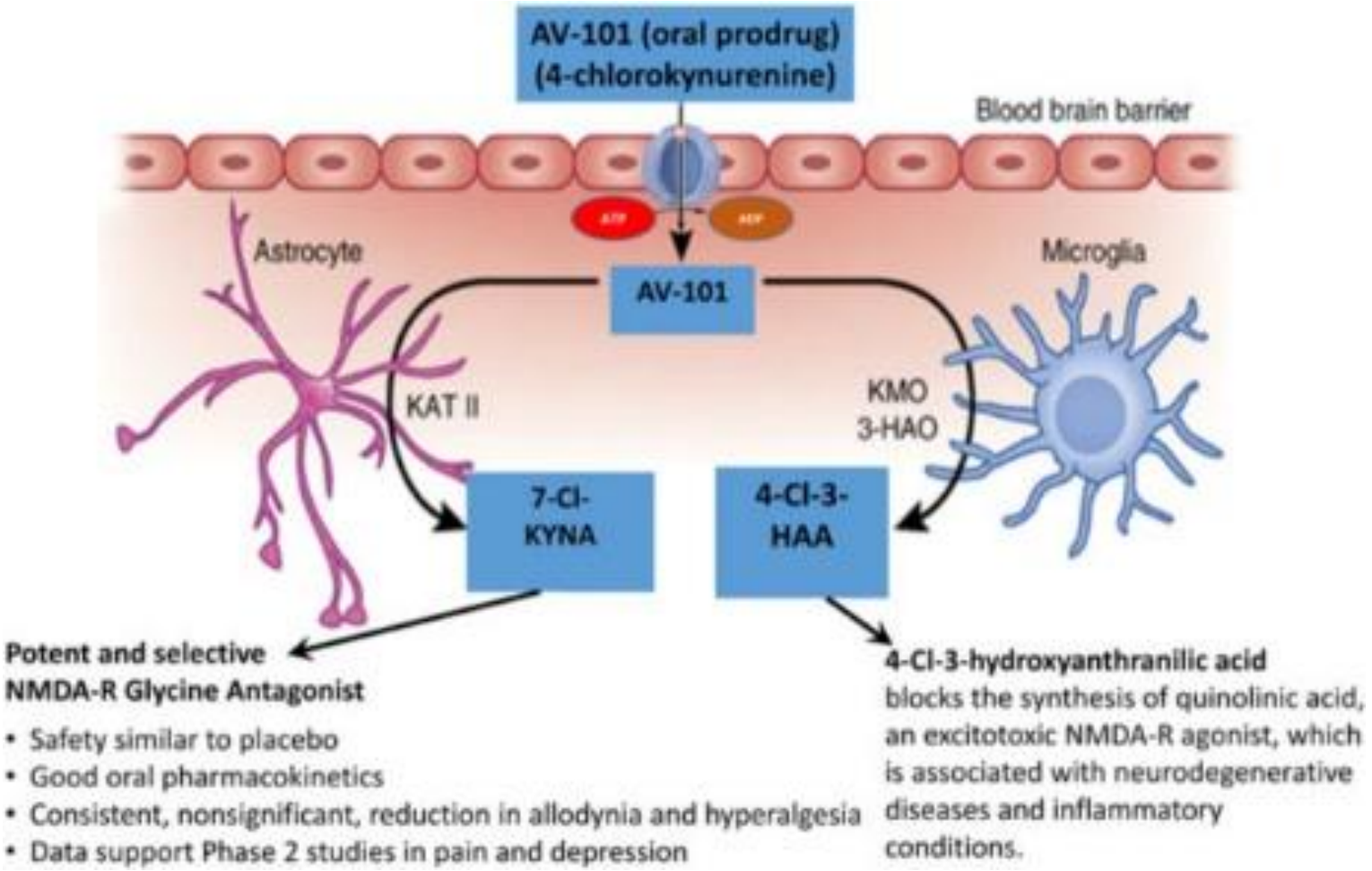


Wallace, Mar et. al. Randomized, double-blind, placebo-controlled, dose-escalation study: Investigation of the safety, pharmacokinetics, and antihyperalgesic activity of l-4-chlorokynurenine in healthy volunteers. Scand J Pain. 2017 Oct;17:243-251

Clinical Trials

- In 2017 following a study of L4 Chlorokynurenine on rodents, a Randomized, double-blind, placebo-controlled, dose-escalation study investigating the safety, pharmacokinetics, and antihyperalgesic activity of L-4-chlorokynurenine took place, lead by Mark Wallace, et. Al.

Objectives

- The primary objective of this study was to evaluate the safety and PK
  - Phase 1A single dose escalation study,
  - Phase 1B study of three doses of orally-administered AV-101 given once daily for 14 days in healthy volunteers.

Objectives

- The secondary objective was to examine the antihyperalgesic effects of 14-daily doses of orally-administered AV-101 versus placebo in healthy volunteers using the intradermal capsaicin model.

Methods

- The Phase 1A study was a single-site, randomized, double-blind, placebo-controlled**, single oral ascending dose** (30–1800 mg) study involving 36 normal healthy volunteers.
  - Seven cohorts (30, 120, 360, 720,1080, 1440, and 1800 mg) with six subjects per cohort (1:1, AV-101:placebo)
  - AV-101 was administered as a single oral dose, and subjects were dosed only once

Methods

- The Phase 1B study was a single-site randomized, double-blind, placebo-controlled, study of **multiple ascending doses** (360,1080, and 1440 mg/day) of AV-101 involving 50 normal healthy volunteers, to whom AV-101 or placebo were administered orally daily **for 14 consecutive days**.
  - Three cohorts (AV-101 at 360, 1080, and1440 mg/day) were enrolled in this study.
  - Each cohort had 12 subjects on active drug and 4 subjects on placebo (oral capsules matching the AV-101 in appearance).
  - Study drug was orally administered daily for 14 consecutive days.

Methods

- Screening visit subjects underwent laboratory assessments, physical examination, 12-lead ECG, ophthalmological examination, and neurological assessment (included tests for mental status, sensory/motor exam, coordination/gait, and ataxia)

Methods

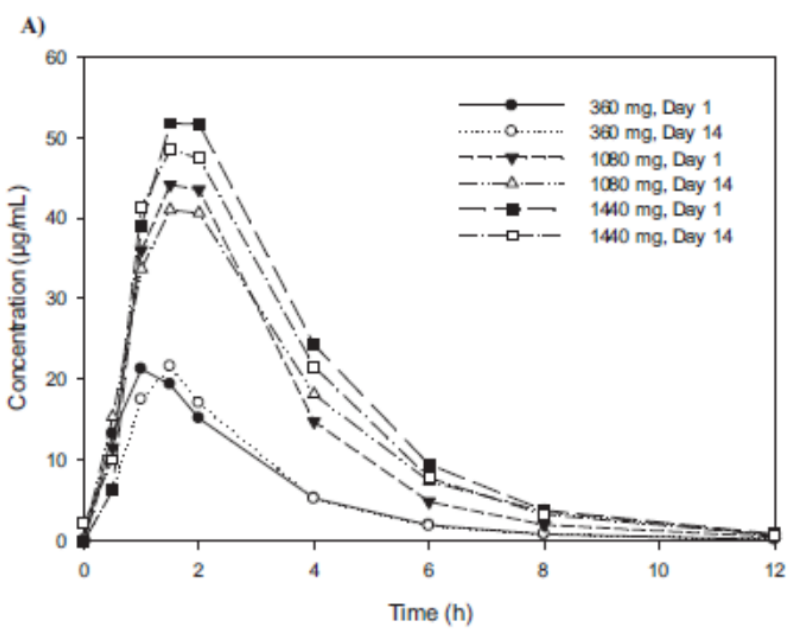
- Capsaicin was delivered intradermally into the volar aspect of one forearm
  - First injection 1 hr after oral administration of AV-101 or placebo
  - Second was given in the other forearm 2 hra after administration of AV-101 or placebo
  - Final capsaicin injection on day 14

Methods

- Patients were provided a paper diary to record dosing, and AEs
- Returned for daily clinical assessment during the 14 days
- PK blood samples were collected on Days 1, 2, 14, and 15

Results: PK Assessments

- Maximum plasma concentrations of AV-101 and 7-Cl-KYNA were reached between 1–2 h
- Mean half-life values of AV-101 were consistent amongst doses ranging from 1.64 h to 1.82 h



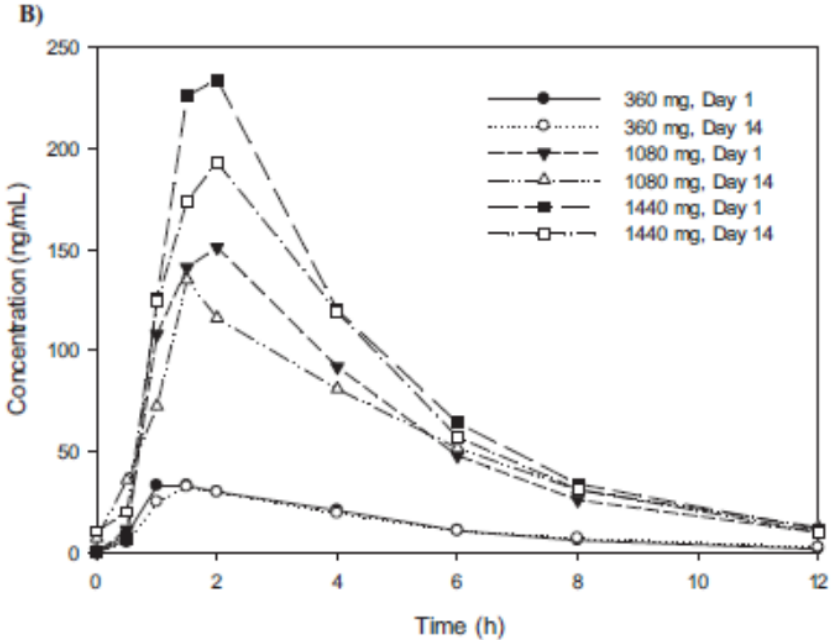


Fig. 1. Mean (n – 12 or 13) plasma concentrations of AV-101 (A) and 7-Cl-KYNA (B) on Days 1 and 14 after oral administration of once-daily doses of AV-101.

Wallace, Mar et. al. Randomized, double-blind, placebo-controlled, dose-escalation study: Investigation of the safety, pharmacokinetics, and antihyperalgesic activity of l-4-chlorokynurenine in healthy volunteers. Scand J Pain. 2017 Oct;17:243-251

Results: PK Assessments

- The highest mean C max and AUC 0–t values for AV-101 occurred in the 1440-mg dose group:
  - 64.4 g/mL (Cmax)
  - 196 g h/mL (AUC 0–t)

- Results were significantly lower than the stopping criteria values

Table 2
Phase 1B summary statistics for AV-101 pharmacokinetic parameters by dose group from treatment Day 1 (B) or Day 14 (B) for subjects receiving a daily oral dose of AV-101 for 14 days.

Subject	T_{max} (h)	C_{max} (μg/mL)	$t_{1/2}$ (h)	AUC_{0-t} (μg h/mL)	$AUC_{0-\infty}$ (μg h/mL)
(A)					
Dose – 360 mg					
n	12	12	12	12	12
Mean	1.15	27.7	1.74	64	64
SD	0.54	6.11	0.37	10	10
Dose – 1080 mg					
n	13	13	13	13	13
Mean	1.52	52.5	1.64	148	149
SD	0.48	14	0.35	32	32
Dose – 1440 mg					
n	12	12	12	12	12
Mean	1.97	64.4	1.66	196	198
SD	1.05	13.6	0.35	41	42
(B)					
Dose – 360 mg					
n	12	12	12	12	12
Mean	1.33	25.5	1.82	62	63
SD	0.44	6.1	0.31	8	8
Dose – 1080 mg					
n	11	11	11	11	11
Mean	1.73	49.2	1.66	162	162
SD	0.89	13.3	0.44	33	33
Dose – 1440 mg					
n	12	12	12	12	12
Mean	1.98	57.7	1.76	183	184
SD	1.18	15.7	0.34	42	42

Objectives

- The primary efficacy endpoint was the analgesic response to spontaneous pain at each dose level of AV-101, 120–180 min after dosing on Day 14
- Secondary endpoints included effects from 60 to 180 min.

Results

- There was no significant change in the area under the pain time curve (AUPC) for the spontaneous pain assessment between the treatment and the placebo groups at 60–180 min after dosing on either Day 1 or 14
- Similarly, there was no significant change at 120–180 min on Day 1 or 14

Results

- There were consistent reductions at 60–180 min after dosing at Day 1 in subjects that received 1080 mg AV-101 for allodynia pain

Results

- There was reduction in the AUPC for heat hyperalgesia pain observed at Day 14 between subjects that received 1080 mg of AV-101

Results

- Plasma concentration–time profiles obtained for AV-101 after administration of once-daily oral doses demonstrated
  - Rapid absorption of the oral dose
  - First-order elimination of both AV-101 and 7-Cl-KYNA
  - Evidence of multicompartment kinetics.

Results

- The mean C max and AUC $_{0-t}$ values for both AV-101 and 7-Cl-KYNA are approximately dose linear but not dose proportional on both Days 1 and 14

Results

- Since the primary objective of the study was safety and PK, a power calculation was not performed.
- However, based on previous studies using the intradermal capsaicin model, 3 cohorts of 16 subjects was considered adequate to fulfill the secondary objective of the study.

Results

- Initial Phase 1A results demonstrated the drug to be well-tolerated
- The frequency and mild severity of the adverse events (AEs) were similar among the placebo and drug cohorts.
- Similar to Phase 1A results, the Phase 1B study demonstrated that AV-101 was well-tolerated, with no serious adverse events

Results

- Thirty-four of 50 subjects in the study reported a total of 57 AEs
- Forty-nine (85.9%) of the AEs were mild, and 8 were moderate.

Table 1
Overall summary of adverse events observed in the Phase 1B study.

	Dose cohorts			
	360 mg AV-101 (*N* = 12) [*n* (%)]	1080 mg AV-101 (*N* = 13) [*n* (%)]	1440 mg AV-101 (*N* = 12) [*n* (%)]	Pooled placebo (*N* = 13) [*n* (%)]
Number of AEs	16	14	10	17
Number of subjects with any AE	9 (75.0%)	8 (61.5%)	7 (58.3%)	10 (76.9%)
Number of SAEs	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Number of AEs resulting in death	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Number of AEs leading to discontinuation of study drug	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)

AE, adverse event; SAE, serious adverse event.

Table S3: Summary of Adverse Events by System Organ Class and Dose Cohort in Phase 1B

Preferred Term	Dose Cohorts*			
	360 mg AV-101 (N = 12) [n (%)]	1,080 mg AV-101 (N = 13) [n (%)]	1,440 mg AV-101 (N = 12) [n (%)]	Pooled Placebo (N = 13) [n (%)]
Cardiac Disorders	0 (0%)	1 (7.7%)	0 (0%)	0 (0%)
Palpitations	0 (0%)	1 (7.7%)	0 (0%)	0 (0%)
Eye Disorders	1 (8.3%)	1 (7.7%)	0 (0%)	1 (7.7%)
Eye discharge	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)
Vision blurred	1 (8.3%)	1 (7.7%)	0 (0%)	0 (0%)
Gastrointestinal Disorders	3 (25.0)	1 (7.7%)	1 (8.3%)	2 (15.4%)
Abdominal pain, upper	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)
Constipation	0 (0%)	1 (7.7%)	0 (0%)	0 (0%)
Diarrhea	1 (8.3%)	0 (0%)	1 (8.3)	0 (0%)
Flatulence	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)
Lip swelling	1 (8.3%)	0 (0%)	0 (0%)	0 (0%)
Nausea	1 (8.3%)	0 (0%)	1 (8.3%)	1 (7.7%)
Vomiting	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)
General Disorders and Administration Site Conditions	0 (0%)	1 (7.7%)	0 (0%)	1 (7.7%)
Chills	0 (0%)	1 (7.7%)	0 (0%)	0 (0%)
Fatigue	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)
Infections and Infestations	1 (8.3%)	0 (0%)	0 (0%)	0 (0%)
Gastroenteritis, viral	1 (8.3%)	0 (0%)	0 (0%)	0 (0%)
Musculoskeletal and Connective Tissue Disorders	1 (8.3)	0 (0%)	0 (0%)	0 (0%)
Pain in extremity	1 (8.3)	0 (0%)	0 (0%)	0 (0%)
Nervous System Disorders	4 (33.3)	6 (46.2)	5 (41.7%)	8 (61.5)
Dizziness	1 (8.3)	1 (7.7%)	0 (0%)	0 (0%)
Dysgeusia	0 (0%)	0 (0%)	1 (8.3%)	0 (0%)
Headache	1 (8.3%)	6 (46.2%)	3 (25.0%)	8 (61.5%)
Hypoesthesia	1 (8.3%)	0 (0%)	0 (0%)	0 (0%)
Somnolence	1 (8.3%)	0 (0%)	1 (8.3%)	0 (0%)
Psychiatric Disorders	1 (8.3%)	1 (7.7%)	1 (8.3%)	0 (0%)
Euphoric mood	1 (8.3%)	1 (7.7%)	1 (8.3%)	0 (0%)
Renal and Urinary Disorders	1 (8.3%)	0 (0%)	0 (0%)	0 (0%)
Pollakiuria	1 (8.3%)	0 (0%)	0 (0%)	0 (0%)
Respiratory, Thoracic, and Mediastinal Disorders	1 (8.3%)	0 (0%)	1 (8.3%)	1 (7.7%)
Nasal congestion	1 (8.3)	0 (0%)	0 (0%)	0 (0%)
Oropharyngeal pain	0 (0%)	0 (0%)	1 (8.3%)	1 (7.7%)
Sinus congestion	0 (0%)	0 (0%)	1 (8.3%)	0 (0%)
Skin and Subcutaneous Tissue Disorders	1 (8.3%)	0 (0%)	0 (0%)	2 (15.4%)
Acne	1 (8.3%)	0 (0%)	0 (0%)	1 (7.7%)
Rash	0 (0%)	0 (0%)	0 (0%)	1 (7.7%)

*** Subjects experiencing more than one AE in a given SOC or PT are counted only once in that particular SOC or PT.**

Results

- The 2 moderate intensity AEs were in the 360-mg AV-101 group: one was unrelated pain in the right foot, and one was a possibly related headache.
- All other moderate AEs occurred in the placebo group and included nausea or vomiting (2 AEs), headache (2 AEs), and rash around the neck (1AE)

Safety

- No clinically significant abnormal measurements in the vital signs, physical examinations, 12-lead ECGs, ataxia tests, ophthalmological examinations, capsaicin injection site examinations, or interactions with concomitant medications during the study, and no significant changes in neurocognitive scores between Days 1 and 14.

Conclusions

- 4-Cl-KYN is an oral prodrug producing a potent brain NMDAR GlyB-site antagonism.
- Antagonism of Glycine-B site with 4-Cl-KYN does not appear to elicit the detrimental side effects (e.g., psychotomimetic and rewarding) associated with the use of ketamine or other NMDA channel blockers
- Good safety profile, better than placebo.
- Exhibited non significant decreases in allodynia and hyperalgesia.
- Reports of feelings of well-being with 4-Cl-KYN suggest anti-depressant activity

Discussion

- Use of capsaicin to accurately predict antinociceptive effects of a drug in humans may vary
- Although no effect on capsaicin induced pain, can have significant therapeutic benefit in reducing clinical pain states, as seen in Duloxetine, amitriptyline and desipramine[8,9]
- Head to head studies

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