Received: 28 July 2021 | Revised: 31 December 2021 | Accepted: 3 January 2022

DOI: 10.1111/bcp.15229

ORIGINAL ARTICLE

Pharmacological profile of ALKS 7119, an investigational compound evaluated for the treatment of neuropsychiatric disorders, in healthy volunteers

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Funding information
Alkermes, Inc

Aims: ALKS 7119 is a novel compound with in vitro affinity highest for the SERT, and for μ receptor, α_{1A}-adrenoceptor, α_{1B}-adrenoceptor, NMDA receptor and sigma non-opioid intracellular receptor 1. This first-in-human study evaluated safety and PK/PD effects of single ascending doses (SAD) of ALKS 7119 in healthy males and compared effects with neurotransmitter modulators with partially overlapping targets.

Methods: In 10 cohorts ($n = 10$ subjects each), PK, safety and PD (NeuroCart tests, measuring neurophysiologic effects [pupillometry, pharmaco-EEG (pEEG)], visuomotor coordination, alertness, [sustained] attention [saccadic peak velocity, adaptive tracking], subjective drug effects [VAS Bowdle and VAS Bond and Lader] and postural stability [body sway]) were evaluated. Neuroendocrine effects (cortisol, prolactin, growth hormone) were measured. Data were analysed over the 12-hour post-dose period using mixed-effects model for repeated measure (MMRM) with baseline as covariate.

Results: ALKS 7119 demonstrated linear PK and was generally well tolerated. QTcF interval increases of 30–60 ms compared to baseline were observed with ALKS 7119 doses of ≥50 mg without related adverse events. Significant increases in left and right pupil/iris ratio were observed at dose levels ≥50 mg (estimate of difference [95% CI], *P*-value) (0.04 [0.01; 0.07], $P < .01$) and (0.06 [0.03; 0.09], $P = .01$), respectively. From dose levels ≥50 mg significant increases (% change) of serum cortisol (51.7 [8.4; 112.3], $P = .02$) and prolactin (77.9 [34.2; 135.8], $P < .01$) were observed.

Conclusion: In line with ALKS 7119's in vitro pharmacological profile, the clinical profile observed in this study is most comparable to SERT inhibition.

KEYWORDS
drug development, neuropsychiatric disorders, pharmacological effects, SERT

The authors confirm that the Principal Investigator for this paper is J.M.A. van Gerven and that he had direct clinical responsibility for research subjects.

1 | INTRODUCTION

ALKS 7119 is an investigational compound that has been evaluated for the potential treatment of neuropsychiatric disorders. Preclinical tests using a panel of in vitro receptor, transporter, enzyme binding and functional assays showed that ALKS 7119 has high affinity for the SERT (Ki = 0.035 μM), and lower affinities for the μ receptor (Ki = 0.6 μM), α_{1A}-adrenoceptor (Ki = 0.98 μM), α_{1B}-adrenoceptor (Ki = 1.8 μM), NMDA receptor (Ki = 7.44 μM) and sigma non-opioid intracellular receptor 1 (Ki = 33.0 μM). In vivo pharmacology studies in rats demonstrated that an oral dose of 10 mg/kg ALKS 7119 completely blocked dopamine release induced by infusion of the synthetic glutamate agonist NMDA in the striatum. In preclinical pharmacokinetic studies with both dogs and rats, the median time to maximum plasma concentration (t_{max}) of ALKS 7119 was 0.5 hour after oral administration. Binding to plasma protein ranged from 11% to 28%, oral bioavailability from 43% to 76% and mean elimination half-life ($t_{1/2}$) from 2.9 to 5.6 hours across species.

Preclinical multiple dose toxicology studies with ALKS 7119 demonstrated a no adverse effect level (NOAEL) of 10 mg/kg in rats. At higher doses symptoms of decreased body weight and hepatocellular vacuolation and hypertrophy and changes in behaviour, such as arousal and decreased mobility in open-field observations, were observed. The NOAEL corresponded to a human equivalent dose (HED) of 96 mg, calculated per FDA guidelines (using body surface area extrapolation).[1] Preclinical multiple dose toxicology studies in dogs demonstrated a NOAEL of 3 mg/kg, corresponding to a HED of 97.2 mg.[1] At a HED of 324 mg, decreases in systolic, diastolic, and mean blood pressure as well as compensatory increases in heart rate occurred. In addition, symptoms of recumbency and decreased activity were observed. No changes in QT intervals at any dose of ALKS 7119 were observed in the preclinical dog cardiovascular study, a finding consistent with in vitro hERG channel testing, showing no effects up to high concentrations (IC_{50} of 191.1 μM or 54348.8 ng/mL). ALKS 7119 showed no potential to induce neuronal abnormalities up to a HED of 3360 mg. Based on these preclinical safety data, it was decided to continue to an FIH study, starting with a dose of 3 mg (32 times lower than the NOAEL in rats, the most sensitive species).

The relatively high affinity of ALKS 7119 for several distinct receptor types offered the potential to evaluate ALKS 7119 for the treatment of various neuropsychiatric conditions, ranging from neuropathic pain and brainstem behavioural disorder to schizophrenia and depression. Further exploration of these indications would have required a large series of preclinical disease models, which all have limited predictive power for compounds with novel and complex profiles of pharmacologic action. It was therefore decided to not only characterize the PK and safety in this FIH study, but to also include a wide range of different CNS functions, which could provide indications for BBB penetration and target engagement profiles in humans.

This approach is in line with the "question-based drug development (QBDD)" method, which is developed to investigate novel compounds in a structured way to prevent late stage drug development failures.[2] According to QBDD, studies must be designed to answer important questions about novel compounds.[2] In the case of a CNS drug, such as ALKS 7119, it was considered important to know whether the drug crosses the BBB and on which receptors it mainly acted.[2] To answer these questions, the current study utilized the NeuroCart, which consists of a battery of drug-sensitive CNS tests, measuring effects on different CNS domains, such as neurophysiologic functioning, visuomotor coordination, balance and subjective feelings.[3] Several CNS-active compounds, including compounds influencing serotonergic, opioid, GABA-ergic and glutamatergic (via NMDA-antagonism) networks, have been profiled using the NeuroCart. This allowed a comparison of the functional profile of this new pharmacologically heterogeneous compound to other known drug profiles, and consequently to obtain a better understanding of the underlying pharmacological effects.[3,4]

What is already known about this subject

- ALKS 7119 [(9α, 13α)-17-methylmorphinan-3-carboxamide] is a novel compound with in vitro binding affinity for the serotonin [5HT] transporter (SERT), μ receptor, α_{1A}-adrenoceptor, α_{1B}-adrenoceptor, N-methyl-D-aspartate-(NMDA) receptor and sigma non-opioid intracellular receptor 1.
- The SERT and NMDA receptor are involved in the pathophysiology of neuropsychiatric disorders, such as depression.
- It is important to demonstrate blood–brain-barrier (BBB) penetration and target engagement of novel central nervous system (CNS) active compounds in early drug development stages.

What this study adds

- This study shows how drug-sensitive CNS tests can be applied in first-in-human (FIH) studies to measure BBB penetration and pharmacological effects at different target receptors.
- ALKS 7119 demonstrated a linear pharmacokinetic (PK) profile with dose-proportional increases in systemic exposure and was generally well tolerated at single doses up to 200 mg.
- ALKS 7119 did not cause any subjective, behavioural or neurophysiological changes, but demonstrated significant increases of pupil size and dose-dependent increases of serum cortisol and prolactin levels. This profile is indicative of BBB penetration and most comparable to SERT inhibition.

The preclinical pharmacological profile of ALKS 7119, with relatively high affinity for different receptor types, required NeuroCart testing at dose levels with small increments, to be able to disentangle ALKS 7119's effects on distinct receptors with different affinities. Specific NeuroCart tests were selected based on ALKS 7119's pharmacological profile. Pupil size measurements were included to measure serotonergic and μ receptor effects, as opioids are known to induce pupil constriction, whilst most studies with selective serotonin reuptake inhibitors (SSRIs) show pupil dilation.[5,6] In addition, pharmaco-EEG (pEEG) was included because this is a potential biomarker for SERT engagement.[6] Ketamine, a well-known NMDA receptor antagonist, demonstrated decreased saccadic peak velocity, adaptive tracking and alertness and increased body sway and psychedelic effects as measured by visual analogue scale (VAS) Bowdle on the NeuroCart.[7] Buprenorphine, a partial μ receptor agonist, decreased adaptive tracking and saccadic peak velocity and increased body sway.[8]

In addition to the NeuroCart tests, serum cortisol and prolactin levels were measured as biomarkers for serotonergic effects, as escitalopram and citalopram are known to increase levels of these hormones.[9] Although it is uncertain whether **growth hormone** levels are influenced by serotonergic compounds,[10] serum growth hormone levels were also measured.

There are no established CNS tests for mild adrenergic modulation. The CNS effects of strong noradrenalin release stimulators like dexamphetamine,[11] or the potent inhibitory effects of presynaptic α_2-adrenoceptor agonists and imidazoline modulators like clonidine[12] or rilmenidine,[13] can be readily shown with several NeuroCart tests. However, demonstration of more subtle noradrenergic modification does not cause spontaneous changes of NeuroCart tests in healthy subjects,[13] but requires more elaborate tests of cognition or pain.[14] The current study did not include any such specific biomarkers for modest α_{1A}- or α_{1B}-adrenergic receptor modulation, other than the safety blood pressure measurements. Similarly, no specific tests for sigma non-opioid intracellular receptor 1 modulation could be identified for inclusion in the study.

The aim of this study was to profile single ascending doses of ALKS 7119 in terms of safety, tolerability, PK and PD effects in healthy male volunteers, and to compare these effects with known functional effects of different neurotransmitter modulators with partially overlapping mechanisms of action.

2 | METHODS

2.1 | General

The study was registered at ToetsingOnline under number NL155561.056.15 and approved by Foundation Beoordeling Ethiek Biomedisch Onderzoek (BEBO), Assen, the Netherlands. All subjects gave written informed consent prior to study start. The study was performed according to ICH GCP guidelines as laid down in the Declaration of Helsinki and its latest amendments. Alkermes Inc. sponsored the study, and the study was conducted from 4 January 2016 to 13 July 2016 at the Centre for Human Drug Research (CHDR), Leiden, the Netherlands.

2.2 | Design

This was a single-centre, randomized, double-blind, placebo-controlled, single ascending dose study in 100 healthy male adults. Due to the exploratory character of this FIH study, the sample size was based on clinical considerations rather than power calculations. Subjects were divided over 10 cohorts (active:placebo ratio of 8:2) where each cohort represented a different dose level: 3, 10, 25, 50, 75, 100, 125, 150, 175 and 200 mg. Sentinel dosing was performed on the first two subjects in cohort 1. Before ascending to the next dose level, all available safety, PK and PD data of the preceding dose level(s) were reviewed.

The study consisted of a medical screening visit, an inpatient study visit and an inpatient follow-up visit. Study visits consisted of 3 inpatient days; subjects arrived the day prior to dosing, were dosed the following day and were discharged the day after dosing (Figure 1). At check-in, eligibility was checked based on physical exam, including weight, laboratory testing including urinalysis, urine drug screen, electrocardiogram (ECG), breath alcohol test, concomitant medication, adverse event (AE) review and vital sign measurement including tympanic temperature measurement, pulse rate and (orthostatic) blood

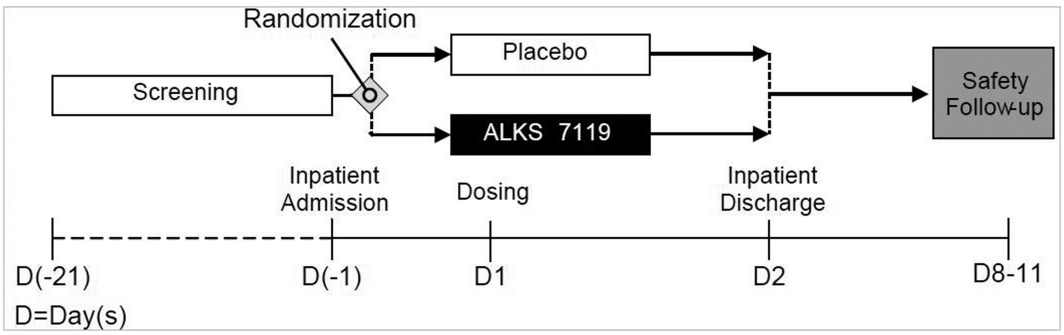


FIGURE 1 Study design

pressure measurements. Blood pressure and pulse rate measurements were performed after subjects had been in supine position for 5 minutes. For orthostatic blood pressure measurements, subjects were then instructed to stand up and after 2 minutes, blood pressure and pulse rate were measured again. Orthostatic hypotension was defined as ≥ 20 mmHg decrease in systolic blood pressure and ≥ 10 mmHg decrease of diastolic blood pressure. The safety measurements were repeated throughout the study at set times.

Blood samples for measurement of plasma concentrations of ALKS 7119 and serum neuro-endocrine hormone levels were collected within 1 hour pre-dose and at 0.25, 0.5, 1, 2, 4, 6, 8, 12, 16, 24 and 36 hours post-dose. NeuroCart assessments, consisting of saccadic eye movements, smooth pursuit eye movements, adaptive tracking, body sway, pupillometry, pharmaco-electroencephalography (pEEG), visual analogue scales (VAS) according to Bond and Lader and Bowdle were performed pre-dose (twice) and 0.5, 1, 1.5, 2, 3, 4, 6 and 10 hours post-dose.

2.3 | Electrocardiogram (ECG) acquisition and analysis

At scheduled time points, standard 12-lead ECG recordings were performed in triplicate with 1 minute in between each replicate. Recordings were made after a 5 minute resting period and in semi-recumbent position. The ECGs were recorded using an electrocardiograph (Marquette 800/5500/2000 or Dash 3000; General Electric Healthcare, Milwaukee, WI, USA) and ten disposable electrodes placed in the standard anatomical position. ECG data were uploaded into the ECG warehouse, which automatically assesses interval, including QTc intervals, and amplitude data from the digital ECGs with the Marquette 12SL algorithm (Muse Cardiology Data Management System v7, General Electric Healthcare, Chicago, IL, USA). The Marquette Cubic Spline filter and Finite Residual Filter were used for artefact and noise management. A physician manually reviewed all ECGs for quality, legibility and abnormalities.

2.4 | Subjects

Healthy male subjects between 18 and 45 years of age at screening were selected. Subjects were not allowed to use medication within 7 days prior to screening or inpatient admission and during the study days. Subjects were asked not to consume any alcohol, caffeine or xanthine-containing beverages within 24 hours and not to use any nicotine-containing products within 30 days prior to inpatient admission and during the study days.

2.5 | Treatments

ALKS 7119 was provided as size 0 Swedish orange, opaque, hard gelatin capsules compounded at target strengths (i.e., 3 mg to 200 mg) for oral use. Placebo consisted of identical, empty capsules. Subjects began fasting the night before study drug administration until 4 hours thereafter. Subjects were allowed water ad libitum except for 1 hour before and 1 hour after study drug administration.

2.6 | Pharmacokinetic assessments

Plasma samples were analysed by an independent bioanalytical laboratory (Analytisch Biochemisch Laboratorium BV, Assen, The Netherlands). Concentrations of ALKS 7119 were quantified using a validated liquid chromatography with tandem mass spectrometry (LC–MS/MS) method with lower limit of quantification (LLOQ) of 1.00 ng/mL and coefficient of variation (CV) between 1.9 and 4.6%.

2.7 | NeuroCart assessments

All tests were performed in a quiet room with subdued illumination with only one subject in the same room per session. A NeuroCart test training was performed during the screening visit, to prevent learning effects during study execution.

2.7.1 | Saccadic eye movement

The primary outcome of saccadic eye movement measurement is saccadic peak velocity (SPV) in degrees per second (deg/s), a sensitive parameter for numerous sedative compounds.[4,15,16] Tests were performed as described in previous publications.[15–18] Subjects were instructed to follow a dot jumping approximately 15 degrees to either side on a computer screen with their eyes, while head movements were restrained using a fixed head support at 58 cm from the computer screen. Fifteen saccades were recorded with interstimulus intervals varying randomly between 3 and 6 seconds. Average values of saccadic peak velocity were calculated for all artefact-free saccades.

2.7.2 | Smooth pursuit

Smooth pursuit was performed as described in previous publications.[16,17,19,20] In short, smooth pursuit measurements were performed using the same set-up as for saccadic eye movements, but the dot was moving continuously at a frequency ranging from 0.3 to 1.1 Hz, by steps of 0.1 Hz instead of jumping on the screen. The amplitude of target displacement corresponds to 22.5 degrees eyeball rotation to both sides. Four cycles are recorded for each stimulus frequency. The target parameter was the average percentage of smooth pursuit for all stimulus frequencies.

2.7.3 | Adaptive tracking

The adaptive tracking test was performed as originally described by Borland and Nicholson,[21] using customized equipment and software (based on TrackerUSB hard-/software [Hobbs, 2004, Hertfordshire, UK]). Adaptive tracking is a pursuit-tracking task that is highly sensitive to a wide range of psychoactive drugs.[15,17,22–24] During the test, a circle moves randomly on a screen and the subject is instructed to try to keep a dot inside the moving circle by operating a joystick. If this effort is successful, the speed of the moving circle increases. Conversely, the velocity is reduced if the test subject cannot maintain the dot inside the circle. The average speed of the moving circle as a percentage of the maximum speed of the circle over a 3.5-minute period was used for analysis.

2.7.4 | Body sway

Postural stability was assessed by body sway as previously described by others.[15,25] Anteroposterior body sway was measured with closed eyes, using a body sway meter (Celesco) based on Wright ataxiameter.[26] All body movements over a 2-minute period were integrated and expressed as millimetres of sway and recorded.

2.7.5 | Pupillometry

Pupillometry was performed as described previously.[27] While subjects were sitting in a chair with their head resting in a head support system, a picture was taken from both eyes simultaneously. The ratio between pupil and iris diameter was measured using Qpupil (radiology department, LUMC, the Netherlands). This ratio was used to make sure that pupil size measurement was independent of distance between camera and subject.

2.7.6 | Pharmaco-EEG

Continuous EEG recordings were made using a 40-channel recording system (Refa-40, TMSi B.V., the Netherlands). EEGs were recorded using 21 electrodes, which were placed according to the international 10–20 system, except that electrodes near the mastoids replaced those on the earlobes. The scalp electrode impedance was kept below 5 kΩ. The ground electrode was placed at AFz (Auricular Frontal midline). Additionally, to detect ocular artefacts, vertical and horizontal electro-oculo-graphic (EOG) signals were also recorded. Two Ag/AgCl electrodes were placed at the outer canthi of both eyes, and two Ag/AgCl electrodes were placed approximately 2 cm above and below the right eye. The derivations of interest for this study were midline frontal-central (Fz-Cz) and midline parietal-occipital left (Pz-O1) and right (Pz-O2).

EEGs were recorded and analysed in line with guidelines described by the international pharmaco-EEG society (IPEG).[28] Subjects were instructed not to stare, to limit their head and eye movements, and to suppress eye blinks. Resting-state EEG recordings with open and closed eyes for 5 minutes in each eye state were performed. All signals were sampled at a sampling rate of 1024 Hz and filtered prior to storage using a first order recursive high-pass filter with a cut-off frequency at 0.1 Hz. Digital markers were recorded by the amplifier indicating the start and end of each eye state.

Recorded channels were band-pass filtered using a third order Butterworth filter with cut-off frequencies at 0.1 and 45.0 Hz. The filtered signals were divided into 4 second epochs. Epochs containing ocular artifacts were removed for further analysis. A power spectrum density (PSD) was calculated for each epoch and averaged for each eye state. The resulting PSDs were subdivided into bands and the total power per band was calculated. The following parameters (all μV) were collected: Alpha-power Fz-Cz, Alpha-power Pz-Oz, Beta-power Fz-Cz, Beta-power Pz-Oz, Gamma-power Fz-Cz, Gamma-power Pz-Oz, Delta-power Fz-Cz, Delta-power Pz-Oz, Theta-power Fz-Cz, Theta-power Pz-Oz.

2.7.7 | Visual analogue scales (VAS)

VAS in this study were used as originally described by Norris.[29] Dutch versions of the scales have frequently been employed at CHDR, for a variety of sedative agents[15] and circumstances.[16] For VAS Bond and Lader, subjects indicate (with vertical marks) on 16 horizontal 100 mm VAS how they feel. From these measurements, three main factors are calculated as described by Bond and Lader.[30] These three factors are "subjective alertness" (from nine scores), "contentedness or mood" (from five scores) and "calmness" (from two scores).[30] VAS Bowdle evaluates psychedelic effects with 13 10 cm VAS lines ranging from 0 (not at all) to 100 mm (extremely).[31] These scores are clustered into three distinct total sum scores: "internal perception" (reflects inner feelings that do not correspond with reality, including mistrustful

TABLE 1 Demographics

Characteristic	ALKS 7119 ($n = 80$)	Placebo ($n = 20$)
Age, years		
Mean (SD)	24.1 (4.6)	24.6 (5.6)
Height (cm)		
Mean (SD)	182.6 (7.5)	181.8 (8.8)
Weight (kg)		
Mean (SD)	76.6 (11.8)	78.8 (12.6)
Body mass index (kg/m^2)		
Mean (SD)	22.8 (2.8)	23.7 (3.2)
Race, *n* (%)		
White	69 (86.3)	16 (80)
Other	7 (8.8)	3 (15)
Asian	2 (2.5)	0 (0)
Black or African American	2 (2.5)	1 (5.0)

TABLE 2 Pharmacokinetic characteristics of ALKS 7119

Parameter statistic	ALKS 7119 3 mg ($n = 8$)	ALKS 7119 10 mg ($n = 8$)	ALKS 7119 25 mg ($n = 8$)	ALKS 7119 50 mg ($n = 8$)	ALKS 7119 75 mg ($n = 8$)	ALKS 7119 100 mg ($n = 8$)	ALKS 7119 125 mg ($n = 8$)	ALKS 7119 150 mg ($n = 8$)	ALKS 7119 175 mg ($n = 8$)	ALKS 7119 200 mg ($n = 8$)
C_{max} (ng/ml)										
Mean (SD)	8.33 (2.78)	25.33 (3.91)	74.51 (16.19)	140.00 (32.44)	225.50 (40.59)	307.63 (75.28)	447.38 (133.80	413.38 (65.38)	660.63 (167.66)	656.00 (104.74)
Range	5.87–13.20	20.40–32.30	51.00–93.00	103.00–190.00	190.00–302.00	224.00–465.00	315.00–745.00	314.00–540.00	451.00–893.00	491.00–795.00
C_{max}/dose	2.78	2.53	2.98	2.80	3.01	3.08	3.58	2.76	3.78	3.28
t_{max} (h)										
Median	2.02	2.00	1.00	1.53	1.50	1.50	2.00	2.00	2.01	3.00
Range	2.00–4.15	1.00–2.00	1.00–2.00	1.00–4.00	0.50–4.00	1.00–4.08	2.00–4.00	1.00–4.00	1.02–4.00	1.00–4.00
AUC_{∞} (hr*ng/ml)										
Mean (SD)	100.03 (25.04)	295.38 (32.29)	731.29 (87.27)	1547.06 (196.89)	2548.51 (468.23)	3136.45 (634.60)	4187.59 (840.18)	4533.66 (862.77)	5954.09 (1047.00)	6721.82 (1227.19)
Range	73.65–140.04	233.76–338.34	640.06–883.94	1277.55–1828.84	1772.06–3367.86	2418.74–4525.46	3188.91–5732.16	3790.65–6362.63	4820.27–7488.93	5273.49–9126.15
$AUC_{infinite}$/dose	33.34	29.54	29.25	30.94	33.98	31.36	33.50	30.22	34.02	33.61
AUC_{last} (hr*ng/ml)										
Mean (SD)	85.88 (25.56)	272.60 (35.69)	703.56 (89.80)	1473.57 (203.53)	2406.33 (373.64)	3027.23 (600.53)	4059.17 (809.98)	4361.53 (782.96)	5754.94 (1013.23)	6490.47 (1144.87)
Range	57.37–129.02	198.45–321.20	608.46–864.96	1234.86–1780.97	1732.82–2969.47	2340.29–4326.23	3120.58–5579.28	3622.31–6015.06	4681.45–7356.86	5120.43–8737.25
$t_{1/2}$ (h)										
Mean (SD)	6.98(1.16)	8.51 (0.89)	7.81 (0.83)	7.81 (0.69)	8.42 (1.63)	7.63 (0.93)	7.26 (0.99)	7.53 (0.88)	7.48 (0.87)	7.35 (0.58)
Range	5.20–8.77	7.01–9.45	6.78–9.41	6.99–8.68	6.84–12.01	6.36–8.84	6.06–8.72	6.10–8.51	5.91–8.83	6.72–8.33

feelings), "external perception" (reflects a misperception of an external stimulus or a change in the awareness of the subject's surroundings) and "feeling high".[31]

2.7.8 | Neuro-endocrine hormones

Samples were analysed by an independent bioanalytical laboratory (Analytisch Biochemisch Laboratorium BV, Assen, The Netherlands). Cortisol concentration was determined using a validated LC–MS/MS method LLOQ of 2.00 ng/mL and CV between 2.4% and 9.9% across measurements. Prolactin concentration was determined using a qualified time-resolved fluoroimmunoassay with LLOQ of 0.260 ng/mL and CV between 0.9% and 1.6% across measurements. Growth hormone concentration was determined using a qualified enzyme immunoassay with LLOQ of 0.550 μIU/mL and CV between −4.8% and 18.6% across measurements.

2.8 | Analysis

2.8.1 | Pharmacokinetics

PK parameters were calculated from concentration data in mass/volume units. Parameters were calculated using noncompartmental analysis, using actual elapsed time from dosing to estimate individual plasma PK parameters. These parameters were: C_{max}, t_{max}, $t_{1/2}$, area under the concentration–time curve from time zero to the last quantifiable concentration time point (AUC_{last}), area under the concentration–time curve from time zero to infinity (AUC_{∞}). All PK data were summarized by treatment group using descriptive statistics. Values were expressed as the mean ± SD for all parameters except t_{max}, which was presented as the median (range).

2.8.2 | Statistical analysis

Statistical analyses were performed using SAS Version 9.4 (SAS Institute Inc., Cary, NC, USA).

Placebo subjects from all cohorts were pooled together to form a placebo group. Comparisons were then made between active treatment groups and the pooled placebo group. The only exception to this was VAS Bowdle as there were many scores of zero in the placebo arm. Therefore, the lowest dose of ALKS 7119 (3 mg) was used as the reference treatment to which the higher dose levels were compared. Repeatedly measured PD data were summarized by treatment group and time point and analysed with a mixed-effects model for repeated measure (MMRM) with treatment, time point, and the interaction term of treatment by time as fixed factors and subject as a random factor. The baseline measurement was included as a covariate. Baseline was defined as the last non-missing value before randomized study drug administration. MMRM was conducted for the change from baseline over the 12-hour post-dose period as the dependent variable. No adjustment for multiple testing was performed. Treatment effects of each ALKS 7119 dose against placebo were reported using least squares means (LSM), least squares mean difference, 95% confidence interval (CI) and the *P*-value.

Body sway (antero-posterior sway in mm/2 minutes) and pharmaco-EEG endpoints were natural log transformed before entering the MMRM. For these endpoints, LS mean, LS mean difference and 95% CI were transformed back to their original scale (i.e., to geometric mean and geometric mean ratio).

Neuro-endocrine hormones underwent natural log transformation before entering the same MMRM model as was used for the NeuroCart analyses, with the only difference that analyses were performed with the 2 hours post-dose data, the expected t_{max}, instead of the post-dose data over 12 hours. To represent results, the neuro-endocrine hormone data were transformed back to their original scale.

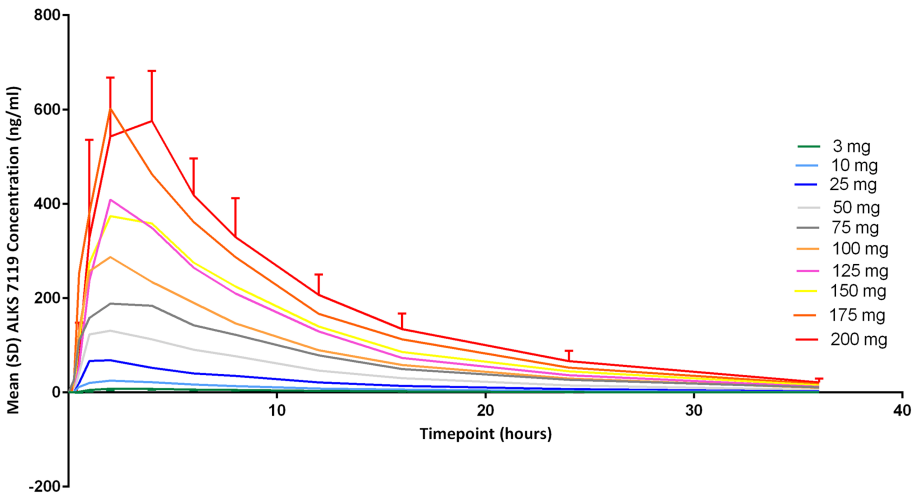


FIGURE 2 Mean (SD) concentration of ALKS 7119 (ng/ml) (linear scale)

TABLE 3 NeuroCart treatment effects compared to placebo. Least squares mean change from baseline over the 12-hour post-dose period. Estimate of difference compared to placebo (standard error)[a] [95% confidence interval], *P*-value

Parameter	Overall	Placebo	ALKS 7119 3 mg	ALKS 7119 10 mg	ALKS 7119 25 mg	ALKS 7119 50 mg	ALKS 7119 75 mg	ALKS 7119 100 mg	ALKS 7119 125 mg	ALKS 7119 150 mg	ALKS 7119 175 mg	ALKS 7119 200 mg
Left pupil/iris ratio	**LSM CFB**	0.01	0.01	0.04	0.04	0.05	0.04	0.04	0.04	0.03	0.05	0.08
	$P < .01^*$		0 (0.01) [−0.02, 0.03] $P = .93$	0.03 (0.01) [0, 0.05] $P = .04^*$	0.03 (0.01) [0, 0.05] $P = .05^*$	0.04 (0.01) [0.01; 0.07] $P < .01^*$	0.03 (0.01) [0, 0.05] $P = .03^*$	0.03 (0.01) [0, 0.05] $P = .03^*$	0.03 (0.01) [0.01, 0.06] $P = .01^*$	0.02 (0.01) [0, 0.05] $P = .08$	0.04 (0.01) [0.05, 0.1] $P < .01^*$	0.07 (0.01) [0.05, 0.1] $P < .01^*$
Right pupil/iris ratio	**LSM CFB**	0.01	0.02	0.03	0.03	0.07	0.03	0.05	0.05	0.05	0.06	0.06
	$P < .01^*$		0.01 (0.01) [−0.02, 0.04] $P = .43$	0.02 (0.01) [−0.01, 0.04] $P = .23$	0.02 (0.01) [−0.01, 0.05] $P = .16$	0.06 (0.01) [0.03, 0.09] $P < .01^*$	0.02 (0.01) [−0.01, 0.05] $P = .12$	0.04 (0.01) [0.01, 0.06] $P = .01^*$	0.03 (0.01) [0.01, 0.06] $P = .01^*$	0.03 (0.01) [0.01, 0.06] $P = .02^*$	0.05 (0.01) [0.02, 0.08] $P < .01^*$	0.05 (0.01) [0.02, 0.08] $P < .01^*$
Saccadic peak velocity (deg/s)	**LSM CFB**	−2.14	−16.15	1.46	6.12	2.91	−5.63	13.25	5.98	18.15	5.26	−0.64
	0.19		−14.01 (9.2) [−32.3, 4.28] $P = .13$	3.6 (9.44) [−15.16, 22.37] $P = .70$	8.26 (9.17) [−9.96, 26.48] $P = .37$	5.05 (9.24) [−13.31, 23.41] $P = .59$	−3.49 (9.17) [−21.71, 14.73] $P = .70$	15.39 (9.23) [−2.95, 33.74] $P = .10$	8.12 (9.14) [−10.03, 26.28] $P = .38$	20.29 (9.14) [2.13, 38.45] $P = .03^*$	7.4 (9.15) [−10.77, 25.57] $P = .42$	1.5 (9.21) [−16.81, 19.81] $P = .87$
Smooth pursuit (%)	**LSM CFB**	−1.17	−0.26	0.06	−1.44	−0.03	−3.44	−0.49	−3.82	−1.14	−1.07	1.24
	0.37		0.91 (1.65) [−2.37, 4.2] $P = .58$	1.23 (1.66) [−2.06, 4.52] $P = .46$	−0.27 (1.65) [−3.56, 3.02] $P = .87$	1.14 (1.66) [−2.16, 4.44] $P = .49$	−2.26 (1.68) [−5.61, 1.08] $P = .18$	0.68 (1.66) [−2.61, 3.97] $P = .68$	−2.64 (1.67) [−5.96, 0.68] $P = .12$	0.03 (1.66) [−3.27, 3.34] $P = .98$	0.1 (1.66) [−3.19, 3.39] $P = .95$	2.41 (1.66) [−0.89, 5.7] $P = .15$
Body sway (LOG mm)	**LSM CFB**	0.03	−0.14	0.16	−0.03	0.12	−0.06	0.08	Not done	Not done	Not done	Not done
	0.34		−0.17 (0.13) [−0.43, 0.08] $P = .18$	0.13 (0.13) [−0.13, 0.39] $P = .33$	−0.06 (0.13) [−0.32, 0.2] $P = .65$	0.09 (0.13) [−0.17, 0.34] $P = .50$	−0.1 (0.13) [−0.35, 0.16] $P = .45$	0.05 (0.13) [−0.21, 0.3] $P = .72$				
Adaptive tracking (%)	**LSM CFB**	1.02	0.15	2.4	0.24	2.04	0.25	1.05	0.8	0.65	0.9	0.72
	0.51		−0.87 (0.88) [−2.61, 0.87] $P = .32$	1.38 (0.87) [−0.36, 3.12] $P = .12$	−0.79 (0.88) [−2.53, 0.96] $P = .37$	1.01 (0.88) [−0.73, 2.75] $P = .25$	−0.78 (0.88) [−2.52, 0.97] $P = .38$	0.03 (0.87) [−1.71, 1.77] $P = .97$	−0.22 (0.88) [−1.97, 1.52] $P = .80$	−0.37 (0.88) [−2.11, 1.37] $P = .67$	−0.12 (0.87) [−1.86, 1.61] $P = .89$	−0.3 (0.87) [−2.04, 1.44] $P = .73$
VAS Bond and Lader	**LSM CFB**	−0.57	1.64	0.03	0.26	0.96	−1.02	0.57	−0.56	1.94	−1.17	0.09

TABLE 3 (Continued)

Parameter	Overall	Placebo	ALKS 7119 3 mg	ALKS 7119 10 mg	ALKS 7119 25 mg	ALKS 7119 50 mg	ALKS 7119 75 mg	ALKS 7119 100 mg	ALKS 7119 125 mg	ALKS 7119 150 mg	ALKS 7119 175 mg	ALKS 7119 200 mg
"subjective alertness" (mm)	0.50		2.21 (1.28) [−0.33, 4.75] $P = .09$	0.61 (1.28) [−1.93, 3.15] $P = .64$	0.84 (1.28) [−1.7, 3.38] $P = .51$	1.53 (1.31) [−1.08, 4.14] $P = .25$	−0.45 (1.28) [−2.99, 2.09] $P = .73$	1.14 (1.29) [−1.41, 3.7) $P = .38$	0.01 (1.28) [−2.53, 2.56] $P = .99$	2.51 (1.28) [−0.03, 5.06] $P = .05$	−0.59 (1.28) [−3.14, 1.96] $P = .65$	0.66 (1.3) [−1.93, 2.28] $P = .61$
VAS Bond and Lader "mood" (mm)	LSM CFB 0.15	−0.33	6.17 6.5 (2.23) [2.06, 10.93] $P < .00$*	−0.04 0.29 (2.23) [−4.13, 4.72] $P = .90$	−0.14 0.19 (2.23) [−4.24, 4.62] $P = .93$	4.47 4.8 (2.36) [0.1, 9.59] $P = .05$*	−0.82 −0.49 (2.22) [−4.91, 3.93] $P = .83$	0.56 0.89 (2.23) [−3.54, 5.32] $P = .69$	1.63 1.96 (2.26) [−2.53, 6.44] $P = .39$	3.35 3.68 (2.23) [−0.74, 8.11] $P = .10$	0.11 0.44 (2.23) [−3.98, 4.86] $P = .84$	0.51 0.84 (2.28) [−3.68, 5.36] $P = .71$
VAS Bond and Lader "calmness" (mm)	LSM CFB 0.33	−0.58	6.65 7.22 (2.55) [2.16, 12.29] $P < .01$*	−0.31 0.27 (2.58) [−4.85, 5.38] $P = .92$	−0.57 0.01 (2.55) [−5.07, 5.08] $P = 1.00$	3.6 4.17 (2.62) [−1.03, 9.37] $P = .11$	0.26 0.83 (2.55) [−4.24, 5.9] $P = .75$	0.58 1.15 (2.55) [−3.92, 6.22] $P = .65$	0.46 1.04 (2.56) [−4.06, 6.13] $P = .69$	2.59 3.16 (2.57) [−1.95, 8.28] $P = .22$	0.08 0.66 (2.61) [−4.52,5.84] $P = .80$	0.68 1.25 (2.61) [−3.94, 6.45] $P = .63$
VAS Bowdle "external perception"	LSM CFB $P = .56$	NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA
VAS Bowdle "internal perception"	LSM CFB 0.73	NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA
VAS Bowdle "feeling high"	LSM CFB 0.35	NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA	0.00 NA
Alpha-power Fz-Cz (Hz)	LSM CFB 0.99	0.06	0.01 0 [−0.97, 0.97] $P = 1.00$	0.09 0.15 [−0.94, 1.24] $P = .79$	−0.01 −0.33 [−1.38, 0.73] $P = .54$	0.08 0.13 [−0.81, 1.07] $P = .79$	0.16 0.32 [−0.64, 1.29] $P = .51$	0.06 0.57 [−0.45, 1.59] $P = .27$	0.05 −0.17 [−1.24,-.89] $P = .75$	0.15 0.99 [0, 1.99] $P = .05$	0.02 −0.15 [−1.19, 0.9] $P = .78$	0.01 −0.28 [−1.39, 0.82] $P = .61$
Alpha-power Pz-Oz (Hz)	LSM CFB 0.63	0.1	−0.06 −0.16 [−0.41, 0.10] $P = .23$	0.11 0.01 [−0.25, 0.27] $P = .94$	−0.01 −0.11 [−0.36, 0.15] $P = .41$	0.03 −0.06 [−0.32, 0.19] $P = .62$	0.14 0.04 [−0.22, 0.30] $P = .75$	0.12 0.02 [−0.23, 0.28] $P = .85$	0.11 0.01 [−0.25, 0.27] $P = .93$	−0.07 −0.17 [−0.43, 0.09] $P = .19$	−0.02 −0.12 [−0.37, 0.14] $P = .36$	0.24 0.14 [−0.11, 0.40] $P = .27$
Beta-power Fz-Cz (Hz)	LSM CFB 0.98	0.12	0.03 −0.09 [−0.31, 0.14] $P = .45$	0.08 −0.03 [−0.26, 0.19] $P = .78$	0.05 −0.06 [−0.28, 0.16] $P = .58$	0.04 −0.07 [−0.31, 0.15] $P = .51$	0.21 0.09 [−0.13, 0.32] $P = .40$	0.08 −0.03 [−0.25, 0.19] $P = .77$	0.12 0.01 [−0.21, 0.23] $P = .93$	0.11 −0.01 [−0.22, 0.22] $P = .97$	0.08 −0.04 [−0.26, 0.18] $P = .73$	0.07 −0.05 [−0.27, 0.17] $P = .67$

(Continues)

TABLE 3 (Continued)

Parameter	Overall	Placebo	ALKS 7119 3 mg	ALKS 7119 10 mg	ALKS 7119 25 mg	ALKS 7119 50 mg	ALKS 7119 75 mg	ALKS 7119 100 mg	ALKS 7119 125 mg	ALKS 7119 150 mg	ALKS 7119 175 mg	ALKS 7119 200 mg
Beta-power Pz-Oz (Hz)	**LSM CFB**	0.13	−0.03	0.14	0.08	0.13	0.17	0.25	0.14	−0.07	0.01	0.2
	0.21		−0.16 [−0.36, 0.04] $P = .11$	0.01 [−0.19, 0.21] $P = .91$	−0.04 [−0.24, 0.15] $P = .66$	0 [−0.20, 0,20] $P = 1.00$	0.04 [−0.16, 0.24] $P = .69$	0.12 [−0.07, 0.32] $P = .22$	0.02 [−0.18, 0.22] $P = .87$	−0.19 [−0.39, 0.01] $P = .06$	−0.12 [−0.32, 0.08] $P = .24$	0.07 [−0.12, 0.27] $P = .46$
Gamma-power Fz-Cz (Hz)	**LSM CFB**	0.1	0.05	0.09	0.06	0.01	0.27	0.1	0.12	0.14	0.16	0.21
	0.70		−0.04 [−0.25, 0.16] $P = .66$	−0.01 [−0.21, 0.21] $P = .95$	−0.04 [−0.04, 0.16] $P = .69$	−0.09 [−0.31, 0.11] $P = .39$	0.17 [−0.03, 0.37] $P = .10$	<−0.01 [−0.20, 0.21] $P = .99$	0.02 [−0.18, 0.22] $P = .85$	0.04 [−0.16, 0.24] $P = .71$	0.06 [−0.14, 0.26] $P = .57$	0.11 [−0.09, 0.31] $P = .29$
Gamma-power Pz-Oz (Hz)	**LSM CFB**	0.21	0.09	0.13	0.16	0.08	0.27	0.27	0.12	0.09	0.03	0.31
	0.67		−0.12 [−0.37, 0.13] $P = .35$	−0.08 [−0.33, 0.17] $P = .54$	−0.05 [−0.30, 0.20] $P = .70$	−0.12 [−0.38, 0.13] $P = .33$	0.06 [−0.19, 0.32] $P = .64$	0.06 [−0.19, 0.32] $P = .63$	−0.09 [−0.35, 0.17] $P = .49$	−0.12 [−0.37, 0.14] $P = .37$	−0.18 [−0.43, 0.08] $P = .17$	0.11 [−0.15, 0.36] $P = .40$
Delta-power Fz-Cz (Hz)	**LSM CFB**	0.1	0	0.05	−0.04	0	0.17	−0.03	0.09	−0.08	0.09	0.02
	0.68		−0.11 [−0.32, 0.10] $P = .32$	−0.05 [−0.26, 0.16] $P = .65$	−0.14 [−0.35, 0.07] $P = .19$	−0.10 [−0.31, 0.11] $P = .34$	0.06 [−0.15, 0.27] $P = .55$	−0.13 [−0.34, 0.08] $P = .22$	−0.01 [0.22, 0.20] $P = .90$	−0.19 [−0.41, 0.03] $P = .08$	−0.01 [−0.23, 0.20] $P = .91$	−0.08 [−0.29, 0.13] $P = .45$
Delta-power Pz-Oz (Hz)	**LSM CFB**	0.06	−0.12	0.07	0	0.08	0.1	0.08	0.22	0.02	−0.07	0.26
	0.28		−0.18 [−0.41, 0.05] $P = .12$	0.01 [−0.23, 0.24] $P = .96$	−0.06 [−0.31, 0.17] $P = .61$	0.02 [−0.21, 0.25] $P = .87$	0.04 [−0.21, 0.27] $P = .76$	0.02 [−0.21, 0.25] $P = .89$	0.15 [−0.08, 0.39] $P = .19$	−0.04 [−0.27, 0.19] $P = .71$	−0.13 [−0.36, 0.11] $P = .26$	0.20 [−0.04, 0.43] $P = .10$
Theta-power Fz-Cz (Hz)	**LSM CFB**	0.07	−0.06	0.02	−0.06	0.02	0.18	0.03	0.1	0	0.08	0.07
			−0.14 [−0.36, 0.09] $P = .23$	−0.05 [−0.28, 0.18] $P = .65$	−0.13 [−0.36, 0.09] $P = .24$	−0.05 [−0.28, 0.17] $P = .64$	0.11 [−0.12, 0.33] $P = .34$	−0.04 [−0.27, 0.19] $P = .72$	0.03 [−0.20, 0.25] $P = .82$	−0.07 [−0.31, 0.16] $P = .54$	−0.01 [−0.22, 0.23] $P = .97$	<−0.01 [−0.23, 0.22] $P = .97$
Theta-power Pz-Oz (Hz)	**LSM CFB**	0.07	−0.1	0.04	−0.04	0.08	0.12	0.14	0.15	0	−0.07	0.25
	0.35		−0.17 [−0.40, 0.06] $P = .15$	−0.03 [−0.26, 0.20] $P = .79$	−0.11 [−0.35, 0.12] $P = .33$	0.01 [−0.23, 0.24] $P = .95$	0.05 [−0.20, 0.28] $P = .69$	0.08 [−0.16, 0.30] $P = .56$	0.08 [−0.15, 0.31] $P = .50$	−0.07 [−0.31, 0.16] $P = .53$	−0.14 [−0.37, 0.10] $P = .25$	0.18 [−0.05, 0.41] $P = .13$

LSM, least squares mean; CFB, change from baseline; NA, not applicable.

[a]For log-transformed variables, no standard error was calculated.

*indicates statistical significance.

2.9 | Nomenclature of targets and ligands

Key protein targets and ligands in this article are hyperlinked to corresponding entries in http://www.guidetopharmacology.org, and are permanently archived in the Concise Guide to PHARMACOLOGY 2019/20.[32–35]

3 | RESULTS

3.1 | Subjects

One hundred healthy male subjects between 18 and 45 years of age were included (Table 1). All except one subject completed the study. This subject discontinued the study for personal reasons unrelated to the study and did not perform the 36-hour post dose assessments. Data obtained for this subject were included in the analysis (Table 1).

3.2 | Pharmacokinetics

Peak plasma concentrations were reached between 0.5 and 4 hours and mean $t_{1/2}$ ranged from 7.0 to 8.5 hours across the dose range of 3 mg to 200 mg. Systemic exposure to ALKS 7119 (C_{max}, AUC_{∞} and AUC_{last}) increased dose proportionally over the evaluated dose range (Table 2, Figure 2).

3.3 | NeuroCart® assessments

A statistically significant overall treatment effect towards increased pupil/iris ratio was observed for both left and right pupil/iris ratio measurements ($P < .01$ and $P < .01$, respectively) (Table 3). In general, this effect was observed with doses of 50 mg or higher (Table 3, Figure 3). Pupil/iris ratio increases were observed from approximately 2 hours post-dose, coinciding with the time when the peak concentrations of ALKS 7119 were observed (Figure 3).

On the other NeuroCart assessments, no statistically significant overall treatment effects were observed (Table 3). Individual treatment effects for VAS Bowdle could not be calculated as too many values under placebo and 3 mg were "0", making the data unsuitable for MMRM analysis.

Of note is that body sway was only performed in cohorts 1–6. After completion of these cohorts, the concern was raised that the test might evoke AEs of postural dizziness in some subjects, leading to the decision not to perform this test in the remaining cohorts.

3.3.1 | Neuro-endocrine hormones

A statistically significant treatment effect for serum cortisol ($P = .02$) and serum prolactin ($P < .01$) levels was observed at 2 hours post-dose, approximately the t_{max} of ALKS 7119, from dose level 50 mg and higher (Table 4, Figures 4 and 5). Growth hormone demonstrated a similar pattern as cortisol and prolactin, but this was not tested for statistical significance due to many values being below the limit of quantification (Figure 6).

3.4 | Safety

Treatment emergent adverse events (TEAEs) were reported in 51 (64%) subjects in the ALKS 7119 group and eight (40%) subjects in the placebo group. The most common reported TEAEs were nausea, presyncope, somnolence, dizziness and vomiting. Nausea and presyncope followed a dose proportional trend with greater incidence in the higher dosing groups and occurrence at or around t_{max} (Table 5). Most TEAEs were of mild severity and none were considered severe. There were no serious adverse events in the study. In general, there were no clinically meaningful findings or trends in changes from baseline for the safety laboratory parameters, urinalysis

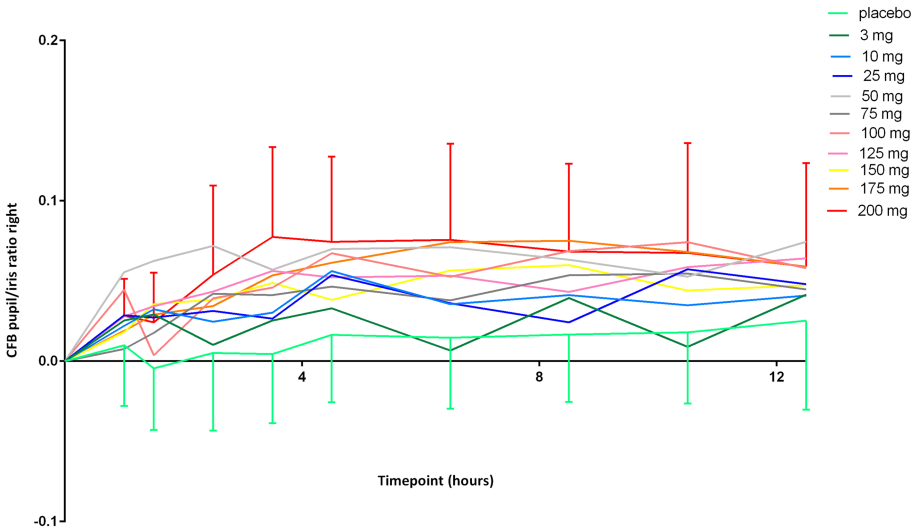


FIGURE 3 Mean (SD) CFB right pupil/iris ratio

TABLE 4 Neuro-endocrine hormone levels compared to placebo at 2 hours post-dose. Estimate of difference (95% CI), *P*-value

	Placebo LSM CFB	ALKS 7119 3 mg	ALKS 7119 10 mg	ALKS 7119 25 mg	ALKS 7119 50 mg	ALKS 7119 75 mg	ALKS 7119 100 mg	ALKS 7119 125 mg	ALKS 7119 150 mg	ALKS 7119 175 mg	ALKS 7119 200 mg
Cortisol (ng/ml) (% change)	−25.7	−22.9	8.9	−20.8	12.7	36.5	70.3	52.6	84.4	53.6	64.7
		−3.6 (−31.1, 34.9) P = .83	−31.8 (−51.3; −4.5) P = .03*	−6.2 (−32.9; 31.3) P = .71	−34.1 (−52.9; −7.8) P = .02*	−45.5 (−61.1; −23.8) P < .01*	−56.4 (−68.8; −38.9) P < .01*	−51.3 (−65.2; −31.8) P < .01*	−59.7 (−71.2; −43.6) P < .01*	−51.6 (−65.4; −32.3) P < .01*	−54.9 (−67.8; −36.9) P < .01*
Prolactin (ng/ml) (% change)	−6.7	−3.6	−15.3	8.0	66.0	63.4	72.0	28.0	106.9	81.7	111.3
		−3.2 (−26.9; 28.3) P = .82	10.1 (−16.9; 45.9) P = .50	−13.6 (−34.8; 14.5) P = .31	−43.8 (−57.6; −25.5) P < .01*	−42.9 (−56.9; −24.3) P < .01*	−45.8 (−59.1; −28.1) P < .01*	−27.1 (−45.1; −3.3) P = .03*	−54.9 (−66.0; −40.2) P < .01*	−48.7 (−61.3; −32.0) P < .01*	−55.8 (−66.7; −41.5) P < .01*

LSM, least squares mean; CFB, change from baseline.
*indicates statistical significance.

or vital signs. For systolic blood pressure, a general trend of a mean increase from baseline was observed at all time points, except at 1.5 hours post-dose, where most of the ALKS 7119 treatment groups, especially the higher dose groups, demonstrated decreases from baseline (Figure 7). On diastolic blood pressure, a trend towards decrease was observed for all treatment groups, which was largest at 1.5 hours post-dose (Figure 8).

The incidence of orthostatic hypotension was assessed as this could be an underlying cause of AEs of postural dizziness that were observed after the body sway test in the first six cohorts. The incidence of orthostatic hypotension was comparable between the placebo group (15%) and all ALKS 7119 dose groups (17.5%). Therefore, there did not seem to be a dose-dependent effect on orthostatic hypotension, with the possible exception of the 200 mg dosing group in which three (37.5%) subjects met the criteria for orthostatic hypotension.

Differences between ALKS 7119 treatment groups and placebo were observed on QT interval corrected according to Fridericia (QTcF). In 8.8% of subjects in the ALKS 7119 treatment groups, QTcF increases from baseline of 30–60 ms were observed, with the largest differences at 3, 6 and 8 hours post-dose, compared to none in the placebo group (Figure 9). Although there were no consistent dose-dependent findings, these changes were only seen at dose levels of 50 mg or higher. The highest individual QTcF value measured was 475 ms, which represented an increase of 33 ms compared to baseline in a subject treated with 200 mg of ALKS 7119. There were no TEAEs related to the changes in QTcF. No clinically meaningful trends were observed on the other ECG parameters (PR interval, QRS duration and RR interval).

4 | DISCUSSION

From dose levels of 50 mg and higher, ALKS 7119 significantly increased pupil size and dose-dependently increased serum levels of cortisol and prolactin at 2 hours post-dose, coinciding with the t_{max} of ALKS 7119. No statistically significant overall treatment effects on the other NeuroCart tests were observed. This profile is most compatible with SERT engagement and suggestive of BBB penetration.

Therapeutic dosages of SSRIs are found to increase pupil size,[6] and to not affect other NeuroCart parameters.[3] It is hypothesized that SSRIs exert their effect on pupil size via serotonergic CNS pathways in the locus coeruleus,[36] but it cannot be completely ruled out that the pupil effects are peripherally mediated as serotonin (5-HT_7 subtype) receptors are also present on the sphincter of the pupil.[37] Binding of serotonin to these receptors leads to pupillary sphincter relaxation and thereby mydriasis.[38] The neuro-endocrine findings also point in the direction of SERT binding, as other serotonergic compounds such as fenfluramine and escitalopram are known to respectively increase cortisol and cortisol and prolactin levels in healthy volunteers.[9,11] It could be argued that the endocrine effects can be caused by pituitary stimulation, outside the BBB and CNS. However, the effects of ALKS 7119 involved several hormones concomitantly.

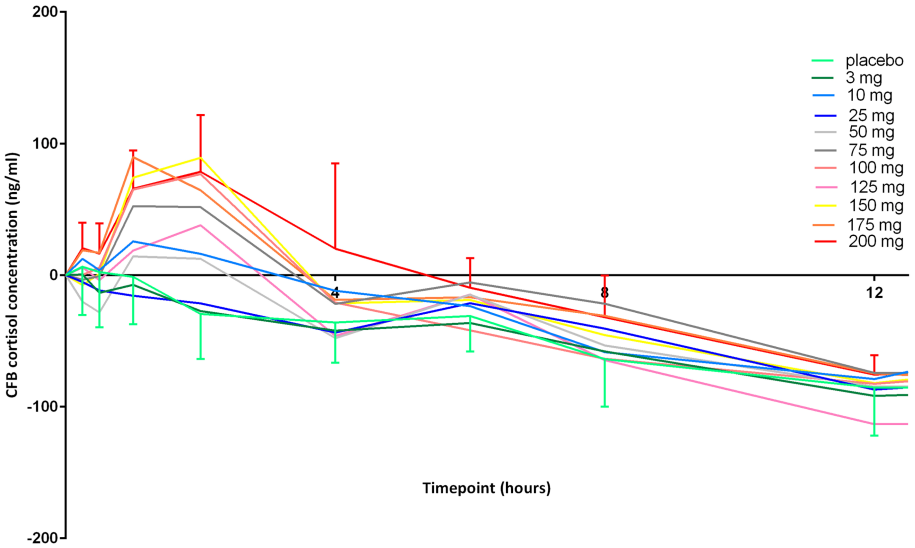


FIGURE 4 Mean (SD) CFB cortisol concentration (ng/ml)

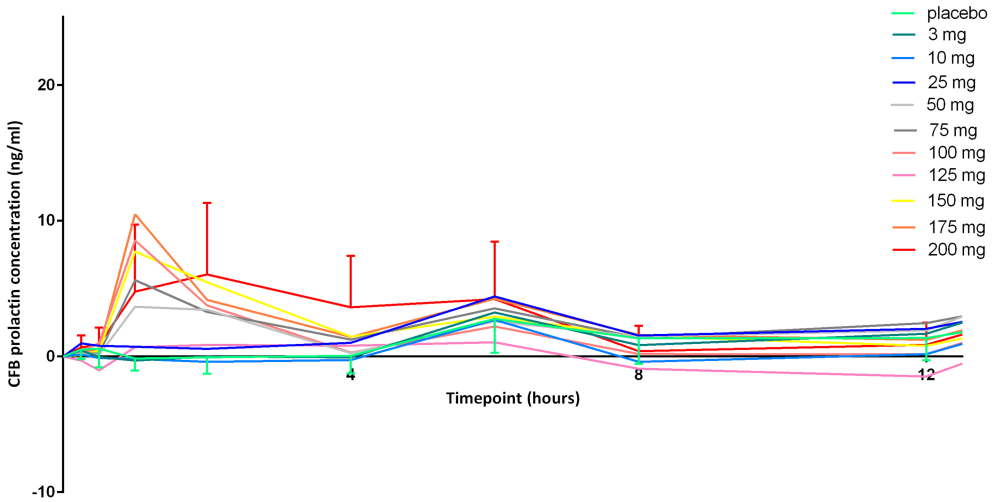


FIGURE 5 Mean (SD) CFB prolactin concentration (ng/ml)

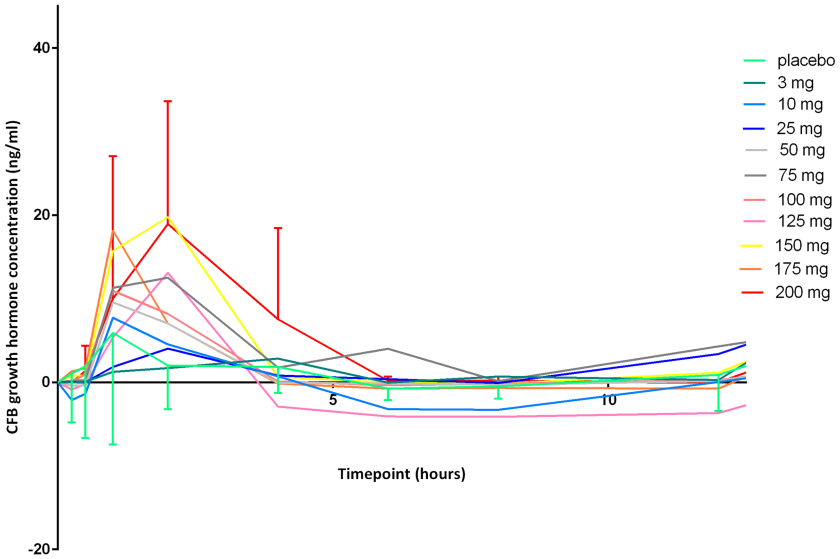


FIGURE 6 Mean (SD) CFB growth hormone concentration (ng/ml)

TABLE 5 Incidence of treatment emergent adverse events per treatment group

TEAE	Placebo ($n = 20$)	ALKS 7119 3 mg ($n = 8$)	ALKS 7119 10 mg ($n = 8$)	ALKS 7119 25 mg ($n = 8$)	ALKS 7119 50 mg ($n = 8$)	ALKS 7119 75 mg ($n = 8$)	ALKS 7119 100 mg ($n = 8$)	ALKS 7119 125 mg ($n = 8$)	ALKS 7119 150 mg ($n = 8$)	ALKS 7119 175 mg ($n = 8$)	ALKS 7119 200 mg ($n = 8$)
Nausea	0	0	0	0	1	3	3	2	5	5	3
Vomiting	0	0	0	0	0	2	1	0	1	0	0
Fatigue	2	3	1	2	0	0	0	0	1	0	1
Headache	6	0	1	0	2	3	3	2	2	0	3
Presyncope	0	0	1	1	1	3	1	0	3	4	1
Somnolence	2	0	1	0	2	1	2	0	1	1	1
Dizziness	0	0	0	0	0	0	0	1	1	0	2
Dizziness postural	0	0	0	0	0	0	2	0	0	0	0
Oropharyngeal pain	0	0	0	0	0	0	0	0	0	0	0
Pruritus	0	0	0	0	0	0	0	1	0	0	2

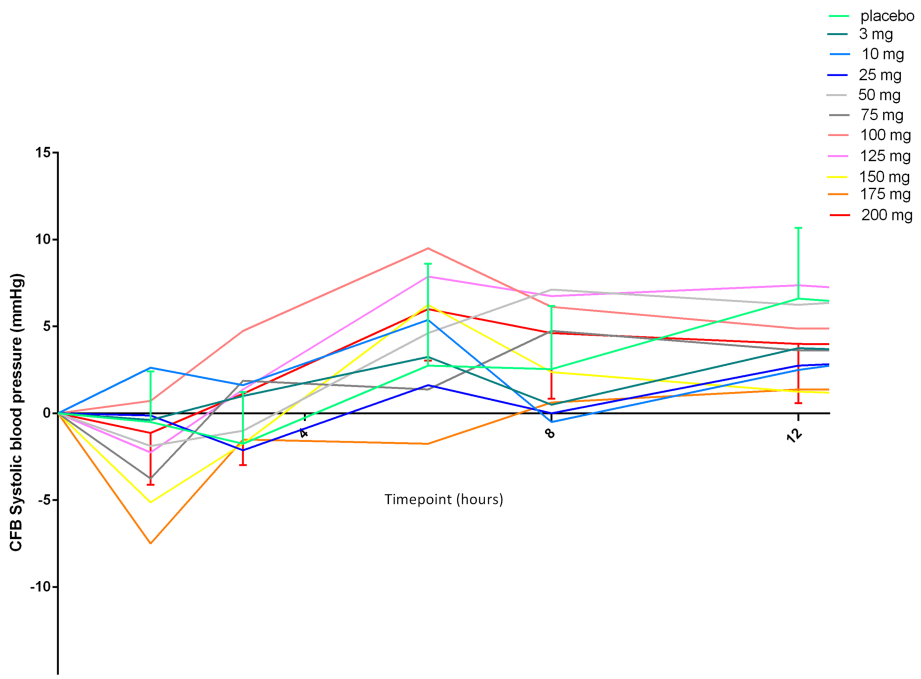


FIGURE 7 Mean (±Standard Error) CFB systolic blood pressure (mmHg)

This is difficult to attribute to a simultaneous effect on different cell populations in the pituitary, which are highly specialized and pharmacologically diverse. A hypothalamic site of action is functionally more plausible if several hormones simultaneously respond to a CNS-active compound, because of the integrative role of the hypothalamus. Since the hypothalamus is the most important autonomic command centre that governs the concerted activity of many autonomic and neuroendocrine processes, the same argument could also be used for a central (hypothalamic) localization of ALKS 7119-induced pupillary dilation.

Of note, no overall treatment effects of ALKS 7119 on pEEG were observed, whereas in a scientific review, it was reported that 100% of studies into the effects of SSRIs on pEEG reported an increase of total EEG power with low-dose SSRIs, whilst high-dose SSRIs increased delta and theta power in 33% of the studies.[6] These apparent complex dose–response relations for SSRIs seem to contrast with the lack of effects of ALKS 7119. This might reflect methodological differences in pEEG recording (e.g., different number of leads and analysis methods),[6] but also limitations of the literature review (e.g., due to publication bias).

In theory, the effects on pupil size and neuroendocrine stimulation can also be caused by NMDA receptor antagonism.[7] However, ALKS 7119 did not match the complete effect profile of NMDA receptor activation, which would also include decrease of saccadic

FIGURE 8 Mean (±Standard Error) CFB diastolic blood pressure (mmHg)

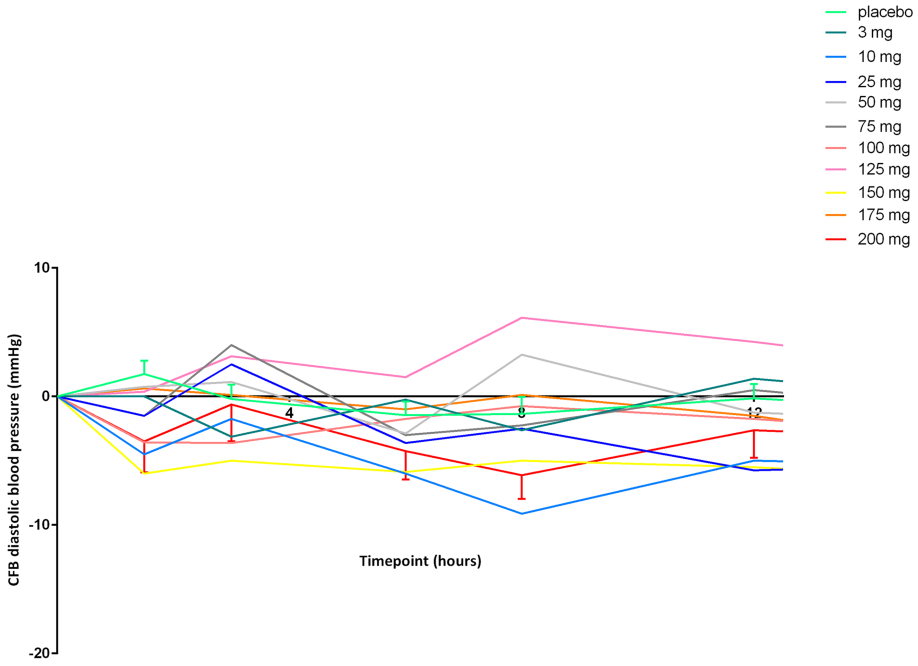
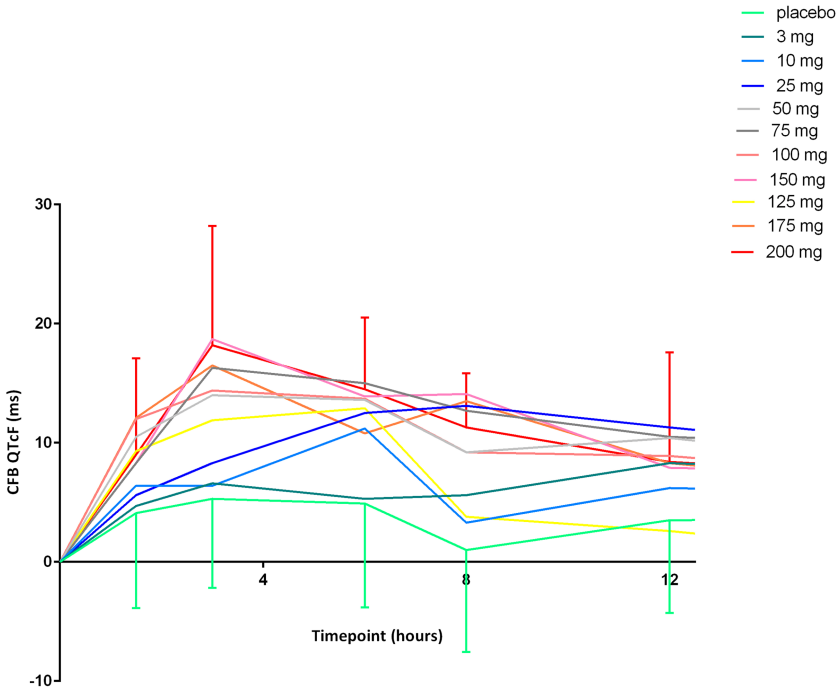


FIGURE 9 Mean (SD) CFB QTcF



peak velocity and adaptive tracking and a variety of other neurophysiological, behavioural and subjective sedative effects.[3,7,39] It is also less likely that the effects were anti-glutamatergic rather than serotonergic, because ALKS 7119 shows a 200-fold lower affinity for the NMDA-receptor than for SERT.

The acute PD effects of mild selective sigma non-opioid intracellular receptor 1 modulation are unknown, so no PD biomarkers for this receptor could be included in our study. However, it is unlikely that the observed effects of ALKS 7119 were caused by sigma non-opioid intracellular receptor 1 modulation since ALKS 7119's affinity for this receptor is 900-fold lower than for the SERT and there was no clear indication for NMDA or even μ receptor activation, with affinities, respectively, 200 and 17-fold lower than for the SERT.

A wide dose range was explored in this study, but it was not possible to escalate the dose of ALKS 7119 to levels expected to influence NMDA receptors or sigma non-opioid intracellular receptor 1. Initially, the maximum planned dose was 100 mg based on the NOAEL in rats. This was increased based on the results of an

interim analysis demonstrating linear PK with dose proportional increase in exposure and no safety or tolerability findings precluding further dose escalation to 200 mg. In the higher dose level groups, a dose-dependent trend towards a decrease in supine systolic and diastolic blood pressure at t_{max} was observed, which most likely was the result of activation of the $\alpha_{1A,B}$ adrenoceptors. SERT inhibition might explain the occurrence of nausea and presyncope at higher dosages.

The effects of ALKS 7119 on QTcF duration were unexpected as no effects on QT intervals were observed in preclinical cardiovascular toxicity studies in dogs up to the highest given dose of 10 mg/kg corresponding to a HED of 324 mg. Moreover, in vitro hERG channel testing showed no effects up to high concentrations (IC_{50} of 191.1 μM or 54348.8 ng/mL). The mechanism underlying the QTcF prolongation observed in this study remains, therefore, unknown. QTcF prolongation occurred in only a small number of subjects in this study, which is reminiscent of the mild prolongations that are reported for most SSRIs.[40]

Taken together, this study demonstrated a CNS effect pattern for ALKS 7119 that is in line with the drug's pharmacological binding profile. These results illustrate how biomarkers, such as the NeuroCart and serum neuro-endocrine hormone levels, can provide important information in early phase drug development to obtain a comprehensive overview of a new compound's clinical pharmacological profile. This knowledge can be used to make rational decisions in early phase clinical trials on dose escalation steps and on the further development of a compound as suggested by the conceptual framework of QBDD.[2]

ACKNOWLEDGEMENT

This study was sponsored by Alkermes, Inc. Alkermes is not further pursuing development activities for the ALKS 7119 program.

COMPETING INTERESTS

R.L.P., L.S., J.M., R.L., L.v.M., and D.R. are all current or former employees of Alkermes, Inc. and may own stock or stock options. The other authors have nothing to declare.

CONTRIBUTORS

F.D., R.Z., P.S., R.L.P., L.S., J.M., M.d.K., R.L., L.v.M., D.R., J.v.G. contributed to the study design. F.D., R.Z., P.S., and J.v.G. contributed to the acquisition of data. All authors contributed to analysis and interpretation of data, drafting and review of the manuscript.

DATA AVAILABILITY STATEMENT

The data collected in this study are proprietary to Alkermes, Inc. Alkermes, Inc. is committed to public sharing of data in accordance with applicable regulations and laws.

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REFERENCES

1. FDA. Guidance for Industry: Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers. https://www.fda.gov/media/72309/download Published 2005. Accessed January 28, 2021.
2. Cohen AF, Burggraaf J, van Gerven JM, Moerland M, Groeneveld GJ. The use of biomarkers in human pharmacology (Phase I) studies. *Annu Rev Pharmacol Toxicol*. 2015;55(1):55-74. doi:10.1146/annurev-pharmtox-011613-135918
3. Groeneveld GJ, Hay JL, Van Gerven JM. Measuring blood–brain barrier penetration using the NeuroCart, a CNS test battery. *Drug Discov Today Technol*. 2016;20:27-34. doi:10.1016/j.ddtec.2016.07.004
4. van Gerven JMAJ. Functional measurements of central nervous system drug effects in early human drug development. In: Nomikos G, Feltner D, eds. *Translational Medicine in CNS Drug Development*. 1st ed. London: Academic Press; 2019:39-62.
5. Murray RB, Adler MW, Korczyn AD. The pupillary effects of opioids. *Life Sci*. 1983;33(6):495-509. doi:10.1016/0024-3205(83)90123-6
6. Dumont GJ, de Visser SJ, Cohen AF, van Gerven JM. Biomarkers for the effects of selective serotonin reuptake inhibitors (SSRIs) in healthy subjects. *Br J Clin Pharmacol*. 2005;59(5):495-510. doi:10.1111/j.1365-2125.2005.02342.x
7. Kleinloog D. Drug-induced psychomimetic effects as a model for psychosis [PhD thesis]. Leiden: Leiden University; 2014. https://chdr.nl/library/drug-induced-psychomimetic-effects-as-a-model-for-psychosis/download. Accessed January 12, 2022.
8. Okkerse P, Alvarez-Jimenez R, Hay JL, et al. No evidence of potentiation of buprenorphine by milnacipran in healthy subjects using a nociceptive test battery. *Eur J Pain*. 2017;21(3):494-506. doi:10.1002/ejp.943
9. Hawken ER, Owen JA, Hudson RW, Delva NJ. Specific effects of escitalopram on neuroendocrine response. *Psychopharmacology (Berl)*. 2009;207(1):27-34. doi:10.1007/s00213-009-1633-1
10. Lewis DA, Sherman BM. Serotonergic regulation of prolactin and growth hormone secretion in man. *Acta Endocrinol*. 1985;110(2):152-157.
11. Recourt K, van der Aart J, Jacobs G, et al. Characterisation of the pharmacodynamic effects of the P2X7 receptor antagonist JNJ-54175446 using an oral dexamphetamine challenge model in healthy males in a randomised, double-blind, placebo-controlled, multiple ascending dose trial. *J Psychopharmacol*. 2020;34(9):1030-1042. doi:10.1177/0269881120914206
12. Hysek CM, Brugger R, Simmler LD, et al. Effects of the α_2-adrenergic agonist clonidine on the pharmacodynamics and pharmacokinetics of 3,4-methylenedioxymethamphetamine in healthy volunteers. *J Pharmacol Exp Ther*. 2012;340(2):286-294. doi:10.1124/jpet.111.188425
13. Jepma M, Te Beek E, Wagenmakers E-J, Van Gerven J, Nieuwenhuis S. The role of the noradrenergic system in the exploration-exploitation trade-off: a pharmacological study. *Front Hum Neurosci*. 2010;4:1-3. doi:10.3389/fnhum.2010.00170
14. Okkerse P, Hay JL, Sitsen E, et al. Pharmacokinetics and pharmacodynamics of intrathecally administered Xen2174, a synthetic conopeptide with norepinephrine reuptake inhibitor and analgesic properties. *Br J Clin Pharmacol*. 2017;83(4):751-763. doi:10.1111/bcp.13176
15. van Steveninck AL, Gieschke R, Schoemaker RC, et al. Pharmacokinetic and pharmacodynamic interactions of bretazenil and diazepam with alcohol. *Br J Clin Pharmacol*. 1996;41(6):565-573. doi:10.1046/j.1365-2125.1996.38514.x
16. van Steveninck AL, van Berckel BN, Schoemaker RC, Breimer DD, van Gerven JM, Cohen AF. The sensitivity of pharmacodynamic tests for the central nervous system effects of drugs on the effects of sleep deprivation. *J Psychopharmacol*. 1999;13(1):10-17. doi:10.1177/026988119901300102

17. van Steveninck AL, Schoemaker HC, Pieters MS, Kroon R, Breimer DD, Cohen AF. A comparison of the sensitivities of adaptive tracking, eye movement analysis and visual analog lines to the effects of incremental doses of temazepam in healthy volunteers. *Clin Pharmacol Ther*. 1991;50(2):172-180.
18. de Haas SL, de Visser SJ, van der Post JP, et al. Pharmacodynamic and pharmacokinetic effects of TPA023, a GABAA α2,3 subtype-selective agonist, compared to lorazepam and placebo in healthy volunteers. *J Psychopharmacol*. 2006;21(4):374-383. doi:10.1177/0269881106072343
19. Baloh RW, Sills AW, Kumley WE, Honrubia V. Quantitative measurement of saccade amplitude, duration, and velocity. *Neurology*. 1975;25(11):1065-1070. doi:10.1212/wnl.25.11.1065
20. Bittencourt PR, Wade P, Smith AT, Richens A. Benzodiazepines impair smooth pursuit eye movements. *Br J Clin Pharmacol*. 1983;15(2):259-262. doi:10.1111/j.1365-2125.1983.tb01495.x
21. Borland RG, Nicholson AN. Comparison of the residual effects of two benzodiazepines (nitrazepam and flurazepam hydrochloride) and pentobarbitone sodium on human performance. *Br J Clin Pharmacol*. 1975;2(1):9-17.
22. de Haas SL, de Visser SJ, van der Post JP, et al. Pharmacodynamic and pharmacokinetic effects of MK-0343, a GABA(A) alpha2,3 subtype selective agonist, compared to lorazepam and placebo in healthy male volunteers. *J Psychopharmacol*. 2008;22(1):24-32. doi:10.1177/0269881107082108
23. Gijsman HJ, Van Gerven JM, Tieleman MC, et al. Pharmacokinetic and pharmacodynamic profile of oral and intravenous meta-chlorophenylpiperazine in healthy volunteers. *J Clin Psychopharmacol*. 1998;18(4):289-295. doi:10.1097/00004714-199808000-00007
24. Cohen AF, Ashby L, Crowley D, Land G, Peck AW, Miller AA. Lamotrigine (BW430C), a potential anticonvulsant. Effects on the central nervous system in comparison with phenytoin and diazepam. *Br J Clin Pharmacol*. 1985;20(6):619-629. doi:10.1111/j.1365-2125.1985.tb05120.x
25. van Steveninck AL, Wallnofer AE, Schoemaker RC, et al. A study of the effects of long-term use on individual sensitivity to temazepam and lorazepam in a clinical population. *Br J Clin Pharmacol*. 1997;44(3):267-275. doi:10.1046/j.1365-2125.1997.t01-1-00580.x
26. Wright BM. A simple mechanical ataxia-meter. *J Physiol*. 1971;218(Suppl):27p-28p.
27. Twa MD, Bailey MD, Hayes J, Bullimore M. Estimation of pupil size by digital photography. *J Cataract Refract Surg*. 2004;30(2):381-389. doi:10.1016/S0886-3350(03)00619-9
28. Jobert M, Wilson FJ, Ruigt GS, Brunovsky M, Prichep LS, Drinkenburg WH. Guidelines for the recording and evaluation of pharmaco-EEG data in man: the International Pharmaco-EEG Society (IPEG). *Neuropsychobiology*. 2012;66(4):201-220. doi:10.1159/000343478
29. Norris H. The action of sedatives on brain stem oculomotor systems in man. *Neuropharmacology*. 1971;10(21):181-191.
30. Bond A, Lader M. The use of analogue scales in rating subjective feelings. *Br J Med Psychol*. 1974;47(3):211-218. doi:10.1111/j.2044-8341.1974.tb02285.x
31. Bowdle AT, Radant AD, Cowley DS, Kharasch ED, Strassman RJ, Roy-Byrne PP. Psychedelic effects of ketamine in healthy volunteers relationship to steady-state plasma concentrations. *Anesthesiology*. 1998;88(1):82-88. doi:10.1097/00000542-199801000-00015
32. Alexander SPH, Christopoulos A, Davenport AP, et al. THE CONCISE GUIDE TO PHARMACOLOGY 2019/20: G protein-coupled receptors. *Br J Pharmacol*. 2019;176(Suppl 1):S21-S141. doi:10.1111/bph.14748
33. Alexander SPH, Kelly E, Mathie A, et al. THE CONCISE GUIDE TO PHARMACOLOGY 2019/20: Transporters. *Br J Pharmacol*. 2019;176(Suppl 1):S397-S493. doi:10.1111/bph.14753
34. Alexander SPH, Cidlowski JA, Kelly E, et al. THE CONCISE GUIDE TO PHARMACOLOGY 2019/20: Introduction and other protein targets. *Br J Pharmacol*. 2019;176(Suppl 1):S1-S20. doi:10.1111/bph.14750
35. Alexander SPH, Mathie A, Peters JA, et al. THE CONCISE GUIDE TO PHARMACOLOGY 2019/20: Ion channels. *Br J Pharmacol*. 2019;176(Suppl 1):S142-S228. doi:10.1111/bph.14749
36. Yu Y, Ramage AG, Koss MC. Pharmacological studies of 8-OH-DPAT-induced pupillary dilation in anesthetized rats. *Eur J Pharmacol*. 2004;489(3):207-213. doi:10.1016/j.ejphar.2004.03.007
37. Costagliola C, Parmeggiani F, Semeraro F, Sebastiani A. Selective serotonin reuptake inhibitors: a review of its effects on intraocular pressure. *Curr Neuropharmacol*. 2008;6(4):293-310. doi:10.2174/157015908787386104
38. Gündüz GU, Parmak Yener N, Kılınçel O, Gündüz C. Effects of selective serotonin reuptake inhibitors on intraocular pressure and anterior segment parameters in open angle eyes. *Cutan Ocul Toxicol*. 2018;37(1):36-40. doi:10.1080/15569527.2017.1330270
39. Krystal JH, Karper LP, Seibyl JP, et al. Subanesthetic effects of the noncompetitive NMDA antagonist, ketamine, in humans: psychotomimetic, perceptual, cognitive, and neuroendocrine responses. *Arch Gen Psychiatry*. 1994;51(3):199-214. doi:10.1001/archpsyc.1994.03950030035004
40. Beach SR, Kostis WJ, Celano CM, et al. Meta-analysis of selective serotonin reuptake inhibitor-associated QTc prolongation. *J Clin Psychiatry*. 2014;75(5):e441-e449. doi:10.4088/JCP.13r08672

How to cite this article: Dijkstra FM, Zuiker RGJA, Siebenga PS, et al. Pharmacological profile of ALKS 7119, an investigational compound evaluated for the treatment of neuropsychiatric disorders, in healthy volunteers. *Br J Clin Pharmacol*. 2022;88(6):2909-2925. doi:10.1111/bcp.15229