Influence of Δ^9-tetrahydrocannabinol (THC) on oxycodone induced ventilatory depression in healthy volunteers

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PROTOCOL SIGNATURE SHEET

Name	Signature	Date
Head of Research Department:	**Prof. Dr. Albert Dahan**	
Principal investigator:	**Dr. Monique van Velzen**	

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LIST OF ABBREVIATIONS AND RELEVANT DEFINITIONS

ABR	General Assessment and Registration form (ABR form), the application form that is required for submission to the accredited Ethics Committee; in Dutch: Algemeen Beoordelings- en Registratieformulier (ABR-formulier)
AE	Adverse Event
AR	Adverse Reaction
CA	Competent Authority
CCMO	Central Committee on Research Involving Human Subjects; in Dutch: Centrale Commissie Mensgebonden Onderzoek
CV	Curriculum Vitae
DSMB	Data Safety Monitoring Board
EU	European Union
EudraCT	European drug regulatory affairs Clinical Trials
ET	End-tidal
GCP	Good Clinical Practice
GDPR	General Data Protection Regulation; in Dutch: Algemene Verordening Gegevensbescherming (AVG)
HCVR	Hypercapnic ventilatory response
IB	Investigator's Brochure
IC	Informed Consent
IMP	Investigational Medicinal Product
IMPD	Investigational Medicinal Product Dossier
IR	Immediate release
METC	Medical research ethics committee (MREC); in Dutch: medisch-ethische toetsingscommissie (METC)
(S)AE	(Serious) Adverse Event
SPC	Summary of Product Characteristics; in Dutch: officiële productinformatie IB1-tekst
SpO_2	Oxygen saturation
SUSAR	Suspected Unexpected Serious Adverse Reaction
THC	Δ^9-tetrahydrocannabinol
UAVG	Dutch Act on Implementation of the General Data Protection Regulation; in Dutch: Uitvoeringswet AVG
WMO	Medical Research Involving Human Subjects Act; in Dutch: Wet Medisch-wetenschappelijk Onderzoek met Mensen

SUMMARY

Rationale: Opioid misuse and abuse are common problems in the Western world. The rate of unintentional drug overdose is rapidly increasing, not only in the Unites States but also in the Netherlands. Additionally, it is well known that opioids are often used (and abused) in combination with other legal or illicit substances, for example cannabis, including medicinal (*i.e.* doctor prescribed) cannabis. A major opioid-induced adverse effect is respiratory depression and there are no data that show how oxycodone interacts with cannabis on the ventilatory control system. An appreciable effect is possible given the sedative effects of cannabis. Moreover, we previously showed that combining even a low dose of oxycodone (20 mg) with ethanol increased the likelihood of an apneic event (van der Schrier et al. Anesthesiology 2017; 102: 115-122). Because of this side effect and also due to the rising number of addicted chronic opioid users, there is an increasing imminent societal, political and medical interest in advancing research on opioids, opioid-drug interaction and alternatives for the treatment of various chronic illnesses and chronic pain.

Hypothesis: We hypothesize that cannabis will amplify the ventilatory depressant effect of oxycodone (primary end-point).

Objective: The objective of the study is to quantify the interactive effect of Δ^9-tetrahydrocannabinol (THC) and oxycodone on ventilatory control.

Study design: Double blind, randomized cross-over, placebo-controlled design.

Study population: Healthy human volunteers between the age of 18 and 45 years old.

Intervention:

Visit A: placebo capsule at t = 0 min + Bedrocan (22.4 mg THC) at t = 90 and 270 min;
Visit B: oxycodone 20 mg at t = 0 min + Bedrocan (22.4 mg THC) at t = 90 and 270 min.

Main study parameters/endpoints:

Primary endpoint: The effect of inhaled THC on ventilation at an end-tidal PCO_2 = 55 mmHg without and with concomitant intake of 20 mg oxycodone immediate release (IR) capsule in healthy volunteers 120 min after oxycodone intake.

Secondary endpoints: (1) Outcome of Bowdle and Bond & Lader questionnaires; (2) Level of sedation; (3) Hemodynamics; (4) Cognition (p-deletion test); (5) Pain Pressure Threshold; (6) slope of the hypercapnic ventilatory response; (7) plasma concentrations of THC, 11-OH-THC and oxycodone; a secondary analysis will be performed on the pharmacokinetic and pharmacodynamic data (PKPD modeling).

Nature and extent of the burden and risks associated with participation, benefit and group relatedness: The burden/risks for the volunteers are the occurrence of side effects from cannabis or oxycodone. Oxycodone may induce opioid-typical side effects such as respiratory depression (research endpoint), nausea/vomiting and sedation, of which nausea and vomiting is most burdensome. Other side effects, which are also cannabis related, include drug high, euphoria/dysphoria, dizziness/lightheadedness. During the study, we will closely monitor the subjects and treat side effects, most importantly nausea by administration of an antiemetic.

1. INTRODUCTION AND RATIONALE

Opioids are commonly prescribed for moderate to severe pain. While initially intended for acute and cancer pain, opioids are currently frequently considered and prescribed in chronic noncancer pain. A consequence of this behavior is the increase in opioid misuse and abuse. The rate of unintentional drug overdose is rapidly increasing, not only in the Unites States but also in the Netherlands. A potential lethal consequence of opioid overdose is opioid-induced respiratory depression. Additionally, it is well known that opioids are often used (and abused) in combination with other legal or illicit substances, for example alcohol, benzodiazepines or cannabis, including medicinal (*i.e.* doctor prescribed) cannabis. We previously showed that combining ethanol with oxycodone (20 mg) increases respiratory depression, indicative of a dangerous alcohol-opioid combination (van der Schrier et al. Anesthesiology 2017; 102: 115-122). There are no data on the interaction between oxycodone and cannabis on the ventilatory control system. In our opinion, there is the false premise that cannabis has no effect on breathing. Apart from a direct effect on receptors in the brainstem, the sedative effects of cannabis may compromise breathing. Because of this side effect and also due to the rising number of addicted chronic opioid users, there is an increasing imminent societal, political and medical interest in furthering research on opioids, opioid-drug interaction and alternatives for the treatment of various chronic illnesses and chronic pain. Additionally, we expect that many patients at home using oxycodone, also use (coffeeshop) cannabis.

In this study we will measure the effect of Bedrocan which contains primarily Δ^9-tetrahydrocannabinol (THC) and a minute quantity cannabidiol (CBD), on ventilation at 55 mmHg end-tidal PCO_2 in 20 healthy volunteers and the combination of THC and 20 mg oral oxycodone immediate release capsules. Primary endpoint is the effect of inhaled THC on ventilation at end-tidal PCO_2 = 55 mmHg without and with concomitant intake of 20 mg oxycodone immediate release (IR) capsule in healthy volunteers 120 min after oxycodone intake. In this study we will use the Volcano cannabis vaporizer to vaporize 100 mg Bedrocan into a 6 L balloon. Volunteers will inhale the cannabis vapor from the balloon.

2. OBJECTIVES

Primary objective: To measure the effect of inhaled THC (100 mg Bedrocan) on ventilation at isohypercapnia (end-tidal PCO_2 = 55 mmHg) without and with concomitant intake of 20 mg oxycodone immediate release (IR) capsule in healthy volunteers 90 min after oxycodone intake.

Secondary objective: To measure the effect of inhaled THC (100 mg Bedrocan) without and with concomitant intake of 20 mg oxycodone immediate release (IR) on
- occurrence of apneic events;
- occurrence of desaturation events;
- slope of the hypercapnic ventilatory response;
- baseline ventilation;
- psychomimetic side effects as measured by Bowdle and Bond & Lader questionnaires;
- cognition (p-deletion test)
- pain pressure threshold
- plasma concentrations of THC and oxycodone;
- blood pressure and heart rate.

3. STUDY DESIGN

3.1 General study design

The design of the study is randomized, placebo-controlled crossover. Each subject will be studied twice, on one occasion he or she will receive THC and a placebo opioid capsule, and on the other occasion THC and an oxycodone capsule. The visits to the lab will be randomized, the washout period between the two occasions is at least 10 days. Subjects will be evaluated by a physician at screening and only healthy volunteers, aged 18-45 years, with a body mass index < 30 $kg.m^{-2}$, are eligible to participate in the study.

Upon arrival in the laboratory (K5-120), we will perform a urinary drug test and breath alcohol test. When these tests are positive, the subject is excluded from further participation. A venous and an arterial line will be placed. The venous line is used for fluid administration (NaCl/Glucose 50-100 ml/h), the arterial line is for blood sample drawing.

Next, the first hypercapnic ventilatory responses (HCVR) will be obtained (t = -30 min). This is the pre-drug baseline measurement. At t = 0, the subjects will receive 20 mg oxycodone immediate release or placebo. Next, we will obtain HCVRs at 1-hour intervals until 6 hours after oxycodone/placebo intake. At t = 1.5 h and at t = 4.5 h the subject will inhale 100 mg Bedrocan. Breathing will be measured using the “dynamic end-tidal forcing” (DEF) system. At specific time points we will draw 10 mL blood for measurement of drug concentrations. After 8 hours of measurement, we will assess whether the HCVR is still depressed. If so, we will take another measurement at t = 9 h and reassess the HCVR. If still depressed, a last response will be obtained at t = 10 h. If necessary, the subject will stay overnight in the hospital. This will be decided by the physician-investigator. For example, the subject may still be sedated.

3.2 The hypercapnic ventilatory response

To obtain the HCVR curve, the subject will rebreathe a gas mixture from a 6 L rebreathing balloon bag containing 7% carbon dioxide in 93% oxygen. The slope of the response and ventilation at 55 mmHg will be used in the analysis.

3.3 Drug intake

3.3.1 Oxycodone intake: At t = 0, the subject will ingest 1 oxycodone 20 mg immediate release or a placebo capsule with 100 mL water. Both drugs will be obtained from the LUMC pharmacy.

3.3.2 Cannabis inhalation: Bedrocan is vaporized using the CE-marked Volcano Medic vaporizer (Storz & Bickel GmbH & Co, Tuttlingen, Germany), a safe and reliable method of intrapulmonary administration of cannabinoids. The Volcano heats the homogenized plant material to 210 °C to allow for conversion of the THC acid and CBD acid into THC and CBD vapor for inhalation. The complete 100 mg from the glass vial will be entered into the vaporizing chamber of the Volcano Medic. The vapor will be collected in a 6-L plastic balloon that, after inflation, is detached from the vaporizer and subsequently equipped with a mouthpiece for inhalation. It is our experience that the full content of the balloon is inhaled without any problems within 3-5 min.

The Bedrocan cannabis variety contains 22% THC (220 mg per gram) and less than 1%CBD. It is

developed in the Netherlands out of a requirement by the Dutch Health Ministry to have a "high THC" variety available to patients. We will administer 100 mg Bedrocan that contains 22.4-mg THC and less than 1-mg CBD, twice. Bedrocan will be obtained via the Bureau voor Medicinale Cannabis (BMO), a governmental organization (VWS) that obtains cannabis from Bedrocan Int. BV in Veenendaal, The Netherlands.

3.4 Blood sampling

Ten mL blood samples will be obtained on 14 occasions on each visit (total volume = 112 mL per visit) at t = 30 and 60 min after oxycodone intake and at t = 5, 20, 40, 60, 120, 180, 185, 200, 220, 240, 300 and 360 min after the first THC inhalation. From these sample, the following drugs will be measured: oxycodone, THC, THC's metabolite 11-OH-THC and cannabidiol (CBD). CBD is measured as it is expected that some minor quantities of CBD are present in Bedrocan.

Arterial blood will be collected in EDTA tubes. After blood collection the tubes will preferably be put in ice water in aluminum foiled containers, will be centrifuged within one hour for 10 minutes at 2000 G at 4 °C. The handling of samples will be done with the lights switched off. The plasma will be equally divided in 2 tubes (Brown Sarstedt) (primary and back-up sample). Plasma samples will be stored at a temperature of -80 °C and blood samples will be sent to Analytical Biochemical Laboratory (ABL) B.V., Assen, The Netherlands on dry-ice. Pharmacokinetic analysis will be performed using a validated assay. Determination of drug concentrations will be performed using liquid chromatography with tandem-mass spectrometer detection (LC-MS/MS). Analysis of the samples and acceptance criteria are indicated in ABL Standard Operating Procedure (SOP) 0251.

3.5 Questionnaires

Psychedelic effects are measured using visual analog scales ranging from 0 (no effect) to 10 cm (maximum effect) of the Bowdle and Bond & Lader questionnaires.

BOWDLE QUESTIONNAIRE: Three factors of psychedelic effects can be derived from the Bowdle questionnaire: drug high, internal perception, and external perception. Internal perception reflects inner feelings that do not correspond with the reality and is derived from questions regarding the hearing of unrealistic voices or sounds and having unrealistic thoughts and paranoid or anxious feelings. The external perception indicates a misperception of an external stimulus or change in the awareness of the subject's surroundings and is derived from questions regarding the change of body parts, the change of surroundings, the altered passing of time, the difficulty of controlling thoughts, and the change in color and sound intensity.

BOND & LADER: The Bond and Lader scales are calculated from sixteen 100 mm visual analog scales. The endpoints are set at antonymous word pairs such as 'alert–drowsy', 'well coordinated–clumsy', 'mentally slow–quick witted' and 'incompetent–proficient'. The study participant's task is to make a mark on each scale at the point that best describes how they currently feel considering that the two anchors reflect the greatest extent they experience each state. Responses from these 16 scales are then scored to yield three main factors of alertness (alert, strong, clear-headed, coordinated, energetic, quick-witted, attentive, proficient, interested), contentment (contented, happy, amicable, gregarious, tranquil), and calmness (calm, relaxed). A high score indicates impairment.

3.6 Pain pressure resting

We will use the FPN 100 N Algometer (FDN 100, Wagner Instruments Inc., Greenwich, CT, USA; Rolke et al., 2005) to deliver pressure pain on a skin area of 1 cm^2 between thumb and index finger. The FDN 100 has a force capacity (± accuracy) of 100 ± 2 N (10 ± 0.2 kgf) and graduation of 1 N (100 gf), respectively. A gradually increasing pressure is manually applied and the subjects are asked to indicate when the procedure becomes painful (pressure pain threshold, PPTh).

4. STUDY POPULATION

4.1 Population (base)

Healthy volunteers of either sex with earlier experience with cannabis (>2 lifetime exposures and <2x/week in the last 12 months).

4.2 Inclusion criteria

- aged 18-45 years,
- body mass index < 30 $kg.m^{-2}$,
- able to understand the written informed consent form,
- able to communicate with the staff,
- able and willing to complete the study procedures,
- signed the informed consent form,
- deemed suitable by the investigators.

4.3 Exclusion criteria

- Presence or history of any medical or psychiatric disease (incl. a history of substance abuse, anxiety, or the presence of a painful syndrome such as fibromyalgia);
- Use of any medication in the three months prior to the study (incl. paracetamol or other pain killers), except for oral contraceptives (females);
- Use of more than 21 alcohol units per week;
- Use of cannabis in the 4 weeks prior to the study;
- A positive urinary drug test or a breath alcohol test at screening or on the morning of the experiment;
- Pregnancy, lactating or a positive pregnancy test on the morning of the experiment;
- Participation in another drug trial in the 60 days prior to dosing.

4.4 Sample size calculation

There are no prior data on the interactive effect of cannabis and oxycodone on isohypercapnic breathing. If we extrapolate results of the ethanol data (van der Schrier et al., 2017) to the current study, we estimate an effect size of 5 L/min of adding THC to oxycodone with SD 6 L/min. We calculated a sample size of 18 subjects with alpha = 0.05 and 1 – beta = 0.90. To take any uncertainties into account we increased the group size to 20.

5. TREATMENT OF SUBJECTS

5.1 Investigational product/treatment

- **Oxycodone (oxycontin)** or placebo will be ingested on t = 0 min. The dose is 20 mg, the formulation an immediate release capsule.

- **Bedrocan™** will be inhaled at 90 min and again at 270 min. The dose is 100 mg containing about 22% THC.

-Placebo capsules will be obtained from the LUMC pharmacy.

5.2 Use of co-intervention

Not applicable.

5.3 Escape medication

In principle no escape medication is needed. However, in case of nausea or vomiting, ondansetron 4 mg iv may be administered (max. dose 12 mg). Additionally, in case of severe respiratory depression (as judged by the investigator), naloxone 400 mug may be given intravenously (possibly repetitive, no. max. dose).

6. INVESTIGATIONAL PRODUCT

6.1 Name and description of investigational product

- Oxycodone (oxycontin) is an opioid analgesic and registered in the Netherlands for the treatment of pain. The SpC is submitted with the protocol. An oxycodone immediate release capsule will be administered orally in a dose of 20 mg. We have done previous studies with oxycodone and know that its respiratory effects are moderate with plasma concentrations not exceeding 50 ng/mL. In the graph below, first results from P18.212 are given, showing the mean ± SD concentration of oxycodone immediate 20 are shown. An important observation is that after 24 h no oxycodone is present in the body and that after 5 hours the concentration dropped to levels < 15 ng/mL.

- **Bedrocan™** variety is a strain of cannabis characterized by circa 22% THC (220 mg per gram) and less than 1% CBD. Bedrocan® was developed in the Netherlands out of a requirement by the Dutch Health Ministry to have a "high THC" variety available to patients. It is a plant type bred because of its high yield, optimal growth characteristics, and derived from the popularly known "Jack Herer" cannabis variety. 100 mg of this study drug contains approx. 22 mg of THC for inhalation.

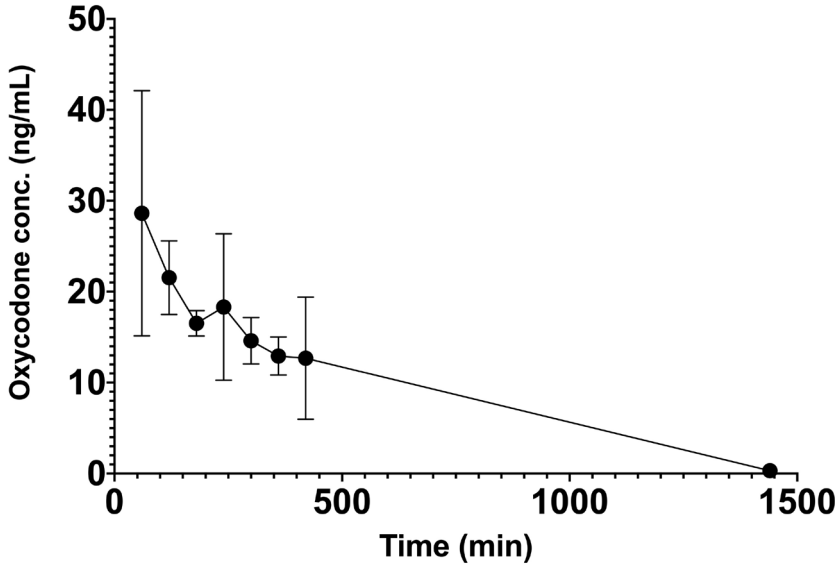

Figure 1. Oxycodone concentration following intake of 20 mg immediate release oral capsule at t = 0 min. Data are mean ± SD of 6 subjects and are from protocol P18.212.

Bedrocan is vaporized using the CE-marked Volcano Medic vaporizer (Storz & Bickel GmbH & Co, Tuttlingen, Germany), a safe and reliable method of intrapulmonary administration of cannabinoids. The Volcano heats the homogenized plant material to 210 °C to allow for conversion of the THC acid and CBD acid into THC and CBD vapor for inhalation. The vapor will be collected in an 8-L plastic balloon that, after inflation, is detached from the vaporizer and subsequently equipped with a mouthpiece for inhalation. It is our experience that the full content of the balloon is inhaled without any problems within 3-5 min.

We performed an earlier study with Bedrocan in 20 chronic pain patients, see Figure 2 (P15.316).THC concentrations (green symbols) reach about 100 ng/mL just after inhalation followed by an

exponential decay towards concentrations < 1 ng/mL after 3 hours. This indicates that at the end of the experiments no active levels of cannabis remain in plasma.

- **Placebo** capsules will be obtained from the LUMC pharmacy.

6.2 Summary of findings from non-clinical studies

Oxycodone, see submitted SPC.

Bedrocan, see submitted Investigators Brochure and IMPD.

Placebo, see submitted SPC.

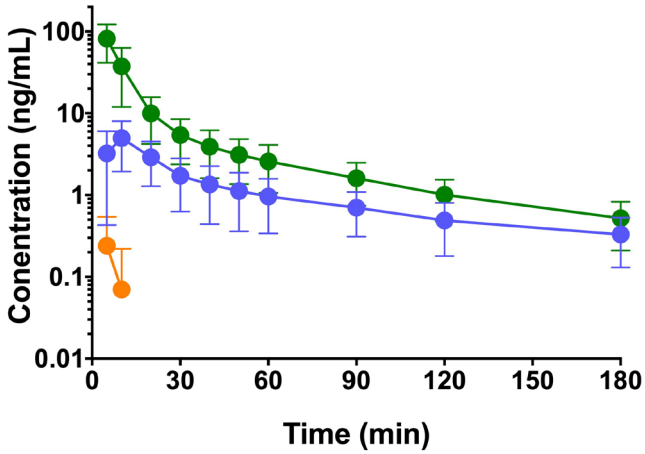

Figure 2. THC (green), 11-OH-THC (blue) and CBD (orange) concentrations following inhalation of 100 mg Bedrocan. Data from P15.316 obtained in 20 chronic pain patients. Data are mean ± 95% confidence interval.

6.3 Summary of findings from clinical studies

- Oxycodone, see submitted SPC and publications from van der Schrier et al. (Anesthesiology 2017 and Br J Anaesth, 2019). These publications describe the safety of 20 mg oral oxycodone treatment in healthy volunteers in a similar population of subjects as being tested in the current study.

- Bedrocan, see submitted Investigators Brochure and IMPD and publication from van den Donk et al. (Pain, 2019). This publication shows the safety of 100 mg Bedrocan in a more vulnerable population than studied by us.

- Placebo, see submitted SPC.

6.4 Summary of known and potential risks and benefits

This interventional proof-of-concept study has no intended benefit in terms of disease load reduction. The study in healthy volunteers attempts to understand the interaction between oxycodone and cannabis on ventilatory control. The gained information is of extreme importance given the current opioid epidemic and high number of opioid toxicities in the world, including the Netherlands.

Opioids produce several side effects. These side effects are the topic of the current study. Since the study is performed under highly controlled and monitored conditions, the occurrence of temporary side effects will have limited impact on the subject. We expect and will measure respiratory depression. In case of severe respiratory depression, we can administer naloxone. In the last 25 years that we have performed these studies, we never needed to administer naloxone because of respiratory depression. Additional side effects include nausea and vomiting (to be treated with an antiemetic) and sedation.

Cannabis does produce side effects, most importantly nausea/vomiting (to be treated with an antiemetic), sedation and psychomimetic side effects of which drug high is most important. These side effects decline rapidly within 2-3 hours after inhalation (See van de Donk et al., Pain 2019).

6.5 Description and justification of route of administration and dosage

Oxycodone (and placebo) will be administered orally. This is a routine form of administration. Bedrocan will be inhaled, a form of administration that is highly effective with an uptake > 66% of vapor.

6.6 Dosages, dosage modifications and method of administration

See 6.1

6.7 Preparation and labelling of Investigational Medicinal Product

The IMPs will be obtained from the LUMC pharmacy. They are responsible for labelling and packaging.

6.8 Drug accountability

All study products are obtained from the pharmacy. Remaining products will be destroyed by the researchers, and noted on the Case Record Form.

7. NON-INVESTIGATIONAL PRODUCT

7.1 Name and description of non-investigational product(s)

7.1.1 The Volcano Vaporizer

The Volcano Vaporizer is a CE-marked device. It is produced by Storz & Bickel GmbH & Co. (Tuttlingen, Germany).

7.1.2 Ventilation measurement

Ventilation will be measured with the CE-marked ExSpiron device; see cosubmitted files.

Name of device	**Manufacturer**	**Model**	**CE conform 93/42/EEG of EU/2017/745 (Medical devices)**	**Conform intended use as described in the instruction manual?**
Volcano Vaporizer	Storz & Bickel GmbH & Co	Mighty Medic 2 Inventory number: will follow	Yes	Yes
ExSpiron™ Minute Ventilation Monitor	Innomed Benelux	1Xi Inventory number: 20-800-1045	Yes	Yes

7.2 Summary of findings from non-clinical studies

NA

7.3 Summary of findings from clinical studies

NA

7.4 Summary of known and potential risks and benefits

NA

7.5 Description and justification of route of administration

NA

7.6 Dosage, dosage modification and method of administration

NA

7.7 Preparation and labelling of Investigational Medicinal Product

NA

7.8 Drug accountability

NA

8. METHODS

8.1 Study parameters/endpoints

8.1.1 Main study parameter/endpoint

Primary endpoint is the effect of inhaled THC on ventilation at end-tidal PCO_2 = 55 mmHg without and with concomitant intake of 20 mg oxycodone immediate release (IR) capsule in healthy volunteers 120 min after oxycodone intake.

8.1.2 Secondary study parameters/endpoints (if applicable)

- occurrence of apneic events;
- occurrence of desaturation events;
- slope of the hypercapnic ventilatory response;
- baseline ventilation;
- psychomimetic side effects as measured by Bowdle and Bond & Lader questionnaires; plasma concentrations of THC and oxycodone;
- blood pressure and heart rate.

8.1.3 Other study parameters (if applicable)

NA

8.2 Randomization, blinding and treatment allocation

The sequence of visit will be randomized using a randomization list generated within CASTOR EDC prior to dosing.

8.3 Study procedures

The estimated time line of the study is as follows:

8:30 arrival in the laboratory at K5-120; urine drug and alcohol breath testing;
8:45 placement of intravenous and arterial line.
9:00 baseline pain pressure test, baseline questionnaires, baseline blood sample
9:30 baseline Hypercapnic ventilatory response (HCVR);
10:00 intake of placebo/oxycodone
11:00 HCVR #2
11:30 inhalation of 100 mg Bedrocan
12:00 HCVR #3
13:00 HCVR #4
14:00 HCVR #5
14:30 inhalation of 100 mg Bedrocan
15:00 HCVR #6
16:00 HCVR #7
17:00 HCVR #8

Assessment of the subject at 17:30. If HCVR is still depressed two additional HCVR may be obtained at 18:00 and 19:00 hours.
Subject returns home or stays overnight (decided upon the discretion of the attending physician-investigator). Our experience is that most if not all subjects will be sufficiently fit to go home.

Blood samples will be drawn according to the following scheme:
10:30; 11:00, 11:35; 11:50; 12:10; 12:30; 13:30; 14:30; 14:35; 14:50; 15:10; 15:30; 16:30; 17:30.

Bowdle and Bond & Lader Questionnaires are taken at 10:30; 11:30; 12:30; 13:30; 14:30; 15:30; 16:30; 17:30. All questionnaires are taken 5 min prior to blood sampling.

Pain pressure threshold will be measured at 10:25, 10:55, 11:30; 11:45; 12:05; 12:25; 13:25; 14:25; 14:30; 14:45; 15:05; 15:25; 16:25; 17:25.

8.4 Withdrawal of individual subjects
Subjects can leave the study at any time for any reason if they wish to do so without any consequences. The investigator can decide to withdraw a subject from the study for any reasons.

8.4.1 Specific criteria for withdrawal (if applicable)
The most important criterium for withdrawal is that the subject indicates that he or she want to terminate the experiment. Additionally, we will terminate the experiment in case of a life-threatening adverse event (to be judged by the physician-investigator) or any other condition that may affect the health of wellbeing of the subject.

Rules for administration of naloxone in case of a respiratory event are:
- loss of respiratory activity for 3 min or longer, despite active stimulation of the subject;
- and increase in end-tidal PCO_2 to 9 kPa or greater for more than 3 min;
- SpO_2 < 85% for more than 3 min despite administration of supplemental oxygen.

In all three cases, when respiration is resumed, respiratory measurements will continue for as long as needed to ensure complete return of respiration to control levels. However, if this occurs prior to the second Bedrocan inhalation, the second inhalation will not be performed. Given the ample experience that the researchers have with performing studies on opioid effect on breathing, we expect that the probability of such events are extremely small.

8.5 Replacement of individual subjects after withdrawal
Subjects will be replaced in case of withdrawal.

8.6 Follow-up of subjects withdrawn from treatment
As indicated above, in case stopping rules were met and naloxone was administered, respiratory measurements will continue for as long as possible to ensure complete return of respiration to control levels. It may additionally be decided to keep the subject overnight. Only when respiration has returned to baseline levels, the subject will be sent home (under supervision).

8.7 Premature termination of the study
When a serious adverse event occurs related to treatment, the ethics committee will be notified and further experiments will be put on hold until the ethics committee has re-evaluated the experiments in close cooperation with the researchers. We do not plan to institute a Data Safety Committee for the reason that the researchers are all experts on the topic of opioid-induced respiratory depression and the probability of such events are extremely small. Overall, given our experience and expertise, we judge that this experiment to have a negligible to moderate risk.

9. SAFETY REPORTING

9.1 Temporary halt for reasons of subject safety

In accordance to section 10, subsection 4, of the WMO (Medical Research Involving Human Subjects Act), the sponsor will suspend the study if there is sufficient ground that continuation of the study will jeopardize subject health or safety. The sponsor will notify the accredited METC (Human Ethics Committee) without undue delay of a temporary halt including the reason for such an action. The study will be suspended pending a further positive decision by the accredited METC. The investigator will take care that all subjects are kept informed.

9.2 AEs, SAEs and SUSARs

9.2.1 Adverse events (AEs)

Adverse events are defined as any undesirable experience occurring to a subject during the study, whether or not considered related to the investigational product, trial procedure/ the experimental intervention. All adverse events reported spontaneously by the subject or observed by the investigator or their staff will be recorded.

9.2.2. Serious adverse events (SAEs)

A serious adverse event is any untoward medical occurrence or effect that

- results in death;
- is life threatening (at the time of the event);
- requires hospitalization or prolongation of existing inpatients' hospitalization;
- results in persistent or significant disability or incapacity;
- is a congenital anomaly or birth defect; or
- any other important medical event that did not result in any of the outcomes listed above due to medical or surgical intervention but could have been based upon appropriate judgement by the investigator.

An elective hospital admission will not be considered as a serious adverse event.

The investigator will report all SAEs to the sponsor (LUMC) without undue delay after obtaining knowledge of the events, except for the following SAEs: any SAE not related to study medication or study procedures.

The sponsor will report the SAEs through the web portal *ToetsingOnline* to the accredited METC that approved the protocol, within 7 days of first knowledge for SAEs that result in death or are life threatening followed by a period of maximum of 8 days to complete the initial preliminary report. All other SAEs will be reported within a period of maximum 15 days after the sponsor has first knowledge of the serious adverse events. SAEs that do not require reporting will be reported in the annual safety report.

9.2.3. Suspected unexpected serious adverse reactions (SUSARs)

Adverse reactions are all untoward and unintended responses to an investigational product related to any dose administered.

Unexpected adverse reactions are SUSARs if the following three conditions are met:
1. the event must be serious (see chapter 9.2.2);
2. there must be a certain degree of probability that the event is a harmful and an undesirable reaction to the medicinal product under investigation, regardless of the administered dose;
3. the adverse reaction must be unexpected, that is to say, the nature and severity of the adverse reaction are not in agreement with the product information as recorded in:
- Summary of Product Characteristics (SPC) for an authorized medicinal product;
- Investigator's Brochure for an unauthorized medicinal product.

The sponsor will report expedited the following SUSARs to the METC:
- SUSARs that have arisen in the clinical trial that was assessed by the METC;
- SUSARs that have arisen in other clinical trials of the same sponsor and with the same medicinal product, and that could have consequences for the safety of the subjects involved in the clinical trial that was assessed by the METC.

The remaining SUSARs are recorded in an overview list (line-listing) that will be submitted once every half year to the METC. This line-listing provides an overview of all SUSARs from the study medicine, accompanied by a brief report highlighting the main points of concern.
The expedited reporting of SUSARs through the web portal Eudravigilance or ToetsingOnline is sufficient as notification to the competent authority.

The sponsor will report expedited all SUSARs to the competent authorities in other Member States, according to the requirements of the Member States. The expedited reporting will occur not later than 15 days after the sponsor has first knowledge of the adverse reactions. For fatal or life-threatening cases the term will be maximal 7 days for a preliminary report with another 8 days for completion of the report.

9.3 Annual safety report
The annual safety report will be combined with the annual progress report. In addition to the expedited reporting of SUSARs, the sponsor will submit, once a year throughout the clinical trial, a safety report to the accredited METC, competent authority, and competent authorities of the concerned Member States. This safety report consists of:
- a list of all suspected (unexpected or expected) serious adverse reactions, along with an aggregated summary table of all reported serious adverse reactions, ordered by organ system, per study;
- a report concerning the safety of the subjects, consisting of a complete safety analysis and an evaluation of the balance between the efficacy and the harmfulness of the medicine under investigation.

9.4 Follow-up of adverse events
All AEs will be followed until they have abated, or until a stable situation has been reached. Depending on the event, follow up may require additional tests or medical procedures as indicated, and/or referral to the general physician or a medical specialist. SAEs need to be reported until the end of study, as defined in the protocol.

10. STATISTICAL ANALYSIS

10.1 Primary study parameter

The primary end-point is ventilation at isohypercapnia (VE_{55}). To obtain VE_{55} we will estimate the slope of the HCVR R (The R Foundation for Statistical Computing, www.r-project.org, accessed January 1, 2017) and next calculate ventilation at an end-tidal carbon dioxide concentration of 55 mmHg.

We with compare VE_{55} between the two treatment arms by considering baseline values (prior to any drug use) and VE_{55} at t = 120 min after oxycodone or placebo intake with visit, and oxycodone treatment as fixed factors and subject as a repeated statement and baseline value as covariate.
$P < 0.05$ is considered significant.

Additionally, we will perform a pharmacokinetic-pharmacodynamic data analysis in NONMEM. The model to be used in the NONMEM analysis has not been specified as yet but will be of the form:

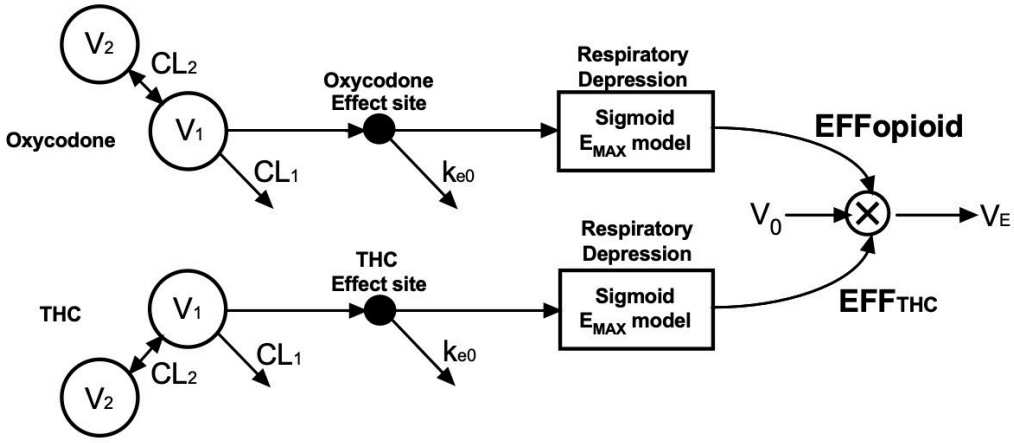

Figure 6. Schematic diagram of a possible NONMEM pharmacokinetic-pharmacodynamic model of the interactive effects of oxycodone and THC on ventilation. Vo is baseline value, V1 and V2 the volumes of the pharmacokinetic compartments 1 and 2, with corresponding clearances CL. Ke0 is the blood-effect-site equilibration rate constant.

10.2 Secondary study parameter(s)

All secondary parameters will be presented as mean ± SD or 95% confidence interval, median (interquartile range). Data will be analyzed using an explorative approach.

10.3 Other study parameters

NA.

10.4 Interim analysis (if applicable)

NA.

11. ETHICAL CONSIDERATIONS

11.1 Regulation statement

The study will be conducted according to the principles of the Declaration of Helsinki (version amended in Fortaleza, Brasil, 2013) and in accordance with the Medical Research Involving Human Subjects Act (WMO). The PI and junior researcher are BROK certified. No additional harm or benefit is expected upon participation within this study.

11.2 Recruitment and consent

Eligible subjects will be approached before inclusion by telephone for brief oral information by one of the researchers. If the subject is willing to participate, written information will be provided and informed consent will be obtained prior to further screening.

11.3 Objection by minors or incapacitated subjects (if applicable)

NA.

11.4 Benefits and risks assessment, group relatedness

This interventional proof-of-concept study has no intended benefit in terms of disease load reduction. The study in healthy volunteers attempts to understand the interaction between oxycodone and cannabis on ventilatory control. The gained information is of importance given the current opioid epidemic and high number of opioid toxicities in the world, including the Netherlands. We contend that the results of this study may be extrapolated to the general population.

Opioids produce several side effects. These side effects are the topic of the current study. Since the study is performed under highly controlled and monitored conditions, the occurrence of temporary side effects will have limited impact on the subject. We expect and will measure respiratory depression. In case of severe respiratory depression, we can administer naloxone. In the last 25 years that we perform these studies, we never needed to administer naloxone because of respiratory depression. Additional side effects include nausea and vomiting (to be treated with an antiemetic) and sedation.

Cannabis does produce side effects, most importantly nausea/vomiting (to be treated with an antiemetic), sedation and psychomimetic side effects of which drug high is most important. These side effects decline rapidly within 2-3 hours after inhalation (See van de Donk et al., Pain 2019).

10.5 Compensation for injury

The risk of injury as a result of this study is considered small. However, the LUMC has insured this risk by a liability insurance, in accordance with article 7, subsection 6 of the WMO.

The sponsor (also) has an insurance policy in accordance, with the legal requirements in the Netherlands (Article 7 WMO and the Measure regarding Compulsory Insurance for Clinical Research in Humans of 23th June 2003). This insurance provides cover for damage to research patients through injury or death caused by the study:

1. € 650.000,-- (i.e. six hundred and fifty thousand Euro) for death or injury for each subject who participates in the Research;

2. € 5.000.000,-- (i.e. five million five hundred thousand Euro) for death or injury for all subjects who participate in the Research;
3. € 7.500.000,-- (i.e. seven and a half million Euro) for the total damage incurred by the organisation for all damage disclosed by scientific research by the sponsor in each year of insurance coverage.

10.6 Incentives (if applicable)

Volunteers will be reimbursed for their participation in the study. They will receive euro 400 for completion of all 2 sessions (or 20 euros/h) or 10 euros per hour participated in the study when the experiment is prematurely terminated for any reason. per session.

12. ADMINISTRATIVE ASPECTS, MONITORING AND PUBLICATION

12.1 Handling and storage of data and documents

All data from the subjects will be stored under code. Subject-related identifying data will be omitted from all secondary documents. The code will be stored digitally on a secured partition of the i:-disk at LUMC that is accessible by just the investigators. Other involved parties (monitor, Inspectie, Gezondheidszorg en Jeugd) may be granted access to subject data, also subject identifying data, to review the research. These involved parties will handle the patient identifying data in a confidential manner. The sponsor, local researchers and project leader are responsible for data processing. When a subject withdraws consent, data collected until that moment will be used. All data will be stored for the length of the study and for 25 years afterwards, for further publication. Blood will be stored until analysis and will be destroyed afterwards. All handling of personal data will comply with the Dutch Personal Data Protection Act. The Functionaris Gegevensbescherming from the LUMC has been informed about the data handling in the study. When subjects have questions or complaints about data handling they can contact the Functionaris Gegevensbescherming (contact information is mentioned in the patient information letter). Only data needed to assess study objectives will be collected.

12.2 Monitoring and Quality Assurance

Monitoring will be executed by (internal) monitors of the LUMC according to the monitor plan.

12.3 Amendments

A 'substantial amendment' is defined as an amendment to the terms of the METC application, or to the protocol or any other supporting documentation, that is likely to affect to a significant degree:
- the safety or physical or mental integrity of the subjects of the trial;
- the scientific value of the trial;
- the conduct or management of the trial; or
- the quality or safety of any intervention used in the trial.

All substantial amendments will be notified to the METC and to the competent authority. Non substantial amendments will not be notified to the accredited METC and the competent authority, but will be recorded and filed by the sponsor.

12.4 Annual progress report

The sponsor/investigator will submit a summary of the progress of the trial to the accredited METC once a year. Information will be provided on the date of inclusion of the first subject, numbers of subjects included and numbers of subjects that have completed the trial, serious adverse events/ serious adverse reactions, other problems, and amendments.

12.5 Temporary halt and (prematurely) end of study report

The sponsor will notify the accredited METC and the competent authority of the end of the study within a period of 90 days. The end of the study is defined by the completion of data analysis.

The sponsor will notify the METC immediately of a temporary halt of the study, including the reason of such an action. In case the study is ended prematurely, the sponsor will notify the accredited METC and the competent authority within 15 days, including the reasons for the premature

termination. Within one year after the end of the study, the investigator/sponsor will submit a final study report with the results of the study, including any publications/abstracts of the study, to the accredited METC and the Competent Authority.

12.6 Public disclosure and publication policy

The study will be registered at trialregister.nl prior to recruitment of subjects. The results of the trial will be published in a peer-review scientific journal and will be presented on (inter)national scientific conferences and meetings. This will be in accordance to the CCMO statement on publication policy.

13. STRUCTURED RISK ANALYSIS

13.1 Potential issues of concern

Most important risks related to oxycodone include: sedation, nausea and vomiting, respiratory depression. However, we consider none of these a potential issue of concern, since (1) we will monitor all subjects continuously during the study, (2) we have ample knowledge on pharmacokinetics and pharmacodynamics of oxycodone and consequently on the occurrence, risk and treatment of these effects, (3) since all adverse effects are related to drug plasma concentration, these events dissipate over time (see for the PK profile Figure 1). Additionally, we have ample knowledge on the mechanism of action of oxycodone and are able to terminate all of these effects by administration of naloxone. This will immediately resolve respiratory depression and sedation. Nausea/vomiting may be treated with antiemetics.

Most important risk of THC include: sedation, nausea and vomiting, and psychomimetic side effects, most importantly drug high. However, we consider none of these a potential issue of concern, since (1) we will monitor all subjects continuously during the study, (2) we have ample knowledge on pharmacokinetics and pharmacodynamics of THC and consequently on the occurrence, risk and treatment of these side effects, (3) since all adverse are related to plasma concentration, these events dissipate over within 3 hours following inhalation (see for the PK profile Figure 2). Additionally, we have ample knowledge on the mechanism of action of THC. Nausea/vomiting may be treated with antiemetics.

Most important risks of the combination of THC and oxycodone include: sedation, nausea and vomiting, respiratory depression and psychomimetic side effects, most importantly drug high. However, we consider none of these a potential issue of concern, since (1) we will monitor all subjects continuously during the study, (2) we have ample knowledge on the occurrence, risk and treatment of these effects in separate administrations, (3) all adverse events dissipate over time. The combination of oxycodone and THC may cause additive effects on sedation, respiratory depression or nausea/vomiting. We are able to terminate the opioid part in case of severe side effect by administration of naloxone; additionally, we can treat nausea/vomiting with antiemetics.

13.2 Synthesis

First, the population in which we will perform these experiments (healthy, young volunteers) is not considered frail or at risk, and it is our experience that these subjects will be able to complete both experimental sessions without consequences.

Second, we will continuously monitor the subjects following administration of oxycodone and/or THC. In case the physician-investigator has doubts whether the subject can return home, we will continue monitoring during an overnight stay.

Third, in case of serious effects on respiration, the opioid effect may be reversed by administration of naloxone.

Fourth, the researchers have ample experience with the administration of even higher doses of opioids and THC in volunteers.

Fifth, the occurrence of side effects is the topic of the current study. Hence all effects of treatment on respiration will be logged and is available for analysis.

14. REFERENCES

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