Randomized placebo-controlled phase II cross-over study on the influence of fampridine on working memory in healthy subjects

Study-ID: FamH

Clinical Study Protocol

Study Type	Clinical Trial with Investigational Medicinal Product (IMP)
Study Categorization	Risk category B according to HRA
Study Registration	NCT04652557
Study ID	FamH
Sponsor-Investigator	Prof. Dominique de Quervain, MD University of Basel Co-Director Transfaculty Research Platform Molecular and Cognitive Neurosciences Director Division of Cognitive Neuroscience Birmannsgasse 8, 4055 Basel E-Mail: dominique.dequervain@unibas.ch Phone: +41 61 207 02 37
Sponsor	Prof. Andreas Papassotiropoulos, MD University of Basel Co-Director Transfaculty Research Platform Molecular and Cognitive Neurosciences Director Division of Molecular Neuroscience Birmannsgasse 8, 4055 Basel E-Mail: andreas.papas@unibas.ch Phone: +41 61 207 05 99
Investigational Product	Fampridine (Fampyra®)
Protocol Version and Date	Version 7 (dated 05/04/2022)

PROTOCOL SIGNATURE FORM

Study Title	Randomized placebo-controlled phase II cross-over study on the influence of fampridine on working memory in healthy subjects
Study number	NCT04652557
Protocol Version	Version 7 (dated 05/04/2022)

Sponsors

The sponsors have approved and confirm hereby to conduct the study according to the protocol, current version of the World Medical Association Declaration of Helsinki, and ICH-GCP guidelines as well as the local legally applicable requirements.

Prof. Dominique de Quervain, MD
Sponsor-Investigator

Basel, 7th April 2022 — Signature
Place/Date

Prof. Andreas Papassotiropoulos, MD
Sponsor

Basel, 7th April 2022 — Signature
Place/Date

Local Investigators and trial statistician at study site

I have read and understood this trial protocol and agree to conduct the trial as set out in this study protocol, the current version of the World Medical Association Declaration of Helsinki, ICH-GCP guidelines and the local legally applicable requirements.

Study Site: University of Basel, Division of Cognitive Neuroscience, Birmannsgasse 8, CH-4055 Basel

Christiane Gerhards, MD
Investigator

Basel, 11/4/22 — Signature
Place/Date

Annette Harings-Kaim, MD
Investigator

Basel, 07.04.2022 — Signature
Place/Date

Nathalie Schicktanz, PhD
Trial statistician

Basel, 7th April 2022 — Signature
Place/Date

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STUDY SYNOPSIS

Sponsor-Investigator	Prof. Dominique de Quervain, MD, Transfaculty Research Platform Molecular and Cognitive Neurosciences, University of Basel.
Sponsor	Prof. Andreas Papassotiropoulos, MD Transfaculty Research Platform Molecular and Cognitive Neurosciences, University of Basel.
Study Title	Randomized placebo-controlled phase II cross-over study on the influence of fampridine on working memory in healthy subjects
Study ID	FamH
Protocol Version and Date	Version 7 (dated 05/04/2022)
Trial Registration	NCT04652557 (Clinicaltrials.gov)
Study Category and Rationale	Risk category B according to ClinO Art. 19 Fampridine is approved in Switzerland for treatment of gait problems in patients with MS. We will use fampridine for a different indication.
Clinical Phase	Phase II, proof of concept
Background and Rationale	Large genome-wide association studies for schizophrenia suggest that genetic variations (single nucleotide polymorphisms) in genes encoding potassium channels modify the risk for the development of this disorder. In healthy humans we found that genetic variations in the same genes are also associated with working memory, an established heritable cognitive component (endophenotype) of schizophrenia.
Objectives	To investigate the effects of fampridine - an inhibitor of voltage-gated potassium (Kv) channels - on working memory in healthy participants.
Outcomes	Primary outcome will be • High-load working-memory performance: 3-back (d') as assessed by a letter n-back task (Heck, Fastenrath et al. 2014) after repeated intake Secondary outcomes will be • High-load working-memory performance in a 3-back (d') task after acute intake • Reaction time (for correct 3-back responses) after acute and repeated intake • Performance in a 0-back (d') task as a measure of attention after acute and repeated intake • Processing speed task (Symbol Digit Modalities Test, SDMT; Smith 1973) after acute and repeated intake • BOMAT - advanced (Hossiep/Turck/Hasella, 1999) after acute and repeated intake • Digit span forward and backward performance (von Aster 2006) after acute and repeated intake • Resting motor threshold (rMT) after repeated intake, measured by transcranial magnetic stimulation (TMS) Safety outcomes will be • Headache, gastrointestinal discomfort, dizziness • AE-recording • Vital signs
Study Design	Randomized, placebo-controlled, double blind, cross-over

Inclusion- / Exclusion Criteria	Inclusion criteria: • male or female • generally healthy • normotensive (BP between 90/60mmHg and 140/90mmHg) • BMI between 19 and 29,9 kg/m^2 • aged between 18 and 30 years • fluent German-speaking • IC as documented by signature • At least double vaccination against Covid-19 Exclusion criteria: • contraindications to the class of drugs under study, e.g. known hypersensitivity or allergy to 4-aminopyridine • use of potassium channel blockers within the last 3 months • concomitant treatment with OCT 2 inhibitors and substrates (e.g. cimetidine, propranolol) • acute or chronic psychiatric disorder (e.g. major depression, psychoses, somatoform disorder, suicidal tendency) • acute cerebrovascular condition • history of seizures • risk of lowered seizure threshold (due to e.g. sleep deprivation, withdrawal of alcohol after alcohol abuse) • renal impairment • history of malignant cancers • walking problems (e.g. due to dizziness) • other clinically significant concomitant disease states (e.g. hepatic dysfunction, cardiovascular disease, diabetes, asthma) • bradycardia < 50/min during clinical examination • clinically significant laboratory or ECG abnormality that could be a safety issue in the study • known or suspected non-compliance • drug or alcohol abuse • inability to follow the procedures of the study, e.g. due to language or psychological problems of the participant • participation in another study with an investigational drug within the 30 days preceding and during the present study • prior participation (less than two years ago) in a study investigating working memory (notably the n-back task) • enrolment of the investigator, his/her family members, employees and other dependent persons • smoking (>3 cigarettes per day) • intake of psychoactive drugs (e.g. benzodiazepines, antidepressants, neuroleptics) • pregnancy or breast feeding • experiencing a syncope during basal rMT measuring Exclusion Criteria concerning TMS measurement • metal in the brain, skull or elsewhere in the body (e.g., splinters, fragments, clips, etc.) • implanted neurostimulator (e.g., DBS, epidural/subdural, VNS) • cardiac pacemaker or intracardiac lines • medication infusion device • piercings, pivot teeth (retainers are no exclusion criterion) • tattoos (head area) less than 3 months old or older than 20 years • condition after neurosurgery

	• hearing problems or tinnitus • not able to sit still due to tremor, tics, itching • history of repeated syncope • head trauma diagnosed as concussion or associated with loss of consciousness • diagnosis of epilepsy, or a convulsion or a seizure in the past of the participant or his family • TMS in the past showing problems • MRI in the past showing problems • surgical procedures to spinal cord • spinal or ventricular derivations
Measurements and procedures	Assessment of efficacy: Performance in cognitive tasks at two time points (after acute and repeated administration) and rMT at one time point (after repeated administration) • Effect after acute administration (visits 2 and 4): the assessment will be performed 4 hours after intake of 10 mg fampridine or placebo. • Effect after repeated administration (visits 3 and 5): 3.5 days after intake of twice daily doses of 10 mg fampridine or placebo. The assessment will be performed 4 hours after the last intake of study medication • Assessment of safety: AE recording, vital signs
Study Product	Twice daily oral administration of 10 mg fampridine (Fampyra®) for 3.5 days
Control Intervention	Twice daily oral administration of placebo for 3.5 days
Number of Participants with Rationale	Total of 44 participants, approx. 22 male and 22 females. There will be replacement of Drop-Outs until data from 44 participants are completed. The estimation of N=44 is based on a power analysis assuming to detect a medium effect size of a drug with a power of 90% at α=0.05.
Study Duration	Study duration is estimated to be 18 months.
Study Schedule	First participant in 09/2021 (planned) Last participant out 02/2023 (planned)
Investigators	Christiane Gerhards, MD Transfaculty Research Platform Division of Cognitive Neuroscience University of Basel Birmannsgasse 8 4055 Basel Phone: +41 61 207 0244 Fax: +41 61 207 0241 Email: christiane.gerhards@unibas.ch Annette Harings-Kaim, MD Transfaculty Research Platform Division of Cognitive Neuroscience University of Basel Birmannsgasse 8 4055 Basel Phone: +41 61 207 0251 Fax: +41 61 207 0241 E-Mail: annette.harings@unibas.ch

Study Centre	University of Basel Transfaculty Research Platform Molecular and Cognitive Neurosciences Birmannsgasse 8 4055 Basel
Statistical Considerations	Linear mixed models will be used to compare the effect of fampridine and placebo on primary and secondary outcome variables.
GCP Statement	This study will be conducted in compliance with the protocol, the current version of the Declaration of Helsinki, the ICH-GCP as well as all national legal and regulatory requirements.

GLOSSARY OF ABBREVIATIONS

AE	Adverse Event
ALT	Alanine Aminotransferase
4-AP	4-Aminopyridine
ASR	Annual Safety Repot
AST	Aspartate Aminotransferase
BASEC	Business Administration System for Ethical Committees
BMI	Body Mass Index
BOMAT	Bochumer Matrizentest
BP	Blood Pressure
Bpm	Beats per minute
CA	Competent Authority
CEC	Competent Ethics Committee
CTCAE	Common Terminology Criteria for Adverse Events
ClinO	Ordinance on Clinical Trials in Human Research
CRF	Case Report Form
CTU	Clinical Trial Unit
DS	Digit Span Task
ECG	Electrocardiography
eCRF	Electronic Case Report Form
EKNZ	Ethikkommission Nordwest- und Zentralschweiz
EMG	Electromyography
FADP	Federal Act on Data Protection
FOPH	Federal Office of Public Health
GCP	Good Clinical Practice
Gf	Fluid Intelligence
GGT	Gamma-Glutamyltransferase
GMP	Good Manufacturing Practice
GWAS	Genome-wide association studies
Ho	Null hypothesis
H1	Alternative hypothesis
hCG	Human choriogonadotropin
HR	Heart Rate
HRA	Federal Act on Research involving Human Beings
IC	Informed Consent
ICH	International Conference on Harmonisation
IEC	Independent Ethics Committee
IIT	Investigator-initiated trial
IMP	Investigational Medicinal Product
IR	Immediate Release
Kv	Voltage-gated potassium channel
LDH	Lactatdehydrogenase
LTP	Long-Term-Potentiation
MADRS	Montgomery Åsberg Depression Rating Scale

MEP	Motor Evoked Potential
MNI	Montreal Neurological Institute
MRHD	Maximum Recommended Human Dose
MS	Multiple Sclerosis
MSO	Maximum Stimulator Output
MTI	Maximum Tolerated Imbalance
OCT 2	Organic Cation Transporter 2
PI	Principal Investigator
PR	Prolonged Release
rMT	resting Motor Threshold
SADRS	Serious Adverse Drug Reaction
SAE	Serious Adverse Event
SD	Source Data
SDMT	Symbol Digit Modalities Test
SDV	Source Data Verification
SNCTP	Swiss National Clinical Trials Portal
SOP	Standard Operating Procedure
SUSAR	Suspected Unexpected Serious Adverse Reaction
SR	Sustained Release
TMF	Trial Master File
TMS	Transcranial Magnetic Stimulation
V/P	Verum/Placebo
VAS	Visual Analog Scale
WI	Working Instruction
WIE	Wechsler Intelligenztest für Erwachsene

STUDY SCHEDULE

	Duration		**Treatment period 1** (3.5 days)			**Wash-out period** 8-82 days between V3 and V4	**Treatment period 2** (3.5 days)		
Visit		**Visit 1**	**Visit 2**		**Visit 3**		**Visit 4**		**Visit 5**
Description		Screening	Test day 1	P/V or V/P	Test day 2	Wash-out period	Test day 3	V/P or P/V	Test day 4
Day		within 14 days before Visit 2	1		4		12-30		15-33
Screening • Written informed consent, assignment of the subject number • Personal, family and medication history • Self-assessment mental health and sleeping habits, MADRS-S, EHI • Physical examination • ECG, Blood sample	1.5 h	x							
Treatment periods • Electronic intake diary • Electronic AE diary (VAS headache, gastrointestinal discomfort, dizziness; free text)	3.5 days			x				x	
Wash-out period (8-82 days between V3 and V4)	8-82 days					x			
Test days									
Pre-examination test days • Vital signs • Flu-like symptoms, common cold symptoms • Pregnancy test (Visits 2 and 4) • Questionnaire drugs and alcohol consumption during last 3 days resp. past 12 hours • Sleep duration last 24 hours • AE recording • VAS headache, gastrointestinal discomfort, dizziness; VAS tiredness, motivation	15 min		x		x		x		x
Assignment of the randomization number	5 min		x						
Intake of study medication	5 min		x	x	x		x	x	x
Waiting time (incl. continental breakfast 2 hours after intake of study medication)	4 h		x		x		x		x
Before start of testing • AE recording • VAS headache, gastrointestinal discomfort, dizziness; VAS tiredness, motivation	15 min		x		x		x		x
Testing Phase – part 1 • n-Back • Processing speed task SDMT • Digit Span	45 min		x		x		x		x
Break	15 min		x		x		x		x
Testing Phase – part 2 • BOMAT	15 min		x		x		x		x
rMT measurement using TMS	15 min	x			x				x
Final Examination • AE recording • VAS headache, gastrointestinal discomfort, dizziness; VAS tiredness, motivation	15-20 min		x		x		x		x

1 STUDY ADMINISTRATIVE STRUCTURE

Function	Address
Sponsor Investigator	Prof. Dominique de Quervain, MD University of Basel Co-Director Transfaculty Research Platform Molecular and Cognitive Neurosciences Director Division of Cognitive Neuroscience Birmannsgasse 8, 4055 Basel E-Mail: dominique.dequervain@unibas.ch Phone: +41 61 207 0237
Sponsor	Prof. Andreas Papassotiropoulos, MD University of Basel Co-Director Transfaculty Research Platform Molecular and Cognitive Neurosciences Director Division of Molecular Neuroscience Birmannsgasse 8, 4055 Basel E-Mail: andreas.papas@unibas.ch Phone: +41 61 207 0599
Investigators	Christiane Gerhards, MD Transfaculty Research Platform Division of Cognitive Neuroscience University of Basel Birmannsgasse 8 4055 Basel E-Mail: christiane.gerhards@unibas.ch Phone: +41 61 207 0244 Annette Harings-Kaim, MD Transfaculty Research Platform Division of Cognitive Neuroscience University of Basel Birmannsgasse 8 4055 Basel E-Mail: annette.harings@unibas.ch Phone: +41 61 207 0251
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2 ETHICAL AND REGULATORY ASPECTS

The decision of the competent ethics committee (CEC) and the competent authority (CA) concerning the conduct of the study will be made in writing to the Sponsor-Investigator before commencement of this study. The clinical study can only begin once approval from all required authorities has been received. Any additional requirements imposed by the authorities shall be implemented.

2.1 Study registration

The study will be registered in the registry clinicaltrial.gov and in the Swiss National Clinical Trials Portal (SNCTP) before the start of recruitment.

2.2 Categorization of study

This clinical trial comes under Category B as fampridine (Fampyra®) is authorized in Switzerland and it is used in an indication different from that specified in the prescribing information.

2.3 Competent Ethics Committee (CEC)

The sponsor-investigator will obtain approval from CEC (EKNZ) before the start of the clinical trial.

No changes are made to the protocol without prior sponsor-investigator and CEC approval, except where necessary to eliminate apparent immediate hazards to study participants.

Serious Adverse Events (SAE) and Suspected Unexpected Serious Adverse Reactions (SUSAR) are reported according to chapter 10.3.

Amendments are reported according to chapter 2.10

2.3.1 Notification and reporting upon completion, discontinuation or interruption of the clinical trial (ClinO, Art. 38)

The following notifications have to be made by the sponsor investigator to the CEC (EKNZ):

- The completion of the study within 90 days. Completion of a clinical trial is marked by the last participant's final visit.
- The discontinuation or interruption of the clinical trial within 15 days. In the notification, the reasons for the discontinuation or interruption shall be stated.
- The final report shall be submitted within two years after completion or discontinuation of the clinical trial.

2.3.2 Notification and reporting of safety and protective measures (ClinO, Art. 37 and 43)

The following notifications and reporting have to be made by the sponsor-investigator to the CEC (EKNZ):

- If immediate safety and protective measures have to be taken during the conduct of the clinical trial, a notification shall be submitted within 7 days. In the notification, these measures and the circumstances necessitating them shall be stated.
- Annual safety report once a year.

2.4 Competent Authority (CA)

The sponsor will obtain approval from the CA (Swissmedic) before the start of the clinical study.

No changes are made to the protocol without prior Sponsor and CA approval, except where necessary to eliminate apparent immediate hazards to study participants.

Serious Adverse Events (SAE) and Suspected Unexpected Serious Adverse Reactions (SUSAR) are reported according to chapter 10.3.

Amendments are reported according to chapter 2.10.

2.4.1 Notification and reporting upon completion, discontinuation or interruption of the clinical trial (ClinO, Art. 38)

The following notifications have to be made by the sponsor investigator to the CA (Swissmedic):

- The completion of the study within 90 days. Completion of a clinical trial is marked by the last participant's final visit.

- The discontinuation or interruption of the clinical trial within 15 days. In the notification, the reasons for the discontinuation or interruption shall be stated.
- The final report shall be submitted within two years after completion or discontinuation of the clinical trial.

2.4.2 Notification and reporting of safety and protective measures (ClinO, Art. 37 and 43)

The following notifications and reporting have to be made by the sponsor investigator to the CA (Swissmedic):

- if immediate safety and protective measures have to be taken during the conduct of the clinical trial, a notification shall be submitted within 7 days. In the notification, these measures and the circumstances necessitating them shall be stated.
- Annual Safety Report (ASR).

2.5 Ethical Conduct of the Study

The study will be carried out in accordance to the protocol and with principles enunciated in the current version of the Declaration of Helsinki, the guidelines of Good Clinical Practice (GCP) issued by ICH, the Swiss Law and Swiss regulatory authority's requirements. The CEC and the CA will receive annual safety and interim reports and they will be informed about study stop/end.

2.6 Declaration of interest

The authors certify that they have no intellectual, financial or proprietary conflict and are independent.

2.7 Patient Information and Informed Consent

The investigators will explain to each participant the nature of the study, its purpose, the procedures involved, the expected duration, the potential risks and benefits and any discomfort it may entail. Each participant will be informed that the participation in the study is voluntary and that he/she may withdraw from the study at any time and that withdrawal of consent will not affect his/her subsequent medical assistance and treatment.

The participant must be informed that his/her medical records may be examined by authorized individuals other than their treating physician.

All interested persons will be provided with sufficient information to make an informed decision about their participation in the study. The "study information and informed consent form" will be sent by e-mail to interested persons before telephone screening. They will be given time (at least 24 hours) between the receipt of the "patient information and informed consent form" and screening for their decision to participate. For details of the recruiting procedures see chapter 7.2.

The formal consent of a participant, using the approved consent form, must be obtained before the participant is submitted to any study procedure.

The participant should read and consider the statement before signing and dating the informed consent form, and should be given a copy of the signed document. The consent form must also be signed and dated by the investigator (or his designee) at the same time as the participant sign, and it will be retained as part of the study records.

2.8 Participant privacy and confidentiality

The investigators affirm and uphold the principle of the participant's right to privacy and that they shall comply with applicable privacy laws. Especially, anonymity of the participants shall be guaranteed when presenting the data at scientific meetings or publishing them in scientific journals.

Individual subject medical information obtained as a result of this study is considered confidential and disclosure to third parties is prohibited. Subject confidentiality will be further ensured by utilizing subject identification code numbers to correspond to treatment data in the computer files.

For data verification purposes, authorized representatives of the sponsor-investigator, a competent authority (e.g. Swissmedic), or the ethics committee may require direct access to parts of the medical records relevant to the study, including participants' medical history.

2.9 Early termination of the study

The sponsor-investigator may terminate the study prematurely according to certain circumstances, for example:

- ethical concerns;
- insufficient participant recruitment;
- when the safety of the participants is doubtful or at risk, respectively;
- alterations in accepted clinical practice that make the continuation of a clinical trial unwise;
- early evidence of benefit or harm of the experimental intervention.

2.10 Protocol amendments

Substantial amendments are only implemented after approval of the CEC and the CA respectively.

Under emergency circumstances, deviations from the protocol to protect the rights, safety and well-being of participants may proceed without prior approval of the sponsor-investigator, the CEC and the CA. Such deviations shall be documented and reported to the sponsor-investigator, the CEC and the CA as soon as possible.

Other reporting commitments are reported according to the chapters 2.3 and 2.4 and safety issues according to chapter 10.

2.10.1 Reporting to the Competent Ethics Committee (CEC)

Substantial amendments: The following changes are considered to be significant and have to be reported to the CEC by the sponsor investigator (ClinO, Art. 29):

- amendments with effect on the safety and health of the human participants as well as on their rights and obligations;
- deviations from the protocol due to new scientific findings regarding the experimental design, methods, target criteria or statistical evaluation concepts;
- changes of locations or use of additional locations for the implementation of the clinical trial;
- changes of sponsor or investigators.

The sponsor-investigator shall submit to the CEC any application documents specified in Annex 3 of the ClinO, which are affected by the change. At the same time, the sponsor-investigator shall provide information on the reasons for the change. The ethics committee shall reach a decision within 30 days.

Non-substantial changes must be notified to the CEC in the Annual Safety Report (ASR).

2.10.2 Reporting to the Competent Authority (CA)

Substantial amendments: The following changes are considered to be significant and have to be reported to the CA by the sponsor-investigator (ClinO, Art. 34):

- changes of the study medication, its application or administration;
- changes based on new preclinical or clinical data which may affect product safety; or
- changes concerning the production of the IMP, which may affect product safety.

The sponsor shall submit to the CA any application documents specified in Annex 4 of the ClinO, which are affected by the change. At the same time, the sponsor shall provide information on the reasons for the change. The CA shall reach a decision within 30 days after receipt of the complete application documents affected by the change.

Non-substantial changes, which affect the documents submitted to CA, must be communicated to CA as soon as possible.

3 BACKGROUND AND RATIONALE

3.1 Background and Rationale

We have shown previously that combining genome-wide association studies (GWAS) of disease with genetic studies of cognitive and behavioral endophenotypes (i.e. heritable disease components observable also in non-affected individuals) may lead to the identification of compounds that modulate cognitive and behavioral traits relevant to the respective disease (Papassotiropoulos, Gerhards et al. 2013).

Large GWAS of schizophrenia suggest that genetic variations (single nucleotide polymorphisms) in genes encoding potassium channels modify the risk for the development of this disorder (Pardinas, Nalmpanti et al. 2019)

In healthy humans (for methods see (Papassotiropoulos, Stephan et al. 2006, Papassotiropoulos, Henke et al. 2009)) we found that genetic variations in the same genes are also associated significantly with working memory performance, an established cognitive endophenotype of schizophrenia. These findings suggest that this class of genes may play a role in working memory and, consequently, that a pharmacological agent that interacts with the respective gene products may modulate this cognitive trait in health and disease. For phenotype description see (de Quervain, Henke et al. 2003, de Quervain, Kolassa et al. 2007, Heck, Fastenrath et al. 2014).

Here, we aim at investigating if the administration of Fampyra®, an inhibitor of voltage gated potassium channels, affects working memory in healthy humans.

Furthermore we want to investigate whether fampridine has an influence on resting motor threshold (rMT). For the rMT measurement we will use single pulse TMS.

In a study of our research group we could show that resting motor threshold correlated with working memory performance in healthy humans (Schicktanz et al. 2014). In a study investigating rMT in patients with schizophrenia a negative correlation between rMT and working memory was confirmed (Bridgman, Barr et al. 2016).

Literature in relation to the effect of fampridine SR on rMT is scarce, and to our knowledge there is no study published examining healthy participants.

Neurophysiological studies in MS patients are controversial. One placebo controlled study of 40 MS patients found no effect for any of the clinical or electrophysiological measures and esp. on rMT after 8 weeks treatment of fampridine 10 mg bd (Marion, Leonid et al. 2020). Another one failed to show an effect on rMT after 14 days treatment with 10 mg fampridine bd in 20 MS patients (Ahdab, Shatila et al. 2019) Krushkov on the other hand found significant effects on rMT after treating 18 MS patients for 5 days with 2x10mg fampridine i.m. (Krushkov, Shotekov et al. 2006).

3.2 Investigational Product (treatment) and Indication

Active study medication consists of 7 tablets of fampridine 10mg sustained release (SR) formulated for oral administration (Fampyra®), manufactured by

Alkermes Pharma Ireland Ltd, Monksland,
Athlone, Co.
Westmeath, Ireland

Biogen Idec (Denmark)
Manufacturing ApS, Biogen Idec Allé 1, Hillerod,
DK-3400, Denmark

Fampridine is a potassium channel blocker indicated to improve walking in patients with multiple sclerosis. This was demonstrated by an increase in walking speed (Jeffery and Pharr 2010). Fampyra® whose active ingredient is prolonged release 4-aminopyridine (4-AP), is a broad-spectrum voltage-dependent K+ channel blocker, which can increase action potential propagation in demyelinated axons (Chwieduk and Keating 2010). The compound, which is lipid soluble, crosses the axon membrane, blocking the exposed K+ channels; this decreases K+ loss, increases action potentials and conduction, and promotes synaptic and neuromuscular transmission of nerve signals (Chwieduk and Keating 2010). Prolonged release

fampridine is completely absorbed from the gastrointestinal tract and this more slowly than immediate release (IR) fampridine (Fernandez, Berger et al. 2012). This decreases peak plasma concentration by 50%, which consequently reduces dose-related adverse effects. (Fernandez, Berger et al. 2012). SR fampridine should be administered under fasting conditions: administration with food increases peak plasma concentration, leading to increased risk of adverse effects (Weir, Torkin et al. 2013), see also prescribing information of Fampyra® SR. Tablets should only be taken whole; not divided, crushed, chew, or dissolved.

Apparent first-order terminal elimination half-life t1/2 of fampridine SR is 5.2 to 6.5 h (Henney, Faust et al. 2011). There will be a wash out period of at least 8 days equalling over 30 half-lives of the active substance fampridine SR ($t_{1/2}$=6h).

3.3 Preclinical Evidence

In rats, fampridine was found to improve short-term memory performance of old, but not young, animals (Barnes, Eppich et al. 1989).

Further it was found to be able to enhance memory as demonstrated in a 72-h retention of passive avoidance task (Haroutunian, Barnes et al. 1985). The effect of fampridine was also investigated in a mouse model of HIV-1 encephalitis. These mice show impaired spatial memory in radial arm water maze tests. Electrophysiology studies revealed a reduction of long-term potentiation (LTP) in the CA1 region of the hippocampus. Systemic administration of fampridine blocked HIV-1-associated reduction of LTP and improved animal performance in the radial arm water maze (Keblesh, Dou et al. 2009).

3.4 Clinical Evidence to Date

Fampridine was investigated in patients with Alzheimer disease. In one study it partially reversed memory deficits (Wesseling, Agoston et al. 1984). In a second study, results indicated no significant difference in total Alzheimer Disease Assessment Scale Scores (Davidson, Zemishlany et al. 1988).

In patients with Multiple Sclerosis fampridine was investigated in several clinical studies. MS can cause progressive walking impairments, which are improved by Fampridine. Through a blockade of potassium channels, fampridine restores nerve impulse conduction (Tseng, Li et al. 2016), facilitates and enhances synaptic transmission, retards the postsynaptic action potential repolarization (Kasatkina 2016), and increases the release of acetylcholine (Lugaresi 2015).

Fampridine also improves cognitive functions in MS. In a clinical trial MS patients receiving a maximum dose of 0.7 mg/kg of weight per day achieved significantly higher scores in attention span, verbal fluency, planning and graphics and constructive motion (Arreola-Mora, Silva-Pereyra et al. 2019). Another study showed improvement in information processing speed in MS patients receiving 10 mg twice daily for 12 weeks (De Giglio, De Luca et al. 2019). In an earlier study the application of 7.5-52.5 mg of immediate release fampridine over 1-5 days showed motor and visual improvements in MS (Stefoski, Davis et al. 1991).

These findings are consistent with another study, where 40 mg/day led to improvements in attention and concentration in MS patients (Smits et al. 1994). In contrast, no cognitive improvement was reported with a dose of 30–32 mg/day (Rossini et al. 2001; Romani et al. 2004). Considering a long period of administration (9–12 months) at a dose of 20 mg per day, a beneficial effect on both processing speed and working memory has been observed (Pavsic, Pelicon et al. 2015).

3.5 Dose Rationale

The maximum recommended dose of fampridine SR is 10 mg twice daily (approximately 12 hours apart) without food (see prescribing information Fampyra®).

In healthy individuals a single dose of fampridine SR 10 mg achieved a mean maximum plasma concentration (C_{max}) of 21.6 ± 3.89 ng/ mL, with a mean time to maximum plasma concentration (T_{max}) of 3.2 ± 1.5 hours (Smith, Swan et al. 2010). Steady state of fampridine SR will be reached after 5 half-lives of fampridine equalling approx. 30 h (Arzneimittelinformation Swissmedic Fampyra®).

Fampridine is lipid soluble and able to pass the blood–brain barrier (Lemeignan, Millart et al. 1984) and therefore able to reach structures of the prefrontal cortex involved in working memory functions.

Cerebrospinal fluid levels of fampridine peak 30–60 min following peak serum concentration (Donovan, Halter et al. 2000).

Previous studies examining long-term use up to several weeks reported pro-cognitive effects with 10 mg fampridine SR twice daily (see 3.4).

We are interested in the effects on working memory after acute administration of 10 mg fampridine SR as well as in effects after repeated administration of 10 mg fampridine SR twice daily.

We will therefore examine working memory after the first dose of 10 mg fampridine SR (i.e. after a 4 h waiting time to reach maximum plasma concentration)).

Second testing will take place after 3.5 days of twice daily application of 10 mg fampridine SR.

3.6 Explanation for choice of placebo

Placebo is chosen to assess effects of fampridine on memory processes in a cross-over design. Placebo consists of an identically looking tablet manufactured by

Losan PharmaLosan Pharma GmbH
Otto-Hahn-Straße 13
79395 Neuenburg am Rhein
Germany

consisting of additives that are very similar to those used for the verum formulation:

Placebo Tabletten zu Fampyra® 10 mg

Beschreibung:			
Spezifikation:	**Bezeichnung Inhaltsstoff:**		**Menge/Einheit:**
Tablettenkern:			
Ph. Eur.	Cellulose mikrokristallin 200		289 mg
Ph. Eur.	Cellulose mikrokristallin 101		122 mg
Ph. Eur.	Hypromellose 6		47 mg
Ph. Eur.	Magnesiumstearat pflanzlich		9,4 mg
Ph. Eur.	Siliciumdioxid hochdispers		2,4 mg
		Summe:	470 mg
Tablettenfilm:			
Ph. Eur.	Hypromellose 6		5,90 mg
Ph. Eur.	Polyethylenglycol		0,90 mg
Ph. Eur.	Titandioxid		1,20 mg
		Summe:	8 mg
		Gesamtsumme:	478 mg

3.7 Risks / Benefits

Fampridine application

Although a short term application of 10 mg fampridine twice daily in healthy participants is generally well tolerated (March and Cardi 2009), participants will be informed of the possible risks associated with taking fampridine. Specific risks that participants need to be aware of are listed below:

- The most frequently occurring AEs included dizziness, headache, insomnia and gastrointestinal discomfort. No discontinuations due to AEs were reported in a study with healthy subjects after administration of a single 10 mg dose (Smith, Swan et al. 2010) as well as after 5 days of twice daily fampridine SR 10 mg in healthy subjects (March and Cardi 2009).
- Fampridine is excreted primarily unchanged in the urine (Blight and Henney 2009). Tolerability and incidence of AEs appear to be dose related, occurring more frequently at plasma concentrations > 100 ng/mL (Van Diemen, Polman et al. 1993). We will therefore exclude subjects with renal impairment to avoid higher plasma levels. To assess renal impairment, we will perform creatinine and uric acid levels in plasma.

- Fampridine is excreted primarily through the organic cation transporter 2 (OCT 2). OCT 2 inhibitors or substrates (e.g. Cimetidine, Carvedilol, Propranolol and Metformin) may inhibit elimination of fampridine and increase the risk of adverse events. Subjects with concomitant use of these drugs are excluded from participation.
- Fampridine can cause seizures. We will exclude people with a history of seizures and subjects at risk of lowered seizure threshold due to e.g. sleep deprivation or alcohol withdrawal after alcohol abuse.
- Despite the fact that fampridine at therapeutic and supratherapeutic doses is not associated with QT prolongation in healthy subjects (March and Cardi 2009) we will exclude subjects with any clinically significant ECG abnormalities or history of cardiovascular disease.
- Fampridine might cause nausea and dizziness and have an additive effect with alcohol. Participants will be advised to avoid alcohol, illegal substances, and any medication (especially psychoactive) due to possible interactions during the course of the study.
- An increased risk of bacterial and viral infections is described in the prescribing information. In a study with 47 healthy subjects receiving 10 mg twice daily doses of fampridine SR over 5 days no viral or bacterial infections were reported (March and Cardi 2009).
- Subjects will be warned about engaging in activities requiring mental alertness, such as driving a car or operating appliances, machinery, etc. Driving a motor vehicle or operating machinery is not allowed during each intervention period of 4 days (96 h) and subjects have to agree to use public transportation or taxi after medication intake for this time period.
- In several in vivo and in vitro studies there was no evidence of drug-related carcinogenicity or mutagenicity (prescribing information Fampyra®).
- Administration of fampridine to animals during pregnancy and lactation resulted in decreased offspring viability and growth at doses 6.8 times the maximum recommended human dose (MRHD) of 20 mg/day. In developmental toxicity studies in rats and rabbits, fampridine was administered orally at doses up to 10 and 5 mg/kg/day, respectively, during the period of organogenesis. These doses are approximately 5 times the MRHD. No evidence of developmental toxicity was found in either species at the highest doses tested, which were maternally toxic. In addition, a case study about a favorable outcome of a pregnancy after exposure with fampridine in the first month of conception is available (Maillart, Gout et al, 2016) For additional safety data, we refer to the prescribing information Fampyra®.

  All female of childbearing potential should use adequate contraception starting 10 days before each intervention period and during intervention period, such as an oral, injectable, or implantable contraceptive or contraceptive devices (IUP). To minimize the risk of exposure to IMP we will perform a pregnancy test before administration of study drug on test day 1 and 3.
- All subjects will be informed about possible side effects of medication and advised to inform the investigator about observed side effects due to possible individual differences in metabolism of study medication.

rMT measurement

- Seizures are the most serious possible TMS-related adverse events. Only few cases of TMS induced seizures have been reported so far out of hundreds of thousands examined subjects, and the vast majority of seizures were induced during repetitive TMS (Rossi, Antal et al. 2021). Single-pulse TMS, the method used here, is considered to have no significant risk (Rossi et al., 2009) and studies investigating physiological mechanisms of corticocortical plasticity in healthy subjects, which did not require repeated sessions over several days in order to reach a clinical effect did not show major AE, including seizure occurrence, Thus, there should not be any special concern in studies of this type (Rossi, Antal et al. 2021). We will exclude all persons having experienced a seizure in the past and/or with positive family history (first-degree relatives) of seizures.
- A number of medications have been reported to increase risk of seizure in clinical populations (Stultz, Osburn, et al. 2020) and it was previously assumed that their use in combination with repetitive TMS may confer heightened risk for seizure induction (Rossi et al., 2009). However, empirical evidence for this risk is lacking, and the observed seizure rate even in repetitive TMS patients is extremely low overall despite that the majority of them were on CNS-active medications (Rossi, Antal et al. 2021).

- Risks concerning single pulse rMT measurement in combination with fampridine SR were monitored in MS patients: In recent studies no SAEs during TMS measurements were reported (Marion, Leonid et al 2020), (Ahdab, Shatila et al. 2019), (Krushkov, Shotekov et al. 2006).
- Compared to seizure, syncope is more likely to occur during a TMS investigation, but this is a rare event too. No systematic studies addressed the relative incidence of the two phenomena during TMS, but this is a common experience in many labs (Groppa, 2012). Vasodepressor (neurocardiogenic) syncope is a common reaction to anxiety and physical discomfort and it can take place following TMS, as with many other non-invasive or minimally invasive medical procedures. The cardinal feature that distinguishes syncope from seizure is rapid recovery of full consciousness within few seconds and not minutes (Lin, Ziegler et al. 1982). We will exclude individuals with a history of repeated syncope.
- Other side effects of single-pulse TMS investigations are mild and, when present, transient. Most of subjects do not complain about discomfort, especially when they are familiarized with the TMS procedure. Some may experience surprise looking at their hand/arm twitching (personal communication Schicktanz 2021).
- Both, patient and examiner will always wear earplugs during diagnostic TMS to prevent transient auditory threshold changes, which are more likely to occur during repetitive TMS, but are theoretically possible even with single pulse application (Rossi, Antal et al. 2021). We will exclude persons with hearing impairment or tinnitus.
- For safety reasons we added a list of exclusion criteria concerning TMS in general (single-pulse, paired-pulse and repetitive TMS). We will use the questionnaire for screening of subjects before TMS investigations that has been developed on a consensus conference by considering the safety and ethical guidelines for the use of TMS in clinical practice and research (Rossi, Hallett et al. 2011) (Groppa, Oliviero et al. 2012).
- All TMS investigations will be performed by specially trained persons.
- The participant is always allowed to stop the TMS measuring if he/she feels ill at ease.

Benefits

The benefit of the study lies in the gain of knowledge regarding the translation of human genetic studies in pharmacological intervention studies and, more specifically, in the potential to find working memory improvements by fampridine. Such a finding may have important clinical implications with regard to treatment of working memory deficits in neuropsychiatric disorders.

3.8 Justification of choice of study population

Genetic variability in genes encoding potassium channels are related to differences in working memory performance in healthy young subjects (Heck, Fastenrath et al. 2014). We therefore decided to investigate the effects of fampridine on working memory in healthy young subjects.

4 STUDY OBJECTIVES

4.1 Overall Objective

The overall objective of this study is to evaluate if fampridine has an influence on cognitive functions in healthy young participants.

4.2 Primary Objective

The primary objective of this study is to evaluate if fampridine has an influence on working memory in healthy young participants.

4.3 Secondary Objectives

The secondary objectives are to assess the influence of fampridine on different working memory functions, attention, processing speed, fluid intelligence and resting motor treshold (rMT).

4.4 Safety Objectives

The study aims to assess tolerability of study medication in terms of incidence of side effects e.g. headache, gastrointestinal discomfort and dizziness and the rate of AEs.

5 STUDY OUTCOMES

5.1 Primary Outcome

- High-load working memory performance (3-back (d') as assessed by a letter n-back task) after repeated intake of study medications (visit 3 and 5).

5.2 Secondary Outcomes

- High-load working memory performance (3-back (d') as assessed by a letter n-back task) after acute intake of study medication (visit 2 and 4)
- rMT measurement using TMS after repeated intake of study medication (visit 3 and 5)

Performance in other working memory-related tasks and other n-back outcomes after acute and repeated intake of study medication

- Reaction time (for correct 3-back responses)
- Performance in a 0 back task as a measure of attention
- Symbol Digit Modalities Test, SDMT (Smith 2013, 13th edition)
- BOMAT - advanced (Hossiep/Turck/Hasella, 1999)
- Digit span forward and backward performance (von Aster 2006)

5.3 Safety Outcomes

- Headache, dizziness and gastrointestinal discomfort (see chapter 10.1.1) will be recorded and rated by the participants using visual analogue scales.
- AE-recording
- Vital signs

5.4 Assessment of potential confounders

- Tiredness and motivation will be rated by the participant using visual analogue scales.

6 STUDY DESIGN

6.1 General study design and justification of design

Pharmacological proof-of-concept study on the effects of 10 mg fampridine (twice daily oral administration) on physiological processes, in particular working memory, in healthy participants using the following design:

- randomized
- double-blind (participant, study team)
- placebo-controlled
- cross-over
- counter-balanced

The duration of the study from first participant in to last participant out is approx. 24 months. Duration for an individual study participant will be at most 7 to max. 15 weeks, depending on the duration of the wash out period.

There will be two intervention periods of 3.5 days during which medication (fampridine /placebo) will be applied separated by a wash out period of 8 (days between Visit 3 and Visit 4; equivaling over 30 half-lives of the study drug) up to 82 days (12 weeks).

Two test days will take place during each intervention period: one on the first day and one on the last day of each period during which cognitive functions under the influence of fampridine/placebo will be assessed using parallel versions of tests.

At the beginning of test day 1 (visit 2), each subject will be randomized to receive either placebo or fampridine first.

After the administration of study medication (visits 2-5) there will be a 4-h waiting period allowing the medication to reach peak cerebrospinal fluid levels (Hayes 2004). After this time period, the cognitive test battery will be administered.

At the end of each visit, participants will be asked about their actual wellbeing. If there are no complaints or symptoms, participants can be dismissed. In case of minor health problems, the participant will have to stay in the study center until recovery under medical supervision. In case of more severe incidents staff acts according to chapter 10.4.6.

The study will end for the subjects in the evening of the second test day of the second intervention period (visit 5). The duration for the different study parts see table 1 (Duration of Study parts, page 12).

6.2 Methods of minimizing bias

To minimize bias, participants will be randomly allocated to treatment groups (e.g. starting with fampridine or placebo) and study will be double-blinded.

6.2.1 Randomization

After pre-examination on visit 2, each participant will be assigned to a randomization number. Each eligible participant will be allocated to one of the two randomization groups (Placebo/Verum or Verum/Placebo) according to the maximum tolerated imbalance (MTI) procedures. This allocation will be generated automatically by secuTrial®. This procedure ensures that participants will be randomly allocated to the two groups, while accounting for sex as a covariate of potential importance.

The replacement of all Drop-Outs will be carried out after assignment of all 44 medication kits is completed. There will be replacement of Drop-Outs until data of approximately 44 participants are completed.

6.2.2 Blinding procedures

The IMP and the placebo are identically looking tablets to ensure double-blind conditions.

The pharmacy will prepare one kit of study medication per participant. Each kit will consist of two intransparent plastic containers, each containing either seven tablets of blistered fampridine and a drying agent or unblistered placebo and a drying agent. It is necessary to keep Fampyra® blistered as long as possible to prevent any contact of fampridine with humidity. After unblistering Fampridin® will stay stable

for seven days in a closed container while adding a drying agent (professional / prescribing information Germany Fampyra® SR). Each kit of study medication will be labelled with a randomization number. A staff member not involved in the conduct of the study will open the kit and the plastic container corresponding to the respective intervention period in a separate room. If Verum is inside he will unblister all the verum tablets and hand over the first tablet to the subject, who is waiting in a different room. If Placebo is inside the staff member may directly hand over the first tablet to the subject, who is waiting in a different room. Afterwards the subject will be handed over the reclosed container for the intake of the study medication at home. During each intervention period a tablet must be taken every 12 hours for 5 times at home. The last (7^{th}) intake of the study medication will be again in the study center. Therefore, the subject must bring along the container including the last tablet at the second study day of each intervention period.

6.2.3 Other methods of minimizing bias

The investigators of the test days will be trained and will follow detailed working instructions to ensure that each test day will be conducted in a highly standardized way.

Study team members involved in testing, data acquisition or data analysis are blinded. The study team member preparing the IMP at the testing days has no other outcome-related tasks in the study.

The administration of medication will take place at the same time frame (8-9 a.m.) for each participant. Testing will take place in the same location at the premises of the Division of Cognitive Neuroscience of the University of Basel for each participant and during the same time of the day to minimize the influence of diurnal differences in cognitive test performance.

6.3 Unblinding Procedures (Code break)

Unblinding will be permitted under these circumstances

- SAE
- AE if knowledge is crucial for further medical approach

The investigator or a representative can perform the unblinding of the participant at any time. To ensure that unblinding is possible at any time, there is a unblinding procedure implemented in secuTrial®. The investigator or a representative will report to the sponsor-investigator and document the unblinding. Not involved study team members will not be informed about the result.

7 STUDY POPULATION

7.1 Eligibility criteria

Participants fulfilling all of the following inclusion criteria are eligible for the study:

- male or female
- generally healthy
- normotensive (BP between 90/60mmHg and 140/90mmHg)
- BMI between 19 and 29,9 kg/m^2
- aged between 18 and 30 years
- fluent German-speaking
- IC as documented by signature
- At least double vaccination against Covid-19

The presence of any one of the following exclusion criteria will lead to exclusion of the participant:

- contraindications to the class of drugs under study, e.g. known hypersensitivity or allergy to 4-aminopyridine
- use of potassium channel blockers within the last 3 months
- concomitant treatment with OCT 2 inhibitors and -substrates (e.g. cimetidine, propranolol)
- acute or chronic psychiatric disorder (e.g. major depression, psychosis, somatoform disorder, suicidal tendency)
- acute cerebrovascular condition
- history of seizures
- risk of lowered seizure threshold (due to e.g. sleep deprivation, withdrawal of alcohol after alcohol abuse)
- renal impairment
- history of malignant cancers
- walking problems (e.g. due to dizziness)
- other clinically significant concomitant disease states (e.g. hepatic dysfunction, cardiovascular disease, diabetes, asthma,
- bradycardia < 50/min during clinical examination
- clinically significant laboratory or ECG abnormality that could be a safety issue in the study
- known or suspected non-compliance (e.g. missing more than one dose of study medication per intervention phase)
- drug or alcohol abuse
- inability to follow the procedures of the study, e.g. due to language or psychological problems of the participant
- participation in another study with an investigational drug within the 30 days preceding and during the present study
- prior participation (less than two years ago) in a study investigating working memory (notably the n-back task)
- enrolment of the investigator, his/her family members, employees and other dependent persons
- smoking (>3 cigarettes per day)
- intake of psychoactive drugs (e.g. benzodiazepines, antidepressants, neuroleptics)
- pregnancy or breast feeding
- experiencing a syncope during basal rMT measuring

Exclusion Criteria concerning TMS measurement

- metal in the brain, skull or elsewhere in the body (e.g., splinters, fragments, clips, etc.)
- implanted neurostimulator (e.g., DBS, epidural/subdural, VNS)
- cardiac pacemaker or intracardiac lines
- medication infusion device
- piercings, pivot teeth (retainers are no exclusion criterion)
- tattoos (head area) less than 3 months old or older than 20 years
- condition after neurosurgery
- hearing problems or tinnitus

- not able to sit still due to tremor, tics, itching
- History of repeated syncope
- head trauma diagnosed as concussion or associated with loss of consciousness
- diagnosis of epilepsy, or a convulsion or a seizure in the past of the participant or his family
- TMS in the past showing problems
- MRI in the past showing problems
- surgical procedures to spinal cord
- spinal or ventricular derivations

Remark: It is necessary to ensure that the metal nose piece of surgical masks, if they have to be worn as protection against Covid-19 infection, is not ferromagnetic (Bikson, Hanlon et al. 2020).

7.2 Recruitment and screening

Study participants will be searched in the German speaking part of Switzerland using the following websites mcn.unibas.ch and markt.unibas.ch,.

We may also invite former participants of unrelated studies of the Divisions of Cognitive and Molecular Neuroscience interested in further study participation by e-mail.

After contacting us, the interested person will be sent the “Participant information and informed consent form” together with some administrative information. If the person is still interested in participating, a study team member will call the interested person to inform him/her about the study and answer his/her questions. Afterwards, a pre-screening with a check of the main in- and exclusion criteria will be performed. If the interested person meets all inclusion criteria and none of the exclusion criteria and is interested in participating, the screening visit will be fixed.

All interested persons will be advised not to come to the study center in case of common cold or flu like symptoms but fix an alternative date. The telephone screening documents will be filed as part of the Source Data (SD) in the participant’s dossier. The telephone screening documents of participants not eligible for a study participation will be shredded. All steps will be documented and we will assess the number of participants, who were not eligible during pre-screening.

For screening procedures see chapter 9.4.1.

A study compensation of CHF 800 including travel expenses will be paid at the end of the second test period. Screening Failures and Drop-Outs receive a pro rata temporis compensation.

7.3 Assignment to study groups

After informed consent, each participant will be assigned to a participant number. Then, eligible participants will be allocated to the treatment group (i.e. either fampridine or placebo first) following the order of the randomization list. The randomization number will be listed in the SD, in the eCRF and in the Drug Accountability Log.

7.4 Criteria for withdrawal / discontinuation of participants

Participants have the right to withdraw from the study at any time for any reason without being obliged to give reason. There will be a final examination after withdrawal for whatever reason. The investigator also has the right to withdraw participants from the study if it is in the best interest of the participant.

The following reasons result in withdrawal:

- Adverse events challenging the health of the participant if continuing the study
- Adverse events prohibiting cognitive testing or rMT-measurement, e.g.
    - headache, dizziness and nausea more than mild or
    - having slept less than 4 hours during the last night (Visits 3 and 5).
- Participant being ill at ease during rMT measuring
- Intake of psychoactive substances within 3 days before test days (e.g. benzodiazepines, antidepressants, neuroleptics, cannabis)
- Non-Compliance
- Severe protocol violations
- Administrative troubles
- Positive pregnancy test (test day 1 or test day 3)

- Alcohol intake within 12 hours before start of all visits

Withdrawal date and reason will be listed in the participant enrolment log. Safety data will be analyzed for all participants, who received at least one dose of study medication. There will be no anonymization of the data. For information about data handling see chapter 12.3.

7.5 Contraception and pregnancy

All female participants of childbearing potential should use adequate contraception starting 10 days before each intervention period and during intervention period, such as an oral, injectable, or implantable contraceptives or contraceptive devices (IUP). To minimize the risk of exposure to IMP we will perform a pregnancy test before administration of study drug on test day 1 and 3.

If against all demanded contraception, a pregnancy should occur the pregnant participant must be withdrawn immediately from the clinical study. Any pregnancy during the treatment phase of the study and within 30 days after discontinuation of study medication will be reported to the Sponsor-Investigator within 24 hours.

The pregnancy will be followed-up like an SAE. A document for the initial registration is available, as well as one for the end of the first trimenon. All other relevant information that we should receive from the participant will be documented. Information about course and outcome of pregnancy from the treating gynecologist might be asked after release from the confidentiality by the participant.

A follow-up of mother and child after the first trimenon is not foreseen, as no risks are expected for the development of the unborn thereafter.

8 STUDY INTERVENTION

8.1 Identity of Investigational Products

8.1.1 Experimental Intervention

Active study medication consists of 7 tablets of fampridine 10 mg formulated for oral administration taken in the morning and evening 12 h apart without food. Tablets must be administered whole: do not divide, crush, chew, or dissolve Fampyra® retard tablets.

There will be a washout period of at least 8 days equaling over 30 half-lives of the active substance fampridine ($t_{½}$ = 6 h) between experimental and control intervention and up to 82 days (12 weeks) depending on the individual scheduling of each subject.

8.1.2 Control Intervention

Identically looking placebo tablets consisting of widely identical additives formulated for oral administration.

8.1.3 Packaging, Labelling and Supply (re-supply)

Packaging, labelling and supply will be provided by the Pharmacy of the University Hospital Basel (see chapter 6.2.2).

8.1.4 Storage Conditions

The IMP will be stored under 25° C and protected from light and humidity until initiation at the pharmacy Thereafter the IMP will be stored for the entire study until last participant out at the same conditions in our division. The storage temperature will be controlled with LogTag temperature recorder.

8.2 Administration of experimental and control interventions

8.2.1 Experimental Intervention

See chapter 8.1.1

8.2.2 Control Intervention

See chapter 8.1.2.

8.3 Dose

No modifications needed.

8.4 Compliance with study intervention

Each subject will be provided with a personal login to secuTrial® to be able to document the intake of the investigational product at a time. A study team member will send a note if entry is missing to remind the subject of intake of the investigational product.

All subjects have to return the container on the following test days in order to control the container for not used tablets and for correct disposal of remaining tablets after study end.

Blinding is warranted (see chapter 7.6, Randomization and Blinding).

8.5 Data Collection and Follow-up for withdrawn participants

Not needed. Subject withdrawn because of (S)AE will be followed up until resolution or stabilization.

8.6 Trial specific preventive measures

In case of headache, pain or fever, paracetamol is allowed and has to be recorded as concomitant medication.

8.7 Concomitant Interventions (treatments)

Treatments with OCT 2 inhibitors or substrates (e.g. cimetidine, propranolol) are prohibited during the course of the study as they may inhibit elimination of fampridine and increase the risk of adverse events.

All concomitant treatment will be recorded in the eCRF.

8.8 Study Drug Accountability

The supplies from and the returns to the pharmacy of the USB for destruction will be documented at the pharmacy and also by the study coordinator on the site in the IMP accountability log. This log documents the IMPs received from the pharmacy, the kits used by each participant, and the return to the pharmacy of used and unused kits.

This documentation includes the following information: delivery date, delivered kits, batch number, expiry date.

The administration of the IMP to the participants is recorded in the participant dossier.

8.9 Return or Destruction of Study Drug

Used and unused kits will be returned to the pharmacy of the USB for destruction. The pharmacy will destroy and dispose them duly in accordance to hospital policy and the local directives.

9 STUDY ASSESSMENTS

9.1 Study flow chart(s) / table of study procedures and assessments

See study schedule (Table 1, Duration of study parts, page 12)

9.2 Assessments of outcomes

9.2.1 Assessment of primary outcome

- High-load working memory performance will be measured as follows:
  We will use the letter n-back task (Heck, Fastenrath et al. 2014)) which includes a 3-back task assessing working memory. The 3-back task requires participants to respond to a letter repeat with two intervening letters (for example, S−m−b−s−g…). Performance will be quantified with the d' measure controlling for false positives. We will use parallel versions (different sequences) for the four test days. Primary outcome will be performance after repeated intake of study medication.

9.2.2 Assessment of secondary outcomes

- High-load working memory performance(see above) after acute intake of study medication.
- Resting motor threshold (rMT) will be measured during screening procedures and after repeated intake with a biphasic magstim Rapid2 stimulator (The MAGSTIM® Company Ltd, Whitland, UK) and a 70 mm figure-of-eight coil.
  rMT will be determined by measuring the motor evoked potential (MEP) in the abductor digiti minimi according to (Rossini 2001). rMT will be defined as the lowest stimulation intensity by stimulating the primary motor cortex of the left or right hemisphere required to induce an MEP in the abductor digiti minimi of the dominant hand in at least 5 out of 10 trials. In case of a relaxed target muscle, a positive MEP is defined as an MEP with $\geqslant$50 µV peak-to-peak amplitude (Rossini 2001).
  The coil will be held tangentially to the skull over M1, approximately four cm lateral and one cm anterior to the Vertex. Additionally, the coil-handle points backwards at an angle of 45° to the corresponding parasagittal line. Then the coil will be shifted systematically in steps of approximately one cm in anterior, posterior, lateral and medial direction. Stimulation will be performed at an initial output intensity of 30% of maximum stimulator's output (MSO) and will be increased in steps of 10% until a twitch is detected. Thereafter, 10 pulses will be applied. If less than five MEPs are observed, stimulator output intensity will be increased by 5 %. Depending on the result, intensity will be further decreased or again increased by steps of two and then one percent until the lowest stimulator intensity is found. Otherwise stimulator output intensity is first decreased 5% and thereafter increased or decreased by steps of two and then one percent until rMT is found. The participant is always allowed to stop the measuring, if he/she feels ill at ease.

Performance in other working memory-related tasks and other n-back outcomes will be measured both after acute and repeated study medication as follows:

- Reaction time (for correct 3-back responses).
- Performance in a 0-back task (d' measure controlled) as a measure of attention. We will use parallel versions (different sequences) for the four test days
- Symbol Digit Modalities Test, SDMT (Smith 2013, 13th edition), a processing speed test. The test consists of the presentation of a series of 9 symbols, each of them is paired with a single digit, labelled 1–9, in a key at the top of a sheet. The remainder of the page has a pseudorandomized sequence of the symbols and the participant must respond with the digit associated with each of these as quickly as possible. The score is the number of correct answers in 90 seconds. The administration of SDMT will be preceded by a learning sequence at both timepoints. We will use parallel versions for all test days.
- Bochumer Matrizentest (BOMAT – advanced; Hossiep/Turck/Hasella, 1999, 1st edition), matrix reasoning. We will administer the BOMAT to measure fluid intelligence (Gf) consisting of 40 items. Parallel versions will be used for the four test days. To create four versions, we divided the original BOMAT versions A and B each into two versions (every second item) with 20 items each. We will use a time-limited version (15 instead of 40 minutes) to avoid a ceiling effect. The total score is calculated by summing the correct solutions, ranging between 0 to 20.

- Working memory will be also assessed with the digit span task, a subtest of the “Wechsler Intelligenztest für Erwachsene“ (WIE; (von Aster 2006)). Total scores for digit span forward and backward will be calculated as described in the manual of the WIE. Parallel version will be used for the four test days.

9.3 Assessment of safety outcomes

The following known side effects will be recorded using visual analog scales: headache, gastrointestinal discomfort, dizziness

Adverse events: All clinically relevant (S)AEs occurring after the participant has signed the informed consent until follow up visit will be fully recorded in the participants eCRF. Description of (S)AE, time of onset, duration and resolution. Assessment of intensity, relationship to study drug and measures taken. For documentation of (S)AEs see section 10.

Laboratory parameters: No laboratory parameters will be assessed after screening.

Vital signs: Vital signs (BP, HR) will be taken as described in screening procedures. Blood pressure and heart rate will be used as baseline before intake of study medication, during waiting period, before testing and before end of visit to control for side effects of the medication.

9.4 Procedures at each visit and during treatment periods

9.4.1 Screening visit

During the screening visit the investigator explains to the participant the aims of the study, the study procedures, the drug under investigation and potential risks (see also chapter 3.7 risks and benefits). Written informed consent will be obtained from all participants without limiting time for signature. Participants who are candidates for enrolment into the study will be evaluated for eligibility during the screening visits by the investigator (inclusion, exclusion criteria). Screening visit will take place within 2 weeks before first test visit and consists of

- Assessment of personal and family history
- Psychosocial assessment
- Assessment of medication history
- Physical examination

The following questionnaires will be used to assess psychiatric disorders:

- Depressive symptoms will be assessed with the self-rating questionnaire MADRS-S based on the MADRS (Schmidtke, 1988). This scale consists of 9 items assessing subjects' mood, feelings of unease, sleep, appetite, ability to concentrate, initiative, emotional involvement, pessimism and zest for life. Each item is scored between 0 and 3. The total score is calculated by summing the answers of the nine items, ranging between 0 and 27 (higher scores indicate increased impairment). A score of ≥8 results in exclusion. In case of suicidal tendency (Item 9 > 1), the subject is excluded and advised to consult a psychologist/psychiatrist. We will provide subjects with addresses.
- Self-assessment questionnaire covering mental health, drug and alcohol consumption, sleeping habits and some sociodemographic information.

The investigator decides, whether there are contraindications for the administration of study medication.

The medical examination performed covers:
Vital signs (blood pressure / heart rate) will be taken with the participant having been in a seated position for at least 5 minutes.

- Height / weight (for calculation of BMI)
- Physical examination; findings will be recorded as “normal” or “abnormal”.
- Twelve lead ECG with formal readings (Wilson, Einthoven, Goldberger), device: Schiller AT10, will be taken and rated by a specialist familiar with the study medication at the Phase I Research Unit (PRU) University Hospital Basel (see contract with PRU).
- Venous blood samples (2,7 ml and 4.7 ml) will be taken at the Phase I Research Unit University Hospital Basel to measure:

- Blood count (incl. platelets) / hemoglobin / hematocrit
- Blood chemistry including: sodium, potassium, chloride, calcium, phosphate, creatinine, uric acid, bilirubin, alkaline phosphatase, LDH, AST, ALT, GGT, pancreatic amylase, albumin, total protein (+Alb./Glob.), C-reactive protein, creatine kinase.

Laboratory values outside the reference range of the laboratory and considered clinically relevant will lead to exclusion of the subject from study participation.

Blood samples will be analyzed by the laboratory of University Hospital Basel. Blood samples will be discarded directly after analysis.

Clinically significant abnormal ECG readings will result in exclusion of the subject.

Resting Motor Threshold (rMT) will be determined as described in chapter 9.2.2. During screening a handedness questionnaire (short version of Edinburgh Handedness Inventory (EHI)) will be administered, as rMT will be measured at the dominant hand.

All female of childbearing potential will be requested to use adequate contraception starting 10 days before each intervention period and during intervention period, such as an oral, injectable, or implantable contraceptive or contraceptive devices (IUP). To minimize the risk of exposure to IMP we will perform a pregnancy test before administration of study drug on test day 1 and 3.

Screening failures equal to participants not meeting all inclusion criteria or meeting one or more of the exclusion criteria. Excluded participants will be listed on a screening failure log. The whole screening will be repeated if the wash out period (time between test day 2 and 3) lasts more than 26 days.

9.4.2 Test days (visits 2-5)

At the beginning of the test days 1 and 3, all female participants will perform a pregnancy test. Pregnancy test will be performed in a urine sample according to instructions of Axaclear, Axapharm. Axaclear is a one-step hCG (monoclonal anti-hCG antibodies) urine pregnancy test used for qualitative (visual) determination of hCG in urine specimen for early detection of pregnancy. The pregnancy tests will be stored with IMP at the same conditions. Additionally women will be requested again to use an adequate contraception during the wash-out period.

There will be a short examination consisting of vital signs, assessment of concomitant medication, sleep duration, self-assessment questionnaire covering drugs and alcohol consumption and adverse events.

Intake of study medication will take place at the same time point for each subject. We will provide a continental breakfast to be eaten 2 hours after intake of study medication. The breakfast consists of two rolls, butter and jam. After breakfast study participants will be provided with fruits and crackers (e.g. Darvida). Whole day water and herbal and fruit tea (no green or black tea) will be available. Caffeine containing beverages are only allowed during breakfast.

Testing will take place at the same time for each subject to minimize the influence of diurnal differences in cognitive test performance.

There will be a 4-h waiting period allowing the medication to reach high cerebrospinal fluid concentration.

After this time period, the cognitive test battery will be administered. A parallel version of the test battery will be administered on each test day.

For the duration of the different study parts see table 1 (Duration of Study parts, page 12).

At the end of each test day, participants will be asked about their actual wellbeing. If there are no complaints / symptoms they will be dismissed. In case of minor health problems the participant will have to stay in the study center until recovery under medical control. In case of more severe incidents staff will act according to chapter 10.

Assessments in participants who prematurely stop the study: There will be a final examination after withdrawal during test days for whatever reason. The examination consists of recording adverse events and assessing vital signs.

The study including assessment of inclusion and exclusion criteria (other than laboratory parameters and ECG that will be assessed at the Phase I Research Unit of the University Hospital Basel), medication intake and assessment of study outcomes will be conducted at the Transfaculty Research Platform Molecular and Cognitive Neurosciences, University of Basel.

9.4.3 Treatment periods

During the visits 2 and 4 (test days 1 and 3) every subject will be supplied with a plastic container containing the remaining 6 tablets of IP to be taken twice per day during the following 3 days, the first tablet in the evening of visit 1 (resp. 3) and the last tablet in the morning of the visits 3 and 5 (test days 2 and 4). Each subject receives a total of 7 verum tablets and 7 placebo tablets.

During the treatment days, the participants document the intake of the tablets and – if any – adverse events via a separate personal login directly in secuTrial® (s. chapter 8.4, Compliance with study intervention).

10 SAFETY

10.1 Definitions

10.1.1 Adverse Event (AE)

An Adverse Event (AE) is any untoward medical occurrence in a patient or a clinical investigation participant administered a pharmaceutical product and which does not necessarily have a causal relationship with the study procedure. An adverse event can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal investigational product, whether or not related to the medicinal investigational product.

In this study, the following very common AEs being already described in the prescribing information Fampyra® (version from May 2019) are excluded from recording, but will be measured as possible confounding factor using VAS:

- headache
- gastrointestinal disturbances such as dyspepsia, abdominal pain, constipation and nausea
- dizziness

All other clinically relevant AEs will be fully recorded by describing the AE, time of onset, duration and resolution, assessment of intensity, relationship to study drug and measures taken

10.1.2 Serious Adverse Event (SAE)

A Serious Adverse Event (SAE) is classified as any untoward medical occurrence that:

- results in death,
- is life-threatening,
- requires in-patient hospitalization or prolongation of existing hospitalization,
- results in persistent or significant disability/incapacity, or
- is a congenital anomaly/birth defect.

In addition, important medical events that may not be immediately life-threatening or result in death, or require hospitalization, but may jeopardize the patient or may require intervention to prevent one of the other outcomes listed above should also usually be considered serious. (ICH E2A)

SAEs should be followed until resolution or stabilization. Participants with ongoing SAEs at study termination (including safety visit) will be further followed up until recovery or until stabilization of the disease after termination.

10.1.3 Unexpected Adverse Drug Reaction

An “unexpected” adverse drug reaction is an adverse reaction, the nature or severity of which is not consistent with the applicable product information (e.g. investigator’s brochure for drugs that are not yet approved and prescribing information for approved drugs, respectively). (ICH E2A)

10.1.4 Suspected Unexpected Serious Adverse Reactions (SUSARs)

The Sponsor-Investigator evaluates any SAE that has been reported regarding seriousness, causality and expectedness. If the event is related to the investigational product and is both serious and unexpected, it is classified as a SUSAR. In order to determine a SUSAR unblinding is needed.

10.2 Assessment of (Serious) Adverse Events and other safety related events

10.2.1 Assessment of causality

Both Investigator and Sponsor-investigator make a causality assessment of the event to the study drug, based on the criteria listed in the ICH E2A guidelines:

Relationship	Description
Definitely	Temporal relationship Improvement after dechallenge* Recurrence after rechallenge (or other proof of drug cause)
Probably	Temporal relationship Improvement after dechallenge No other cause evident
Possibly	Temporal relationship Other cause possible
Unlikely	Any assessable reaction that does not fulfil the above conditions
Not related	Causal relationship can be ruled out
*Improvement after dechallenge (stop administering a drug, to see whether its suspected effects cease to occur) only taken into consideration, if applicable to reaction	

10.2.2 Assessment of Severity

Adverse events will be categorized as 1=mild, 2= moderate, 3=severe, 4=life-threatening, 5= death.

10.3 Documentation

10.3.1 Serious Adverse Events (SAE)

The sponsor's SOPs provide more detail on safety reporting.

During the entire duration of the study all serious adverse events (SAEs) are collected, fully investigated and documented in source documents and electronic case report forms (eCRF). Study duration encompassed the time from when the participant signs the informed consent until the final examination.

10.3.2 Adverse Events (AE)

The following information needs to be collected for adverse events:

- time of onset,
- duration,
- resolution,
- action to be taken,
- assessment of intensity, and
- relationship with study treatment.

For assessment of safety outcomes see

chapters 9.3

10.4 Reporting of serious adverse events (SAE) and other safety related events

10.4.1 Reporting of SAEs

All SAEs must be reported immediately and within a maximum of 24 hours to the sponsor-investigator of the study. The sponsor-investigator will re-evaluate the SAE and return the form to the site.

SAEs resulting in death are reported to the EKNZ within 7 days.

10.4.2 Reporting of SUSARs

A SUSAR needs to be reported to the CEC (EKNZ) and to the CA (Swissmedic) (via Sponsor-Investigator) within 7 days, if the event is fatal, or within 15 days (all other events).

10.4.3 Reporting of safety signals

All suspected new risks and relevant new aspects of known adverse reactions that require safety-related measures, i.e. so called safety signals, must be reported to the sponsor-investigator within 24 hours. The sponsor-investigator must report the safety signals within 7 days to the CEC (EKNZ) and to the CA (Swissmedic).

10.4.4 Reporting and handling of pregnancies

Pregnant participants must be withdrawn immediately from the clinical study. Any pregnancy during the treatment phase of the study and within 30 days after discontinuation of study medication will be reported to the Sponsor-Investigator within 24 hours.

The course and outcome of the pregnancy will be followed up until the end of the first trimenon, and any abnormal outcome regarding the mother or the child be documented and reported. For further information see Chapter 7.5.

10.4.5 Periodic reporting of safety

An annual safety report is submitted once a year to the local Ethics Committee via BASEC and to the CA via sponsor-investigator.

10.4.6 Follow-up of (Serious) Adverse Events (AE)

In case of minor health problems the participant will have to stay under medical control in our department.

In case of major health problems the participant will be transferred to the emergency department in the University Hospital of Basel (USB). The responsible investigator will inform the health care provider about the participation in the study. For unblinding see 6.3.

Information about the outcome in all above-mentioned cases will be collected until full recovery.

11 STATISTICAL METHODS

11.1 Hypothesis

Fampridine improves working memory performance in healthy humans.

11.2 Determination of Sample Size

We are interested to detect a drug effect with at least medium effect size. The estimation of N=44 is based on a power analysis using dependent two-tailed t-tests assuming to detect a medium effect size (Cohens dz = 0.5) with a power of 90% at α=0.05 (software: G-power 3).

11.3 Statistical criteria of termination of trial

As soon as 44 participants will have completed the entire study. For subjects still enrolled at that time point see 11.4.1.

11.4 Planned Analyses

All analyses will be performed with the software R. The differences between the experimental conditions (placebo, verum) will be analyzed using linear mixed models in combination with analysis of variance (SS II). Subjects will be included as the random effect of the mixed model. Sex and age will be inserted as covariates.

In case of significant interactions between covariates and experimental condition, post-hoc tests will be applied to describe the interaction.

To control for an influence of other variables, possible confounders will be entered into the model as covariate (each possible confounder entering the statistical analyses separately and corrected for multiple comparisons with the Bonferroni method).

11.4.1 Datasets to be analyzed, analysis populations

We will apply a Per-Protocol-Analysis, i.e. the analysis of the primary and secondary outcome measures will be only performed with those participants, who completed all tests and investigations. There will be replacement of Drop-Outs until data of 44 participants are completed. Drop-Outs will be thoroughly described to assess the reason(s) for dropping out.

We will recruit up to 4 subjects more towards the end of the trial to prevent under-recruitment due to Drop-Outs. If more then 44 subjects will complete all tests and investigations their data will also be analyzed.

11.4.2 Analysis for the primary outcome

A significance level of $p < 0.05$ will be considered as significant.

The dependent variable is the primary outcome (i.e.3-back (d`)) after repeated administration of study medication). The independent variable of interest is the experimental condition (placebo or verum).

To control for possible confounders or moderators, we will also consider tiredness, motivation, headache, gastrointestinal discomfort, dizziness, mood as assessed by MADRS-S, potassium blood concentration at screening, body weight, duration of washout period and vital signs as covariates (each covariate entering the statistical analysis separately). In case of a significant (i.e. after Bonferroni correction for multiple comparisons) interaction between independent variables of interest and covariates, post-hoc analysis will be applied to describe the interaction effect.

11.4.3 Analyses for secondary outcomes

Behavioral secondary outcomes will be treated statistically the same way as the primary outcome, but a Bonferroni correction (i.e. corrected for the number of behavioral secondary outcomes) will apply separately for the main effect (i.e. drug vs. placebo) and the interaction effect (i.e. main effect x acute vs. repeated administration). In the case of a significant (i.e. Bonferroni-corrected) interaction effect, post-hoc analysis will be applied to describe the interaction effect.

The measure of the non-behavioral secondary outcome (i.e. rMT) will be treated statistically the same way as the primary outcome measure.

11.4.4 Interim analyses

No interim analyses are planned.

11.4.5 Safety analysis

Safety data will be analyzed for all participants, who received at least one dose of study medication.

11.4.6 Deviation(s) from the original statistical plan

Deviations from the original statistical plan will be justified and reported to the ethical committee and regulatory authorities.

Deviation from the original statistical plan will be performed, if reviewers demand specific analyses. If in the meantime other studies find important effects or confounding effects related to our study, we will include these found confounders (if we have assessed those) as an additional analysis in our statistical plan beside our planned analyses.

11.4.7 Handling of missing data and drop-outs

Missing data will be recorded as NA.

Drop-Outs see chapter 11.4.1.

12 QUALITY ASSURANCE AND CONTROL

12.1 Responsibilities

The sponsor-investigator has an overall responsibility for the implementation and conduct of the study. He is allowed to delegate tasks to the research team as specified in the authorization list. Adequate information and training of the involved staff is in his charge and should be documented.

12.2 Data handling and record keeping / archiving

12.2.1 Case Report Forms (CRF)

Study data is recorded with electronic Case Report Forms (eCRF). secuTrial® will be used for the eCRF (see contract with CTU). For each enrolled study participant a eCRF will be maintained. The eCRF will be kept current, as it has to reflect participant status at each phase during the course of study. For confidentiality reasons eCRFs must not contain any personal data of study participants. It will be used a coded identification consisting of a participant number (see chapter 12.3.1). Authorized for eCRF entries is mainly the study coordinator and additionally the investigators (especially for medical issues). secuTrial® has a detailed audit trail so that every relevant change is traceable and assignable to the person who made it. Data is entered into the eCRF and can be validated for completeness and discrepancies automatically.

We will perform single data entry. Entries in the eCRF must be consistent with information recorded in the source documents. eCRF data should be accurate, consistent, complete and reliable.

The n-back Tasks and the Digit Span Task will not be entered in the eCRF (secuTrial®). These tests will be performed with Presentation® respectively SoSci-Survey and the generated logfiles will be transferred to a secure electronical archive (LabKey®) to guarantee originality and to prepare the data for data analysis. The data collected with Presentation® (n-Back Task) and SoSci-Survey (Digit Span) will be transferred to LabKey® according to the procedures described in chapter 12.3.3.

12.2.2 Specification of source documents

Source data are all information in original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Source data are contained in source documents (e.g. hospital records, laboratory notes, recorded data from automated instruments, and records kept at the pharmacy and other departments involved in the clinical trial). (GCP 1.51/2)

Source data will consist of the following documents:

Paper documentation:

- Informed Consent Form
- Screening documentation, incl. telephone screening
- Sociodemographic questionnaire
- AE-log, concomitant medication
- SDMT (visits 2 to 5)
- Administrative documents: visit checklists, visit plan, randomization number, rMT-value, etc.

Direct data entry with secuTrial®:

- Questionnaire at the beginning of the test days (visits 2 to 5)
- Questionnaires during the test days (before and after testing; visits 2 to 5)
- BOMAT (visits 2 to 5)
- Visual Analog Scales and documentation of the intake of the IP at home (treatment periods)

Direct data entry with Presentation® (https://www.neurobs.com/):

- n-back tasks

Direct data entry with SoSci-Survey:

- Digit Span task

Data recording with Brainsight® (TMS-software):

- Location of rMT assessment (MNI-coordinates)
- Electromyography (EMG) during rMT (MEP information: peak-to-peak amplitude, EMG waveform information; accuracy specifications such as: distance to associated target, target error).

12.2.3 Record keeping / Archiving

All study related data (essential documents and site documents) will be archived for a minimum of 10 years after study termination or premature termination of the clinical trial in the archives of Division of Cognitive and Molecular Neuroscience and in case of the eCRF in the electronic archive-system of the University Hospital Basel. Electronically captured data with presentation® (n-back task), SoSci-Survey (Digit Span) and Brainsight® (EMG) will be archived in a read only status in LabKey® for at least 10 years (for further information to LabKey® see chapter 12.3.4).

The study team will archive all essential data, such as a printout version of the eCRFs, medical records including logs for concomitant medication, laboratory reports, informed consent documents, IP disposition records including randomization and decryption lists, safety reports, information regarding participants who discontinued, and other pertinent data.

12.3 Data management

12.3.1 Pseudonymization and Coding

Each subject will be assigned to a numeric code in the following way: 81-0001-01. The first two digits identify the study, the 4 four digits in the middle are the subject number and the last two digits are reserved for the sample number.

The telephone screening will be performed before assignment of the subject code as described above. This document will be coded with a separate numeric code and the date of birth.

The participant identification list will be kept under lock and key. During the course of the study the study coordinator is responsible for the list. Access to the participant identification list will have only authorized study team members. After the monitoring close out visit the study documentation will be transferred to our archive (s. 12.3). Access will have only persons authorized by the sponsor-investigator of the project.

12.3.2 Data Security and Access

Trial and participant data will be handled with uttermost discretion and is only accessible to authorized personnel who require the data to fulfil their duties within the scope of the study. On the eCRFs, participants are only identified by a unique participant number and additionally the year of birth can be noted (see chapter 12.3.1).

All Source Data are kept under lock and key. All electronic systems used in this study are password protected, to ensure that only authorized persons can enter the system to view, add or modify data according to their permissions within the scope of the study.

The investigators will permit study related monitoring visits, audits, EC reviews, and regulatory inspections, and provide direct access to all source data.

12.3.3 Data Management Systems, Back-up

SecuTrial® (eCRF) runs on a server maintained by the IT-department of the University Hospital Basel. The data management group at the Clinical Trial Unit (CTU) of the University Hospital Basel implements the eCRF. The user administration and user training is performed by the CTU according to predefined processes. Back-up of eCRF data is performed according to the processes of the IT-department of the University Hospital Basel. SecuTrial® has a detailed audit trail so that every relevant change is traceable and assignable to the person who made it.

LabKey® (https://www.labkey.com; nBack tasks (Presentation), Digit Span task (SoSci-Survey), TMS-Logfiles (Brainsight®)) runs on a server maintained by the IT-department of the University of Basel. The data manager of the Division of Cognitive Neuroscience is responsible for the implementation of the tasks, for the user administration and for the user training. Back-up is performed according to processes of the IT-department of the University of Basel. LabKey® has a detailed audit trail so that every relevant change is traceable and assignable to the person who made it. Back-up of the LabKey® database is done by sciCORE, University of Basel, on daily bases. An additional text-file-based database dump is stored within the Linux or Macintosh file-system deployed by the Psychology IT-Department. This is done occasionally, e.g. when the data collection is finished.

The *source data (including Presentation®-, SoSci-Survey- and TMS-logfiles*) will be stored on a Linux or Macintosh file-system with restricted user access (main file-system). The meta-information of the source

data (SHA-1 hashes as file id, file modification time, path to the file on the file-system, date and time information logged in the log-file) is stored in LabKey® in a study specific folder (main study folder). The relevant content of the source data that is necessary for creating an analytical database is additionally uploaded to LabKey® (main study folder). The analytical database is created as text-files with time-stamps based on the uploaded or manually entered raw data. These text-files are stored and accessible within the LabKey® file-system in the main study folder.

For source data, we don't expect that any changes will be done. Therefore, we store the meta-information of the source data to be able to verify that the files are in the original state. All other information is documented and stored within LabKey®.

For source data (including the Presentation®-, SoSci-Survey- and Brainsight®-logfiles) we use different levels of *validation*: As a first step, we evaluate for each subject, visit and computer if all expected files or entries are available and stored in the correct sequence (via the time-stamp). If this basis checks fail, we manually curate the source data, if possible; manual data curation is documented in text-files stored together with the source data or in LabKey®. After performing these basic checks, the data is copied and stored in the final storage space of a study in the main file-system (deployed by Psychology IT-department). At the same time-point the meta-information of each file is stored in LabKey®. When uploading the relevant content of the raw data, we further validate if the file-content corresponds to the expected design of a task or survey, if possible (this is data-dependent). Furthermore, within LabKey® we track for each subject, visit and task if there are exclusion reasons (filter-variables). While creating the final analytical database we apply these filter-variables to the data. An audit trail system maintains a record of initial entries and changes (time and date of changes, user identification of entry and changes). Reasons for changes can be added by a commentary. The data entered or uploaded in the LabKey® study folder will be reviewed by the investigator.

12.3.4 Analysis and Archiving

The eCRF will be locked after all data was monitored and all raised queries have been resolved. Data is exported and transferred to the investigator by the CTU according to internally defined processes. The exported data will be archived by the study coordinator (see 12.2.3).

12.3.5 Electronic and central data validation

Data is entered into the eCRF and can be validated for completeness and discrepancies automatically. An audit trail system maintains a record of initial entries and changes (reasons for changes, time and date of changes, user identification of entry and changes).

An independent monitor from the CTU Basel will review the data entered into the eCRF. The monitor will raise queries using the query management system implemented. Designated investigators and the study coordinator have to respond to the query and confirm or correct the corresponding data. Thereafter the monitor can close the query.

12.4 Monitoring

The aim of monitoring is to evaluate the progress of the study, to verify the accuracy and completeness of CRFs, to ensure that all protocol requirements, applicable local authority regulations and investigator's obligations are being fulfilled, and to resolve any inconsistencies in the study records. The investigator will allow the sponsor to periodically monitor at mutually convenient times during and after the study and they will answer questions during monitoring.

Monitoring will be provided by the CTU Basel (see contract). Monitoring will be performed according to the separate monitoring plan.

12.5 Audits and inspections

Audits by the CEC or the sponsor or inspections by regulatory authorities during study or after study closure may be performed to ensure proper study conduct and data handling procedures according to ICH-GCP guidelines and regulatory requirements. Audits and inspections may include verification of all source documents, check of CRFs and site files and a visual inspection of the study site. Direct access to all documents and places at study site is mandatory. In case of an announced audit or inspection immediate notification of the respective other party is necessary.

The investigators will permit study related monitoring visits, audits, IEC reviews, and regulatory inspections, and provide direct access to all essential documents including the source data. Essential documents permit individually and collectively an evaluation of the conduct of a study and the quality of the data produced. Source data are all information, original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial.

All involved parties must keep the participant data strictly confidential.

12.6 Confidentiality and Data Protection

Participant's confidentiality will be maintained at all times. Personnel from the sponsor, CTU Basel, from the Department of Pharmacology (PRU USB) and Toxicology (USB), laboratory (USB), from regulatory authorities and members of IEC are obliged to respect medical secrecy and to refrain from divulging the participant's identity or any other personal information they might fortuitously be aware of.

The participants name or other personal identifiable data are not recorded in the eCRF.

Direct access to source documents will be permitted for purposes of monitoring (chapter 12.4), audits and inspections (chapter 12.5). People who will have access to protocol, dataset, statistical code, etc. during and after the study (publication, dissemination) will be declared.

13 PUBLICATION AND DISSEMINATION POLICY

The main publication will be created by Prof. Dominique de Quervain and Prof. Andreas Papassotiropoulos. Subsequent publications of subgroups can follow thereafter and will have to be approved by Prof. de Quervain and Prof. Andreas Papassotiropoulos.

No unpublished data given to the investigator may be transmitted to a third party without prior written approval by Prof. de Quervain and Prof. Andreas Papassotiropoulos. No publication or communication involving the results of the study is authorized without prior written consent from the Prof. de Quervain and Prof. Andreas Papassotiropoulos. In view of patent and confidentiality issues, however, the investigator must accept requirements on the timing of early publication. The investigator's name should not be used in any publication without the prior written permission of Prof. de Quervain and Prof. Andreas Papassotiropoulos.

14 FUNDING AND SUPPORT

Funding the trial: Funds of the Swiss National Science Foundation and the Transfaculty Research Platform, University of Basel.

15 INSURANCE

The sponsor-investigator declares that he has taken out an insurance policy for the total study length, covering the participants in respect of the risks involved in this study. In case of injury or disability deriving from participation in the study, the subject is requested to inform without delay the investigator responsible for the study.

16 REFERENCES

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