clorazepate (20mg 2×/d), and pregabalin (100 mg 3×/d). Because of resurgence of severe anxio-depressive symptoms, without any change of the treatment, the patient was readmitted 2 months later. Despite increasing the dose of clomipramine up to 225 mg/d, there was no clinical improvement, **and the patient finally attempted to her life by abusing** drugs. She then improved after 2 weeks on clomipramine IV (50 mg/d). Compliance was estimated good and no pharmacokinetic interactions with the rest of the treatment were found. C and DC plasma levels were measured, and CYP2D6/CYP2C19 genotype analyzed.

Results: The plasma levels of C and DC are given in the **Table** below. Measures were done at the steady state and at trough concentration for IV treatment and 10 hours after the last dose for oral treatment.

Table. Doses and plasma levels.

Posology (mg/d)			
Route of administration	150	50	
	oral	intravenous	
C (micromol/l)	0.3-0.8	0.17	0.28
DC (micromol/l)		0.14	0.27
C + DC (micromol/l)	0.8-1.6	0.31	0.55

Clomipramine bioavailability, 50%.

The CYP2D6 genotyping was CYP2D6 *1/*2xN, compatible with an ultrarapid metabolizer. The CYP2C19 genotyping was CYP2C19*1/*1, compatible with an extensive metabolizer.

Conclusion: The lack of clinical effect of oral clomipramine after a dramatic response to IV administration and the low per os plasma levels of clomipramine and its active metabolite, desmethylclomipramine, suggested a rapid phenotype for CYP2D6. This was confirmed by genotyping. This case stresses the value of genotype determination to assess treatment failures in a population of patient wherein lack of compliance is often mentioned.

Disclosure of Interest: None declared.

PP270—COMPUTATIONAL MODELING OF DRAVET SYNDROME

P. Kurbatova[1*] on behalf of Cresim project; P. Benquet[2]; F. Wendling[2]; C. Cornu[3]; R. Guerrini[4]; A. Kaminska[5]; O. Dulac[6]; G. Pons[5]; R. Nabbout[5]; C. Chiron[5]; and P. Nony[7]

[1]UMR 5558, University of Lyon 1, Lyon; [2]UMR 1099, Inserm-University Rennes1, Rennes; [3]Centre d'Investigation Clinique, INSERM CIC201/UMR5558, Hôpital Louis Pradel, Bron, France; [4]Pediatric Neurology Unit and Laboratories, Children's Hospital A. Meyer-University of Florence, Florence, Italy; [5]UMR 663, Inserm-University Paris Descartes-CEA; [6]UMR 663, InUniversity Paris Descartes-CEA, Paris; and [7]Service de Pharmacologie Clinique, CHU Lyon, Lyon, France

Introduction: Dravet syndrome (DS) is a rare pediatric epilepsy of early life onset characterized by pharmacoresistant hemi- or generalized (tonic)-clonic seizures and severe cognitive prognosis. The voltage-gated sodium channel SCN1A gene is mutated in 85% of the patients. Transgenic mice models of DS suggest that the mutation specifically involves GABAergic interneurons and leads to selective loss of sodium current affecting their firing properties. However, the mechanisms leading from interictal activity to seizure generation and the subtype of interneurons involved are not known. Computational modeling of neuronal network is currently used to analyze these mechanisms. The inconsistency of firing rate in GABAergic interneurons can be used as a starting point to build a computational model of DS.

Objectives: To find parameters and connections in the model that reproduce EEG patterns of DS patients.

Patients (or Materials) and Methods: We first made a quantitative and qualitative analysis of EEG recorded in DS patients during interictal, preictal, and ictal periods. We next used a lumped-parameter approach (mean-field model) lying at the level of neuronal population and able to represent the generation of spontaneous EEG. This computational model includes 1 subpopulation of pyramidal cells and 2 subpopulations of interneurons (mediating fast and slow GABAergic currents).

Results: EEG signals were characterized by slow background activity (1–4 Hz), multifocal interictal epileptic spikes and, as far as hemi-clonic seizures were concerned, by fast oscillations at the onset of seizures. To mimic the effect of a SCN1A mutation, the firing rate of GABAergic current was modified in slow interneurons. Preliminary results show that appropriate alterations in the strengths of GABAergic and glutamatergic connections, and in the amplitudes of average EPSPs/IPSPs in the model successfully lead to slow background activity (1–4 Hz), generation of interictal epileptic spikes, and fast onset activity and seizure-like activity.

Conclusion: Our computational modeling of DS is therefore promising. Further optimization is needed for reproducing all the features of the real EEG from patients and identifying the key parameters of specific EEG patterns.

Disclosure of Interest: None declared.

PP271—NOVEL THERAPEUTIC STRATEGY TO PREVENT CHEMOTHERAPY-INDUCED PERSISTENT SENSORY NEUROPATHY BY TRPA1 BLOCKADE

G. Trevisan[1]; S. Materazzi[2]; C. Fusi[3]; A. Altomare[4]; G. Aldini[4]; R. Patacchini[5]; P. Geppetti[6]; and R. Nassini[7*]

[1]Federal University of Santa Maria (UFSM), Santa Maria, Brazil; [2]Department Health Sciences–Clinical Pharmacology and Oncology unit; [3]Health Sciences–Clinical Pharmacology and Oncology Unit, University of Florence, Florence; [4]Department of Pharmaceutical Science, University of Milan, Milan; [5]Pharmacology Department, Chiesi Farmaceutici spa, Parma; [6]Department of Health Sciences–Clinical Pharmacology and Oncology Unit; and [7]Department of Health Sciences–Clinical Pharmacology and Oncology Unit, University of Florence, Florence, Italy

Introduction: Several anticancer medicines evoke sensory adverse events, collectively referred to as chemotherapy-induced peripheral neuropathy (CIPN), which are represented by sensory symptoms. No effective therapy is currently available to treat or prevent CIPN, most likely because the underlying mechanisms are poorly understood. A host of hypotheses has been proposed to explain CIPN, but nonetheless no unified mechanism that may reconcile results of clinical investigation and findings obtained in experimental animals has been advanced so far. Chemotherapeutic drugs, which produce CIPN, are known to increase oxidative stress and reactive oxygen, nitrogen, or carbonyl species (ROS, RNS, and RCS, respectively) and treatment with antioxidant substances has been shown to reduce sensory hypersensitivity in experimental animals and to exhibit some degree of protection in patients with CIPN. The transient potential receptor ankyrin 1 (TRPA1) is a nonselective cation channel, coexpressed with TRP vanilloid 1 (TRPV1) in a subset of C-fiber nociceptors, where it functions as a multimodal sensor to noxious stimuli. TRPA1 shows a unique sensitivity for an unprecedented number of endogenous reactive molecules produced at sites of tissue injury or inflammation, which include ROS, RNS and RCS. Bortezomib is a proteasome inhibitor used indifferent types of cancer. CIPN has

emerged as a major complication of bortezomib therapy, which usually appears in the first courses of therapy with a number of sensory and painful symptoms, including reduced threshold to mechanical and cold stimuli. No satisfactory explanation or effective treatment exists for bortezomib-evoked CIPN.

Patients (or Materials) and Methods: In this study, we evaluated whether TRPA1 acted as a critical mediator of CIPN by bortezomib or oxaliplatin in a mouse model system.

Results: Our data demonstrated that CIPN hypersensitivity phenotype that was stably established by bortezomib could be transiently reverted by systemic or local treatment with the TRPA1 antagonist HC-030031. A similar effect was produced by the oxidative stress scavenger α-lipoic acid. Notably, the CIPN phenotype was abolished completely in mice that were genetically deficient in TRPA1, highlighting its essential role. Administration of bortezomib or oxaliplatin, which also elicits TRPA1-dependent hypersensitivity, produced a rapid, transient increase in plasma of carboxy-methyllysine, a by-product of oxidative stress. Short-term systemic treatment with either HC-030031 or α -lipoic acid could completely prevent hypersensitivity if administered before the cytotoxic drug.

Conclusion: Our findings highlight a key role for early activation/sensitization of TRPA1 by oxidative stress by-products in producing CIPN. Furthermore, they suggest prevention strategies for CIPN in patients through the use of early, short-term treatments with TRPA1 antagonists.

Disclosure of Interest: None declared.

PP272—MIGRAINE AND PARTHENOLIDE INHIBITION OF TRANSIENT RECEPTOR POTENTIAL ANKYRIN 1

S. Materazzi[1]; S. Benemei[1*]; C. Fusi[1]; G. De Siena[1]; G. Trevisan dos Santos[1]; G. Dussor[2]; F. Pollastro[3]; R. Patacchini[4]; G. Appendino[3]; P. Geppetti[1]; and R. Nassini[1]

[1]Department of Health Sciences, University of Florence, Florence, Italy; [2]Department of Pharmacology, University of Arizona, Tucson, United States; [3]Department of Pharmaceutical Sciences, University of Eastern Piedmont, Novara; and [4]Department of Pharmacology, Chiesi Farmaceutici SpA, Parma, Italy

Introduction: *Tanacetum parthenium* L. (feverfew) has long been known as a migraine remedy and, according to positive results of clinical trials, it is currently recommended for migraine prevention. However, the mechanism responsible for such protective action remains unknown. Parthenolide, a major ingredient of feverfew, is a reactive molecule that can interact with nucleophilic sites of transient receptor potential ankyrin 1 (TRPA1). Thus, we hypothesized that parthenolide inhibits TRPA1 channel on peptidergic trigeminal nerves.

Patients (or Materials) and Methods: Experiments were performed in vitro in human and mouse cultured cells/neurons and rat isolated tissues, and in in vivo in rats and wild-type and TRPA1-deficient mice. Electrophysiologic, calcium, neuropeptide release, smooth muscle motility, allodynic and nociceptive responses, and changes in meningeal blood flow were evaluated.

Results: Parthenolide selectively activates recombinant (transfected cells) or natively expressed (rat/mouse trigeminal neurons) TRPA1, and, by targeting TRPA1, activates trigeminal nerve endings. However, parthenolide behaves as a partial agonist at neuronal TRPA1 of the rat urinary bladder, desensitizes the recombinant TRPA1, and, after initial stimulation, renders peptidergic, TRPA1-expressing nerve terminals unresponsive to any stimulus. These effects causes abrogation of nociceptive responses evoked by TRPA1 agonists, and inhibition of calcitonin gene-related peptide (CGRP) release from trigeminal neurons, and, in particular, of CGRP-mediated meningeal vasodilatation evoked by TRPA1 stimulants and other mechanisms.

Conclusion: Peculiar features of parthenolide (TRPA1 partial agonism, channel desensitization, and defunctionalisation of peptidergic primary sensory neurons), ultimately resulting in the inhibition of CGRP release from trigeminal neurons, may contribute to the anti-migraine effect of feverfew.

Disclosure of Interest: None declared.

PP273—EFFECT OF THE GABAA LIGANDS CLOBAZAM AND CLONAZEPAM ON THE MODULATION OF PAIN TRANSMISSION IN HUMANS: A PK-PD STUDY

M. Besson[1*]; A. Matthey[1]; D. Youssef[1]; P. Vuillemier[2]; M. Curatolo[2]; P. Dayer[1]; H.U. Zeilhofer[3]; and J. Desmeules[1]

[1]Clinical Pharmacology and toxicology, Geneva University Hospitals, Geneva; [2]University Department of Anesthesiology and Pain Therapy, Inselspital, Bern, Bern; and [3]Institute of Pharmacology and Toxicology, University of Zurich, Zurich, Switzerland

Introduction: Facilitation of spinal GABAergic inhibition with benzodiazepines (BZD) reverses pain sensitization in rodents. In human, the use of BZD in pain is limited by their sedative effect. We previously demonstrated the antihyperalgesic effect of clobazam, a 1,5-BZD, in mice and its lack of sedation at effective doses. Hence we designed a pharmacokinetic-pharmacodynamic study to explore the effect of antihyperalgesic effect of BZD in healthy volunteers.

Patients (or Materials) and Methods: Randomized, double-blind, crossover controlled study in 25 healthy volunteers comparing clobazam 20 mg with clonazepam 1 mg (positive control) and tolterodine 1.37 mg (active placebo) 2 weeks apart. The primary outcome was the effect on the size of secondary hyperalgesia elicited by the UVB irradiation of the skin of the forearm (sunburn model). Quantitative sensory testing, nociceptive flexion reflex, and the cold pressor test were also performed. Sedation was measured by a visual analog scale (VAS), the digit substitution symbol test (DSST), and saccadic eye movements (SEM) recording. Blood samples were taken to determine the pharmacokinetic of clobazam.

Results: We observed a reduction of the area of the secondary hyperalgesia with clobazam and clonazepam. The maximum of the effect was seen at t = 2 hours (median of the difference between t = 2 and baseline [MOD2-0] [+/-CI] vs placebo: 19.2 cm^2 (–2.7-37.9) $P = 0.07$ for clobazam and 29.6 cm2 (2.0-55.1), $P = 0.03$, for clonazepam), in line with the expected T_{max} of the compounds. Regarding sedation, at t = 2 hours, we saw an effect of the 2 active compounds on the VAS (MOD2-0 [+/-CI] vs placebo: 14 mm (1.0-21.5), $P = 0.03$ for clobazam and 26 mm (14.0-37.5), $P < 0.001$ for clonazepam) and on the peak velocity of the SEM (MOD2-0 [+/-CI] vs placebo: 30.8 deg./s. (6.6-60.6), $P < 0.01$ for clobazam and 55.2 deg./s.(24.5–85.0), $P < 0.01$ for clonazepam]. The DSST was only impaired by clonazepam (MOD2-0 [+/-CI] vs placebo: 11.0 (5.0–18.0), $P = 0.03$). These effects disappeared at t = 8 hours except that clonazepam still impaired SEM (median of the difference between t = 8 and baseline [+/-CI] vs placebo: 28.3 deg./s. (5.3–52.7), $P = 0.01$).

Conclusion: Clobazam and clonazepam decreased the area of secondary hyperalgesia in the sunburn model, which suggests that GABAA receptor ligands are involved in the modulation of pain sensitization in human. Clobazam was less sedative than clonazepam and therefore a suitable "tool compound" to assess the role of GABAergic pathways in human.

Disclosure of Interest: None declared.