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Predicting Drug Efficacy in Chronic Low-Back Pain by Quantitative Sensory Tests

Drug prediction by QST in chronic low-back pain

Juerg Schliessbach[1,2], Andreas Siegenthaler[3], Lukas Bütikofer[4], Pascal Vuilleumier[1], Peter Juni[5], Ulrike Stamer[1], Lars Arendt-Nielsen[6], Michele Curatolo[7,6]

[1]Department of Anesthesiology and Pain Medicine, Inselspital, Bern University Hospital, University of Bern, Switzerland

[2]Institute of Anesthesiology, University Hospital Zurich, Zürich, Switzerland

[3]Chronic Pain Management, Lindenhof Hospital, Lindenhof Group Bern, Switzerland

[4]CTU Bern, and Institute of Social and Preventive Medicine (ISPM), University of Bern, Switzerland

[5]Applied Health Research Centre (AHRC), Li Ka Shing Knowledge Institute of St. Michael's Hospital, Department of Medicine, University of Toronto, Canada

[6]Centre of Sensory Motor Interaction SMI, University of Aalborg, Denmark

[7]Department of Anesthesiology and Pain Medicine, University of Washington, Seattle WA, USA

- Corresponding author: Juerg Schliessbach, M.D., Institute of Anesthesiology, University Hospital Zurich, Rämistrasse 100, 8091 Zürich, Switzerland. Phone: +41 44 255 48 35, e-mail: juerg.schliessbach@usz.ch

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- Significance: Predicting drug efficacy in chronic low-back pain remains difficult. There is some evidence that patients more sensitive to heat and cold pain respond better to imipramine.

Introduction

Pharmacotherapy is a mainstay of chronic pain treatment. In current practice, there is no way to reliably predict the effect of a medication, so that patients are frequently exposed to long trials of different compounds and experience of side effects in the absence of efficacy. Quantitative sensory testing (QST) has been investigated in the past years as a tool to discriminate patients according to sensory phenotype (Edwards et al., 2016; Maier et al., 2010) and to detect differences in nociceptive processing within patients suffering from the same pain syndrome (Baron et al., 2017). If medications target these different nociceptive processes in a specific way, QST may have the potential to identify groups of patients that respond or do not respond to certain pharmacologic treatments. Few investigations have been conducted in healthy volunteers, neuropathic pain and chronic pancreatitis (Attal et al., 2004; Demant et al., 2014; Edwards et al., 2006; Eisenberg et al., 2010; Olesen et al., 2013; Yarnitsky et al., 2012). These studies identified a selection of QST to predict treatment response, but the sample sizes were generally small and the results are not consistent across studies. The most recent evidence (Grosen et al., 2017) showed that opioid efficacy was predicted by low levels of pain catastrophizing, low pain intensity during cold pressor stimulus of the hand and certain EEG patterns. The patient population in this study was very heterogeneous in terms of pain syndrome and pain location. To our knowledge, there is no specific investigation on the predictive ability of QST for pharmacological treatment of chronic low-back pain, which is one of the most common and challenging pain conditions.

There is evidence that chronic low-back pain is associated with sensory hypersensitivity that extends far beyond the painful region of the back and includes decreased pressure pain thresholds (Giesecke et al., 2004), as well as enlarged receptive fields and enhanced temporal summation (Biurrun Manresa et al., 2013) at distant sites. Furthermore, such generalized

sensory hypersensitivity has been detected in as much as 71-80% of chronic low-back pain patients (Curatolo et al., 2015). Given its high prevalence, generalized sensory hypersensitivity is very likely to be associated with some of the pathogenic processes underlying chronic low-back pain and might therefore be a major determinant of a patient's drug responsiveness.

Genetic variations such as polymorphisms of drug metabolizing enzymes affect drug response as well. A further important question is therefore whether assessing genetic polymorphisms before initiating pharmacological treatment can explain different drug effects and thus help selecting the appropriate therapeutic strategy for individual patients.

The aim of the present study was to investigate whether generalized sensory hypersensitivity measured by QST could predict the analgesic effect of three different drugs in chronic low-back pain: the μ-opioid agonist oxycodone, the tricyclic antidepressant imipramine, and the benzodiazepine clobazam. These drugs were chosen in order to cover multiple modes of analgesic action. Oxycodone is a potent agonist at peripheral and central opioidergic pathways, imipramine is a modulator of noradrenergic and serotonergic neurotransmission in the central nervous system, and clobazam modulates spinal nociceptive inhibitory GABA-ergic pathways (Schliessbach et al., 2017; Vuilleumier et al., 2013; Zeilhofer et al., 2009). Polymorphisms of pain-related genes were examined as co-factors. The μ-opioid receptor variant A118G (Chou et al., 2006) was examined as a possible factor affecting the effect of oxycodone. COMT (catechyl-o-methyltransferase) (Diatchenko et al., 2005), GCH-1 (GTP-Cyclohydroxylase) (Campbell et al., 2009) and the potassium channel subunit KCNS1 (Costigan et al., 2010) are known for influencing pain perception. Finally, the major metabolic pathways for the three drugs were investigated: CYP2C19, which is involved in imipramine and clobazam metabolism, CYP2D6 for imipramine and oxycodone metabolism, and CYP3A4 that mediates oxycodone and clobazam metabolism (Giraud et al., 2004; Kosaki et al., 2004).

Methods

Setting

This randomized placebo-controlled trial in consecutive patients with chronic low-back pain was carried out at the University Department of Anesthesiology and Pain Medicine, Inselspital Bern, Switzerland. The study was approved by the local ethics committee (KEK 213-09), registered with clinicaltrials.gov (NCT01179828) and strictly followed good clinical practice guidelines and the Helsinki declaration. The study protocol has been published previously (Siegenthaler et al., 2015). All participants gave written informed consent prior to inclusion.

Patients

Consecutive patients aged between 18 and 80 years with chronic low-back pain of at least 3 months duration were recruited by advertisement in local newspapers and from the outpatient pain clinic of our department. Exclusion criteria were pain intensity at rest <3 on the numerical rating scale (NRS) at the time of testing (whereby 0 = no pain and 10 = worst pain imaginable), suspected radicular pain (as defined by leg pain associated with an MRI finding of a herniated disc or foraminal stenosis), signs or suspicion of neurological dysfunction at the tested sites, pregnancy (as assessed by pregnancy test in women of fertile age), breast feeding, ongoing treatment with an antidepressant, opioid or anticonvulsant, intake of centrally active substances, drug or alcohol abuse, known allergy or pharmacological contraindications to any of the tested drugs, systemic inflammatory or rheumatologic disease, and major depression (Beck Depression Inventory short form score >9). Current analgesic medication had to be stopped one week before the first experiment. Only acetaminophen and ibuprofen were allowed as rescue medication until 24 hours before the experiment. Patients unable to stop their analgesic regimen were not recruited.

Study medication

A single oral dose of imipramine 75 mg or oxycodone 15 mg or clobazam 20 mg were each compared to active placebo in a cross-over fashion. Because all of the three drugs are likely to be associated with minor central side effects, such as dizziness or sedation, the anti-cholinergic compound tolterodine was chosen as an active placebo. It is usually prescribed for hyperactive bladder syndrome and causes some sedation and dry mouth, but is devoid of analgesic effects. The recommended starting dose is 2 mg twice a day, which can be decreased to 1 mg twice a day. In order to minimize the likelihood of excessive side effects, a dose of 1 mg was chosen for this study. A minimal wash-out period of one week between sessions was ensured.

After completion of one experiment, patients were allowed to cross over to one or both of the remaining drugs, which were each compared to a new placebo session again. Therefore, those patients who took part in all 3 drug tests had a maximum of 6 testing sessions (each of the three drugs vs. placebo). The drugs were administered as identical-looking red gelatin capsules in random order and in a fasting state. Blinding and randomization were provided by the hospital pharmacy. If a patient was re-enrolled to another drug, his sequence number was announced to the pharmacy. Thus, the pharmacist ensured that the patient was not randomized twice to the same drug.

QST

Quantitative sensory testing was performed at baseline as well as one and two hours after drug administration. A complete series of training measurements was performed half an hour before baseline assessments, at the same locations and in the same sequence as the subsequent definite measurements, in order to familiarize patients with the procedure. All tests were performed at the more painful body side. In case of bilateral or midline pain, the side was randomly selected.

The test battery consisted of pressure pain thresholds, meant to assess mechanical nociception, electrical pain thresholds which are thought to bypass peripheral nociceptors and directly stimulate nerve fibers, temporal summation thresholds which reflects central integration of nociceptive stimuli by wide dynamic range neurons, as well as heat and cold pain tests assessing thermally-induced nociceptive processes. The rationale for the multiple testing is the fact that responses to different stimulus modalities reflect different aspect of nociceptive processes (Neziri et al., 2011). Conditioned pain modulation was tested as a feature of endogenous pain inhibitory capacity. Tests were always performed in the order as presented.

Pressure pain detection and tolerance thresholds (PPDT and PPTT)

PPDT and PPTT were recorded at the pulp of the 2nd toe using an electronic pressure algometer (Somedic AB, Horby, Sweden) with a probe tip of 1 cm^2. Pressure was increased at a rate of 30 kPa/s up to a maximum of 1000 kPa. The subject stopped the measurement by pressing a button when the pressure sensation turned to pain (PPDT) and when the painful sensation became intolerable (PPTT), respectively. Both PPDT and PPTT were recorded in intervals of 1 minute between measurements. The 2nd toe was chosen because large differences in pain sensitivity between pain patients and healthy controls can be detected there (Banic et al., 2004) and because it is distant from the painful site, therefore reflecting generalized excitability of the nervous system.

Electrical single and repeated pain thresholds (ESPT and ERPT)

ESPT and ERPT were performed using a computer-controlled constant current stimulator (Digitimer Ltd, Welwyn Garden City, UK). Bursts of five 1 ms square wave impulses within 25 ms (perceived as one single stimulus) were delivered via 2 Ag-AgCl electrodes placed in the innervation area of the sural nerve, directly below the lateral malleolus. The current intensity was increased from 1 mA in steps of 0.5 mA until the sensation was rated as painful (ESPT). For ERPT, the stimuli were repeated five times at a frequency of 2 Hz. Current

intensity of all 5 stimuli was increased in steps of 0.5 mA until the last 2-3 stimuli were perceived as painful, indicating temporal summation threshold. This measure of ESPT has one of the best positive predictive values to discriminate low-back pain patients from healthy controls (Neziri et al., 2012).

Electrical train of twenty

The arithmetical mean of three ERPT assessments at baseline was used to deliver 20 identical stimuli over 10 seconds with a frequency of 2 Hz. This stimulus intensity remained constant over the two subsequent measurements at 60 and 120 minutes. Subjects rated the maximal and final pain intensity during this stimulation on a 0-10 NRS. A decrease in pain intensity in the subsequent measurements would be indicative of an analgesic effect. A decrease from maximal to final pain intensity during the 20 stimulations was considered a feature of pain habituation that might be due to activation of inhibitory neuronal circuits. An increase in pain intensity, on the other hand, was suggestive of pain-facilitatory mechanisms. Patients whose pain ratings decreased during the train-of-twenty stimulation (T20) were defined as T20-decreasers in contrast to those with constant or increasing pain ratings over all 20 stimuli.

Temperature pain thresholds (HPDT, HPTT, CPDT)

Temperature pain thresholds were assessed using a thermode (TSA II, Medoc, Ramat Yishai, Israel) with a probe surface of 3x3 cm. All measurements started at 30.0°C, the rate of temperature change was 1°C/s. Subjects stopped the measurements by pressing a button when the warm sensation turned to pain (HPDT) or when the pain became intolerable (HPTT) or when the cold sensation started to become painful (CPDT). In any case, the measurements were stopped at a temperature of 50.5°C for HPTT or 0°C for CPDT, respectively. Measurements were made first at the lateral aspect of the lower leg (dermatome L5), and then at the radial surface of the proximal forearm (dermatome C6). Because HPTT and CPDT measurements were truncated at 50.5°C and 0°C, respectively, the results were dichotomized for statistical modelling according to whether patients reached the limit or not.

Conditioned pain modulation (CPM)

CPM was assessed using the cold pressor test at the hand contralateral to the tested side. Subjects immersed their hand in ice saturated water (1.5±1°C), until the cold pain reached an intensity of 7/10 on the NRS. Five electrical stimulations at an intensity 1.2 times stronger than the previously measured ERPT were delivered three times in intervals of 10 seconds and rated by the subject on a 0-10 NRS. This was performed before and during the cold pressor test. The percent decrease in pain rating with electrical stimulation during the cold pressor test was calculated as indication measure of CPM. Furthermore, the time until cold pressor pain reached 7/10 NRS was recorded. For all tests but CPM, triplicate measurements were recorded.

Outcome measures

Intensity of low-back pain in the supine position and after sitting for 10 minutes was assessed on a 0-10 NRS at baseline and in intervals of 30 minutes up to 2 hours after drug intake. This was considered sufficient time given that oxycodone starts to be effective 1 hour after intake (Ordonez Gallego et al., 2007) and clobazam peaks around 2 hours after intake (Greenblatt et al., 1983). For imipramine, major anti-nociceptive effects were detected already 90 minutes after intake (Bromm et al., 1986). Patients with ≥30% pain reduction were classified as drug responders. The patients' global impression of change scale (PGIC) (Dworkin et al., 2005) was assessed on a 7 point scale ranging from "1 = very much improved" over "4 = no change" to "7 = very much worse", in intervals of 30 minutes, starting 30 minutes after drug administration. Patients remained in the supine position during the whole experiment, except for those 10-min intervals when sitting pain was assessed. Reading newspapers or magazines was allowed between the measurements.

Descriptive variables

The following descriptive variables were assessed on a questionnaire before the first experiment: age, sex, body mass index (BMI), pain duration in years, history of surgery due to the painful condition, average pain intensity during the last 24 hours on a 0-10 NRS, pain-related life interference from the multidimensional pain inventory (MPI) (Kerns et al., 1985), catastrophizing scale (Keefe et al., 1989) and Beck Depression Inventory (BDI) (Poole et al., 2009).

Genotyping

Genetic analyses were performed for the following candidate genes involved either in drug metabolism or in pain perception: CYP2C19 (involved in imipramine and clobazam metabolism), CYP2D6 (imipramine and oxycodone metabolism), CYP3A4 (oxycodone and clobazam metabolism) (Giraud et al., 2004; Kosaki et al., 2004), the μ-opioid receptor variant A118G (oxycodone binding site) (Chou et al., 2006), COMT (catechyl-o-methyltransferase with 3 categories: low, average or high pain sensitivity) (Diatchenko et al., 2005); GCH-1 (GTP-Cyclohydroxylase with no, one or two pain-protective alleles) (Campbell et al., 2009) and the potassium channel subunit KCNS1 (low, medium and high pain risk for zero, one or two mutant alleles, respectively) (Costigan et al., 2010). Genotyping was performed using real-time polymerase chain reaction (PCR) and identification of specific variants by means of melting curve analysis. For CYP2D6, translation of genotypes into a qualitative measure of phenotype was made according to Gaedigk's system of "activity scores" (Gaedigk et al., 2008): alleles *3,*4,*5,*6,*7, and *8 were assigned a value of 0, alleles *10 and *41 a value of 0.5, the wild type (wt) allele a value of 1, and wtxN (representing multiplication of the wt allele) a value of 2. The sum of the values assigned to each single allele resulted in a CYP2D6 activity score. Activity scores of 0 correspond to poor metabolizers (PM), scores of 0.5-1 to

intermediate metabolizers (IM); scores of 1.5-2 to extensive metabolizers (EM) and scores of 3 to ultra-rapid metabolizers (UM).

Statistical analyses

The predictive effects of individual baseline variables including descriptives, genetics and baseline QST measures were analyzed using linear mixed model with pain intensity (NRS) after 120 minutes as dependent variable. Baseline NRS, type of drug (verum vs. placebo), treatment order (i.e. whether verum or placebo session was first), a baseline variable (e.g. QST measure) and its interaction with the type of drug were used as explanatory variables. Positively skewed QST measures (PPDT, PPTT, ESPT, ERPT, time in ice water) were log-transformed. All continuous explanatory variables were standardized and the z-scores were used in the analyses. To account for intra-subject correlation, a random intercept was added for each subject. The models were fitted via maximum likelihood and likelihood ratio tests were used to compare models with and without interaction. P-values were adjusted according to the Benjamini-Hochberg procedure to control for false-positive results due to the high number of analyzed baseline variables (Benjamini and Hochberg 1995). Adjusted p-values represent the false discovery rate, i.e. the proportion of false discoveries among all significant findings. A false discovery rate of 10% was deemed acceptable for this analysis, thus findings with an adjusted $p<0.1$ were considered significant.

Sample size calculation was performed assuming a correlation of pain scores across active and placebo phase within a patient of 0.65, a prevalence of treatment responders of 40% and a difference in NRS of 2.5 between drug and placebo. Using these parameters, analyzing 50 patients per drug would allow to detect an interaction between treatment effect and QST at a two-sided alpha-level of 5% with a power of 90%.

Statistical analysis were done in Stata 14 (StataCorp, College Station, TX) and R (R Core Team, Vienna, Austria).

Results

Here we present the result pertaining to the aim of the present paper, specifically the ability of baseline QST to predict medication efficacy. Separate papers are under construction or have been published that address the effects of medications on pain and QST. The results of these analyses are mentioned only briefly in the present paper.

Results tables display the interaction of baseline parameters with the effect of each specific drug. A positive interaction term indicates a positive influence of the variable on drug effect, compared to placebo. Z-transformation makes the interaction term independent from the unit of measure (e.g. kPa, mA, °C). Equal interaction terms thus indicate equal effects of the QST parameter on drug response. For example, an interaction term of -0.5 indicates a pain decrease of 0.5 points on the NRS per one standardized unit increase of the covariate. P-values are from likelihood ratio tests comparing models with and without interaction.

Oxycodone

Fifty patients (26 females) were tested in the oxycodone arm (mean age 55 years, SD 15.2). A significant analgesic effect on low-back pain and anti-nociceptive effects on almost all QST parameters were observed. Supine pain decreased from 3.7 (95%-CI 3.4 to 4.1) at baseline to 1.5 (1.1 to 2.0) with oxycodone and from 4.0 (3.5 to 4.5) to 3.0 (2.4 to 3.5) with placebo after 2 hours ($p<0.001$). There were 36 vs. 22 responders in the verum vs. placebo session, respectively. Sitting pain decreased from 4.0 (3.6 to 4.4) at baseline to 1.6 (1.2 to 2.0) with oxycodone and from 4.4 (4.0 to 4.8) to 2.9 (2.4 to 3.3) with placebo after 2 hours ($p<0.001$). There were 44 vs. 25 responders in the verum vs. placebo session, respectively. More detailed results are addressed in a separate publication (Schliessbach et al., Scand J Pain, in press).

Only for the supine position, significant interactions of clinical variables with oxycodone effect were found. Average pain in the last 24 hours (interaction term 0.50, 95%-CI 0.16 to 0.84), catastrophizing score (interaction term 0.45, 95%-CI 0.06 to 0.84) and BDI (interaction

term 0.21, 95%-CI -0.00 to 0.42) showed potential positive influences on the effect of oxycodone after 120 minutes (p=0.005, 0.027 and 0.06, respectively). However, none of these variables remained statistically significant after p-value adjustment for multiple testing (adjusted p=0.20, 0.52, 0.74, respectively). Neither genetics nor the baseline sensory tests were associated with the effect of oxycodone (supplementary tables S1 and S2).

Imipramine

A total of 50 patients underwent the imipramine experiment (32 females, mean age 54.4 years, SD 17.3). The effect of imipramine was at no time point significantly different from placebo, neither in the sitting nor in the supine position. Pain intensity in supine position decreased from 4.2 (95%-CI 3.8 to 4.6) to 2.6 (2.1 to 3.2) after 2 hours in the imipramine arm and from 4.0 (3.5 to 4.5) to 2.5 (2.0 to 3.1) in the placebo arm (treatment effect 0.02 (-0.51 to 0.56), p=0.95). There were 27 responders in the verum vs. 31 responders in the placebo session. Pain intensity in sitting position decreased from 4.7 (4.1 to 5.1) to 2.9 (2.3 to 3.5) after 2 hours in the imipramine arm and from 4.2 (3.8 to 4.6) to 2.7 (2.2 to 3.2) in the placebo arm (treatment effect 0.16 (-0.28 to 0.6), p=0.74). There were 30 responders in the verum vs. 27 responders in the placebo session.

Although imipramine had no overall effect on low back pain, the baseline thermal thresholds significantly interacted with the effect of imipramine on pain intensity compared to placebo after 120 minutes in the sitting and – slightly less – in the supine position. Specifically, patients more sensitive to heat and cold pain experienced a greater reduction of their low-back pain by imipramine. Interaction terms and p-values are summarized in tables 1 and 2; treatment effects are displayed by Forest plots in figures 1 and 2.

Further possible interactions with imipramine-effect on low-back pain were found for the μ-opioid receptor A118G allele (interaction term 0.84, 95%-CI 0.03 to 1.66, p=0.047, only in sitting position), the COMT high-pain-sensitivity genotype (1.51, -0-09 to 3.11, p=0.05, only

in sitting position), PPDT (-1.19, -2.23 to -0.14, p=0.03, only in sitting position), but they remained no longer significant after correction for multiple testing. Average pain intensity during 24 hours before the experiment (-0.34, -0.57 to -0.11, p=0.005, p=0.07 after Benjamini-Hochberg correction) showed some trend for interaction with drug effect, but only in the supine position.

Clobazam

Fifty patients were included in the clobazam arm, one of which did not show up for the second test session. Forty-nine patients were therefore analyzed (29 females, mean age 54.3 years, SD 15.8). A significant analgesic effect was found in the supine, but not in the sitting position (treatment effect compared to placebo: 0.7, 95%-CI 0.2 to 1.1, p=0.003), which is the object of a separate publication (Schliessbach et al., 2017). For supine pain, there were 29 responders in the verum session vs. 20 in the placebo session. For sitting pain, there were 28 responders in the verum session vs. 25 in the placebo session.

Baseline heat pain thresholds interacted with clobazam effect after 120 minutes in sitting but not in supine position (table 3 and supplementary table S3). Specifically, patients with baseline HPTT at limit (i.e. relatively insensible to heat) responded better to placebo, whereas more heat-sensitive patients had a better effect of clobazam. Treatment effects are shown in figure 3. In supine position, significant interaction was only found for the KCNS1 gene mutation, with the medium-pain-risk genotype pointing towards a more negative influence and the high-pain-risk genotype towards a positive influence on the effect of clobazam than the low-pain-risk genotype.

Genotyping

Genotyping was successfully performed in all 90 participants except for the rs4680 of the COMT gene and the CYP2D6*41 single-nucleotide polymorphism, each of which had 1 missing value. The results corresponded well with what was expected from a middle

European population. All but the CYP2D6*3A polymorphism were well within the Hardy-Weinberg equilibrium. Detailed allele frequencies are presented in supplementary table S4.

False discovery rate

After adjustment of p-values according to the Benjamini-Hochberg procedure, significant interactions of baseline variables and drug effect were only found in the imipramine experiment. For imipramine in supine position, the following descriptive variables remained significant (with 10% potential false discoveries among them): dichotomized baseline HPTT (leg and arm) and average pain in the last 24 hours. For imipramine in sitting position, the following variables remained significant (with 10% potential false discoveries among them): dichotomized baseline HPTT and CPDT (both leg and arm), both HPDT at leg and arm, CPDT at leg and arm, as well as HPTT at the arm. Among these 12 significant findings, 1-2 may be potential false discoveries.

Discussion

This study found a pronounced analgesic effect of oxycodone on low-back pain, but no evidence for any of the baseline characteristics to predict that effect. For imipramine, the data suggest that thermal sensory tests predict its effect: patients who are more sensitive to heat or cold pain had a better effect of imipramine than patients who were less sensitive to these modalities. While an analgesic effect was found for clobazam, no predictor could be identified.

Oxycodone

Oxycodone is a strong opioid with well documented analgesic effects in various acute and chronic pain conditions. Its short-term effectiveness on chronic low-back pain is therefore not surprising (Chaparro et al., 2013). The fact that average pain during the past 24 hours, catastrophizing and BDI were found to interact with oxycodone effect only in supine position suggests that these may be chance findings. Otherwise, there should have been at least a trend for these interactions in the sitting position as well. After correction for multiple testing, these variables were no longer significantly associated with drug effect. Yet, the study by Grosen et al. (Grosen et al., 2017) identified pain catastrophizing as a significant predictor for opioid efficacy. It must be noted, however, that their study population included patients with various pain syndromes, including head, neck and other musculoskeletal as well as neuropathic pain patients.

Of particular interest is the fact that not even the μ-opioid receptor A118G mutation significantly influenced the analgesic effect of oxycodone. This may partly be due to insufficient sample size, with no homozygous and only 16 heterozygous carriers of the mutant allele among the 50 patients. Another explanation may be that the influence of the genetic variant varies with the type of opioid used. There is evidence that carriers of the mutant G allele seem to have less analgesic effect of morphine (Campa et al., 2008), but in a similar

investigation for oxycodone such an association could not be demonstrated (Zwisler et al., 2012).

As to the prediction of oxycodone effect by QST, there was a previous study in healthy volunteers that found high basal heat pain thresholds and high degrees of temporal summation to be associated with greater oxycodone analgesia (Eisenberg et al., 2010). Neither of those parameters was found to influence oxycodone effect in the present study. These differing results cannot easily be compared, because outcome measures are not the same in pain patients and in volunteers and the study on healthy volunteers had no placebo control.

Another possible explanation may be the quite unanimous response to the drug in our study sample, with up to 88% of patients having significant pain reduction. The number of patients experiencing minimal or no effect may therefore have been too small to allow for sufficient discrimination between responders and non-responders.

Imipramine

The most consistent interactions were found in the imipramine experiment, where almost all thermal tests were associated with the effect of the drug. This was most pronounced for the dichotomized CPDT and HPTT and remained significant even after p-value adjustment for multiple testing. In particular, patients who reached the limits without having pain were less likely to experience a drug effect, whereas patients who did not reach the limits (i.e. who were more sensitive to heat and cold pain) experienced greater drug effect. The same tendency could be observed when thermal QST were analyzed as continuous variables, but less pronounced and only for pain in the sitting position.

Existing literature is mainly based on neuropathic pain patients, but has repeatedly found thermal pain thresholds to predict analgesic effects: Holbech et al. found that neuropathic pain patients with gain-of-function phenotype (including thermal allodynia) were more likely to benefit from imipramine (Holbech et al., 2016), and thermal pain thresholds were identified as

predictors of drug effect in post-herpetic neuralgia and traumatic nerve injury (Attal et al., 2004; Edwards et al., 2006).

It is increasingly recognized that there may be a neuropathic component in low-back pain patients even in the absence of typical radicular pain. However, no gold-standard tests exists to diagnose this reliably (Baron et al., 2016). A neuropathic component in our patient population could partly explain the observed results.

Clobazam

In the clobazam arm, the dichotomized HPTT were found to influence drug effect on pain in the sitting position in a similar way than for imipramine. The results suggested that patients who were more sensitive to heat pain (i.e. HPTT not at limit) experienced a greater analgesic effect of clobazam in sitting position. However, these results were no longer significant after correction for multiple testing, so we cannot rule out that they are chance findings. For pain in the supine position, where an analgesic effect was detected, only KCNS1 showed a significant interaction with drug effect. According to Costigan et al. (Costigan et al., 2010) the presence of one or two valine alleles confers an additive effect on pain threshold. The present results, however, were somewhat contradictory because homozygous (i.e. one valine allele) and heterozygous (i.e. both valine alleles) patients experienced opposite clobazam effects compared to the wild type. This was no longer significant after p-value correction and may therefore be a false-positive finding. Unfortunately, there is no existing literature specifically addressing clobazam in low-back pain to compare these findings to.

Implications of results

The search for parameters predicting the response to analgesic treatment has been of great interest in the past few years. Existing studies have addressed various forms of chronic pain. For instance, duloxetine for diabetic neuropathy seems to be more effective in patients with poor baseline CPM (Yarnitsky et al., 2012). Patients with chronic pancreatitis responded

415 better to treatment with pregabalin when they were hypersensitive to electrical stimulation
416 within the pancreatic dermatome Th10 (Olesen et al., 2013). As mentioned above, heat pain
417 thresholds predicted opioid analgesia in patients with post-herpetic neuralgia (Edwards et al.,
418 2006). It has been proposed that "dynamic" QST (e.g. temporal summation or CPM) are more
419 suitable than "static" paradigms (i.e. simple pain threshold measurements) to predict drug
420 efficacy and to distinguish "pro-nociceptive" and "anti-nociceptive" pain states (Yarnitsky et
421 al., 2012). However, for the prediction of opioid efficacy, both static and dynamic tests seem
422 to be useful (Eisenberg et al., 2010). Of note, static QST probably have a better long-term
423 reliability than dynamic tests (Marcuzzi et al., 2017). In this regard, caution must be taken not
424 to overrate experimental findings that solely rely on one-time assessments of dynamic QST.
425 To the best knowledge of the authors, no study has so far investigated the predictive ability of
426 QST in chronic low-back pain. In this respect, the present study adds important information to
427 the existing evidence, as chronic low-back pain is one of the most common painful disorders
428 in clinical practice.
429 The strict selection criteria of patients give us some confidence that we have enrolled a
430 sample of individuals with relatively homogeneous pathophysiology. Hypothesizing that the
431 majority of our patients had mainly nociceptive and not neuropathic pain might explain why
432 oxycodone but not imipramine showed a profound analgesic effect. Oxycodone has a specific
433 pharmacologic target at the μ-opioid receptor which may lead to pain relief in most patients
434 regardless of their QST-profile. Conversely, imipramine with its multiple pharmacologic
435 actions tended to relieve pain only in a subgroup of more heat- and cold-sensitive patients.
436 The question remains whether these patients had a certain neuropathic component in the
437 pathogenesis of their pain and therefore responded better to imipramine, or whether their
438 relative thermal hypersensitivity was an expression of a specific nociceptive mechanism in
439 which imipramine was particularly effective. It is tempting to speculate that these patients had

some sort of spinal hypersensitivity that responded well to imipramine-mediated modulation of inhibitory noradrenergic and serotonergic neural pathways.

Most studies about prediction of drug response by QST were conducted in neuropathic pain. Unlike low-back pain patients, neuropathic pain patients display a broad clinical picture of sensory alterations of thermal, mechanical or vibratory perception, alone or in combination, with gain or loss of function. According to this variety, three distinct phenotypic groups were identified (Baron et al., 2017): (1) patients with predominant sensory loss, (2) patients with heat hyperalgesia and (3) patients with mechanical hyperalgesia. The authors hypothesized that group 1 might best be treated with oral opioids, group 2 with oxcarbazepine or capsaicin and group 3 with gabapentinoids or lidocaine. These findings are promising, but need to be substantiated in future prospective studies. In the light of the present results, it seems unlikely that similar considerations pertain to chronic low-back pain, most probably because chronic low-back pain patients do not show such clearly distinguishable sensory phenotypes. Conceivably, the broader the spectrum of detectable sensory phenotypes, the greater the chances of identifying one particular phenotype that responds to a given drug. However, even in these cases, the statistical models could barely account for more than about 20% of observed variability (Edwards et al., 2006). Unfortunately, no two studies used the same QST paradigms, drugs or pain syndromes. Because of this methodologic heterogeneity, no firm conclusion about the ability of QST to predict analgesic response can be made at the time (Grosen et al., 2013).

Strengths and limitations

The present study is the first one to investigate the ability of QST to predict drug response in a fairly homogeneous and sufficiently large population of patients with chronic low-back pain. The QST protocol was extensive and included mechanical, thermal and electrical pain threshold as well as dynamic paradigms such as CPM and temporal summation, therefore

reflecting a wide range of nociceptive processes. However, other modalities could be included provide complementary information. Three drugs with different modes of action were studied: oxycodone as a clearly defined μ-opioid agonist, imipramine with multiple pharmacologic actions such as sodium channel blockade and central noradrenergic and serotonergic effects, and clobazam as a modulator of spinal inhibitory GABA-ergic transmission.

A large number of statistical tests had to be performed as a consequence of the extensive protocol, bearing the risk of chance findings. The few statistically significant results have therefore to be interpreted in this context, although the data were corrected for multiple testing. A multivariable model with a combination of predictors was not within the scope of this study and interactions between predictors cannot be excluded. The fact that some patients were randomized to more than one drug may introduce the risk of a selection bias. Finally, this was a single-dose study with an observation time of 2 hours, intended to investigate immediate effects from a mechanistic point of view. Immediate effects could indeed be demonstrated for oxycodone and clobazam. Unfortunately, no immediate effects were seen for imipramine. This does not imply that imipramine is ineffective in low-back pain, as most previous studies investigating tricyclic antidepressants used treatment periods of several weeks.

Conclusion

This is the first study to address the ability of QST to predict drug effect in chronic low-back pain. None of the selected QST measures could be identified as predictor of analgesic effect of oxycodone or clobazam. We found evidence that patients more sensitive to heat and cold pain respond better to imipramine. None of the candidate genes involved in pain sensitivity or drug metabolism seemed to be a predictor of drug effect.

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Author contributions

Conception and design: A.S., M.C., P.J. H.U.Z. and L.A.N.

Data acquisition: J.S. and P.H.V.

Data analyses: L.B. (statistics), U.S. (genotyping)

Interpretation of results: all authors

Manuscript drafting: J.S. and M.C.

All authors critically revised the manuscript and agreed to the final version

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646

647 **Table 1**

	No. of observations	Interaction with drug effect	P-value from LR test	Adjusted p-value
Sex	98		0.15	0.58
male	36	0 (Ref)		
female	62	-0.69 (-1.62 to 0.23)		
Operated due to pain	96		0.59	0.72
no	77	0 (Ref)		
yes	19	-0.31 (-1.42 to 0.81)		
KCNS1	98		0.76	0.83
low pain risk	21	0 (Ref)		
medium pain risk	56	0.04 (-1.04 to 1.13)		
high pain risk	21	-0.36 (-1.68 to 0.95)		
GCH1	98		0.17	0.58
no pain protect	74	0 (Ref)		
one pain protect	18	1.00 (-0.07 to 2.06)		
both pain protect	6	0.67 (-1.09 to 2.44)		
OPRM1	98		0.30	0.72
homozygous wild type	66	0 (Ref)		
1/2 mutant allele	32	0.49 (-0.43 to 1.41)		
COMT	98		0.58	0.72
low pain sensitivity	12	0 (Ref)		
average pain sensitivity	76	0.37 (-1.03 to 1.78)		
high pain sensitivity	10	0.96 (-0.87 to 2.79)		
2D6	98		0.55	0.72
poor metabolizer	6	0 (Ref)		
intermediate metabolizer	39	0.39 (-1.53 to 2.31)		
extensive metabolizer	51	0.59 (-1.24 to 2.42)		
ultrarapid metabolizer	2	2.42 (-0.97 to 5.80)		
2C19	98		0.52	0.72
poor metabolizer	2	0 (Ref)		
intermediate metabolizer	20	1.82 (-1.33 to 4.97)		
extensive metabolizer	76	1.76 (-1.26 to 4.77)		
3A5	98		0.09	0.44
low expressors	81	0 (Ref)		
normal/high expressors	17	0.99 (-0.15 to 2.13)		
T20 decreasers	98		0.32	0.72
No decrease	89	0 (Ref)		
Decrease	9	0.92 (-0.86 to 2.69)		
Baseline HPTT (leg) at limit	97		0.004	0.07
No	58	0 (Ref)		
Yes	39	-1.34 (-2.23 to -0.46)		
Baseline HPTT (arm) at limit	91		0.003	0.07
No	70	0 (Ref)		
yes	21	-1.59 (-2.62 to -0.56)		
Baseline CPDT (leg) at limit	97		0.71	0.82
no	52	0 (Ref)		
yes	45	-0.18 (-1.15 to 0.78)		
Baseline CPDT (arm) at limit	91		0.06	0.42

no	65	0 (Ref)		
yes	26	-1.00 (-2.01 to 0.01)		
Age	98	0.08 (-0.34 to 0.51)	0.70	0.82
BMI	98	-0.19 (-0.62 to 0.24)	0.38	0.72
Pain duration	98	-0.38 (-0.80 to 0.03)	0.08	0.42
Average pain in the last 24h	98	-0.59 (-0.99 to -0.19)	0.005	0.07
Impairment of daily life	98	-0.18 (-0.61 to 0.26)	0.43	0.72
Catastrophizing score	98	-0.26 (-0.69 to 0.17)	0.24	0.62
Beck Depression Index	98	0.12 (-0.32 to 0.57)	0.59	0.72
CPM	97	-0.21 (-0.69 to 0.26)	0.38	0.72
PPDT	98	0.03 (-0.42 to 0.48)	0.90	0.92
PPTT	98	0.21 (-0.26 to 0.67)	0.39	0.72
ESPT	98	-0.06 (-0.54 to 0.41)	0.79	0.84
ERPT	98	-0.02 (-0.50 to 0.46)	0.93	0.93
Iwsec	97	-0.16 (-0.60 to 0.28)	0.48	0.72
HPDT (leg)	97	-0.57 (-1.03 to -0.11)	0.020	0.19
HPDT (arm)	91	-0.33 (-0.81 to 0.15)	0.18	0.58
HPTT (leg)	97	-0.29 (-0.75 to 0.17)	0.22	0.61
HPTT (arm)	91	-0.30 (-0.77 to 0.18)	0.22	0.61
CPDT (leg)	97	0.15 (-0.32 to 0.62)	0.53	0.72
CPDTT (arm)	91	0.43 (-0.01 to 0.86)	0.07	0.42

Table 1: Imipramine in supine position: Interaction of baseline parameters with the effect of imipramine on pain (NRS) at 120 min. A positive interaction term indicates a positive influence on the effect of imipramine compared to placebo. Adjusted p-values are corrected for multiple testing and indicate the proportion of false-positive discoveries. LR-test = likelihood-ratio test, KCNS1 = potassium channel subunit, GCH1 = GTP-cyclohydrolase, OPRM1 = mu-opioid receptor variant A118G, COMT = catechol-O-methyltransferase, 2D6/2C19/3A5 = cytochrome P450 2D6, 2C19, 3A5. T20 = electrical train-of-twenty stimulation. HPDT/HPTT = heat pain detection/tolerance threshold, CPDT = cold pain detection threshold, BMI = body mass index, CPM = conditioned pain modulation, PPDT/PPTT = pressure pain detection/tolerance thresholds, ESPT/ERPT = electrical single and repeated pain threshold, Iwsec = time in seconds during cold pressor test until cold pain reaches 7/10 on the numeric rating scale.

661 **Table 2**

	No. of observations	Interaction with drug effect	P-value from LR test	Adjusted p-value
Sex	98		0.96	0.96
male	36	0 (Ref)		
female	62	-0.02 (-0.84 to 0.80)		
Operated due to pain	96		0.33	0.50
no	77	0 (Ref)		
yes	19	-0.48 (-1.44 to 0.48)		
KCNS1	98		0.75	0.86
low pain risk	21	0 (Ref)		
medium pain risk	56	-0.37 (-1.38 to 0.63)		
high pain risk	21	-0.38 (-1.55 to 0.78)		
GCH1	98		0.22	0.39
no pain protect	74	0 (Ref)		
one pain protect	18	0.88 (-0.12 to 1.88)		
both pain protect	6	0.49 (-1.13 to 2.10)		
OPRM1	98		0.047	0.16
homozygous wild	66	0 (Ref)		
1/2 mutant allele	32	0.84 (0.03 to 1.66)		
COMT	98		0.05	0.16
low pain sensitivity	12	0 (Ref)		
average pain sensitivity	76	-0.02 (-1.24 to 1.20)		
high pain sensitivity	10	1.51 (-0.09 to 3.11)		
2D6	98		0.79	0.86
poor metabolizer	6	0 (Ref)		
intermediate metabolizer	39	0.37 (-1.45 to 2.19)		
extensive metabolizer	51	0.69 (-1.03 to 2.42)		
ultrarapid metabolizer	2	0.49 (-2.62 to 3.60)		
2C19	98		0.46	0.60
poor metabolizer	2	0 (Ref)		
intermediate metabolizer	20	-1.61 (-4.44 to 1.21)		
extensive metabolizer	76	-1.72 (-4.45 to 1.00)		
3A5	98		0.17	0.37
low expressors	81	0 (Ref)		
normal/high expressors	17	0.72 (-0.30 to 1.74)		
T20 decreasers	98		0.41	0.58
No decrease	89	0 (Ref)		
Decrease	9	-0.65 (-2.20 to 0.90)		
Baseline HPTT (leg) at limit	97		0.006	0.027
No	58	0 (Ref)		
Yes	39	-1.18 (-1.96 to -0.39)		
Baseline HPTT (arm) at limit	91		<0.001	0.001
No	70	0 (Ref)		
Yes	21	-1.93 (-2.81 to -1.05)		
Baseline CPDT (leg) at limit	97		0.005	0.027
No	52	0 (Ref)		
yes	45	-1.20 (-2.00 to -0.39)		

Baseline CPDT (arm) at limit	91		<0.001	0.002
no	65	0 (Ref)		
yes	26	-1.72 (-2.55 to -0.89)		
Age	98	0.21 (-0.19 to 0.60)	0.30	0.47
BMI	98	-0.25 (-0.63 to 0.13)	0.20	0.39
Pain duration	98	-0.36 (-0.73 to 0.02)	0.07	0.18
Average pain in the last 24h	98	-0.34 (-0.73 to 0.04)	0.09	0.23
Impairment of daily life	98	-0.01 (-0.40 to 0.38)	0.96	0.96
Catastrophizing score	98	0.02 (-0.37 to 0.41)	0.93	0.96
Beck Depression Index	98	0.24 (-0.15 to 0.62)	0.23	0.39
CPM	97	-0.14 (-0.56 to 0.28)	0.52	0.65
PPDT	98	-0.44 (-0.83 to -0.05)	0.030	0.12
PPTT	98	-0.17 (-0.57 to 0.23)	0.41	0.58
ESPT	98	-0.35 (-0.76 to 0.07)	0.11	0.26
ERPT	98	-0.28 (-0.71 to 0.14)	0.19	0.39
Iwsec	97	-0.24 (-0.63 to 0.16)	0.24	0.39
HPDT (leg)	97	-0.69 (-1.07 to -0.30)	0.001	0.009
HPDT (arm)	91	-0.80 (-1.16 to -0.43)	<0.001	0.001
HPTT (leg)	97	-0.32 (-0.73 to 0.09)	0.13	0.31
HPTT (arm)	91	-0.49 (-0.90 to -0.08)	0.021	0.09
CPDT (leg)	97	0.59 (0.20 to 0.99)	0.005	0.027
CPDTT (arm)	91	0.80 (0.43 to 1.17)	<0.001	0.001

Table 2: Imipramine in sitting position: Interaction of baseline parameters with the effect of imipramine on pain (NRS) at 120 min. A positive interaction term indicates a positive influence on the effect of imipramine compared to placebo.

667 **Table 3**

	No. of observations	Interaction with drug effect	P-value from LR test	Adjusted p-value
Sex	97		0.15	0.84
male	40	0 (Ref)		
female	57	0.55 (-0.19 to 1.29)		
Operated due to pain	95		0.49	0.94
no	79	0 (Ref)		
yes	16	-0.33 (-1.27 to 0.61)		
KCNS1	97		0.14	0.84
low pain risk	18	0 (Ref)		
medium pain risk	46	-0.76 (-1.73 to 0.21)		
high pain risk	33	-0.04 (-1.06 to 0.99)		
GCH1	97		0.85	0.94
no pain protect	75	0 (Ref)		
one pain protect	20	-0.24 (-1.18 to 0.70)		
both pain protect	2	0.37 (-2.20 to 2.94)		
OPRM1	97		0.32	0.84
homozygous wild	66	0 (Ref)		
1/2 mutant allele	31	0.41 (-0.39 to 1.20)		
COMT	97		0.91	0.94
low pain sensitivity	8	0 (Ref)		
average pain sensitivity	81	0.29 (-1.10 to 1.69)		
high pain sensitivity	8	0.34 (-1.45 to 2.13)		
2D6	97		0.28	0.84
poor metabolizer	6	0 (Ref)		
intermediate metabolizer	36	-1.31 (-2.90 to 0.29)		
extensive metabolizer	55	-1.16 (-2.72 to 0.40)		
ultrarapid metabolizer	0			
2C19	97		0.86	0.94
poor metabolizer	0			
intermediate metabolizer	32	0 (Ref)		
extensive metabolizer	65	-0.07 (-0.89 to 0.74)		
3A5	97		0.81	0.94
low expressors	82	0 (Ref)		
normal/high expressors	15	0.13 (-0.96 to 1.23)		
T20 decreasers	95		0.38	0.94
No decrease	83	0 (Ref)		
Decrease	12	-0.62 (-2.00 to 0.75)		
Baseline HPTT (leg) at limit	97		0.011	0.20
No	60	0 (Ref)		
Yes	37	-1.10 (-1.90 to -0.29)		
Baseline HPTT (arm) at limit	92		0.007	0.20
No	69	0 (Ref)		
Yes	23	-1.23 (-2.10 to -0.36)		
Baseline CPDT (leg) at limit	97		0.22	0.84
No	51	0 (Ref)		
yes	46	-0.50 (-1.29 to 0.30)		

Baseline CPDT (arm) at limit	92		0.49	0.94
no	60	0 (Ref)		
yes	32	-0.32 (-1.22 to 0.58)		
Age	97	-0.20 (-0.56 to 0.16)	0.28	0.84
BMI	97	-0.10 (-0.50 to 0.30)	0.63	0.94
Pain duration	95	0.11 (-0.29 to 0.52)	0.58	0.94
Average pain in the last 24h	97	0.20 (-0.18 to 0.58)	0.30	0.84
Impairment of daily life	97	-0.20 (-0.57 to 0.16)	0.29	0.84
Catastrophizing score	97	-0.09 (-0.47 to 0.29)	0.64	0.94
Beck Depression Index	97	-0.06 (-0.43 to 0.31)	0.76	0.94
CPM	94	0.08 (-0.34 to 0.50)	0.70	0.94
PPDT	97	0.14 (-0.23 to 0.52)	0.45	0.94
PPTT	97	-0.06 (-0.44 to 0.32)	0.76	0.94
ESPT	95	-0.00 (-0.43 to 0.43)	0.99	0.99
ERPT	95	-0.13 (-0.58 to 0.31)	0.55	0.94
Iwsec	94	-0.02 (-0.40 to 0.36)	0.92	0.94
HPDT (leg)	97	-0.40 (-0.80 to 0.00)	0.05	0.68
HPDT (arm)	92	-0.08 (-0.50 to 0.33)	0.69	0.94
HPTT (leg)	97	-0.24 (-0.68 to 0.20)	0.30	0.84
HPTT (arm)	92	-0.09 (-0.49 to 0.31)	0.66	0.94
CPDT (leg)	97	0.06 (-0.34 to 0.45)	0.78	0.94
CPDTT (arm)	92	0.06 (-0.34 to 0.47)	0.76	0.94

Table 3: Clobazam in sitting position: Interaction of baseline parameters with the effect of clobazam on pain (NRS) at 120 min. A positive interaction term indicates a positive influence on the effect of clobazam compared to placebo.

673 **Supplementary table S1**

	No. of observations	Interaction with drug effect	p-value from LR test	Adjusted p-value
Sex	97		0.57	0.94
male	45	0 (Ref)		
female	52	0.29 (-0.71 to 1.29)		
Surgery due to pain	97		0.29	0.86
no	79	0 (Ref)		
yes	18	0.72 (-0.60 to 2.04)		
KCNS1	97		0.27	0.86
low pain risk	23	0 (Ref)		
medium pain risk	52	0.22 (-0.99 to 1.43)		
high pain risk	22	-0.80 (-2.25 to 0.66)		
GCH1	97		0.58	0.94
no pain protect	71	0 (Ref)		
one pain protect	22	0.39 (-0.79 to 1.56)		
both pain protect	4	1.14 (-1.34 to 3.61)		
OPRM1	97		0.30	0.86
homozygous wt	65	0 (Ref)		
1/2 mutant allele	32	-0.56 (-1.61 to 0.49)		
COMT	97		0.13	0.86
low pain sens	14	0 (Ref)		
average pain sens	74	1.33 (-0.04 to 2.69)		
high pain senss	9	1.80 (-0.23 to 3.83)		
2D6	97		0.97	0.97
poor metabol	10	0 (Ref)		
intermediate metabol	39	-0.02 (-1.75 to 1.71)		
extensive metabol	44	0.15 (-1.57 to 1.88)		
ultrarapid metabol	4	-0.45 (-3.34 to 2.43)		
2C19	97		0.80	0.94
poor metabol	0			
intermediate metabol	24	0 (Ref)		
extensive metabol	73	-0.16 (-1.33 to 1.02)		
3A5	97		0.23	0.86
low expressors	79	0 (Ref)		
normal/high expressors	18	-0.80 (-2.08 to 0.49)		
T20 decreasers	97		0.95	0.97
max=end	79	0 (Ref)		
end<max	18	-0.05 (-1.42 to 1.33)		
Baseline HPTT (leg) at limit	97		0.73	0.94
no	51	0 (Ref)		
yes	46	0.18 (-0.85 to 1.20)		
Baseline HPTT (arm) at limit	93		0.16	0.86
no	63	0 (Ref)		
yes	30	-0.81 (-1.94 to 0.31)		
Baseline CPDT (leg) at limit	97		0.94	0.97
no	50	0 (Ref)		
yes	47	0.04 (-1.01 to 1.09)		
Baseline CPDT (arm) at limit	93		0.96	0.97
no	65	0 (Ref)		
yes	28	-0.03 (-1.19 to 1.12)		
Age (per decade)	97	0.05 (-0.29 to 0.38)	0.79	0.94
BMI	95	0.03 (-0.09 to 0.15)	0.60	0.94
Pain duration	95	-0.01 (-0.05 to 0.04)	0.77	0.94
Average pain in the last 24h	95	0.50 (0.16 to 0.84)	0.005	0.20
Impairment of daily life	95	0.29 (-0.14 to 0.71)	0.19	0.86

	No. of observations	Interaction with drug effect	p-value from LR test	Adjusted p-value
Catastrophizing score	95	0.45 (0.06 to 0.84)	0.027	0.52
Beck Depression Index	95	0.21 (-0.00 to 0.42)	0.06	0.74
CPM	97	-0.13 (-0.68 to 0.41)	0.63	0.94
PPDT	97	-0.30 (-1.67 to 1.07)	0.67	0.94
PPTT	97	0.51 (-1.22 to 2.24)	0.57	0.94
ESPT	97	-0.57 (-1.61 to 0.46)	0.28	0.86
ERPT	97	-0.43 (-1.46 to 0.60)	0.42	0.94
Iwsec	97	0.12 (-0.61 to 0.85)	0.75	0.94
HPDT (leg)	97	0.04 (-0.12 to 0.21)	0.59	0.94
HPDT (arm)	93	-0.03 (-0.15 to 0.09)	0.60	0.94
HPTT (leg)	97	0.18 (-0.19 to 0.55)	0.35	0.94
HPTT (arm)	93	0.04 (-0.21 to 0.28)	0.78	0.94
CPDT (leg)	97	-0.02 (-0.07 to 0.03)	0.38	0.94
CPDTT (arm)	93	-0.01 (-0.06 to 0.04)	0.70	0.94

Supplementary table S1: Oxycodone in supine position: Interaction of baseline parameters with the effect of oxycodone on pain (NRS) at 120 min. A positive interaction term indicates a positive influence on the effect of oxycodone compared to placebo.

Supplementary table S2

	No. of observations	Interaction with drug effect	p-value from LR test	Adjusted p-value
Sex	98		0.09	0.98
male	46	0 (Ref)		
female	52	0.83 (-0.11 to 1.77)		
Surgery due to pain	98		0.18	0.98
no	79	0 (Ref)		
yes	19	0.89 (-0.41 to 2.18)		
KCNS1	98		0.83	0.98
low pain risk	23	0 (Ref)		
medium pain risk	51	0.16 (-1.05 to 1.37)		
high pain risk	24	-0.21 (-1.61 to 1.20)		
GCH1	98		0.78	0.98
no pain protect	72	0 (Ref)		
one pain protect	22	0.13 (-1.03 to 1.29)		
both pain protect	4	0.87 (-1.59 to 3.33)		
OPRM1	98		0.26	0.98
homozygous wt	66	0 (Ref)		
1/2 mutant allele	32	-0.59 (-1.61 to 0.42)		
COMT	98		0.81	0.98
low pain sens	16	0 (Ref)		
average pain sens	73	-0.15 (-1.47 to 1.16)		
high pain senss	9	0.40 (-1.60 to 2.40)		
2D6	98		0.44	0.98
poor metabol	10	0 (Ref)		
intermediate metabol	41	0.89 (-0.75 to 2.53)		
extensive metabol	43	1.28 (-0.38 to 2.94)		
ultrarapid metabol	4	0.18 (-2.58 to 2.95)		
2C19	98		1.00	1.00
poor metabol	0	0 (Ref)		
intermediate metabol	24			
extensive metabol	74	0.00 (-1.14 to 1.14)		
3A5	98		1.00	1.00
low expressors	80	0 (Ref)		
normal/high expressors	18	-0.00 (-1.26 to 1.26)		
T20 decreasers	98		0.20	0.98
max=end	81	0 (Ref)		
end<max	17	0.88 (-0.44 to 2.20)		
Baseline HPTT (leg) at limit	97		0.51	0.98
no	51	0 (Ref)		
yes	46	0.34 (-0.66 to 1.33)		
Baseline HPTT (arm) at limit	93		0.28	0.98
no	63	0 (Ref)		
yes	30	0.61 (-0.48 to 1.71)		
Baseline CPDT (leg) at limit	97		0.94	1.00
no	49	0 (Ref)		
yes	48	0.04 (-0.99 to 1.07)		
Baseline CPDT (arm) at limit	93		0.89	0.99
no	65	0 (Ref)		
yes	28	0.08 (-1.05 to 1.22)		
Age (per decade)	98	-0.12 (-0.44 to 0.20)	0.45	0.98
BMI	96	-0.07 (-0.18 to 0.05)	0.26	0.98
Pain duration	96	-0.01 (-0.05 to 0.04)	0.80	0.98
Average pain in the last 24h	96	0.14 (-0.20 to 0.49)	0.42	0.98
Impairment of daily life	96	-0.21 (-0.63 to 0.22)	0.34	0.98

	No. of observations	Interaction with drug effect	p-value from LR test	Adjusted p-value
Catastrophizing score	96	-0.12 (-0.52 to 0.28)	0.56	0.98
Beck Depression Index	96	-0.09 (-0.30 to 0.12)	0.38	0.98
CPM	98	-0.16 (-0.67 to 0.36)	0.56	0.98
PPDT	98	-0.20 (-1.55 to 1.15)	0.77	0.98
PPTT	98	-0.03 (-1.74 to 1.67)	0.97	1.00
ESPT	98	-0.26 (-1.27 to 0.75)	0.61	0.98
ERPT	98	-0.09 (-1.10 to 0.92)	0.86	0.99
Iwsec	98	0.20 (-0.50 to 0.91)	0.57	0.98
HPDT (leg)	97	0.05 (-0.11 to 0.21)	0.51	0.98
HPDT (arm)	93	0.04 (-0.08 to 0.16)	0.51	0.98
HPTT (leg)	97	0.05 (-0.32 to 0.42)	0.78	0.98
HPTT (arm)	93	0.09 (-0.15 to 0.33)	0.45	0.98
CPDT (leg)	97	-0.02 (-0.06 to 0.03)	0.49	0.98
CPDTT (arm)	93	-0.02 (-0.07 to 0.03)	0.43	0.98

Supplementary table S2: Oxycodone in sitting position: Interaction of baseline parameters with the effect of oxycodone on pain (NRS) at 120 min. A positive interaction term indicates a positive influence on the effect of oxycodone compared to placebo.

686 **Supplementary table S3**

	No. of observations	Interaction with drug effect	p-value from LR test	Adjusted p-value
Sex	94		0.80	0.90
male	37	0 (Ref)		
female	57	0.12 (-0.80 to 1.04)		
Surgery due to pain	92		0.27	0.71
no	76	0 (Ref)		
yes	16	0.66 (-0.50 to 1.81)		
KCNS1	94		0.007	0.12
low pain risk	18	0 (Ref)		
medium pain risk	46	-0.57 (-1.65 to 0.50)		
high pain risk	30	0.98 (-0.18 to 2.13)		
GCH1	94		0.23	0.71
no pain protect	72	0 (Ref)		
one pain protect	20	-0.91 (-1.97 to 0.14)		
both pain protect	2	0.38 (-2.63 to 3.40)		
OPRM1	94		0.85	0.90
homozygous wt	66	0 (Ref)		
1/2 mutant allele	28	0.09 (-0.88 to 1.06)		
COMT	94		0.84	0.90
low pain sens	8	0 (Ref)		
average pain sens	78	-0.50 (-2.17 to 1.17)		
high pain senss	8	-0.45 (-2.59 to 1.70)		
2D6	94		0.26	0.71
poor metabol	6	0 (Ref)		
intermediate metabol	36	-1.00 (-2.88 to 0.88)		
extensive metabol	52	-0.27 (-2.10 to 1.56)		
ultrarapid metabol	0			
2C19	94		0.40	0.83
poor metabol	0			
intermediate metabol	32	0 (Ref)		
extensive metabol	62	0.42 (-0.56 to 1.39)		
3A5	94		0.32	0.71
low expressors	82	0 (Ref)		
normal/high expressors	12	-0.71 (-2.13 to 0.70)		
T20 decreasers	92		0.24	0.71
max=end	81	0 (Ref)		
end<max	11	-0.88 (-2.33 to 0.56)		
Baseline HPTT (leg) at limit	94		0.11	0.71
no	57	0 (Ref)		
yes	37	-0.78 (-1.71 to 0.15)		
Baseline HPTT (arm) at limit	89		0.29	0.71
no	66	0 (Ref)		
yes	23	-0.57 (-1.60 to 0.46)		
Baseline CPDT (leg) at limit	94		0.64	0.87
no	50	0 (Ref)		
yes	44	-0.22 (-1.15 to 0.70)		
Baseline CPDT (arm) at limit	89		0.82	0.90
no	58	0 (Ref)		
yes	31	-0.11 (-1.12 to 0.89)		
Age (per decade)	94	-0.09 (-0.38 to 0.20)	0.53	0.84
BMI	94	0.02 (-0.08 to 0.12)	0.75	0.90
Pain duration	92	-0.00 (-0.05 to 0.04)	0.85	0.90
Average pain in the last 24h	94	0.23 (-0.01 to 0.46)	0.06	0.70
Impairment of daily life	94	0.01 (-0.31 to 0.33)	0.97	0.97

	No. of observations	Interaction with drug effect	p-value from LR test	Adjusted p-value
Catastrophizing score	94	0.12 (-0.22 to 0.46)	0.48	0.84
Beck Depression Index	94	0.04 (-0.15 to 0.24)	0.66	0.87
CPM	91	0.13 (-0.28 to 0.54)	0.54	0.84
PPDT	94	-0.46 (-1.68 to 0.77)	0.47	0.84
PPTT	94	-0.93 (-2.27 to 0.41)	0.18	0.71
ESPT	92	0.55 (-0.35 to 1.45)	0.24	0.71
ERPT	92	0.48 (-0.47 to 1.44)	0.33	0.71
Iwsec	91	-0.15 (-0.81 to 0.51)	0.66	0.87
HPDT (leg)	94	-0.16 (-0.33 to 0.01)	0.08	0.70
HPDT (arm)	89	-0.03 (-0.14 to 0.08)	0.64	0.87
HPTT (leg)	94	-0.30 (-0.68 to 0.09)	0.14	0.71
HPTT (arm)	89	-0.09 (-0.30 to 0.13)	0.44	0.84
CPDT (leg)	94	0.00 (-0.04 to 0.05)	0.97	0.97
CPDTT (arm)	89	0.01 (-0.03 to 0.06)	0.55	0.84

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688 Supplementary table S3: Clobazam in supine position: Interaction of baseline parameters with
689 the effect of clobazam on pain (NRS) at 120 min. A positive interaction term indicates a
690 positive influence on the effect of clobazam compared to placebo.

691

692

Supplementary table S4

Gene		SNP	Allele Frequency n(%)			Hardy Weinberg X^2	p-value
			11	12	22		
KCNS1		rs734784	28 (31%)	42 (47%)	20 (22%)	0.32	0.57
GCH-1		rs8007267	4 (5%)	22 (24%)	64 (71%)	1.30	0.25
		rs3783641	61 (67%)	24 (27%)	5 (6%)	1.51	0.21
		rs10483639	62 (68%)	23 (26%)	5 (6%)	1.93	0.16
OPRM	A118G	rs1799971	58 (65%)	30 (33%)	2 (2%)	0.69	0.4
COMT		rs6269	12 (13%)	47 (53%)	31 (34%)	0.78	0.37
		rs4633	20 (22%)	50 (56%)	20 (22%)	1.11	0.29
		rs4818	33 (37%)	45 (50%)	12 (13%)	0.30	0.58
		rs4680[1]	20 (22%)	49 (56%)	20 (22%)	0.91	0.34
CYP3A	3A4*1b	rs2740574	83 (92%)	7 (8%)	-	0.15	0.70
	3A5*3	rs776746	1 (1%)	14 (16%)	75 (83%)	0.14	0.70
CYP2D6	CYP2D6*6	rs5030655	90 (100%)	-	-	n/a	-
	CYP2D6*8	rs5030865	90 (100%)	-	-	n/a	-
	CYP2D6*10	rs1065852	53 (59%)	32 (35%)	5 (6%)	0.06	0.95
	CYP2D6*41	rs28371725[1]	76 (85%)	12 (14%)	1 (1%)	0.43	0.51
	CYP2D6*3A	rs35742686	86 (96%)	3 (3%)	1 (1%)	**13.2**	**<0.001**
	CYP2D6*4	rs3892097	57 (63%)	28 (31%)	5 (6%)	0.39	0.53
	CYP2D6*5	Gene deletion	Normal: 82 (91%)		Deleted: 8 (9%)	n/a	-
	CYP2D6*2	Gene multiplication	Normal: 87 (97%)		Multiple: 3 (3%)	n/a	-
CYP2C19	CYP2C19*2	rs4244285	68 (76%)	21 (23%)	1 (1%)	0.20	0.66
	CYP2C19*3	Rs4986893	90 (100%)	-	-	n/a	-

Supplementary table S4: Allele frequencies for each of the genotyped single-nucleotide polymorphisms (n=90). KCNS1 = Potassium voltage-gated channel subfamily S member 1, GCH-1 = GTP-Cyclohydrolase, OPRM = mu opioid receptor, COMT = catechol-O-methyltransferase, CYP = Cytochrome P450. 1One missing value (n=89)

Figure 1

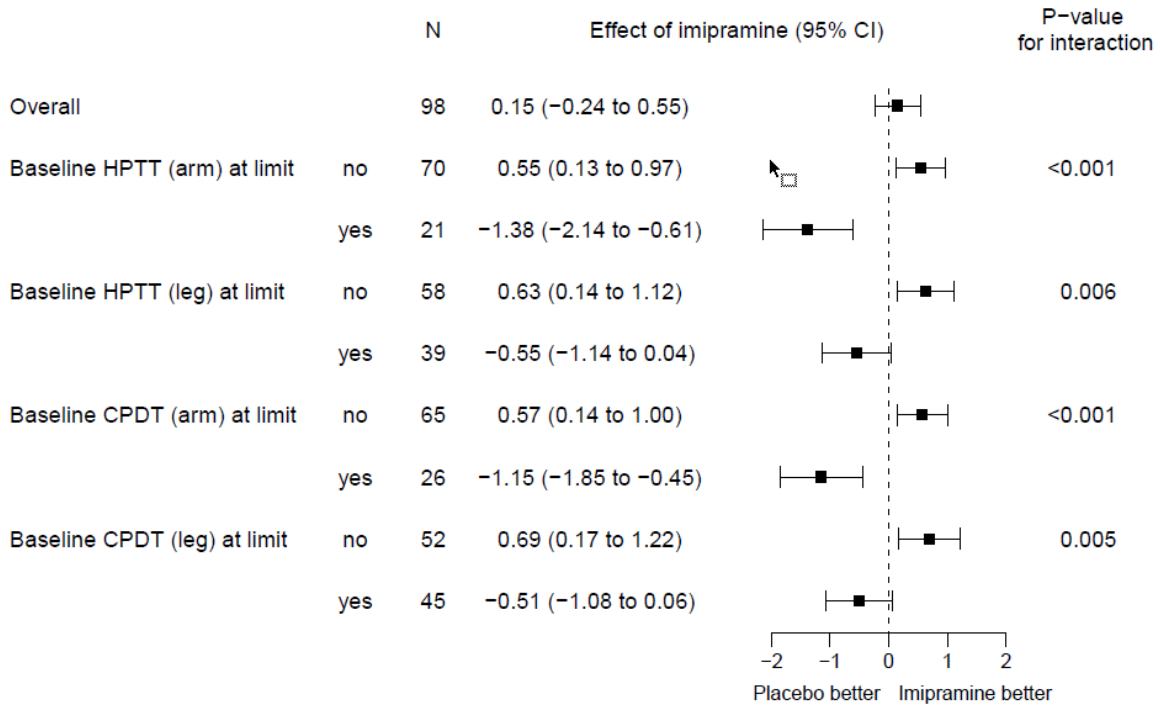

Figure 1: Effect of imipramine versus placebo in sitting position. A positive number indicates a positive effect (i.e. a decrease in pain). Imipramine is more effective in cold/heat sensitive patients. Two patients had missing values for pain in the imipramine phase. NRS = numeric rating scale, HPTT = heat pain tolerance threshold, CPDT = cold pain detection threshold.

Figure 2

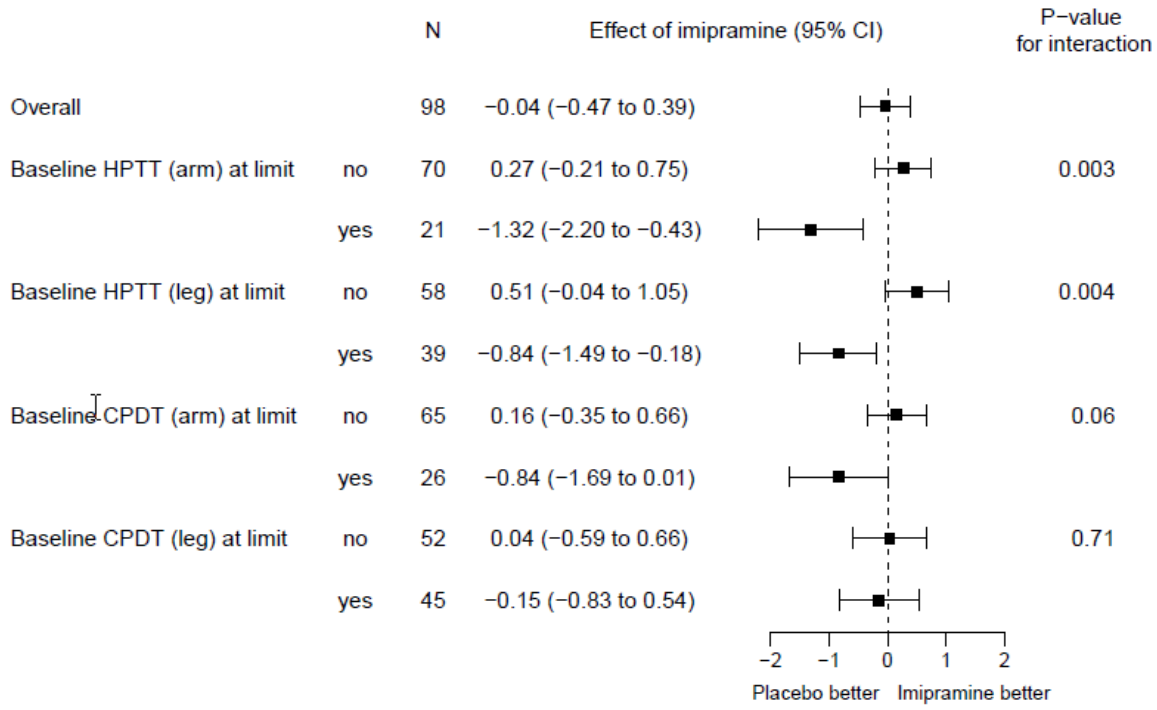

Figure 2: Effect of imipramine versus placebo in supine position. A positive number indicates a positive effect (i.e. a decrease in pain). There is a trend towards better effect of imipramine in heat-sensitive patients. Two patients had missing values for pain in the imipramine phase.

Figure 3

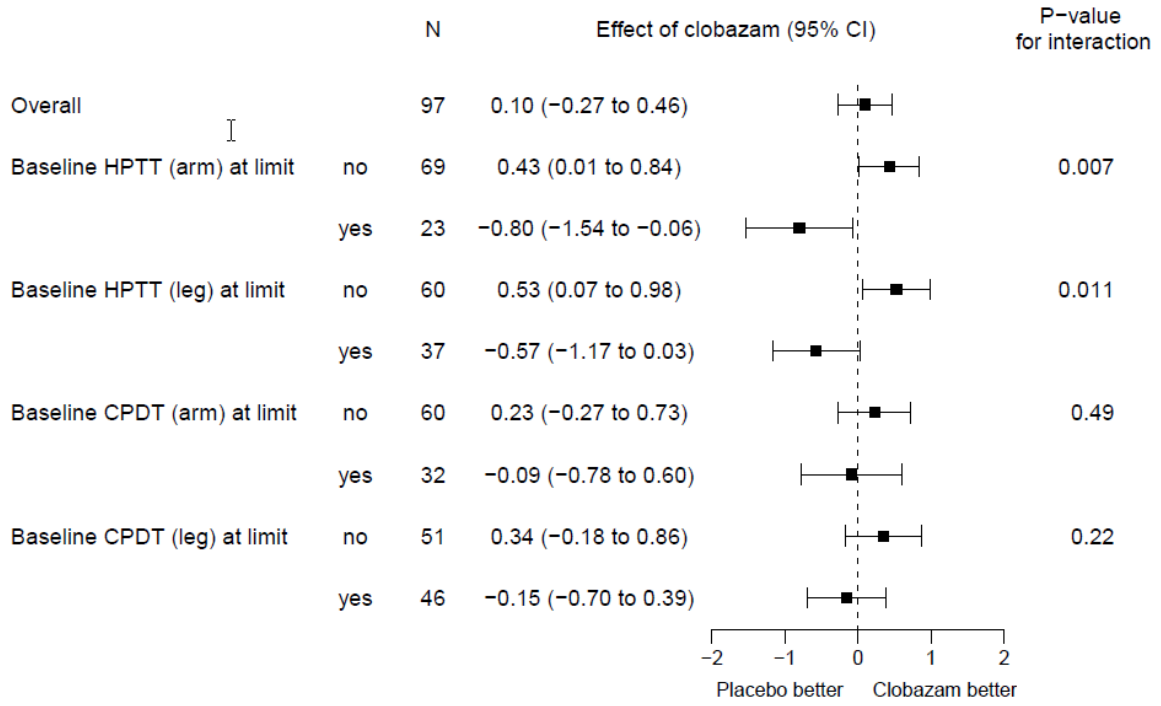

Figure 3: Effect of clobazam versus placebo in sitting position. A positive number indicates a positive effect (i.e. a decrease in pain). One patient had missing values for pain in the placebo phase.