BMJ Open

# Pain Predict Genetics: Protocol for a prospective observational study of clinical and genetic factors to predict the development of postoperative pain.



| | |
|---|---|
| Journal: | *BMJ Open* |
| Manuscript ID | bmjopen-2022-066134 |
| Article Type: | Protocol |
| Date Submitted by the Author: | 28-Jun-2022 |
| Complete List of Authors: | LI, SONG; Radboudumc, Department of Human Genetics<br>van Boekel, Regina L.M.; Radboudumc, Department of Anesthesiology, Pain and Palliative Medicine<br>van den Heuvel, Sandra ; Radboudumc, Department of Anesthesiology, Pain and Palliative Medicine<br>Coenen, Marieke J.H.; Radboudumc, Department of Human Genetics<br>Vissers, Kris ; Radboudumc, Department of Anesthesiology, Pain and Palliative Medicine |
| Keywords: | PAIN MANAGEMENT, GENETICS, SURGERY, EPIDEMIOLOGY |
| | |

# Pain Predict Genetics: Protocol for a prospective observational study of clinical and genetic factors to predict the development of postoperative pain.

Song Li [1], Regina L.M. van Boekel [2], Sandra A.S. van den Heuvel [2], Marieke J.H. Coenen [1], Kris C.P. Vissers [2]

Authors' Affiliations:

[1] Department of Human Genetics, Radboud Institute for Health Sciences, Radboud university medical center, Nijmegen, The Netherlands.

[2] Department of Anesthesiology, Pain and Palliative Medicine, Radboud university medical center, Nijmegen, The Netherlands.

Email:

Song Li: Song.Li@radboudumc.nl

Regina (Rianne) van Boekel: Rianne.vanBoekel@radboudumc.nl

Sandra van den Heuvel Sandra.vandenHeuvel@Radboudumc.nl

Marieke Coenen: Marieke.Coenen@radboudumc.nl

Kris C.P. Vissers Kris.Vissers@radboudumc.nl

Corresponding author:

Regina L.M. van Boekel

Rianne.vanBoekel@radboudumc.nl

Radboud University Medical Center,

Department of Anesthesiology, Pain and Palliative Medicine

PO Box 9101, intern 549

6500 HB, Nijmegen, The Netherlands

Phone: 0031-243668120

Fax: 0031243613585

The number of text pages: 28

The actual number of figures and tables: 1 Figure, 1 Table, and 1 Supplementary file.

## Abstract

**Introduction** Postoperative pain remains a challenging medical condition impacting the quality of life of every patient. Although several predictive factors for postoperative pain have been identified, an adequate prediction of postoperative pain in patients at risk has not been achieved yet.

The primary objective of this study is to identify specific genetic risk factors for the development of acute and chronic postoperative pain to construct a prediction model facilitating a more personalized postoperative pain management for each individual. The secondary objectives are to build a databank enabling researchers to identify other risk factors for postoperative pain, for instance, demographic and clinical outcome indicators; provide insight into (genetic) factors that predict pharmacological pain relief; investigate the relationship between acute and chronic postoperative pain.

**Methods and analysis** In this prospective, observational study, patients who undergo elective surgery will be recruited to a sample size of approximately 10,000 patients. Postoperative acute and chronic pain outcomes will be collected through questionnaires at different time points after surgery in the follow-up of six months. Potential genetic, demographic, and clinical risk factors for prediction model construction will be collected through blood, questionnaires, and electronic health records, respectively.

Genetic factors associated with acute and/or chronic postoperative pain will be identified using a genome-wide association (GWA) analysis. Clinical risk factors as stated in the secondary

objectives will be assessed by multivariable regression. A clinical easy-to-use prediction model will be created for postoperative pain to allow clinical use for the stratification of patients.

**Ethics and dissemination** The Institutional Review Board of the Radboud university medical center approved the study (authorization number: 2012/117). The results of this study will be made available through peer-reviewed scientific journals and presentations at relevant conferences, which will finally contribute to personalized postoperative pain management.

**Trial registration number** NCT02383342

**Keywords**: Postoperative pain, Genome-wide association study (GWAS), Risk factor, Prediction model, Pharmacogenetics

## Strengths and limitations of this study

- This is a large prospective study to identify genetic and other risk factors for postoperative pain.
- We will build a databank with comprehensive interdisciplinary measurements that assess postoperative pain from multiple perspectives.
- Outcome measurements of pain by patient-reported outcomes, rather than evaluated by professionals.
- The primary goal of this study is to identify genetic variants as biomarkers of postoperative pain, but the collected blood samples enable future research to characterize the multi-omics biomarker signatures of postoperative pain.

## Introduction

Pain after surgery remains a challenging medical and societal problem [1]. Pain is one of the most common postsurgical side effects, with moderate to severe acute postoperative pain occurring in about 41% of the patients [2-4]. Severe postoperative pain is associated with an increased incidence of postoperative complications [5], including prolonged hospital stay, readmissions, and significant reduction of patient satisfaction and quality of life [6 7]. Besides, acute postoperative pain is associated with chronic pain development after surgery [8]. A recent position paper from the International Association for the Study of Pain stated that among the almost 40 million people undergoing surgery globally each year, one out of ten develops chronic postsurgical pain (CPSP), and one out of hundred suffers from severe CPSP, which will negatively affect patients' quality of life [9]. In addition, postoperative pain is a considerable burden on health care service costs, both directly due to patients' increased consumption of medical care and indirectly due to absenteeism, reduced productivity, and increased social welfare payments [10-15].

The management of both acute postoperative pain [2 31] and CPSP [2 32] has remained suboptimal. Despite major investments in clinical protocols and guidelines for structural pain management, infrastructure, and acute pain services (APS), no significant outcome improvements in the quality of postoperative pain management for individual patients have been achieved in the last fifteen years [10 11].

Given the high incidence of postoperative pain, identifying patients at risk for CPSP before the operation is important to apply more personalized pain prevention strategies. The most important demographic and clinical risk factors for postoperative pain are younger age, female sex, smoking, history of depressive symptoms, anxiety symptoms, sleep difficulties, higher body

mass index, presence of preoperative pain, and use of preoperative analgesics [16]. Based on these factors, models have been developed to predict severe acute postoperative pain [17 18] and CPSP [19 20]. A recent study has evaluated a presurgical risk score for CPSP in a prospective cohort, and it reliably identified about 70% of the patients undergoing surgeries at risk of CPSP [21 22].

As a multifactorial trait, the incidence variation of CPSP in the population can be explained partly by the demographic and clinical risk factors mentioned above, and partly due to the genetic and epigenetic differences among patients [23 24]. To improve the accuracy and power of prediction, efforts have been made to predict CPSP using genetic variants [19 22]. However, no unequivocal genetic predictors have been found yet. In addition, many exploratory studies investigated the possible role of candidate genes in postoperative pain development. In particular, associations have been found between CPSP and the *μ-opioid receptor (OPRM1)* and *catechol-O-methyl transferase (COMT)* genes [25 26]. Still, these results have not been confirmed by others. *OPRM1* is also associated with basal pain sensitivity differences [27], which could be caused by the altered opioid binding potential in the central nervous system [28]. More recently, hypothesis-free methods, such as genome-wide association studies, have been applied for CPSP to identify markers across the genome [29 30]. One of the studies showed that a genetic variant in the *protein-kinase C* gene is linked to neuropathic pain after complete joint replacement. This gene is involved in long-term potentiation, synaptic plasticity, chronic pain, and memory, indicating that this gene may be relevant for neuropathic pain initiation. The disadvantage of this study is that it was small in terms of patient numbers and only focused on one specific surgical procedure.

Besides genetic variants for altered pain sensitivity, also gene variants in drug metabolism can play a role. Understanding the reasons for ineffective treatment can facilitate the early identification of patients at risk and provide more effective and customized postoperative management. Some associated genes with pain treatment outcomes are also involved in pain development, such as *COMT* [33-35]. Genes involved in the action site of active drugs or the drugs' metabolism might play a role in the therapeutic response of this drug. A well-known example is the *cytochrome P450* (*CYP*) family investigated for several drugs (e.g., codeine and tramadol) [36]. However, this area has never been charted in a large population [37].

To date, adequate prediction of patients at risk for postoperative pain in clinical practice has not been achieved for several reasons. First, although many demographic, clinical, and lifestyle factors of postoperative pain have been reported [16], a lack of consensus on the best outcome indicators for postoperative pain management [38 39] hinders choosing the proper outcome variables for prediction model construction. Second, the potential genetic risk factors of postoperative pain prediction remain obscure. The role of genetic factors in postoperative pain have not been investigated sufficiently, making it challenging to select appropriate genetic risk factors to construct a prediction model. Third, when prediction models are updated, external validation (i.e., in a new population) is important before being implemented in a clinical setting [40-43], which is often difficult due to the lack of validation cohorts. For these reasons, we hypothesize that a global structural multicenter diagnostic program of postoperative pain in a surgical patient population will be valuable for better identifying patients at risk of CPSP and ultimately preventing postoperative pain using individualized pharmacological and non-pharmacological interventions.

## Objectives

The primary objective of the Pain Predict Genetics (PPG) study is to identify genetic risk factors for acute and chronic postoperative pain development and to construct a prediction model for personalized postoperative pain management.

The secondary objectives of the PPG study are to 1) identify other risk factors for the development of acute and chronic postoperative pain; 2) provide insights into complications and other clinical outcome indicators after surgery; 3) provide insights into the relationship between acute and chronic postoperative pain; 4) identify (genetic) factors that predict pharmacological pain relief.

The extensive data collection on (chronic) postoperative pain development of patients undergoing surgery offers many possibilities for additional research questions using conventional statistical methods and artificial intelligence, e.g., machine learning. The cohort could be used to 1) conduct epidemiological studies; 2) investigate other parameters (for example, types of surgery) that are involved in the development of chronic postoperative pain; 3) validate new prediction models for (chronic) postoperative pain; 4) identify factors for the postoperative outcome (for example, death, long-term hospitalization, complications); 5) collaborate with other groups to perform large-scale analysis to identify predictors for the development of (chronic) postoperative pain.

# Methods and analyses

## Study design

A prospective, observational study of 10,000 patients will undergo elective surgery. This study will run for at least ten years, during which period it must be possible to include the intended number of patients. Patient inclusion after CMO (Human Research Committee, in Dutch Commissie Mensgebonden Onderzoek) approval was started in March 2015, and patient inclusion was temporarily stopped in 2020 due to COVID restrictions. In the near future, this study will be continued as a multicenter study; hospitals have already been approached and indicated that they intend to participate.

## Patient and public involvement

During the design of the study the patients aided in the pilot phase of the questionnaires, during the recruitment the patients are informed concerning the project. In addition, patient reported outcomes will be used. Patients will be informed about the outcome of the study at several moments (depending on the obtained results).

## Participants

Patients who undergo electvie surgry and are eligible for this study will be approached before their planned surgery during the preoperative consultation. In this way, potential participants will have sufficient time to consider the study information. If any questions arise, it is possible to contact the researchers by telephone or ask the questions during the preoperative consultation. During the preoperative consultation (outpatient clinic or by telephone), the physician (assistant) will ask the patient if they are interested to participating in the study. If the patient is willing to participate, the informed consent form will be signed and dated. If patients have an

online preoperative consultation, this procedure will take place digitally, and patients receive the study forms (signed in advance) at home to return if they consent.

Patients are eligible for study inclusion if they 1) are older or equal to 16 years; 2) undergo elective surgery with an incision, including cardiothoracic surgery (e.g., cardiomyotomy), general surgery (e.g., breast resection), neurological surgery (e.g., nerve decompression), oral and maxillofacial surgery (e.g., removal of head and neck benign and malignant tumors), otorhinolaryngology (e.g., tympanoplasty), plastic surgery (e.g., breast reconstruction), trauma and orthopedic surgery (e.g., arthroplasty), urology (e.g., prostatectomy) and vascular surgery (e.g., treatment of varicose veins); 3) can read and understand the patient information; 4) will provide informed consent. Patients will be excluded if they 1) intend to undergo another surgery within six months; 2) do not have enough knowledge of the language in words and understanding to complete questionnaires.

## Measurements

### Questionnaires

After written informed consent, participants will be asked to complete questionnaires before and after their surgery. An overview of the study workflow and data collection time points can be found in **Figure 1** and **Table 1**. All patient data will be stored in an online digital database, Castor [44]. The reliability and validity of all questionnaires for measurement collection have been validated in the corresponding populations.

## Table 1: Overview of data collection

| | T0 | Day -1 | Surgery | Day 1 | Day 2 | Day 3 | Week 1 | Week 6 | Month 3 | Month 6 |
|---|---|---|---|---|---|---|---|---|---|---|
| Informed consent | x | | | | | | | | | |
| **Questionnaires** | | | | | | | | | | |
| Demografic data | | x | | | | | | | | |
| Incision size | | x | | x | | | | | | |
| Pain scores | | x | | x | x | x | x | x | x | x |
| Physical activities | | | | x | x | x | x | | | |
| Pain disability index | | x | | | | | x | x | x | x |
| APAIS | | x | | | | | | | | |
| PCS | | x | | | | | | | | |
| PSQ | | x | | | | | | | | |
| Chronic pain | | x | | | | | | | x | x |
| IDS depression | | x | | | | | | | | |
| Brief pain inventory | | | | | | | | | x | x |
| **Data electronic medical file** | | | | | | | | | | |
| Physical status by ASA | | | | | | | | | | x |
| Type of surgery | | | | | | | | | | x |
| Duration of surgery | | | | | | | | | | x |
| Type of anesthesia | | | | | | | | | | x |
| Complications | | | | | | | | | | x |
| Hospital stay | | | | | | | | | | x |
| Pain medication use | | | | | | | | | | x |
| Incision size | | | | | | | | | | x |
| Second surgery within 6 months | | | | | | | | | | x |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| General clinical outcome indicators | | | | | | | | | | x |
| **Body material*** | | | | | | | | | | |
| 1x10 ml blood for DNA | | | x | | | | | | | |

*In the event that it is not possible to collect a blood sample during surgery, the subject may be asked to provide a DNA sample via a saliva collection tube.

APAISI, Amsterdam Pre-operative Anxiety and Information Scale. PCS, Pain Catastrophizing Scale. PSQ, Pain Sensitivity questionnaire. IDS, Inventory of Depressive Symptomatology. ASA, American Society of Anesthesiologists classification.

The first digital questionnaire must be completed the day before the surgery (no longer than one week before). Before surgery, the following parameters will be collected (**Table 1,** Supplementary File 1): demographic characteristics (such as gender, age, BMI), expected incision size in mm, pain intensity, pain disability, preoperative anxiety and need for information, pain catastrophizing, pain sensitivity, preoperative chronic pain characteristics, and depressive symptoms.

After surgery, the following parameters will be collected: actual incision size in mm on day 1; pain intensity on day 1, 2, 3, week 1 and 6, and month 3 and 6; physical activities on day 1, 2, 3, week 1; pain disability on week 1 and 6, and month 3 and 6; postoperative chronic pain characteristics on month 3 and 6; characteristics of pain on month 3 and 6.

Pain intensity will be measured with an 11-point numerical rating scale (NRS) at rest and during a normal patient action at that time [18]. The endpoints represent the extremes of the pain experience: 0 means "no pain at all", and 10 means "worst possible pain".

Pain disability (disability associated with pain) will be measured by the widely used Pain Disability Index Dutch language version (PDI) [45 46]. The PDI is a 7-item questionnaire to investigate the magnitude of the self-reported disability in different situations such as work, leisure time, daily life activities, and sports. The questionnaire is constructed on an 11-point NRS in which 0 means "no disability" and 10 means "maximum disability".

Preoperative anxiety and need for information will be evaluated by the Amsterdam Preoperative Anxiety and Information Scale (APAIS) [47]. The APAIS consists of six questions and each score on a 5-point Likert scale from 1 (not at all) to 5 (extremely), with four questions to

assess the patient's preoperative anxiety score and two questions to assess the patient's need for information regarding the scheduled surgery and anesthesia [18].

Pain catastrophizing is generally described as an absurd negative orientation towards hurtful stimuli and is important in pain coping [48]. It will be measured by the Pain Catastrophizing Scale (PCS), a self-evaluating questionnaire consisting of 13 questions. People are asked to indicate the degree to which they have thoughts and feelings when experiencing pain using the 0 (not at all) to 4 (all the time) scale, and a total score will be yielded (range from 0 to 52).

The Pain Sensitivity Questionnaire (PSQ) will measure patients' preoperative pain sensitivity [49] [50]. The PSQ consists of 17 questions that describe daily life situations; respondents score their pain intensity for these situations on an NRS by scoring 0 (not painful) to 10 (severest pain imaginable).

Chronic pain characteristics will be measured preoperatively and postoperatively by five questions. The definition of chronic pain is in agreement with IASP terminology of chronic postsurgical pain, i.e., "*chronic pain that develops or increases in intensity after a surgical procedure persists beyond the healing process, i.e., at least 3 months after the surgery*" [9]. Patients will be asked to indicate whether they had a recent pain experience, the site of pain and whether it lasted more than three months [51 52].

The severity of overall depressive symptoms will be assessed by the Inventory of Depressive Symptomatology Self Report (IDS-SR) [53 54]. IDS-SR is a 30-item questionnaire, and each item has four statements scored on a four-point scale from 0 to 3. There are two items about either increasing or decreasing appetite and two items about increasing or decreasing weight. Only the

item with the higher score from both pairs will be chosen. The total score is based on 28 items and ranges from 0 to 84.

Physical activities (ability to perform normal activities) will be measured by questions assessing the degree of physical activities interfered by surgery, including bed activities (such as turning), breathing deeply of coughing, sleeping, and activities out of bed. Each item is scored on an 11-point NRS in which 0 means did not interfere and 10 means completely interfered. These questions are derived from the validated International Pain Outcomes questionnaire and are found responsive to asking patients about their ability to perform normal activities directly after surgery [55].

Characteristics of pain will be measured by the Brief Pain Inventory – Short Form (BPI-SF), which is a shortened version of the Brief Pain Inventory [56]. BPI-SF evaluates pain severity during the past 24 hours and current level, with 0 representing "no pain" and 10 "the worst pain imaginable". Seven items in BPI-SF assess interference with daily functioning (such as general activity, walking, and work) on an 11-point scale, where 0 represents "no interference" and 10 "complete interference".

## Collection of body material

One tube of blood will be collected for DNA isolation. The burden for the patient is minimalized as blood will be taken using the intravenous line in place for surgery. If it is impossible to collect blood presurgically or postsurgically, we will collect saliva for DNA isolation (Genefix DNA saliva collectors; GFX-02/50, Isohelix).

## Clinical information

The following clinical information will be collected from the electronic patient file six months after operation (**Table 1**): physical status by The American Society of Anesthesiologists classification (ASA-status); type of surgery; duration of surgery; type of anesthesia; postoperative complications within 30 days after surgery, one-time retrospectively, which is defined as any medical adverse outcome occurring between admission and 30 days after operation. Complications occurring in the operation room and complications directly related to anesthesia (e.g., nausea which resolves immediately after medication in the operation room) will not be included [5 57]. Furthermore, data on pain medication use, before surgery and after surgery; actual incision size in mm; second surgery within 6 months; general clinical outcome indicators, including surgical site infection at 30 days, stroke within 30 days of surgery, death within 30 days of surgery, admission to the intensive care unit within 14 days of surgery, readmission to hospital within 30 days of surgery, and length of hospital stay (with or without in-hospital mortality) will be collected [38].

## Sample size calculation

The power of the genetic study is based on the primary research question investigating which genetic factors are associated with postoperative pain. Power is calculated using the Genetic Power Calculator [58], and the estimated number of patients is based on a GWA approach. For chronic postoperative pain, we assume a case-control analysis for discrete traits (2df test), a risk allele frequency of 30%, a linkage disequilibrium (D') of 0.8, a prevalence of chronic postoperative pain of 15%, and the relative risk of chronic postoperative pain for persons who are heterozygous of 1.5 and for homozygous persons of 2.25. For a power of 80% with a p-value

cut off $5 \times 10^{-8}$ (genome-wide significance threshold), we need 750 patients with chronic postoperative pain and 4,250 people without chronic postoperative pain. For acute pain, the power is even higher. With the same population, we have more than 80% power to detect a relative risk of 1.2 and 1.44 for heterozygous and homozygous patients, respectively. This higher power is due to the higher prevalence of acute (moderate to severe) pain of 55%. Most importantly, results will be replicated in the additional study participants, as the total number of patients included in the study will be 10,000. In addition, we will use cohorts of our collaborators for replication purposes.

## Statistical analysis

The key objective is to identify genetic risk factors that can predict development of acute or chronic postoperative pain and validate previously reported SNPs. A GWA approach will be used as the main analysis. Phenotype data and DNA will be used to identify genetic factors. We will use 5,000 patients for the discovery of genetic variants. Samples will be genotyped with the Infinium Global Screening Array (Illumina). Pre-imputation quality control, principal component analyses, and imputation will follow the RICOPILI pipeline [59]. The 1000 Genomes reference panel will be used for imputation, followed by post-imputation quality control in PLINK [60]. Associations between SNPs and the presence of acute or chronic pain will be performed using cutting-edge methods when data collection is finished. Results will be to ensure validity. SNPs that can be validated will be included in the prediction model described below.

Secondary objectives include identifying other potential risk factors for acute and chronic postoperative pain. Therefore, a univariate association of each potential predictor will be calculated and tested in a multivariable regression model. We will use a least absolute shrinkage

and selection operator (Lasso) regression. Shrinkage is where data values are shrunk towards a central point, like the mean. Lasso is a regression analysis method that performs both variable selection and regularization to enhance the prediction accuracy and interpretability of the statistical model it produces. After identifying these risk factors, a prediction rule will be created for acute and chronic postoperative pain. Based on this prediction rule, a simple, clinically easy applicable tool will be developed to allow clinical use for the stratification of patients. The predictive performance will be studied in another cohort of patients to test whether the rule is generalizable across time and place. Because it appears from the literature that acute and chronic pain are correlated after surgery, additional correlation analysis will be performed to investigate this correlation in the data.

Similar approaches will be followed to identify the clinical and genetic factors that predict pharmacological pain relief. For some pain medicines, genes that impact pain relief are already known (e.g., *CYP2D6* and morphine). We will first investigate those genes to see if these variants indeed contribute to pharmacological pain relief differences.

## Ethics and dissemination:

The study will be conducted according to the principles of the Declaration of Helsinki version 2013 and in accordance with the Medical Research Involving Human Subjects Act and Good Clinical Practice. The study was approved by the local ethics committee for human research in Nijmegen (Medical Review Ethics Committee Region Arnhem-Nijmegen, authorization number: 2012/117). This study was registered on ClinicalTrials.gov (NCT02383342).

The privacy of the participants is guaranteed by storing encrypted data. Every participant will receive a pseudo-anonymous study number. No identifying data is recorded within the meaning of the law. The key is only accessible to the study team and monitors. Data and material will only be used in coded form within possible collaborations.

The results of this study will be made available through peer-reviewed scientific journals and presentations at relevant conferences. After a thorough evaluation, decisions will be made regarding including the identified risk factors and constructed prediction models into clinical guidelines, thus facilitating personalized postoperative pain management.

## Discussion

This cohort will be a large prospective study to identify risk factors for postoperative pain and to build and evaluate dedicated prediction models for postoperative pain in surgical patients. In addition, the comprehensive information collected in this study will also enable us to answer other research questions regarding postoperative pain, such as the relationships between acute and chronic postoperative pain development. Eventually, these results will be applied in the clinical settings to improve the quality of life for patients who develop postoperative pain.

The strengths of this study are that we will include all elective major operations rather than limiting to one specific operation as in previous studies [30], which allows us to investigate the shared genetic background of postoperative pain in different operations. Furthermore, as there are discrepancies in pain intensity scores understanding [61] and pain management decisions [61 62] between patients and caregivers, the patient's perspective should be respected and assessed for pain evaluation and management [63 64]. Therefore, pain assessment will be conducted by patients

themselves (patient-reported outcomes) rather than professionals in this study, leading to a more comprehensive outcome assessment and interpretation [65]. Moreover, the single-use of NRS might be inadequate for patients' pain experience evaluation and pain management decisions [64 66 67]. Thus, another strength of this cohort is that the experience of pain will be estimated by multidimensional measurements focusing on patients' overall functionality rather than merely a NRS pain score. Besides, the comprehensively collected information for postoperative pain in this cohort also empowers analysis that cannot be performed in large-scale registry data (e.g., UK Biobank) as such phenotype data is not available in those datasets. The data collected in this cohort will also enable additional research using conventional and cutting-edge statistical methods like artificial intelligence.

The possible limitations of this study are that we will only investigate DNA variants as biomarkers for pain prediction as our primary research goal. However, other epigenetic [67 68], transcriptomic [68], proteomics [69], and metabolic markers [70] are also potentially involved in (postoperative) pain development. For instance, recent studies indicate that methylation patterns might predict opioid treatment outcomes [67 68]. As the DNA sample of patients is accessible, we will be able to investigate additional related research questions, such as the association between epigenetic changes and postoperative pain in the future. In addition, when prediction tools are applied in clinical settings, the sensitivity and specificity of prediction tools are crucial to evaluate their adequacy and usefulness [71]. Although the measurement tools used in prediction models are well-validated and verified (see methods), our findings could still be subject to false positive or negative errors because all measurement tools have limitations. Furthermore, chronic pain assessment is more complex than acute pain [72], and GWAS findings

are sometimes incidental [73]. We will consider seeking other available cohorts for validation and applying other statistical methods to validate our findings in future studies, such as polygenic risk scores [74]. Another potential limitation is that loss of follow-up of patients might result in lower patient numbers than expected. Despite this potential concern, we still expect a sufficient sample size as additional centres will start patient incudion, and the measurements are mainly from patient-reported outcomes via digital follow-up.

Identifying the genetic background of postoperative pain development may give valuable insights into the mechanisms underlying the relationship between postoperative pain and complications after surgery. This may open the way to identify new targets for treatment and potentially simplify the risk profiling assay for future use, yielding a simpler, more accurate, and cost-efficient assay or product. The contribution of improved prevention and treatment of pain after surgery will benefit many patients undergoing surgery and society by decreasing health care service costs.

## Trial status

Patient recruitment is expected to continue until 2025. Recruitment has already started in Radboud university medical center, with more than 500 patients recruited as of October 2021. National and international collaborations will be greatly accepted after careful consideration.

## Author contributions

All authors were responsible for the study design. SL drafted the manuscript. All authors critically reviewed the manuscript.

## Acknowledgment

SL was supported by China Scholarship Council (CSC) Grant number 201908130179.

## Funding

Departmental funding covers the costs of this study [grant number: N/A].

## Competing interests statement

The authors have no relevant financial or non-financial interests to disclose.

# References

1. Gaskin DJ, Richard P. The economic costs of pain in the United States. *J Pain* 2012;13(8):715-24. doi: 10.1016/j.jpain.2012.03.009 [published Online First: 2012/05/23]
2. Dolin SJ, Cashman JN, Bland JM. Effectiveness of acute postoperative pain management: I. Evidence from published data. *Br J Anaesth* 2002;89(3):409-23.
3. Sommer M, de Rijke JM, van Kleef M, et al. The prevalence of postoperative pain in a sample of 1490 surgical inpatients. *Eur J Anaesthesiol* 2008;25(4):267-74. doi: 10.1017/S0265021507003031
4. Gan TJ. Poorly controlled postoperative pain: prevalence, consequences, and prevention. *J Pain Res* 2017;10:2287-98. doi: 10.2147/JPR.S144066 [published Online First: 2017/10/14]
5. van Boekel RLM, Warle MC, Nielen RGC, et al. Relationship Between Postoperative Pain and Overall 30-Day Complications in a Broad Surgical Population: An Observational Study. *Ann Surg* 2019;269(5):856-65. doi: 10.1097/SLA.0000000000002583 [published Online First: 2017/11/15]
6. Peters CL, Shirley B, Erickson J. The effect of a new multimodal perioperative anesthetic regimen on postoperative pain, side effects, rehabilitation, and length of hospital stay after total joint arthroplasty. *The Journal of arthroplasty* 2006;21(6 Suppl 2):132-8. doi: 10.1016/j.arth.2006.04.017
7. Regenbogen SE, Mullard AJ, Peters N, et al. Hospital Analgesia Practices and Patient-reported Pain After Colorectal Resection. *Ann Surg* 2016;264(6):1044-50. doi: 10.1097/SLA.0000000000001541
8. Katz J, Jackson M, Kavanagh BP, et al. Acute pain after thoracic surgery predicts long-term post-thoracotomy pain. *Clin J Pain* 1996;12(1):50-5. [published Online First: 1996/03/01]
9. Schug SA, Lavand'homme P, Barke A, et al. The IASP classification of chronic pain for ICD-11: chronic postsurgical or posttraumatic pain. *Pain* 2019;160(1):45-52. doi: 10.1097/j.pain.0000000000001413 [published Online First: 2018/12/27]
10. Apfelbaum JL, Chen C, Mehta SS, et al. Postoperative pain experience: results from a national survey suggest postoperative pain continues to be undermanaged. *Anesth Analg* 2003;97(2):534-40, table of contents.
11. Meissner W, Coluzzi F, Fletcher D, et al. Improving the management of post-operative acute pain: priorities for change. *Curr Med Res Opin* 2015;31(11):2131-43. doi: 10.1185/03007995.2015.1092122 [published Online First: 2015/09/12]
12. Zimberg SE. Reducing pain and costs with innovative postoperative pain management. *Managed care quarterly* 2003;11(1):34-6.
13. Morrison RS, Magaziner J, McLaughlin MA, et al. The impact of post-operative pain on outcomes following hip fracture. *Pain* 2003;103(3):303-11.
14. Zoucas E, Lydrup ML. Hospital costs associated with surgical morbidity after elective colorectal procedures: a retrospective observational cohort study in 530 patients. *Patient safety in surgery* 2014;8(1):2. doi: 10.1186/1754-9493-8-2
15. Encinosa WE, Hellinger FJ. The impact of medical errors on ninety-day costs and outcomes: an examination of surgical patients. *Health services research* 2008;43(6):2067-85. doi: 10.1111/j.1475-6773.2008.00882.x
16. Yang MMH, Hartley RL, Leung AA, et al. Preoperative predictors of poor acute postoperative pain control: a systematic review and meta-analysis. *BMJ Open* 2019;9(4):e025091. doi: 10.1136/bmjopen-2018-025091 [published Online First: 2019/04/04]
17. Kalkman CJ, Visser K, Moen J, et al. Preoperative prediction of severe postoperative pain. *Pain* 2003;105(3):415-23. doi: 10.1016/s0304-3959(03)00252-5 [published Online First: 2003/10/07]

18. Janssen KJ, Kalkman CJ, Grobbee DE, et al. The risk of severe postoperative pain: modification and validation of a clinical prediction rule. *Anesth Analg* 2008;107(4):1330-9. doi: 10.1213/ane.0b013e31818227da [published Online First: 2008/09/23]
19. Hoofwijk DMN, van Reij RRI, Rutten BPF, et al. Genetic polymorphisms and prediction of chronic post-surgical pain after hysterectomy-a subgroup analysis of a multicenter cohort study. *Acta Anaesthesiol Scand* 2019;63(8):1063-73. doi: 10.1111/aas.13413 [published Online First: 2019/06/18]
20. Althaus A, Hinrichs-Rocker A, Chapman R, et al. Development of a risk index for the prediction of chronic post-surgical pain. *Eur J Pain* 2012;16(6):901-10. doi: 10.1002/j.1532-2149.2011.00090.x
21. Montes A, Roca G, Cantillo J, et al. Presurgical risk model for chronic postsurgical pain based on 6 clinical predictors: a prospective external validation. *Pain* 2020;161(11):2611-18. doi: 10.1097/j.pain.0000000000001945 [published Online First: 2020/06/17]
22. Montes A, Roca G, Sabate S, et al. Genetic and Clinical Factors Associated with Chronic Postsurgical Pain after Hernia Repair, Hysterectomy, and Thoracotomy: A Two-year Multicenter Cohort Study. *Anesthesiology* 2015;122(5):1123-41. doi: 10.1097/ALN.0000000000000611 [published Online First: 2015/05/20]
23. Mauck M, Van de Ven T, Shaw AD. Epigenetics of chronic pain after thoracic surgery. *Curr Opin Anaesthesiol* 2014;27(1):1-5. doi: 10.1097/ACO.0000000000000030 [published Online First: 2013/12/05]
24. van Reij RRI, Joosten EAJ, van den Hoogen NJ. Dopaminergic neurotransmission and genetic variation in chronification of post-surgical pain. *Br J Anaesth* 2019;123(6):853-64. doi: 10.1016/j.bja.2019.07.028 [published Online First: 2019/09/29]
25. De Gregori M, Diatchenko L, Belfer I, et al. OPRM1 receptor as new biomarker to help the prediction of post mastectomy pain and recurrence in breast cancer. *Minerva Anestesiol* 2015;81(8):894-900. [published Online First: 2014/10/11]
26. Hoofwijk DM, van Reij RR, Rutten BP, et al. Genetic polymorphisms and their association with the prevalence and severity of chronic postsurgical pain: a systematic review. *Br J Anaesth* 2016;117(6):708-19. doi: 10.1093/bja/aew378 [published Online First: 2016/12/14]
27. Kim H, Clark D, Dionne RA. Genetic contributions to clinical pain and analgesia: avoiding pitfalls in genetic research. *J Pain* 2009;10(7):663-93. doi: 10.1016/j.jpain.2009.04.001 [published Online First: 2009/06/30]
28. Mueller C, Klega A, Buchholz HG, et al. Basal opioid receptor binding is associated with differences in sensory perception in healthy human subjects: a [18F]diprenorphine PET study. *Neuroimage* 2010;49(1):731-7. doi: 10.1016/j.neuroimage.2009.08.033 [published Online First: 2009/08/26]
29. van Reij RRI, Hoofwijk DMN, Rutten BPF, et al. The association between genome-wide polymorphisms and chronic postoperative pain: a prospective observational study. *Anaesthesia* 2020;75 Suppl 1:e111-e20. doi: 10.1111/anae.14832 [published Online First: 2020/01/07]
30. Warner SC, van Meurs JB, Schiphof D, et al. Genome-wide association scan of neuropathic pain symptoms post total joint replacement highlights a variant in the protein-kinase C gene. *Eur J Hum Genet* 2017;25(4):446-51. doi: 10.1038/ejhg.2016.196 [published Online First: 2017/01/05]
31. Sinatra R. Causes and consequences of inadequate management of acute pain. *Pain Med* 2010;11(12):1859-71. doi: 10.1111/j.1526-4637.2010.00983.x [published Online First: 2010/11/03]
32. Glare P, Aubrey KR, Myles PS. Transition from acute to chronic pain after surgery. *Lancet* 2019;393(10180):1537-46. doi: 10.1016/S0140-6736(19)30352-6 [published Online First: 2019/04/16]

33. Zubieta JK, Heitzeg MM, Smith YR, et al. COMT val158met genotype affects mu-opioid neurotransmitter responses to a pain stressor. *Science* 2003;299(5610):1240-3. doi: 10.1126/science.1078546 [published Online First: 2003/02/22]
34. Rakvåg TT, Klepstad P, Baar C, et al. The Val158Met polymorphism of the human catechol-O-methyltransferase (COMT) gene may influence morphine requirements in cancer pain patients. *Pain* 2005;116(1-2):73-8. doi: 10.1016/j.pain.2005.03.032 [published Online First: 2005/06/02]
35. Reyes-Gibby CC, Shete S, Rakvåg T, et al. Exploring joint effects of genes and the clinical efficacy of morphine for cancer pain: OPRM1 and COMT gene. *Pain* 2007;130(1-2):25-30. doi: 10.1016/j.pain.2006.10.023 [published Online First: 2006/12/13]
36. Owusu Obeng A, Hamadeh I, Smith M. Review of Opioid Pharmacogenetics and Considerations for Pain Management. *Pharmacotherapy* 2017;37(9):1105-21. doi: 10.1002/phar.1986 [published Online First: 2017/07/13]
37. De Gregori M, Diatchenko L, Ingelmo PM, et al. Human Genetic Variability Contributes to Postoperative Morphine Consumption. *J Pain* 2016;17(5):628-36. doi: 10.1016/j.jpain.2016.02.003 [published Online First: 2016/02/24]
38. Haller G, Bampoe S, Cook T, et al. Systematic review and consensus definitions for the Standardised Endpoints in Perioperative Medicine initiative: clinical indicators. *Br J Anaesth* 2019;123(2):228-37. doi: 10.1016/j.bja.2019.04.041 [published Online First: 2019/05/28]
39. Pogatzki-Zahn E, Schnabel K, Kaiser U. Patient-reported outcome measures for acute and chronic pain: current knowledge and future directions. *Curr Opin Anaesthesiol* 2019;32(5):616-22. doi: 10.1097/ACO.0000000000000780 [published Online First: 2019/08/16]
40. Justice AC, Covinsky KE, Berlin JA. Assessing the generalizability of prognostic information. *Ann Intern Med* 1999;130(6):515-24. doi: 10.7326/0003-4819-130-6-199903160-00016 [published Online First: 1999/03/13]
41. Reilly BM, Evans AT. Translating clinical research into clinical practice: impact of using prediction rules to make decisions. *Ann Intern Med* 2006;144(3):201-9. doi: 10.7326/0003-4819-144-3-200602070-00009 [published Online First: 2006/02/08]
42. Altman DG, Royston P. What do we mean by validating a prognostic model? *Stat Med* 2000;19(4):453-73. doi: 10.1002/(sici)1097-0258(20000229)19:4<453::aid-sim350>3.0.co;2-5 [published Online First: 2000/03/01]
43. Harrell FE, Jr., Lee KL, Mark DB. Multivariable prognostic models: issues in developing models, evaluating assumptions and adequacy, and measuring and reducing errors. *Stat Med* 1996;15(4):361-87. doi: 10.1002/(SICI)1097-0258(19960229)15:4<361::AID-SIM168>3.0.CO;2-4 [published Online First: 1996/02/28]
44. Castor EDC. Castor Electronic Data Capture 2019 [27 Aug. 2019]. Available from: https://castoredc.com accessed August 28, 2019.
45. Soer R, Köke AJ, Vroomen PC, et al. Extensive validation of the pain disability index in 3 groups of patients with musculoskeletal pain. *Spine (Phila Pa 1976)* 2013;38(9):E562-8. doi: 10.1097/BRS.0b013e31828af21f [published Online First: 2013/02/08]
46. Tait RC, Pollard CA, Margolis RB, et al. The Pain Disability Index: psychometric and validity data. *Arch Phys Med Rehabil* 1987;68(7):438-41. [published Online First: 1987/07/01]
47. Moerman N, van Dam FS, Muller MJ, et al. The Amsterdam Preoperative Anxiety and Information Scale (APAIS). *Anesth Analg* 1996;82(3):445-51. doi: 10.1097/00000539-199603000-00002 [published Online First: 1996/03/01]
48. Sullivan MJL, Bishop SR, Pivik J. The Pain Catastrophizing Scale: Development and validation. *Psychological Assessment* 1995;7(4):524-32. doi: 10.1037/1040-3590.7.4.524

49. Ruscheweyh R, Marziniak M, Stumpenhorst F, et al. Pain sensitivity can be assessed by self-rating: Development and validation of the Pain Sensitivity Questionnaire. *Pain* 2009;146(1-2):65-74. doi: 10.1016/j.pain.2009.06.020 [published Online First: 2009/08/12]
50. Van Boekel RLM, Timmerman H, Bronkhorst EM, et al. Translation, Cross-Cultural Adaptation, and Validation of the Pain Sensitivity Questionnaire in Dutch Healthy Volunteers. *Pain Res Manag* 2020;2020:1050935. doi: 10.1155/2020/1050935 [published Online First: 2020/08/11]
51. Macrae WA. Chronic post-surgical pain: 10 years on. *Br J Anaesth* 2008;101(1):77-86. doi: 10.1093/bja/aen099 [published Online First: 2008/04/25]
52. Werner MU, Kongsgaard UE. I. Defining persistent post-surgical pain: is an update required? *Br J Anaesth* 2014;113(1):1-4. doi: 10.1093/bja/aeu012 [published Online First: 2014/02/21]
53. Rush AJ, Giles DE, Schlesser MA, et al. The Inventory for Depressive Symptomatology (IDS): preliminary findings. *Psychiatry Res* 1986;18(1):65-87. doi: 10.1016/0165-1781(86)90060-0 [published Online First: 1986/05/01]
54. Rush AJ, Gullion CM, Basco MR, et al. The Inventory of Depressive Symptomatology (IDS): psychometric properties. *Psychol Med* 1996;26(3):477-86. doi: 10.1017/s0033291700035558 [published Online First: 1996/05/01]
55. Rothaug J, Zaslansky R, Schwenkglenks M, et al. Patients' perception of postoperative pain management: validation of the International Pain Outcomes (IPO) questionnaire. *J Pain* 2013;14(11):1361-70. doi: 10.1016/j.jpain.2013.05.016 [published Online First: 2013/09/12]
56. Cleeland CS, Ryan KM. Pain assessment: global use of the Brief Pain Inventory. *Ann Acad Med Singap* 1994;23(2):129-38. [published Online First: 1994/03/01]
57. Dindo D, Clavien PA. What is a surgical complication? *World J Surg* 2008;32(6):939-41. doi: 10.1007/s00268-008-9584-y [published Online First: 2008/04/17]
58. Purcell S, Cherny SS, Sham PC. Genetic Power Calculator: design of linkage and association genetic mapping studies of complex traits. *Bioinformatics* 2003;19(1):149-50. doi: 10.1093/bioinformatics/19.1.149 [published Online First: 2002/12/25]
59. Lam M, Awasthi S, Watson HJ, et al. RICOPILI: Rapid Imputation for COnsortias PIpeLIne. *Bioinformatics* 2020;36(3):930-33. doi: 10.1093/bioinformatics/btz633 [published Online First: 2019/08/09]
60. Purcell S, Neale B, Todd-Brown K, et al. PLINK: a tool set for whole-genome association and population-based linkage analyses. *Am J Hum Genet* 2007;81(3):559-75. doi: 10.1086/519795 [published Online First: 2007/08/19]
61. van Dijk JF, van Wijck AJ, Kappen TH, et al. Postoperative pain assessment based on numeric ratings is not the same for patients and professionals: a cross-sectional study. *Int J Nurs Stud* 2012;49(1):65-71. doi: 10.1016/j.ijnurstu.2011.07.009 [published Online First: 2011/08/16]
62. Harting B, Johnson T, Abrams R, et al. An exploratory analysis of the correlation of pain scores, patient satisfaction with relief from pain, and a new measure of pain control on the total dose of opioids in pain care. *Qual Manag Health Care* 2013;22(4):322-6. doi: 10.1097/qmh.0000000000000009 [published Online First: 2013/10/04]
63. Raja SN, Carr DB, Cohen M, et al. The revised International Association for the Study of Pain definition of pain: concepts, challenges, and compromises. *Pain* 2020;161(9):1976-82. doi: 10.1097/j.pain.0000000000001939 [published Online First: 2020/07/23]
64. van Boekel RLM, Vissers KCP, van der Sande R, et al. Moving beyond pain scores: Multidimensional pain assessment is essential for adequate pain management after surgery. *PLoS One* 2017;12(5):e0177345. doi: 10.1371/journal.pone.0177345 [published Online First: 2017/05/11]
65. Weldring T, Smith SM. Patient-Reported Outcomes (PROs) and Patient-Reported Outcome Measures (PROMs). *Health Serv Insights* 2013;6:61-8. doi: 10.4137/hsi.S11093 [published Online First: 2013/01/01]

66. Sloman R, Wruble AW, Rosen G, et al. Determination of clinically meaningful levels of pain reduction in patients experiencing acute postoperative pain. *Pain Manag Nurs* 2006;7(4):153-8. doi: 10.1016/j.pmn.2006.09.001 [published Online First: 2006/12/06]
67. Clark CW, Yang JC, Tsui SL, et al. Unidimensional pain rating scales: a multidimensional affect and pain survey (MAPS) analysis of what they really measure. *Pain* 2002;98(3):241-47. doi: 10.1016/s0304-3959(01)00474-2 [published Online First: 2002/07/20]
68. Dorsey SG, Renn CL, Griffioen M, et al. Whole blood transcriptomic profiles can differentiate vulnerability to chronic low back pain. *PLoS One* 2019;14(5):e0216539. doi: 10.1371/journal.pone.0216539 [published Online First: 2019/05/17]
69. Van Der Heijden H, Fatou B, Sibai D, et al. Proteomics based markers of clinical pain severity in juvenile idiopathic arthritis. *Pediatr Rheumatol Online J* 2022;20(1):3. doi: 10.1186/s12969-022-00662-1 [published Online First: 2022/01/17]
70. Jha MK, Song GJ, Lee MG, et al. Metabolic Connection of Inflammatory Pain: Pivotal Role of a Pyruvate Dehydrogenase Kinase-Pyruvate Dehydrogenase-Lactic Acid Axis. *J Neurosci* 2015;35(42):14353-69. doi: 10.1523/jneurosci.1910-15.2015 [published Online First: 2015/10/23]
71. Trevethan R. Sensitivity, Specificity, and Predictive Values: Foundations, Pliabilities, and Pitfalls in Research and Practice. *Front Public Health* 2017;5:307. doi: 10.3389/fpubh.2017.00307 [published Online First: 2017/12/07]
72. Fillingim RB, Loeser JD, Baron R, et al. Assessment of Chronic Pain: Domains, Methods, and Mechanisms. *J Pain* 2016;17(9 Suppl):T10-20. doi: 10.1016/j.jpain.2015.08.010 [published Online First: 2016/09/03]
73. Ioannidis JP. Non-replication and inconsistency in the genome-wide association setting. *Hum Hered* 2007;64(4):203-13. doi: 10.1159/000103512 [published Online First: 2007/06/07]
74. van Reij RRI, Voncken JW, Joosten EAJ, et al. Polygenic risk scores indicates genetic overlap between peripheral pain syndromes and chronic postsurgical pain. *Neurogenetics* 2020;21(3):205-15. doi: 10.1007/s10048-020-00614-5 [published Online First: 2020/05/08]

Figure 1: Pain Predict Genetics study design overview.

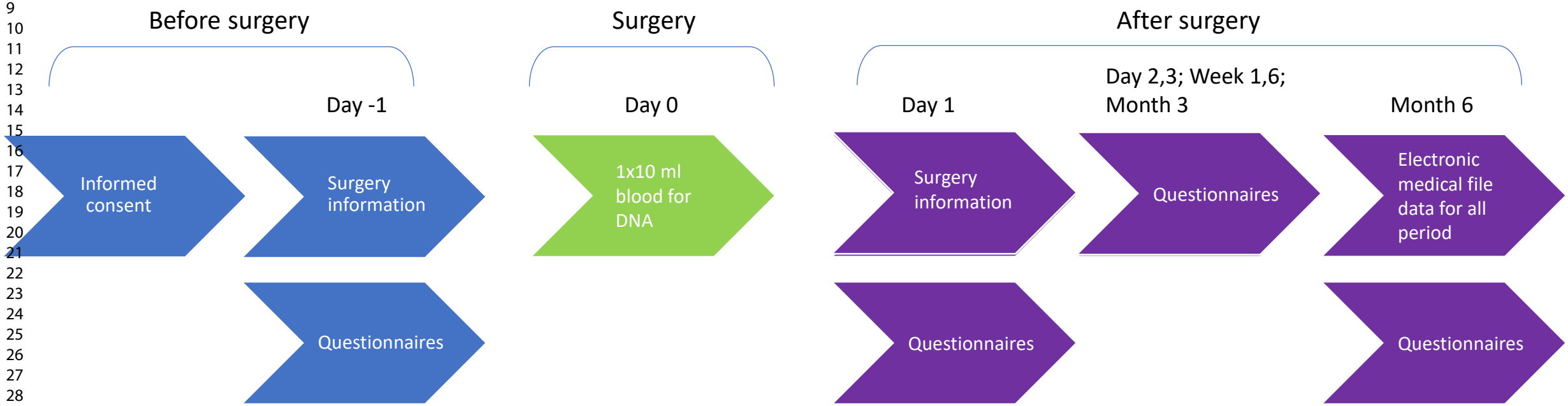


Table of Content

Appendix a: General data

*General data*

- What is your year of birth?
- What is your gender? male/female
- What is your length ? _____ cm
- What is your weight ? _____ kg

- What country were you born in?_____
- What country(ies) were your parents born in?_____
- What country(ies) were your grandparents born in?_____
- What human race are you? (black, white, Asian, etc.)_____

*Data of the surgery:*

*- Would you please describe your surgery*:

| |
|---|
| |

- How much pain do you expect after surgery (0= no pain, 10=worst pain imaginable)
- Will you stay one or more nights in the hospital after surgery? Yes / No

Appendix b: Pain before and after surgery

Pain before and after surgery

Circle how much pain you have, expressed as a number. The pain score means a score between 0 and 10, where 0 means no pain and 10 means the worst pain imaginable. For your pain, consider a figure between 0 and 10. You also tick whether you think the pain is acceptable or not.

| | |
|---|---|
| Pain while being at rest at this moment (0-10) | No pain 0-1-2-3-4-5-6-7-8-9-10 worst pain imaginable |
| Pain score at this moment if you perform a normal effort (0-10) | No pain 0-1-2-3-4-5-6-7-8-9-10 worst pain imaginable |
| Do you think pain is acceptable to you at this moment? | Pain acceptable\|__\| pain not acceptable \|__\| |
| Only pre-operatively: How much pain do you expect after surgery? | No pain 0-1-2-3-4-5-6-7-8-9-10 worst pain imaginable |

Appendix c: Physical activities

Physical activities

Circle the one number below that best describes how much, since your surgery, pain interfered with or prevented you from doing physical activities, expressed by figure. The score means a figure between 0 and 10, where 0 means no interference and 10 means complete interference.

1. How much has pain interfered with or prevented you from doing activities in bed such as turning, sitting up, changing position (0= did not interfere, 10= completely interfered)

0-1-2-3-4-5-6-7-8-9-10

2. How much has pain interfered with or prevented you from breathing deeply of coughing (0= did not interfere, 10= completely interfered)

0-1-2-3-4-5-6-7-8-9-10

3. How much has pain interfered with or prevented you from sleeping (0= did not interfere, 10= completely interfered)

0-1-2-3-4-5-6-7-8-9-10

4. Have you been out of bed since your surgery? Yes/no

0-1-2-3-4-5-6-7-8-9-10

5. If yes, how much did pain interfere or prevent you from doing activities out of bed such as walking, sitting in a chair, standing at the sink (0= did not interfere, 10= completely interfered)

0-1-2-3-4-5-6-7-8-9-10

Appendix d: Pain disability index

Pain disability index

*We would like to know how much pain is preventing you from doing what you would normally do or from doing it as well as you normally would. Respond to each category indicating the overall impact of pain in your life, not just when pain is at its worst.*

*For each of the 7 categories of life activity listed, please circle the number on the scale that describes the level of disability you typically experience. A score of 0 means no disability at all, and a score of 10 signifies that all of the activities in which you would normally be involved have been totally disrupted or prevented by your pain.*

*In case of no pain, please circle "0".*

| | |
|---|---|
| **1. Family/Home Responsibilities**<br><br>This category refers to activities of the home or family. It includes chores or duties performed around the house (e.g. yard work) and errands or favors for other family members (e.g. driving the children to school). | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **2. Recreation**<br><br>This disability includes hobbies, sports, and other similar leisure time activities. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **3. Social activity**<br><br>This category refers to activities, which involve participation with friends and acquaintances other than family members. It includes parties, theater, concerts, dining out, and other social functions. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **4. Occupation**<br><br>This category refers to activities that are part of or directly related to one's job. This includes non-paying jobs as well, such as that of a housewife or volunteer. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **5. Sexual behavior**<br><br>This category refers to the frequency and quality of one's sex life. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **6. Self care** | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |

| | |
|---|---|
| This category includes activities, which involve personal maintenance and independent daily living (e.g. taking a shower, driving, getting dressed, etc.) | |
| **7. Life-support activities**<br><br>This category refers to basic life supporting behaviors such as eating, sleeping and breathing. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |

Appendix e: Anxiety and need for information

Anxiety and need for information

*Please circle the number on the scale that describes your experience:*

| **The Amsterdam Preoperative Anxiety and Information Scale (APAIS):** | Not at all | | | | Extremely |
|---|---|---|---|---|---|
| I am worried about the anesthetic | 1 | 2 | 3 | 4 | 5 |
| The anesthetic is on my mind continually | 1 | 2 | 3 | 4 | 5 |
| I am worried about the procedure | 1 | 2 | 3 | 4 | 5 |
| The procedure is on my mind continually | 1 | 2 | 3 | 4 | 5 |
| I would like to know as much as possible about the anesthetic | 1 | 2 | 3 | 4 | 5 |
| I would like to know as much as possible about the procedure | 1 | 2 | 3 | 4 | 5 |

Appendix f: Pain Catastrophizing Scale (PCS)

Pain Catastrophizing Scale (PCS)

We are interested in the types of thoughts and feelings that you have when you are in pain. Listed below are thirteen statements describing different thoughts and feelings that may be associated with pain. Using the following scale, please indicate the degree to which you have these thoughts and feelings when you are experiencing pain.

0=not at all 1=to a slight degree 2=to a moderate degree 3=to a great degree 4=all the time

When I'm in pain …….

| | | | | | | |
|---|---|---|---|---|---|---|
| 1. | I worry all the time about whether the pain will end | 0 | 1 | 2 | 3 | 4 |
| 2. | I feel I can't go on | 0 | 1 | 2 | 3 | 4 |
| 3. | It's terrible and I think that it's never going to get any better | 0 | 1 | 2 | 3 | 4 |
| 4. | It's awful and I feel that it overwhelms me | 0 | 1 | 2 | 3 | 4 |
| 5. | I feel that I can't stand it any more | 0 | 1 | 2 | 3 | 4 |
| 6. | I become afraid that the pain will get worse | 0 | 1 | 2 | 3 | 4 |
| 7. | I keep thinking of other painful events | 0 | 1 | 2 | 3 | 4 |
| 8. | I anxiously want the pain to go away | 0 | 1 | 2 | 3 | 4 |
| 9. | I can't seem to keep it out of my mind | 0 | 1 | 2 | 3 | 4 |
| 10. | I keep thinking about how much it hurts | 0 | 1 | 2 | 3 | 4 |
| 11. | I keep thinking about how badly I want the pain to stop | 0 | 1 | 2 | 3 | 4 |
| 12. | There's nothing I can do to reduce the intensity of the pain | 0 | 1 | 2 | 3 | 4 |
| 13. | I wonder whether something serious may happen | 0 | 1 | 2 | 3 | 4 |

Appendix g: Pain Sensitivity Questionnaire

Pain Sensitivity Questionnaire

*This questionnaire contains a series of questions in which you should imagine yourself in certain*
*situations. You should then decide if these situations would be painful for you and if yes, how painful*
*they would be.*

***Let 0 stand for no pain; 1 is an only just noticeable pain arid l0 the most severe pain that you can imagine or consider possible.***

*Please mark the scale with a cross on the number that is most true for you. Keep in mind that there are no "right" or "wrong" answers; only your personal assessment of the situation counts. Please try as much as possible not to allow your fear or aversion of the imagined situations affect your assessment of painfulness.*

1. Imagine you bump your shin badly on a hard edge, for example, on the edge of a glass coffee
   table. How painful would that be for you?
   **0 = not at all painful, 10= most severe pain imaginable**

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

2. Imagine you burn your tongue on a very hot drink.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

3. Imagine your muscles are slightly sore as the result of physical activity.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

4. Imagine you trap your finger in a drawer.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

5. Imagine you take a shower with lukewarm water.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

6. Imagine you have mild sunburn on your shoulders.

0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

7. Imagine you grazed your knee falling off your bicycle.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

8. Imagine you accidentally bite your tongue or cheek badly while eating.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

9. Imagine walking across a cool tiled floor with bare feet.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

10. Imagine you have a minor cut on your finger and inadvertently get lemon juice in the wound.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

11. Imagine you prick your fingertip on the thorn of a rose.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

12. Imagine you stick your bare hands in the snow for a couple of minutes or bring your hands
    in contact with snow for some time, for example, while making snowballs.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

13. Imagine you shake hands with someone who has a normal grip.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

14. Imagine you shake hands with someone who has a very strong grip.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

15. Imagine you pick up a hot pot by inadvertently grabbing its equally hot handles.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

16. Imagine you are wearing sandals and someone with heavy boots steps on your foot.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

17. Imagine you bump your elbow on the edge of a table ("funny bone").

0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

Appendix h: Chronic pain

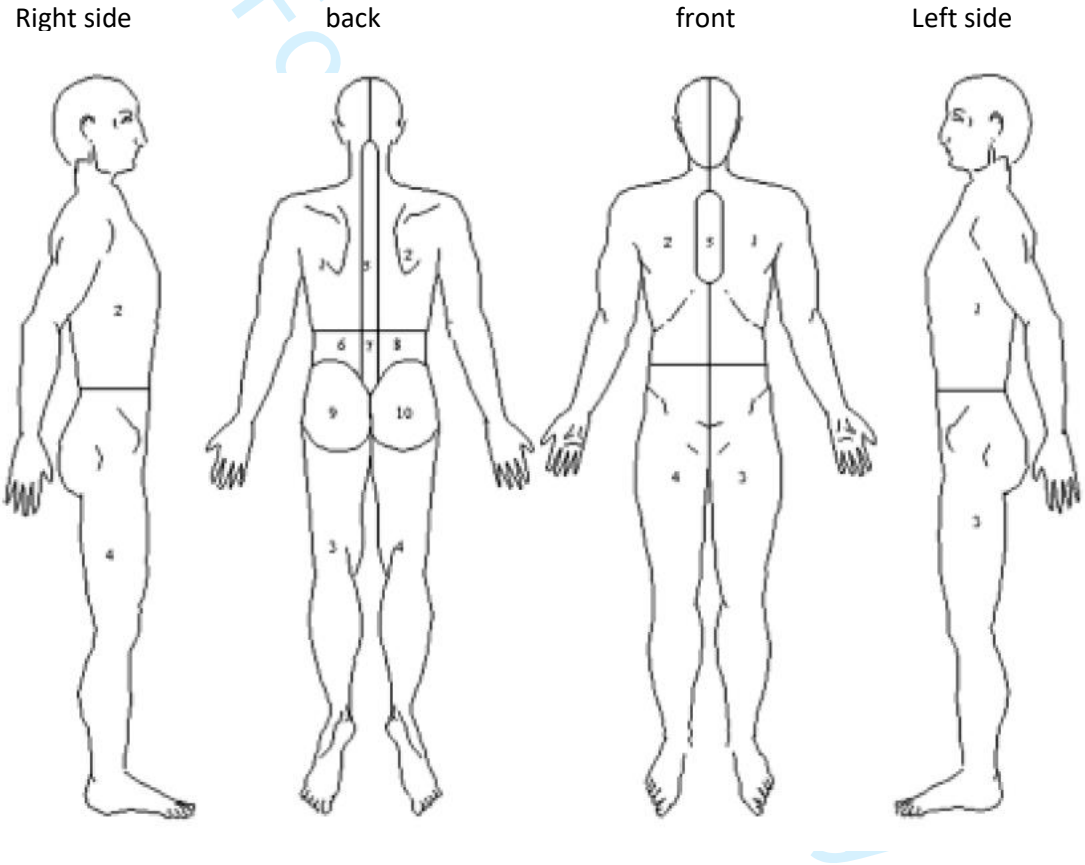
Chronic pain

Did you experience any pain in the last month that lasted for a day or more?

□ Yes, next question

□ No

Can you indicate in the drawings below where you suffer (have suffered) from pain?




Is this the same spot as the spot you are operated on? Yes/no

Does the pain differ from the pain before surgery? Yes/no

How long have you been affected by the above-mentioned pain?

□ Less than three months

□ More than three months

Appendix i: Inventory of depressive symptomatology (self-report) (IDS-SR)

**INVENTORY OF DEPRESSIVE SYMPTOMATOLOGY (SELF-REPORT)**
**(IDS-SR)**

NAME: _____ TODAY'S DATE _____

Please circle the one response to each item that best describes you for the past seven days.

1. Falling Asleep:

   0 I never take longer than 30 minutes to fall asleep.
   1 I take at least 30 minutes to fall asleep, less than half the time.
   2 I take at least 30 minutes to fall asleep, more than half the time.
   3 I take more than 60 minutes to fall asleep, more than half the time.

2. Sleep During the Night:

   0 I do not wake up at night.
   1 I have a restless, light sleep with a few brief awakenings each night.
   2 I wake up at least once a night, but I go back to sleep easily.
   3 I awaken more than once a night and stay awake for 20 minutes or more, more than half the time.

3. Waking Up Too Early:

   0 Most of the time, I awaken no more than 30 minutes before I need to get up.
   1 More than half the time, I awaken more than 30 minutes before I need to get up.
   2 I almost always awaken at least one hour or so before I need to, but I go back to sleep eventually.
   3 I awaken at least one hour before I need to, and can't go back to sleep.

4. Sleeping Too Much:

   0 I sleep no longer than 7-8 hours/night, without napping during the day.
   1 I sleep no longer than 10 hours in a 24-hour period including naps.
   2 I sleep no longer than 12 hours in a 24-hour period including naps.
   3 I sleep longer than 12 hours in a 24-hour period including naps.

5. Feeling Sad:

   0 I do not feel sad
   1 I feel sad less than half the time.
   2 I feel sad more than half the time.
   3 I feel sad nearly all of the time.

6. Feeling Irritable:

   0 I do not feel irritable.
   1 I feel irritable less than half the time.
   2 I feel irritable more than half the time.
   3 I feel extremely irritable nearly all of the time.

7. Feeling Anxious or Tense:

   0 I do not feel anxious or tense.
   1 I feel anxious (tense) less than half the time.
   2 I feel anxious (tense) more than half the time.
   3 I feel extremely anxious (tense) nearly all of the time.

8. Response of Your Mood to Good or Desired Events:

   0 My mood brightens to a normal level which lasts for several hours when good events occur.
   1 My mood brightens but I do not feel like my normal self when good events occur.
   2 My mood brightens only somewhat to a rather limited range of desired events.
   3 My mood does not brighten at all, even when very good or desired events occur in my life.

9. Mood in Relation to the Time of Day:

   0 There is no regular relationship between my mood and the time of day.
   1 My mood often relates to the time of day because of environmental events (e.g., being alone, working).
   2 In general, my mood is more related to the time of day than to environmental events.
   3 My mood is clearly and predictably better or worse at a particular time each day.

   9A. Is your mood typically worse in the morning, afternoon or night? (circle one)

   9B. Is your mood variation attributed to the environment? (yes or no) (circle one)

10. The Quality of Your Mood:

   0 The mood (internal feelings) that I experience is very much a normal mood.
   1 My mood is sad, but this sadness is pretty much like the sad mood I would feel if someone close to me died or left.
   2 My mood is sad, but this sadness has a rather different quality to it than the sadness I would feel if someone close to me died or left.
   3 My mood is sad, but this sadness is different from the type of sadness associated with grief or loss.

**Please complete either 11 or 12 (not both)**

11. Decreased Appetite:

0 There is no change in my usual appetite.
1 I eat somewhat less often or lesser amounts of food than usual.
2 I eat much less than usual and only with personal effort.
3 I rarely eat within a 24-hour period, and only with extreme personal effort or when others persuade me to eat.

12. Increased Appetite:

0 There is no change from my usual appetite.
1 I feel a need to eat more frequently than usual.
2 I regularly eat more often and/or greater amounts of food than usual.
3 I feel driven to overeat both at mealtime and between meals.

**Please complete either 13 or 14 (not both)**

13. Decreased Weight (Within the Last Two Weeks):

0 I have not had a change in my weight.
1 I feel as if I've had a slight weight loss.
2 I have lost 2 pounds or more.
3 I have lost 5 pounds or more.

14. Increased Weight (Within the Last Two Weeks):

0 I have not had a change in my weight.
1 I feel as if I've had a slight weight gain.
2 I have gained 2 pounds or more.
3 I have gained 5 pounds or more.

15. Concentration/Decision Making:

0 There is no change in my usual capacity to concentrate or make decisions.
1 I occasionally feel indecisive or find that my attention wanders.
2 Most of the time, I struggle to focus my attention or to make decisions.
3 I cannot concentrate well enough to read or cannot make even minor decisions.

16. View of Myself:

0 I see myself as equally worthwhile and deserving as other people.
1 I am more self-blaming than usual.
2 I largely believe that I cause problems for others.
3 I think almost constantly about major and minor defects in myself.

17. View of My Future:

0 I have an optimistic view of my future.
1 I am occasionally pessimistic about my future, but for the most part I believe things will get better.
2 I'm pretty certain that my immediate future (1-2 months) does not hold much promise of good things for me.
3 I see no hope of anything good happening to me anytime in the future.

18. Thoughts of Death or Suicide:

0 I do not think of suicide or death.
1 I feel that life is empty or wonder if it's worth living.
2 I think of suicide or death several times a week for several minutes.
3 I think of suicide or death several times a day in some detail, or I have made specific plans for suicide or have actually tried to take my life.

19. General Interest:

0 There is no change from usual in how interested I am in other people or activities.
1 I notice that I am less interested in people or activities.
2 I find I have interest in only one or two of my formerly pursued activities.
3 I have virtually no interest in formerly pursued activities.

20. Energy Level:

0 There is no change in my usual level of energy.
1 I get tired more easily than usual.
2 I have to make a big effort to start or finish my usual daily activities (for example, shopping, homework, cooking or going to work).
3 I really cannot carry out most of my usual daily activities because I just don't have the energy.

21. Capacity for Pleasure or Enjoyment (excluding sex):

0 I enjoy pleasurable activities just as much as usual.
1 I do not feel my usual sense of enjoyment from pleasurable activities.
2 I rarely get a feeling of pleasure from any activity.
3 I am unable to get any pleasure or enjoyment from anything.

22. Interest in Sex (Please Rate Interest, not Activity):

0 I'm just as interested in sex as usual.
1 My interest in sex is somewhat less than usual or I do not get the same pleasure from sex as I used to.
2 I have little desire for or rarely derive pleasure from sex.
3 I have absolutely no interest in or derive no pleasure from sex.

23. Feeling slowed down:

0 I think, speak, and move at my usual rate of speed.
1 I find that my thinking is slowed down or my voice sounds dull or flat.
2 It takes me several seconds to respond to most questions and I'm sure my thinking is slowed.
3 I am often unable to respond to questions without extreme effort.

24. Feeling restless:

0 I do not feel restless.
1 I'm often fidgety, wring my hands, or need to shift how I am sitting.
2 I have impulses to move about and am quite restless.
3 At times, I am unable to stay seated and need to pace around.

25. Aches and pains:

0 I don't have any feeling of heaviness in my arms or legs and don't have any aches or pains.
1 Sometimes I get headaches or pains in my stomach, back or joints but these pains are only sometime present and they don't stop me from doing what I need to do.
2 I have these sorts of pains most of the time.
3 These pains are so bad they force me to stop what I am doing.

26. Other bodily symptoms:

0 I don't have any of these symptoms: heart pounding fast, blurred vision, sweating, hot and cold flashes, chest pain, heart turning over in my chest, ringing in my ears, or shaking.
1 I have some of these symptoms but they are mild and are present only sometimes.
2 I have several of these symptoms and they bother me quite a bit.
3 I have several of these symptoms and when they occur I have to stop doing whatever I am doing.

Range 0-84 Score: _____

27. Panic/Phobic symptoms:

0 I have no spells of panic or specific fears (phobia) (such as animals or heights).
1 I have mild panic episodes or fears that do not usually change my behavior or stop me from functioning.
2 I have significant panic episodes or fears that force me to change my behavior but do not stop me from functioning.
3 I have panic episodes at least once a week or severe fears that stop me from carrying on my daily activities.

28. Constipation/diarrhea:

0 There is no change in my usual bowel habits.
1 I have intermittent constipation or diarrhea which is mild.
2 I have diarrhea or constipation most of the time but it does not interfere with my day-to-day functioning.
3 I have constipation or diarrhea for which I take medicine or which interferes with my day-to-day activities.

29. Interpersonal Sensitivity:

0 I have not felt easily rejected, slighted, criticized or hurt by others at all.
1 I have occasionally felt rejected, slighted, criticized or hurt by others.
2 I have often felt rejected, slighted, criticized or hurt by others, but these feelings have had only slight effects on my relationships or work.
3 I have often felt rejected, slighted, criticized or hurt by others and these feelings have impaired my relationships and work.

30. Leaden Paralysis/Physical Energy:

0 I have not experienced the physical sensation of feeling weighted down and without physical energy.
1 I have occasionally experienced periods of feeling physically weighted down and without physical energy, but without a negative effect on work, school, or activity level.
2 I feel physically weighted down (without physical energy) more than half the time.
3 I feel physically weighted down (without physical energy) most of the time, several hours per day, several days per week.

Appendix j: Brief Pain Inventory

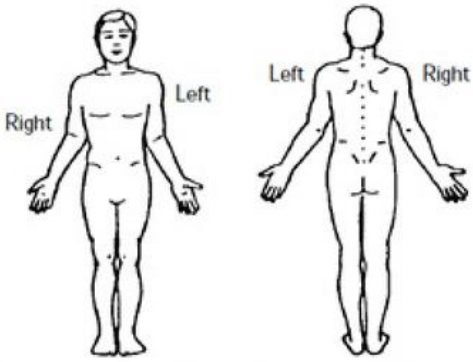
Date: _____ /_____ /_____ Time: _____

Name: _____ _____ _____
Last First Middle intial

1) Throughout our lives, most of us have had pain from time to time (such as minor headaches, sprains, and toothaches). Have you had pain other than these everyday kinds of pain today?

1. Yes 2. No

2) On the diagram, shade in the areas where you feel pain. Put an X on the area that hurts the most.




3) Please rate your pain by circling the one number that best describes your pain at its **worst** in the past 24 hours.

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| No pain | | | | | | | | | | Pain as bad as you can imagine |

4) Please rate your pain by circling the one number that best describes your pain at its **least** in the past 24 hours.

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| No pain | | | | | | | | | | Pain as bad as you can imagine |

5) Please rate your pain by circling the one number that best describes your pain on **average**.

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| No pain | | | | | | | | | | Pain as bad as you can imagine |

6) Please rate your pain by circling the one number that tells how much pain you have **right now**.

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| No pain | | | | | | | | | | Pain as bad as you can imagine |

7) What treatments or medications are you receiving for your pain?

_____

_____

8) In the past 24 hours, how much **relief** have pain treatments or medications provided? Please circle the one percentage that most shows how much relief you have received.

| 0% | 10 | 20 | 30 | 40 | 50 | 60 | 70 | 80 | 90 | 100% |
|---|---|---|---|---|---|---|---|---|---|---|
| No relief | | | | | | | | | | Complete relief |

9) Circle the one number that describes how, during the past 24 hours, pain has **interfered** with your:

A. General activity

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

B. Mood

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

C. Walking ability

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

D. Normal work (includes both work outside the home and housework)

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

E. Relations with other people

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

F. Sleep

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

G. Enjoyment of life

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

# Reporting checklist for genetic association study.

Based on the STREGA guidelines.

## Instructions to authors

Complete this checklist by entering the page numbers from your manuscript where readers will find each of the items listed below.

Your article may not currently address all the items on the checklist. Please modify your text to include the missing information. If you are certain that an item does not apply, please write "n/a" and provide a short explanation.

Upload your completed checklist as an extra file when you submit to a journal.

In your methods section, say that you used the STREGAreporting guidelines, and cite them as:

Little J, Higgins JP, Ioannidis JP, Moher D, Gagnon F, von Elm E, Khoury MJ, Cohen B, Davey-Smith G, Grimshaw J, Scheet P, Gwinn M, Williamson RE, Zou GY, Hutchings K, Johnson CY, Tait V, Wiens M, Golding J, van Duijn C, McLaughlin J, Paterson A, Wells G, Fortier I, Freedman M, Zecevic M, King R, Infante-Rivard C, Stewart A, Birkett N; STrengthening the REporting of Genetic Association Studies. STrengthening the REporting of Genetic Association Studies (STREGA): An Extension of the STROBE Statement.

| | | Reporting Item | Page Number |
|---|---|---|---|
| **Title and abstract** | | | |
| Title | #1a | Indicate the study's design with a commonly used term in the title or the abstract | 1 |

| | | | |
|---|---|---|---|
| Abstract | #1b | Provide in the abstract an informative and balanced summary of what was done and what was found | 3 |
| **Background/rationale** | | | |
| | #2 | Explain the scientific background and rationale for the investigation being reported | 6 |
| **Objectives** | | | |
| | #3 | State specific objectives, including any prespecified hypotheses. State if the study is the first report of a genetic association, a replication effort, or both. | 9 |
| **Study design** | | | |
| | #4 | Present key elements of study design early in the paper | 10 |
| **Setting** | | | |
| | #5 | Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection | 10 |
| **Eligibility criteria** | | | |
| | #6a | Cohort study – Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up. Case-control | 11 |

| | | | |
|---|---|---|---|
| | | study – Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls. Cross-sectional study – Give the eligibility criteria, and the sources and methods of selection of participants. Give information on the criteria and methods for selection of subsets of participants from a larger study, when relevant. | |
| | #6b | Cohort study – For matched studies, give matching criteria and number of exposed and unexposed. Case-control study – For matched studies, give matching criteria and the number of controls per case. | n/a, not matched study |
| **Variables** | | | |
| | #7a | Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable | 11, 14-16 |
| | #7b | Clearly define genetic exposures (genetic variants) using a widely-used nomenclature system. Identify variables likely to be associated with population stratification (confounding by ethnic origin). | 16 |
| **Data** | | | |

| | | | |
|---|---|---|---|
| sources/measurement | | | |
| | #8a | For each variable of interest give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group. Give information separately for for exposed and unexposed groups if applicable. | 11, 14-16 |
| | #8b | Describe laboratory methods, including source and storage of DNA, genotyping methods and platforms (including the allele calling algorithm used, and its version), error rates and call rates. State the laboratory / centre where genotyping was done. Describe comparability of laboratory methods if there is more than one group. Specify whether genotypes were assigned using all of the data from the study simultaneously or in smaller batches. | 16 |
| Bias | | | |
| | #9a | Describe any efforts to address potential sources of bias | 21 |
| | #9b | Describe any efforts to address potential sources of bias | 21 |
| Study size | | | |

| | | | |
|---|---|---|---|
| | #10 | Explain how the study size was arrived at | 17 |
| **Quantitative variables** | | | |
| | #11 | Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen, and why. If applicable, describe how effects of treatment were dealt with. | 18 |
| **Statistical methods** | | | |
| | #12a | Describe all statistical methods, including those used to control for confounding. State software version used and options (or settings) chosen. | 18-19 |
| | #12b | Describe any methods used to examine subgroups and interactions | n/a to protocol paper. |
| | #12c | Explain how missing data were addressed | n/a to protocol paper. |
| | #12d | If applicable, explain how loss to follow-up was addressed | n/a to protocol paper. |
| | #12e | Describe any sensitivity analyses | n/a to protocol paper. |
| | #12f | State whether Hardy-Weinberg equilibrium was considered and, if so, how. | 18 |
| | #12g | Describe any methods used for inferring genotypes or haplotypes | 18 |

| | | | |
|---|---|---|---|
| | #12h | Describe any methods used to assess or address population stratification. | 18 |
| | #12i | Describe any methods used to address multiple comparisons or to control risk of false positive findings. | 18 |
| | #12j | Describe any methods used to address and correct for relatedness among subjects | 18 |
| **Participants** | | | |
| | #13a | Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed. Give information separately for for exposed and unexposed groups if applicable. Report numbers of individuals in whom genotyping was attempted and numbers of individuals in whom genotyping was successful. | n/a to protocol paper. Participant enrollment is not finished yet |
| | #13b | Give reasons for non-participation at each stage | n/a to protocol paper. Participant enrollment is not finished yet |
| | #13c | Consider use of a flow diagram | See figure 1 |

| | | | |
|---|---|---|---|
| **Descriptive data** | | | |
| | #14a | Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders. Give information separately for exposed and unexposed groups if applicable. Consider giving information by genotype | n/a to protocol paper. Participant enrollment is not finished yet |
| | #14b | Indicate number of participants with missing data for each variable of interest | |
| | #14c | Cohort study – Summarize follow-up time, e.g. average and total amount. | |
| **Outcome data** | | | |
| | #15 | Cohort study Report numbers of outcome events or summary measures over time.Give information separately for exposed and unexposed groups if applicable. Report outcomes (phenotypes) for each genotype category over time Case-control study – Report numbers in each exposure category, or summary measures of exposure.Give information separately for cases and controls . Report numbers in each genotype category. Cross-sectional study – Report numbers of outcome events or summary measures. Give information separately for exposed and | n/a to protocol paper. Participant enrollment is not finished yet |

| | | | |
|---|---|---|---|
| | | unexposed groups if applicable. Report outcomes (phenotypes) for each genotype category | |
| **Main results** | | | |
| | #16a | Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included | n/a to protocol paper. Participant enrollment is not finished yet |
| | #16b | Report category boundaries when continuous variables were categorized | |
| | #16c | If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period | |
| | #16d | Report results of any adjustments for multiple comparisons | |
| **Other analyses** | | | |
| | #17a | Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity analyses | n/a to protocol paper. Participant enrollment is not finished yet |
| | #17b | Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity | |

| | | | |
|---|---|---|---|
| | | analyses | |
| | #17c | Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity analyses | |
| **Key results** | | | |
| | #18 | Summarise key results with reference to study objectives | n/a to protocol paper. Participant enrollment is not finished yet |
| **Limitations** | | | |
| | #19 | Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias. | 21 |
| **Interpretation** | | | |
| | #20 | Give a cautious overall interpretation considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence. | 21 |
| **Generalisability** | | | |
| | #21 | Discuss the generalisability (external validity) of the study results | 21 |

## Funding

| | | |
|---|---|---|
| #22 | Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based | 23 |

BMJ Open

# Pain Predict Genetics: Protocol for a prospective observational study of clinical and genetic factors to predict the development of postoperative pain.



| | |
|---|---|
| Journal: | *BMJ Open* |
| Manuscript ID | bmjopen-2022-066134.R1 |
| Article Type: | Protocol |
| Date Submitted by the Author: | 12-Sep-2022 |
| Complete List of Authors: | LI, SONG; Radboudumc Radboud Institute for Health Sciences, Department of Human Genetics<br>van Boekel, Regina L.M.; Radboudumc, Department of Anesthesiology, Pain and Palliative Medicine<br>van den Heuvel, Sandra ; Radboudumc, Department of Anesthesiology, Pain and Palliative Medicine<br>Coenen, Marieke J.H.; Radboudumc Radboud Institute for Health Sciences, Department of Human Genetics<br>Vissers, Kris ; Radboudumc, Department of Anesthesiology, Pain and Palliative Medicine |
| Primary Subject Heading: | Surgery |
| Secondary Subject Heading: | Epidemiology, Genetics and genomics, Surgery |
| Keywords: | PAIN MANAGEMENT, GENETICS, SURGERY, EPIDEMIOLOGY |
| | |

# Pain Predict Genetics: Protocol for a prospective observational study of clinical and genetic factors to predict the development of postoperative pain.

Song Li [1], Regina L.M. van Boekel [2], Sandra A.S. van den Heuvel [2], Marieke J.H. Coenen [1], Kris C.P. Vissers [2]

Authors' Affiliations:

[1] Department of Human Genetics, Radboud Institute for Health Sciences, Radboud university medical center, Nijmegen, The Netherlands.

[2] Department of Anesthesiology, Pain and Palliative Medicine, Radboud university medical center, Nijmegen, The Netherlands.

Email:

Song Li: Song.Li@radboudumc.nl

Regina (Rianne) van Boekel: Rianne.vanBoekel@radboudumc.nl

Sandra van den Heuvel Sandra.vandenHeuvel@Radboudumc.nl

Marieke Coenen: Marieke.Coenen@radboudumc.nl

Kris C.P. Vissers Kris.Vissers@radboudumc.nl

Corresponding author:

Regina L.M. van Boekel

Rianne.vanBoekel@radboudumc.nl

Radboud University Medical Center,

Department of Anesthesiology, Pain and Palliative Medicine

PO Box 9101, intern 549

6500 HB, Nijmegen, The Netherlands

Phone: 0031-243668120

Fax: 0031243613585

The number of text pages: 28

The actual number of figures and tables: 1 Figure, 1 Table, and 1 Supplementary file.

## Abstract

**Introduction** Postoperative pain remains a challenging medical condition impacting the quality of life of every patient. Although several predictive factors for postoperative pain have been identified, an adequate prediction of postoperative pain in patients at risk has not been achieved yet.

The primary objective of this study is to identify specific genetic risk factors for the development of acute and chronic postoperative pain to construct a prediction model facilitating a more personalized postoperative pain management for each individual. The secondary objectives are to build a databank enabling researchers to identify other risk factors for postoperative pain, for instance, demographic and clinical outcome indicators; provide insight into (genetic) factors that predict pharmacological pain relief; investigate the relationship between acute and chronic postoperative pain.

**Methods and analysis** In this prospective, observational study, patients who undergo elective surgery will be recruited to a sample size of approximately 10,000 patients. Postoperative acute and chronic pain outcomes will be collected through questionnaires at different time points after surgery in the follow-up of six months. Potential genetic, demographic, and clinical risk factors for prediction model construction will be collected through blood, questionnaires, and electronic health records, respectively.

Genetic factors associated with acute and/or chronic postoperative pain will be identified using a genome-wide association (GWA) analysis. Clinical risk factors as stated in the secondary

objectives will be assessed by multivariable regression. A clinical easy-to-use prediction model will be created for postoperative pain to allow clinical use for the stratification of patients.

**Ethics and dissemination** The Institutional Review Board of the Radboud university medical center approved the study (authorization number: 2012/117). The results of this study will be made available through peer-reviewed scientific journals and presentations at relevant conferences, which will finally contribute to personalized postoperative pain management.

**Trial registration number** NCT02383342

**Keywords**: Postoperative pain, Genome-wide association study (GWAS), Risk factor, Prediction model, Pharmacogenetics

## Strengths and limitations of this study

- This is a large prospective study to identify genetic and other risk factors for postoperative pain.
- We will build a databank with comprehensive interdisciplinary measurements that assess postoperative pain from multiple perspectives.
- Outcome measurements of pain by patient-reported outcomes, rather than evaluated by professionals.
- The investigating biomarkers of postoperative pain are limited to genetic variants.

## Introduction

Pain after surgery remains a challenging medical and societal problem [1]. Pain is one of the most common postsurgical side effects, with moderate to severe acute postoperative pain occurring in about 41% of the patients [2-4]. Severe postoperative pain is associated with an increased incidence of postoperative complications [5], including prolonged hospital stay, readmissions, and significant reduction of patient satisfaction and quality of life [6 7]. Besides, acute postoperative pain is associated with chronic pain development after surgery [8]. A recent position paper from the International Association for the Study of Pain stated that among the almost 40 million people undergoing surgery globally each year, one out of ten develops chronic postsurgical pain (CPSP), and one out of hundred suffers from severe CPSP, which will negatively affect patients' quality of life [9]. In addition, postoperative pain is a considerable burden on health care service costs, both directly due to patients' increased consumption of medical care and indirectly due to absenteeism, reduced productivity, and increased social welfare payments [10-15].

The management of both acute postoperative pain [2 16] and CPSP [2 17] has remained suboptimal. Despite major investments in clinical protocols and guidelines for structural pain management, infrastructure, and acute pain services (APS), no significant outcome improvements in the quality of postoperative pain management for individual patients have been achieved in the last fifteen years [10 11].

Given the high incidence of postoperative pain, identifying patients at risk for CPSP before the operation is important to apply more personalized pain prevention strategies. The most important demographic and clinical risk factors for postoperative pain are younger age, female sex, smoking, history of depressive symptoms, anxiety symptoms, sleep difficulties, higher body

mass index, presence of preoperative pain, and use of preoperative analgesics [18]. Based on these factors, models have been developed to predict severe acute postoperative pain [19 20] and CPSP [21 22]. A recent study has evaluated a presurgical risk score for CPSP in a prospective cohort, and it reliably identified about 70% of the patients undergoing surgeries at risk of CPSP [23 24].

As a multifactorial trait, the incidence variation of CPSP in the population can be explained partly by the demographic and clinical risk factors mentioned above, and partly due to the genetic and epigenetic differences among patients [25 26]. To improve the accuracy and power of prediction, efforts have been made to predict CPSP using genetic variants [21 24]. However, no unequivocal genetic predictors have been found yet. In addition, many exploratory studies investigated the possible role of candidate genes in postoperative pain development. In particular, associations have been found between CPSP and the *μ-opioid receptor (OPRM1)* and *catechol-O-methyl transferase (COMT)* genes [27 28]. Still, these results have not been confirmed by others. *OPRM1* is also associated with basal pain sensitivity differences [29], which could be caused by the altered opioid binding potential in the central nervous system [30]. More recently, hypothesis-free methods, such as genome-wide association studies, have been applied for CPSP to identify markers across the genome [31 32]. One of the studies showed that a genetic variant in the *protein-kinase C* gene is linked to neuropathic pain after complete joint replacement. This gene is involved in long-term potentiation, synaptic plasticity, chronic pain, and memory, indicating that this gene may be relevant for neuropathic pain initiation. The disadvantage of this study is that it was small in terms of patient numbers and only focused on one specific surgical procedure.

Besides genetic variants for altered pain sensitivity, also gene variants in drug metabolism can play a role. Understanding the reasons for ineffective treatment can facilitate the early identification of patients at risk and provide more effective and customized postoperative management. Some associated genes with pain treatment outcomes are also involved in pain development, such as *COMT* [33-35]. Genes involved in the action site of active drugs or the drugs' metabolism might play a role in the therapeutic response of this drug. A well-known example is the *cytochrome P450* (*CYP*) family investigated for several drugs (e.g., codeine and tramadol) [36]. However, this area has never been charted in a large population [37].

To date, adequate prediction of patients at risk for postoperative pain in clinical practice has not been achieved for several reasons. First, although many demographic, clinical, and lifestyle factors of postoperative pain have been reported [18], a lack of consensus on the best outcome indicators for postoperative pain management [38 39] hinders choosing the proper outcome variables for prediction model construction. Second, the potential genetic risk factors of postoperative pain prediction remain obscure. The role of genetic factors in postoperative pain have not been investigated sufficiently, making it challenging to select appropriate genetic risk factors to construct a prediction model. Third, when prediction models are updated, external validation (i.e., in a new population) is important before being implemented in a clinical setting [40-43], which is often difficult due to the lack of validation cohorts. For these reasons, we hypothesize that a global structural multicenter diagnostic program of postoperative pain in a surgical patient population will be valuable for better identifying patients at risk of CPSP and ultimately preventing postoperative pain using individualized pharmacological and non-pharmacological interventions.

## Objectives

The primary objective of the Pain Predict Genetics (PPG) study is to identify genetic risk factors for acute and chronic postoperative pain development and to construct a prediction model for personalized postoperative pain management.

The secondary objectives of the PPG study are to build a databank enabling researchers to 1) identify other risk factors for the development of acute and chronic postoperative pain; 2) provide insights into complications and other clinical outcome indicators after surgery; 3) provide insights into the relationship between acute and chronic postoperative pain; 4) identify (genetic) factors that predict pharmacological pain relief. The databank will be open to the public with access fees, and reasonable requests will be discussed in the research group before approval.

The extensive data collection on (chronic) postoperative pain development of patients undergoing surgery offers many possibilities for additional research questions using conventional statistical methods and artificial intelligence, e.g., machine learning. The cohort could be used to 1) conduct epidemiological studies; 2) investigate other parameters (for example, types of surgery) that are involved in the development of chronic postoperative pain; 3) validate new prediction models for (chronic) postoperative pain; 4) identify factors for the postoperative outcome (for example, death, long-term hospitalization, complications); 5) collaborate with other groups to perform large-scale analysis to identify predictors for the development of (chronic) postoperative pain.

# Methods and analyses

## Study design

A prospective, observational study of 10,000 patients will undergo elective surgery. This study will run for at least ten years, during which period it must be possible to include the intended number of patients. Patient inclusion after CMO (Human Research Committee, in Dutch Commissie Mensgebonden Onderzoek) approval was started in March 2015, and patient inclusion was temporarily stopped in 2020 due to COVID restrictions. In the near future, this study will be continued as a multicenter study; hospitals have already been approached and indicated that they intend to participate.

## Patient and public involvement

During the design of the study the patients aided in the pilot phase of the questionnaires, during the recruitment the patients are informed concerning the project. In addition, patient reported outcomes will be used. Patients will be informed about the outcome of the study at several moments (depending on the obtained results).

## Participants

Patients who undergo electvie surgry and are eligible for this study will be approached before their planned surgery during the preoperative consultation. In this way, potential participants will have sufficient time to consider the study information. If any questions arise, it is possible to contact the researchers by telephone or ask the questions during the preoperative consultation. During the preoperative consultation (outpatient clinic or by telephone), the physician (assistant) will ask the patient if they are interested to participating in the study. If the patient is willing to participate, the informed consent form will be signed and dated. If patients have an

online preoperative consultation, this procedure will take place digitally, and patients receive the study forms (signed in advance) at home to return if they consent.

Patients are eligible for study inclusion if they 1) are older or equal to 16 years; 2) undergo elective surgery with an incision, including cardiothoracic surgery (e.g., cardiomyotomy), general surgery (e.g., breast resection), neurological surgery (e.g., nerve decompression), oral and maxillofacial surgery (e.g., removal of head and neck benign and malignant tumors), otorhinolaryngology (e.g., tympanoplasty), plastic surgery (e.g., breast reconstruction), trauma and orthopedic surgery (e.g., arthroplasty), urology (e.g., prostatectomy) and vascular surgery (e.g., treatment of varicose veins); 3) can read and understand the patient information; 4) will provide informed consent. Patients will be excluded if they 1) intend to undergo another surgery within six months; 2) do not have enough knowledge of the language in words and understanding to complete questionnaires.

## Measurements

### Questionnaires

After written informed consent, participants will be asked to complete questionnaires before and after their surgery. An overview of the study workflow and data collection time points can be found in **Figure 1** and **Table 1**. All patient data will be stored in an online digital database, Castor [44]. The reliability and validity of all questionnaires for measurement collection have been validated in the corresponding populations.

## Table 1: Overview of data collection

| | T0 | Day -1 | Surgery | Day 1 | Day 2 | Day 3 | Week 1 | Week 6 | Month 3 | Month 6 |
|---|---|---|---|---|---|---|---|---|---|---|
| Informed consent | x | | | | | | | | | |
| **Questionnaires** | | | | | | | | | | |
| Demografic data | | x | | | | | | | | |
| Incision size | | x | | x | | | | | | |
| Pain scores | | x | | x | x | x | x | x | x | x |
| Physical activities | | | | x | x | x | x | | | |
| Pain disability index | | x | | | | | x | x | x | x |
| APAIS | | x | | | | | | | | |
| PCS | | x | | | | | | | | |
| PSQ | | x | | | | | | | | |
| Chronic pain | | x | | | | | | | x | x |
| IDS depression | | x | | | | | | | | |
| Brief pain inventory | | | | | | | | | x | x |
| **Data electronic medical file** | | | | | | | | | | |
| Physical status by ASA | | | | | | | | | | x |
| Type of surgery | | | | | | | | | | x |
| Duration of surgery | | | | | | | | | | x |
| Type of anesthesia | | | | | | | | | | x |
| Complications | | | | | | | | | | x |
| Hospital stay | | | | | | | | | | x |
| Pain medication use | | | | | | | | | | x |
| Incision size | | | | | | | | | | x |
| Second surgery within 6 months | | | | | | | | | | x |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| General clinical outcome indicators | | | | | | | | | | x |
| **Body material*** | | | | | | | | | | |
| 1x10 ml blood for DNA | | | x | | | | | | | |

*In the event that it is not possible to collect a blood sample during surgery, the subject may be asked to provide a DNA sample via a saliva collection tube.

APAISI, Amsterdam Pre-operative Anxiety and Information Scale. PCS, Pain Catastrophizing Scale. PSQ, Pain Sensitivity questionnaire. IDS, Inventory of Depressive Symptomatology. ASA, American Society of Anesthesiologists classification.

The first digital questionnaire must be completed the day before the surgery (no longer than one week before). Before surgery, the following parameters will be collected (**Table 1,** Supplementary File 1): demographic characteristics (such as gender, age, BMI), expected incision size in mm, pain intensity, pain disability, preoperative anxiety and need for information, pain catastrophizing, pain sensitivity, preoperative chronic pain characteristics, and depressive symptoms.

After surgery, the following parameters will be collected: actual incision size in mm on day 1; pain intensity on day 1, 2, 3, week 1 and 6, and month 3 and 6; physical activities on day 1, 2, 3, week 1; pain disability on week 1 and 6, and month 3 and 6; postoperative chronic pain characteristics on month 3 and 6; characteristics of pain on month 3 and 6.

Pain intensity will be measured with an 11-point numerical rating scale (NRS) at rest and during a normal patient action at that time [20]. The endpoints represent the extremes of the pain experience: 0 means "no pain at all", and 10 means "worst possible pain".

Pain disability (disability associated with pain) will be measured by the widely used Pain Disability Index Dutch language version (PDI) [45 46]. The PDI is a 7-item questionnaire to investigate the magnitude of the self-reported disability in different situations such as work, leisure time, daily life activities, and sports. The questionnaire is constructed on an 11-point NRS in which 0 means "no disability" and 10 means "maximum disability".

Preoperative anxiety and need for information will be evaluated by the Amsterdam Preoperative Anxiety and Information Scale (APAIS) [47]. The APAIS consists of six questions and each score on a 5-point Likert scale from 1 (not at all) to 5 (extremely), with four questions to

assess the patient's preoperative anxiety score and two questions to assess the patient's need for information regarding the scheduled surgery and anesthesia [20].

Pain catastrophizing is generally described as an absurd negative orientation towards hurtful stimuli and is important in pain coping [48]. It will be measured by the Pain Catastrophizing Scale (PCS), a self-evaluating questionnaire consisting of 13 questions. People are asked to indicate the degree to which they have thoughts and feelings when experiencing pain using the 0 (not at all) to 4 (all the time) scale, and a total score will be yielded (range from 0 to 52).

The Pain Sensitivity Questionnaire (PSQ) will measure patients' preoperative pain sensitivity [49] [50]. The PSQ consists of 17 questions that describe daily life situations; respondents score their pain intensity for these situations on an NRS by scoring 0 (not painful) to 10 (severest pain imaginable).

The severity of overall depressive symptoms will be assessed by the Inventory of Depressive Symptomatology Self Report (IDS-SR) [51 52]. IDS-SR is a 30-item questionnaire, and each item has four statements scored on a four-point scale from 0 to 3. There are two items about either increasing or decreasing appetite and two items about increasing or decreasing weight. Only the item with the higher score from both pairs will be chosen. The total score is based on 28 items and ranges from 0 to 84.

Physical activities (ability to perform normal activities) will be measured by questions assessing the degree of physical activities interfered by surgery, including bed activities (such as turning), breathing deeply of coughing, sleeping, and activities out of bed. Each item is scored on an 11-point NRS in which 0 means did not interfere and 10 means completely interfered. These

questions are derived from the validated International Pain Outcomes questionnaire and are found responsive to asking patients about their ability to perform normal activities directly after surgery [53].

Characteristics of pain will be measured by the Brief Pain Inventory – Short Form (BPI-SF), which is a shortened version of the Brief Pain Inventory [54]. BPI-SF evaluates pain severity during the past 24 hours and current level, with 0 representing "no pain" and 10 "the worst pain imaginable". Seven items in BPI-SF assess interference with daily functioning (such as general activity, walking, and work) on an 11-point scale, where 0 represents "no interference" and 10 "complete interference".

## Collection of body material

One tube of blood will be collected for DNA isolation. The burden for the patient is minimalized as blood will be taken using the intravenous line in place for surgery. If it is impossible to collect blood presurgically or postsurgically, we will collect saliva for DNA isolation (Genefix DNA saliva collectors; GFX-02/50, Isohelix).

## Clinical information

The following clinical information will be collected from the electronic patient file six months after operation (**Table 1**): physical status by The American Society of Anesthesiologists classification (ASA-status); type of surgery; duration of surgery; type of anesthesia; postoperative complications within 30 days after surgery, one-time retrospectively, which is defined as any medical adverse outcome occurring between admission and 30 days after operation. Complications occurring in the operation room and complications directly related to anesthesia (e.g., nausea which resolves immediately after medication in the operation room)

will not be included [5 55]. Furthermore, data on pain medication use, before surgery and after surgery; actual incision size in mm; second surgery within 6 months; general clinical outcome indicators, including surgical site infection at 30 days, stroke within 30 days of surgery, death within 30 days of surgery, admission to the intensive care unit within 14 days of surgery, readmission to hospital within 30 days of surgery, and length of hospital stay (with or without in-hospital mortality) will be collected [38].

## Outcome measures

The outcome measures are acute postoperative pain and chronic postoperative pain. Acute postoperative pain is defined as pain experienced directly after surgery. Thresholds or cut-off points of the pain intensity are set as none to mild (0-3), moderate (4-7), and severe (8-10) [56 57]. The definition of CPSP is in agreement with IASP terminology of chronic postsurgical pain, i.e., "*chronic pain that develops or increases in intensity after a surgical procedure persists beyond the healing process, i.e., at least 3 months after the surgery*" [9]. CPSP will be measured by a chronic pain characteristics questionnaire postoperatively at three and six months. Patients will be asked to indicate whether they had a recent pain experience, the site of pain, and whether it lasted more than three months [58 59]. The intensity of CPSP will also be characterized by the pain scores questionnaire using the same threshold as acute postoperative pain. The influence of pain on functional and mood changes will be measured by the PDI and the BPI-SF.

## Sample size calculation

The power of the genetic study is based on the primary research question investigating which genetic factors are associated with postoperative pain. Power is calculated using the Genetic Power Calculator [60], and the estimated number of patients is based on a GWA approach. For

chronic postoperative pain, we assume a case-control analysis for discrete traits (2df test), a risk allele frequency of 30%, a linkage disequilibrium (D') of 0.8, a prevalence of chronic postoperative pain of 15%, and the relative risk of chronic postoperative pain for persons who are heterozygous of 1.5 and for homozygous persons of 2.25. For a power of 80% with a p-value cut off $5 \times 10^{-8}$ (genome-wide significance threshold), we need 750 patients with chronic postoperative pain and 4,250 people without chronic postoperative pain. For acute pain, the power is even higher. With the same population, we have more than 80% power to detect a relative risk of 1.2 and 1.44 for heterozygous and homozygous patients, respectively. This higher power is due to the higher prevalence of acute (moderate to severe) pain of 55%. Most importantly, results will be replicated in the additional study participants, as the total number of patients included in the study will be 10,000. In addition, we will use cohorts of our collaborators for replication purposes.

## Statistical analysis

The key objective is to identify genetic risk factors that can predict development of acute or chronic postoperative pain and validate previously reported SNPs. A GWA approach will be used as the main analysis. Phenotype data and DNA will be used to identify genetic factors. We will use 5,000 patients for the discovery of genetic variants. Samples will be genotyped with the Infinium Global Screening Array (Illumina). Pre-imputation quality control, principal component analyses, and imputation will follow the RICOPILI pipeline [61]. Potential confounding by ethnic origin will be corrected by principal component analyses. The 1000 Genomes reference panel will be used for imputation, followed by post-imputation quality control in PLINK [62]. Associations between SNPs and the presence of acute or chronic pain will be performed using cutting-edge

methods when data collection is finished. Results will be to ensure validity. SNPs that can be validated will be included in the prediction model described below.

Secondary objectives include identifying other potential risk factors for acute and chronic postoperative pain. Therefore, a univariate association of each potential predictor will be calculated and tested in a multivariable regression model. We will use a least absolute shrinkage and selection operator (Lasso) regression. Shrinkage is where data values are shrunk towards a central point, like the mean. Lasso is a regression analysis method that performs both variable selection and regularization to enhance the prediction accuracy and interpretability of the statistical model it produces. After identifying these risk factors, a prediction rule will be created for (moderate to severe) acute and chronic postoperative pain. Based on this prediction rule, a simple, clinically easy applicable tool will be developed to allow clinical use for the stratification of patients. The predictive performance will be studied in another cohort of patients to test whether the rule is generalizable across time and place. Because it appears from the literature that acute and chronic pain are correlated after surgery, additional correlation analysis will be performed to investigate this correlation in the data.

Similar approaches will be followed to identify the clinical and genetic factors that predict pharmacological pain relief. For some pain medicines, genes that impact pain relief are already known (e.g., *CYP2D6* and morphine). We will first investigate those genes to see if these variants indeed contribute to pharmacological pain relief differences.

## Ethics and dissemination:

The study will be conducted according to the principles of the Declaration of Helsinki version 2013 and in accordance with the Medical Research Involving Human Subjects Act and Good Clinical Practice. The study was approved by the local ethics committee for human research in Nijmegen (Medical Review Ethics Committee Region Arnhem-Nijmegen, authorization number: 2012/117). This study was registered on ClinicalTrials.gov (NCT02383342).

The privacy of the participants is guaranteed by storing encrypted data. Every participant will receive a pseudo-anonymous study number. No identifying data is recorded within the meaning of the law. The key is only accessible to the study team and monitors. Data and material will only be used in coded form within possible collaborations.

The results of this study will be made available through peer-reviewed scientific journals and presentations at relevant conferences. After a thorough evaluation, decisions will be made regarding including the identified risk factors and constructed prediction models into clinical guidelines, thus facilitating personalized postoperative pain management.

## Discussion

This cohort will be a large prospective study to identify risk factors for postoperative pain and to build and evaluate dedicated prediction models for postoperative pain in surgical patients. In addition, the comprehensive information collected in this study will also enable us to answer other research questions regarding postoperative pain, such as the relationships between acute and chronic postoperative pain development. Eventually, these results will be applied in the clinical settings to improve the quality of life for patients who develop postoperative pain.

The strengths of this study are that we will include all elective major operations rather than limiting to one specific operation as in previous studies [32], which allows us to investigate the shared genetic background of postoperative pain in different operations. Furthermore, as there are discrepancies in pain intensity scores understanding [63] and pain management decisions [63 64] between patients and caregivers, the patient's perspective should be respected and assessed for pain evaluation and management [65 66]. Therefore, pain assessment will be conducted by patients themselves (patient-reported outcomes) rather than professionals in this study, leading to a more comprehensive outcome assessment and interpretation [67]. Moreover, the single-use of NRS might be inadequate for patients' pain experience evaluation and pain management decisions [66 68 69]. Thus, another strength of this cohort is that the experience of pain will be estimated by multidimensional measurements focusing on patients' overall functionality rather than merely a NRS pain score. Besides, the comprehensively collected information for postoperative pain in this cohort also empowers analysis that cannot be performed in large-scale registry data (e.g., UK Biobank) as such phenotype data is not available in those datasets. The data collected in this cohort will also enable additional research using conventional and cutting-edge statistical methods like artificial intelligence.

The possible limitations of this study are that we will only investigate DNA variants as biomarkers for pain prediction as our primary research goal. However, other epigenetic [69 70], transcriptomic [70], proteomics [71], and metabolic markers [72] are also potentially involved in (postoperative) pain development. For instance, recent studies indicate that methylation patterns might predict opioid treatment outcomes [69 70]. As the DNA sample of patients is accessible, we will be able to characterize the multi-omics biomarker signatures of

postoperative pain in future researchs, such as investigating the association between epigenetic changes and postoperative pain. In addition, when prediction tools are applied in clinical settings, the sensitivity and specificity of prediction tools are crucial to evaluate their adequacy and usefulness [73]. Although the measurement tools used in prediction models are well-validated and verified (see methods), our findings could still be subject to false positive or negative errors because all measurement tools have limitations. Furthermore, chronic pain assessment is more complex than acute pain [74], and GWAS findings are sometimes incidental [75]. We will consider seeking other available cohorts for validation and applying other statistical methods to validate our findings in future studies, such as polygenic risk scores [76]. Another potential limitation is that loss of follow-up of patients might result in lower patient numbers than expected. Despite this potential concern, we still expect a sufficient sample size as additional centres will start patient inclusion, and the measurements are mainly from patient-reported outcomes via digital follow-up.

Identifying the genetic background of postoperative pain development may give valuable insights into the mechanisms underlying the relationship between postoperative pain and complications after surgery. This may open the way to identify new targets for treatment and potentially simplify the risk profiling assay for future use, yielding a simpler, more accurate, and cost-efficient assay or product. The contribution of improved prevention and treatment of pain after surgery will benefit many patients undergoing surgery and society by decreasing health care service costs.

## Trial status

Patient recruitment is expected to continue until 2025. Recruitment has already started in Radboud university medical center, with more than 500 patients recruited as of October 2021. National and international collaborations will be greatly accepted after careful consideration.

## Author contributions

RvB, MC and KV are responsible for overall planning and execution, formulation and evolution of overarching research goals and aims, development and design of the methodology. RvB, MC, and SvdH will be responsible for project management and coordination responsibility. Analyses and data visualization will be conducted by SL, RvB, and MC. SL prepared the draft of the manuscript, and all authors critically revised the manuscript.

## Acknowledgment

SL was supported by China Scholarship Council (CSC) Grant number 201908130179.

## Funding

Departmental funding covers the costs of this study [grant number: N/A]. We aim to apply for extra grants to cover the potential cost of including more patients and the cost of databank maintenance.

## Competing interests statement

The authors have no relevant financial or non-financial interests to disclose.

# References

1. Gaskin DJ, Richard P. The economic costs of pain in the United States. *J Pain* 2012;13(8):715-24. doi: 10.1016/j.jpain.2012.03.009 [published Online First: 2012/05/23]
2. Dolin SJ, Cashman JN, Bland JM. Effectiveness of acute postoperative pain management: I. Evidence from published data. *Br J Anaesth* 2002;89(3):409-23.
3. Sommer M, de Rijke JM, van Kleef M, et al. The prevalence of postoperative pain in a sample of 1490 surgical inpatients. *Eur J Anaesthesiol* 2008;25(4):267-74. doi: 10.1017/S0265021507003031
4. Gan TJ. Poorly controlled postoperative pain: prevalence, consequences, and prevention. *J Pain Res* 2017;10:2287-98. doi: 10.2147/JPR.S144066 [published Online First: 2017/10/14]
5. van Boekel RLM, Warle MC, Nielen RGC, et al. Relationship Between Postoperative Pain and Overall 30-Day Complications in a Broad Surgical Population: An Observational Study. *Ann Surg* 2019;269(5):856-65. doi: 10.1097/SLA.0000000000002583 [published Online First: 2017/11/15]
6. Peters CL, Shirley B, Erickson J. The effect of a new multimodal perioperative anesthetic regimen on postoperative pain, side effects, rehabilitation, and length of hospital stay after total joint arthroplasty. *The Journal of arthroplasty* 2006;21(6 Suppl 2):132-8. doi: 10.1016/j.arth.2006.04.017
7. Regenbogen SE, Mullard AJ, Peters N, et al. Hospital Analgesia Practices and Patient-reported Pain After Colorectal Resection. *Ann Surg* 2016;264(6):1044-50. doi: 10.1097/SLA.0000000000001541
8. Katz J, Jackson M, Kavanagh BP, et al. Acute pain after thoracic surgery predicts long-term post-thoracotomy pain. *Clin J Pain* 1996;12(1):50-5. [published Online First: 1996/03/01]
9. Schug SA, Lavand'homme P, Barke A, et al. The IASP classification of chronic pain for ICD-11: chronic postsurgical or posttraumatic pain. *Pain* 2019;160(1):45-52. doi: 10.1097/j.pain.0000000000001413 [published Online First: 2018/12/27]
10. Apfelbaum JL, Chen C, Mehta SS, et al. Postoperative pain experience: results from a national survey suggest postoperative pain continues to be undermanaged. *Anesth Analg* 2003;97(2):534-40, table of contents.
11. Meissner W, Coluzzi F, Fletcher D, et al. Improving the management of post-operative acute pain: priorities for change. *Curr Med Res Opin* 2015;31(11):2131-43. doi: 10.1185/03007995.2015.1092122 [published Online First: 2015/09/12]
12. Zimberg SE. Reducing pain and costs with innovative postoperative pain management. *Managed care quarterly* 2003;11(1):34-6.
13. Morrison RS, Magaziner J, McLaughlin MA, et al. The impact of post-operative pain on outcomes following hip fracture. *Pain* 2003;103(3):303-11.
14. Zoucas E, Lydrup ML. Hospital costs associated with surgical morbidity after elective colorectal procedures: a retrospective observational cohort study in 530 patients. *Patient safety in surgery* 2014;8(1):2. doi: 10.1186/1754-9493-8-2
15. Encinosa WE, Hellinger FJ. The impact of medical errors on ninety-day costs and outcomes: an examination of surgical patients. *Health services research* 2008;43(6):2067-85. doi: 10.1111/j.1475-6773.2008.00882.x
16. Sinatra R. Causes and consequences of inadequate management of acute pain. *Pain Med* 2010;11(12):1859-71. doi: 10.1111/j.1526-4637.2010.00983.x [published Online First: 2010/11/03]
17. Glare P, Aubrey KR, Myles PS. Transition from acute to chronic pain after surgery. *Lancet* 2019;393(10180):1537-46. doi: 10.1016/S0140-6736(19)30352-6 [published Online First: 2019/04/16]

18. Yang MMH, Hartley RL, Leung AA, et al. Preoperative predictors of poor acute postoperative pain control: a systematic review and meta-analysis. *BMJ Open* 2019;9(4):e025091. doi: 10.1136/bmjopen-2018-025091 [published Online First: 2019/04/04]
19. Kalkman CJ, Visser K, Moen J, et al. Preoperative prediction of severe postoperative pain. *Pain* 2003;105(3):415-23. doi: 10.1016/s0304-3959(03)00252-5 [published Online First: 2003/10/07]
20. Janssen KJ, Kalkman CJ, Grobbee DE, et al. The risk of severe postoperative pain: modification and validation of a clinical prediction rule. *Anesth Analg* 2008;107(4):1330-9. doi: 10.1213/ane.0b013e31818227da [published Online First: 2008/09/23]
21. Hoofwijk DMN, van Reij RRI, Rutten BPF, et al. Genetic polymorphisms and prediction of chronic post-surgical pain after hysterectomy-a subgroup analysis of a multicenter cohort study. *Acta Anaesthesiol Scand* 2019;63(8):1063-73. doi: 10.1111/aas.13413 [published Online First: 2019/06/18]
22. Althaus A, Hinrichs-Rocker A, Chapman R, et al. Development of a risk index for the prediction of chronic post-surgical pain. *Eur J Pain* 2012;16(6):901-10. doi: 10.1002/j.1532-2149.2011.00090.x
23. Montes A, Roca G, Cantillo J, et al. Presurgical risk model for chronic postsurgical pain based on 6 clinical predictors: a prospective external validation. *Pain* 2020;161(11):2611-18. doi: 10.1097/j.pain.0000000000001945 [published Online First: 2020/06/17]
24. Montes A, Roca G, Sabate S, et al. Genetic and Clinical Factors Associated with Chronic Postsurgical Pain after Hernia Repair, Hysterectomy, and Thoracotomy: A Two-year Multicenter Cohort Study. *Anesthesiology* 2015;122(5):1123-41. doi: 10.1097/ALN.0000000000000611 [published Online First: 2015/05/20]
25. Mauck M, Van de Ven T, Shaw AD. Epigenetics of chronic pain after thoracic surgery. *Curr Opin Anaesthesiol* 2014;27(1):1-5. doi: 10.1097/ACO.0000000000000030 [published Online First: 2013/12/05]
26. van Reij RRI, Joosten EAJ, van den Hoogen NJ. Dopaminergic neurotransmission and genetic variation in chronification of post-surgical pain. *Br J Anaesth* 2019;123(6):853-64. doi: 10.1016/j.bja.2019.07.028 [published Online First: 2019/09/29]
27. De Gregori M, Diatchenko L, Belfer I, et al. OPRM1 receptor as new biomarker to help the prediction of post mastectomy pain and recurrence in breast cancer. *Minerva Anestesiol* 2015;81(8):894-900. [published Online First: 2014/10/11]
28. Hoofwijk DM, van Reij RR, Rutten BP, et al. Genetic polymorphisms and their association with the prevalence and severity of chronic postsurgical pain: a systematic review. *Br J Anaesth* 2016;117(6):708-19. doi: 10.1093/bja/aew378 [published Online First: 2016/12/14]
29. Kim H, Clark D, Dionne RA. Genetic contributions to clinical pain and analgesia: avoiding pitfalls in genetic research. *J Pain* 2009;10(7):663-93. doi: 10.1016/j.jpain.2009.04.001 [published Online First: 2009/06/30]
30. Mueller C, Klega A, Buchholz HG, et al. Basal opioid receptor binding is associated with differences in sensory perception in healthy human subjects: a [18F]diprenorphine PET study. *Neuroimage* 2010;49(1):731-7. doi: 10.1016/j.neuroimage.2009.08.033 [published Online First: 2009/08/26]
31. van Reij RRI, Hoofwijk DMN, Rutten BPF, et al. The association between genome-wide polymorphisms and chronic postoperative pain: a prospective observational study. *Anaesthesia* 2020;75 Suppl 1:e111-e20. doi: 10.1111/anae.14832 [published Online First: 2020/01/07]
32. Warner SC, van Meurs JB, Schiphof D, et al. Genome-wide association scan of neuropathic pain symptoms post total joint replacement highlights a variant in the protein-kinase C gene. *Eur J Hum Genet* 2017;25(4):446-51. doi: 10.1038/ejhg.2016.196 [published Online First: 2017/01/05]
33. Zubieta JK, Heitzeg MM, Smith YR, et al. COMT val158met genotype affects mu-opioid neurotransmitter responses to a pain stressor. *Science* 2003;299(5610):1240-3. doi: 10.1126/science.1078546 [published Online First: 2003/02/22]

34. Rakvåg TT, Klepstad P, Baar C, et al. The Val158Met polymorphism of the human catechol-O-methyltransferase (COMT) gene may influence morphine requirements in cancer pain patients. *Pain* 2005;116(1-2):73-8. doi: 10.1016/j.pain.2005.03.032 [published Online First: 2005/06/02]
35. Reyes-Gibby CC, Shete S, Rakvåg T, et al. Exploring joint effects of genes and the clinical efficacy of morphine for cancer pain: OPRM1 and COMT gene. *Pain* 2007;130(1-2):25-30. doi: 10.1016/j.pain.2006.10.023 [published Online First: 2006/12/13]
36. Owusu Obeng A, Hamadeh I, Smith M. Review of Opioid Pharmacogenetics and Considerations for Pain Management. *Pharmacotherapy* 2017;37(9):1105-21. doi: 10.1002/phar.1986 [published Online First: 2017/07/13]
37. De Gregori M, Diatchenko L, Ingelmo PM, et al. Human Genetic Variability Contributes to Postoperative Morphine Consumption. *J Pain* 2016;17(5):628-36. doi: 10.1016/j.jpain.2016.02.003 [published Online First: 2016/02/24]
38. Haller G, Bampoe S, Cook T, et al. Systematic review and consensus definitions for the Standardised Endpoints in Perioperative Medicine initiative: clinical indicators. *Br J Anaesth* 2019;123(2):228-37. doi: 10.1016/j.bja.2019.04.041 [published Online First: 2019/05/28]
39. Pogatzki-Zahn E, Schnabel K, Kaiser U. Patient-reported outcome measures for acute and chronic pain: current knowledge and future directions. *Curr Opin Anaesthesiol* 2019;32(5):616-22. doi: 10.1097/ACO.0000000000000780 [published Online First: 2019/08/16]
40. Justice AC, Covinsky KE, Berlin JA. Assessing the generalizability of prognostic information. *Ann Intern Med* 1999;130(6):515-24. doi: 10.7326/0003-4819-130-6-199903160-00016 [published Online First: 1999/03/13]
41. Reilly BM, Evans AT. Translating clinical research into clinical practice: impact of using prediction rules to make decisions. *Ann Intern Med* 2006;144(3):201-9. doi: 10.7326/0003-4819-144-3-200602070-00009 [published Online First: 2006/02/08]
42. Altman DG, Royston P. What do we mean by validating a prognostic model? *Stat Med* 2000;19(4):453-73. doi: 10.1002/(sici)1097-0258(20000229)19:4<453::aid-sim350>3.0.co;2-5 [published Online First: 2000/03/01]
43. Harrell FE, Jr., Lee KL, Mark DB. Multivariable prognostic models: issues in developing models, evaluating assumptions and adequacy, and measuring and reducing errors. *Stat Med* 1996;15(4):361-87. doi: 10.1002/(SICI)1097-0258(19960229)15:4<361::AID-SIM168>3.0.CO;2-4 [published Online First: 1996/02/28]
44. Castor EDC. Castor Electronic Data Capture 2019 [27 Aug. 2019]. Available from: https://castoredc.com accessed August 28, 2019.
45. Soer R, Köke AJ, Vroomen PC, et al. Extensive validation of the pain disability index in 3 groups of patients with musculoskeletal pain. *Spine (Phila Pa 1976)* 2013;38(9):E562-8. doi: 10.1097/BRS.0b013e31828af21f [published Online First: 2013/02/08]
46. Tait RC, Pollard CA, Margolis RB, et al. The Pain Disability Index: psychometric and validity data. *Arch Phys Med Rehabil* 1987;68(7):438-41. [published Online First: 1987/07/01]
47. Moerman N, van Dam FS, Muller MJ, et al. The Amsterdam Preoperative Anxiety and Information Scale (APAIS). *Anesth Analg* 1996;82(3):445-51. doi: 10.1097/00000539-199603000-00002 [published Online First: 1996/03/01]
48. Sullivan MJL, Bishop SR, Pivik J. The Pain Catastrophizing Scale: Development and validation. *Psychological Assessment* 1995;7(4):524-32. doi: 10.1037/1040-3590.7.4.524
49. Ruscheweyh R, Marziniak M, Stumpenhorst F, et al. Pain sensitivity can be assessed by self-rating: Development and validation of the Pain Sensitivity Questionnaire. *Pain* 2009;146(1-2):65-74. doi: 10.1016/j.pain.2009.06.020 [published Online First: 2009/08/12]

50. Van Boekel RLM, Timmerman H, Bronkhorst EM, et al. Translation, Cross-Cultural Adaptation, and Validation of the Pain Sensitivity Questionnaire in Dutch Healthy Volunteers. *Pain Res Manag* 2020;2020:1050935. doi: 10.1155/2020/1050935 [published Online First: 2020/08/11]
51. Rush AJ, Giles DE, Schlesser MA, et al. The Inventory for Depressive Symptomatology (IDS): preliminary findings. *Psychiatry Res* 1986;18(1):65-87. doi: 10.1016/0165-1781(86)90060-0 [published Online First: 1986/05/01]
52. Rush AJ, Gullion CM, Basco MR, et al. The Inventory of Depressive Symptomatology (IDS): psychometric properties. *Psychol Med* 1996;26(3):477-86. doi: 10.1017/s0033291700035558 [published Online First: 1996/05/01]
53. Rothaug J, Zaslansky R, Schwenkglenks M, et al. Patients' perception of postoperative pain management: validation of the International Pain Outcomes (IPO) questionnaire. *J Pain* 2013;14(11):1361-70. doi: 10.1016/j.jpain.2013.05.016 [published Online First: 2013/09/12]
54. Cleeland CS, Ryan KM. Pain assessment: global use of the Brief Pain Inventory. *Ann Acad Med Singap* 1994;23(2):129-38. [published Online First: 1994/03/01]
55. Dindo D, Clavien PA. What is a surgical complication? *World J Surg* 2008;32(6):939-41. doi: 10.1007/s00268-008-9584-y [published Online First: 2008/04/17]
56. Gerbershagen HJ, Rothaug J, Kalkman CJ, et al. Determination of moderate-to-severe postoperative pain on the numeric rating scale: a cut-off point analysis applying four different methods. *Br J Anaesth* 2011;107(4):619-26. doi: 10.1093/bja/aer195 [published Online First: 20110630]
57. Treede RD, Rief W, Barke A, et al. Chronic pain as a symptom or a disease: the IASP Classification of Chronic Pain for the International Classification of Diseases (ICD-11). *Pain* 2019;160(1):19-27. doi: 10.1097/j.pain.0000000000001384
58. Macrae WA. Chronic post-surgical pain: 10 years on. *Br J Anaesth* 2008;101(1):77-86. doi: 10.1093/bja/aen099 [published Online First: 2008/04/25]
59. Werner MU, Kongsgaard UE. I. Defining persistent post-surgical pain: is an update required? *Br J Anaesth* 2014;113(1):1-4. doi: 10.1093/bja/aeu012 [published Online First: 2014/02/21]
60. Purcell S, Cherny SS, Sham PC. Genetic Power Calculator: design of linkage and association genetic mapping studies of complex traits. *Bioinformatics* 2003;19(1):149-50. doi: 10.1093/bioinformatics/19.1.149 [published Online First: 2002/12/25]
61. Lam M, Awasthi S, Watson HJ, et al. RICOPILI: Rapid Imputation for COnsortias PIpeLIne. *Bioinformatics* 2020;36(3):930-33. doi: 10.1093/bioinformatics/btz633 [published Online First: 2019/08/09]
62. Purcell S, Neale B, Todd-Brown K, et al. PLINK: a tool set for whole-genome association and population-based linkage analyses. *Am J Hum Genet* 2007;81(3):559-75. doi: 10.1086/519795 [published Online First: 2007/08/19]
63. van Dijk JF, van Wijck AJ, Kappen TH, et al. Postoperative pain assessment based on numeric ratings is not the same for patients and professionals: a cross-sectional study. *Int J Nurs Stud* 2012;49(1):65-71. doi: 10.1016/j.ijnurstu.2011.07.009 [published Online First: 2011/08/16]
64. Harting B, Johnson T, Abrams R, et al. An exploratory analysis of the correlation of pain scores, patient satisfaction with relief from pain, and a new measure of pain control on the total dose of opioids in pain care. *Qual Manag Health Care* 2013;22(4):322-6. doi: 10.1097/qmh.0000000000000009 [published Online First: 2013/10/04]
65. Raja SN, Carr DB, Cohen M, et al. The revised International Association for the Study of Pain definition of pain: concepts, challenges, and compromises. *Pain* 2020;161(9):1976-82. doi: 10.1097/j.pain.0000000000001939 [published Online First: 2020/07/23]
66. van Boekel RLM, Vissers KCP, van der Sande R, et al. Moving beyond pain scores: Multidimensional pain assessment is essential for adequate pain management after surgery. *PLoS One* 2017;12(5):e0177345. doi: 10.1371/journal.pone.0177345 [published Online First: 2017/05/11]

67. Weldring T, Smith SM. Patient-Reported Outcomes (PROs) and Patient-Reported Outcome Measures (PROMs). *Health Serv Insights* 2013;6:61-8. doi: 10.4137/hsi.S11093 [published Online First: 2013/01/01]
68. Sloman R, Wruble AW, Rosen G, et al. Determination of clinically meaningful levels of pain reduction in patients experiencing acute postoperative pain. *Pain Manag Nurs* 2006;7(4):153-8. doi: 10.1016/j.pmn.2006.09.001 [published Online First: 2006/12/06]
69. Clark CW, Yang JC, Tsui SL, et al. Unidimensional pain rating scales: a multidimensional affect and pain survey (MAPS) analysis of what they really measure. *Pain* 2002;98(3):241-47. doi: 10.1016/s0304-3959(01)00474-2 [published Online First: 2002/07/20]
70. Dorsey SG, Renn CL, Griffioen M, et al. Whole blood transcriptomic profiles can differentiate vulnerability to chronic low back pain. *PLoS One* 2019;14(5):e0216539. doi: 10.1371/journal.pone.0216539 [published Online First: 2019/05/17]
71. Van Der Heijden H, Fatou B, Sibai D, et al. Proteomics based markers of clinical pain severity in juvenile idiopathic arthritis. *Pediatr Rheumatol Online J* 2022;20(1):3. doi: 10.1186/s12969-022-00662-1 [published Online First: 2022/01/17]
72. Jha MK, Song GJ, Lee MG, et al. Metabolic Connection of Inflammatory Pain: Pivotal Role of a Pyruvate Dehydrogenase Kinase-Pyruvate Dehydrogenase-Lactic Acid Axis. *J Neurosci* 2015;35(42):14353-69. doi: 10.1523/jneurosci.1910-15.2015 [published Online First: 2015/10/23]
73. Trevethan R. Sensitivity, Specificity, and Predictive Values: Foundations, Pliabilities, and Pitfalls in Research and Practice. *Front Public Health* 2017;5:307. doi: 10.3389/fpubh.2017.00307 [published Online First: 2017/12/07]
74. Fillingim RB, Loeser JD, Baron R, et al. Assessment of Chronic Pain: Domains, Methods, and Mechanisms. *J Pain* 2016;17(9 Suppl):T10-20. doi: 10.1016/j.jpain.2015.08.010 [published Online First: 2016/09/03]
75. Ioannidis JP. Non-replication and inconsistency in the genome-wide association setting. *Hum Hered* 2007;64(4):203-13. doi: 10.1159/000103512 [published Online First: 2007/06/07]
76. van Reij RRI, Voncken JW, Joosten EAJ, et al. Polygenic risk scores indicates genetic overlap between peripheral pain syndromes and chronic postsurgical pain. *Neurogenetics* 2020;21(3):205-15. doi: 10.1007/s10048-020-00614-5 [published Online First: 2020/05/08]

Figure 1: Pain Predict Genetics study design overview. After written informed consent, participants will be asked to complete questionnaires before and after their surgery. One tube of blood will be collected for DNA isolation using the intravenous line in place for surgery. Clinical information will be collected from the electronic patient file after the operation.

Figure 1: Pain Predict Genetics study design overview.

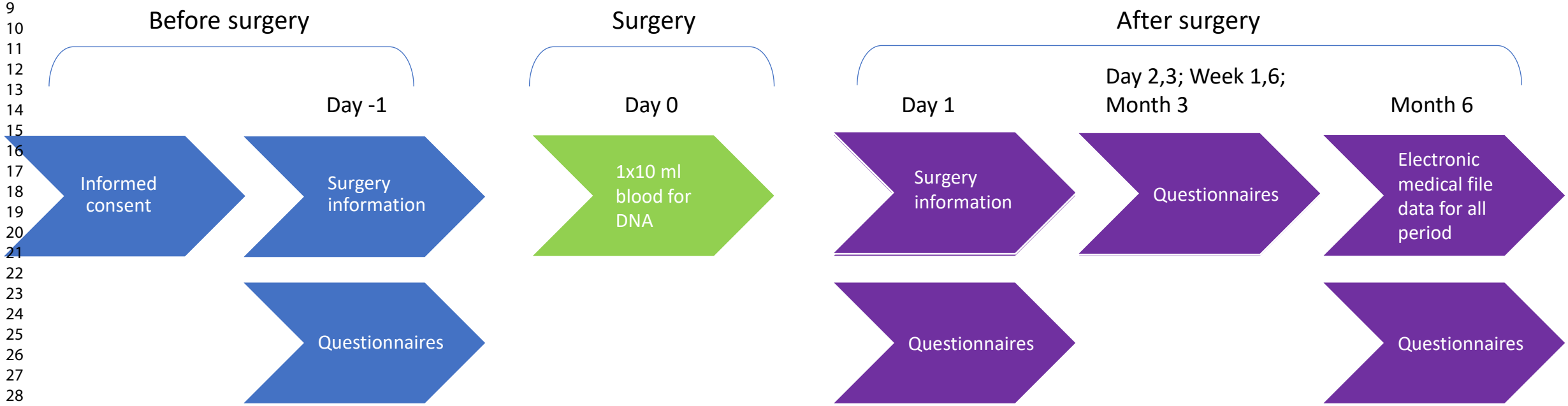


Table of Content

Appendix a: General data

*General data*

- What is your year of birth?
- What is your gender? male/female
- What is your length ? _____ cm
- What is your weight ? _____ kg

- What country were you born in?_____
- What country(ies) were your parents born in?_____
- What country(ies) were your grandparents born in?_____
- What human race are you? (black, white, Asian, etc.)_____

*Data of the surgery:*

*- Would you please describe your surgery*:

| |
|---|
| |

- How much pain do you expect after surgery (0= no pain, 10=worst pain imaginable)
- Will you stay one or more nights in the hospital after surgery? Yes / No

Appendix b: Pain before and after surgery

Pain before and after surgery

Circle how much pain you have, expressed as a number. The pain score means a score between 0 and 10, where 0 means no pain and 10 means the worst pain imaginable. For your pain, consider a figure between 0 and 10. You also tick whether you think the pain is acceptable or not.

| | |
|---|---|
| Pain while being at rest at this moment (0-10) | No pain 0-1-2-3-4-5-6-7-8-9-10 worst pain imaginable |
| Pain score at this moment if you perform a normal effort (0-10) | No pain 0-1-2-3-4-5-6-7-8-9-10 worst pain imaginable |
| Do you think pain is acceptable to you at this moment? | Pain acceptable\|__\| pain not acceptable \|__\| |
| Only pre-operatively: How much pain do you expect after surgery? | No pain 0-1-2-3-4-5-6-7-8-9-10 worst pain imaginable |

Appendix c: Physical activities

Physical activities

Circle the one number below that best describes how much, since your surgery, pain interfered with or prevented you from doing physical activities, expressed by figure. The score means a figure between 0 and 10, where 0 means no interference and 10 means complete interference.

1. How much has pain interfered with or prevented you from doing activities in bed such as turning, sitting up, changing position (0= did not interfere, 10= completely interfered)

0-1-2-3-4-5-6-7-8-9-10

2. How much has pain interfered with or prevented you from breathing deeply of coughing (0= did not interfere, 10= completely interfered)

0-1-2-3-4-5-6-7-8-9-10

3. How much has pain interfered with or prevented you from sleeping (0= did not interfere, 10= completely interfered)

0-1-2-3-4-5-6-7-8-9-10

4. Have you been out of bed since your surgery?

Yes/no

5. If yes, how much did pain interfere or prevent you from doing activities out of bed such as walking, sitting in a chair, standing at the sink (0= did not interfere, 10= completely interfered)

0-1-2-3-4-5-6-7-8-9-10

Appendix d: Pain disability index

Pain disability index

*We would like to know how much pain is preventing you from doing what you would normally do or from doing it as well as you normally would. Respond to each category indicating the overall impact of pain in your life, not just when pain is at its worst.*

*For each of the 7 categories of life activity listed, please circle the number on the scale that describes the level of disability you typically experience. A score of 0 means no disability at all, and a score of 10 signifies that all of the activities in which you would normally be involved have been totally disrupted or prevented by your pain.*

*In case of no pain, please circle "0".*

| | |
|---|---|
| **1. Family/Home Responsibilities**<br><br>This category refers to activities of the home or family. It includes chores or duties performed around the house (e.g. yard work) and errands or favors for other family members (e.g. driving the children to school). | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **2. Recreation**<br><br>This disability includes hobbies, sports, and other similar leisure time activities. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **3. Social activity**<br><br>This category refers to activities, which involve participation with friends and acquaintances other than family members. It includes parties, theater, concerts, dining out, and other social functions. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **4. Occupation**<br><br>This category refers to activities that are part of or directly related to one's job. This includes non-paying jobs as well, such as that of a housewife or volunteer. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **5. Sexual behavior**<br><br>This category refers to the frequency and quality of one's sex life. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |
| **6. Self care** | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |

| | |
|---|---|
| This category includes activities, which involve personal maintenance and independent daily living (e.g. taking a shower, driving, getting dressed, etc.) | |
| **7. Life-support activities**<br><br>This category refers to basic life supporting behaviors such as eating, sleeping and breathing. | No disability 0-1-2-3-4-5-6-7-8-9-10 Worst disability |

Appendix e: Anxiety and need for information

Anxiety and need for information

*Please circle the number on the scale that describes your experience:*

| **The Amsterdam Preoperative Anxiety and Information Scale (APAIS):** | Not at all | | | | Extremely |
|---|---|---|---|---|---|
| I am worried about the anesthetic | 1 | 2 | 3 | 4 | 5 |
| The anesthetic is on my mind continually | 1 | 2 | 3 | 4 | 5 |
| I am worried about the procedure | 1 | 2 | 3 | 4 | 5 |
| The procedure is on my mind continually | 1 | 2 | 3 | 4 | 5 |
| I would like to know as much as possible about the anesthetic | 1 | 2 | 3 | 4 | 5 |
| I would like to know as much as possible about the procedure | 1 | 2 | 3 | 4 | 5 |

Appendix f: Pain Catastrophizing Scale (PCS)

Pain Catastrophizing Scale (PCS)

We are interested in the types of thoughts and feelings that you have when you are in pain. Listed below are thirteen statements describing different thoughts and feelings that may be associated with pain. Using the following scale, please indicate the degree to which you have these thoughts and feelings when you are experiencing pain.

0=not at all 1=to a slight degree 2=to a moderate degree 3=to a great degree 4=all the time

When I'm in pain …….

| | | | | | | |
|---|---|---|---|---|---|---|
| 1. | I worry all the time about whether the pain will end | 0 | 1 | 2 | 3 | 4 |
| 2. | I feel I can't go on | 0 | 1 | 2 | 3 | 4 |
| 3. | It's terrible and I think that it's never going to get any better | 0 | 1 | 2 | 3 | 4 |
| 4. | It's awful and I feel that it overwhelms me | 0 | 1 | 2 | 3 | 4 |
| 5. | I feel that I can't stand it any more | 0 | 1 | 2 | 3 | 4 |
| 6. | I become afraid that the pain will get worse | 0 | 1 | 2 | 3 | 4 |
| 7. | I keep thinking of other painful events | 0 | 1 | 2 | 3 | 4 |
| 8. | I anxiously want the pain to go away | 0 | 1 | 2 | 3 | 4 |
| 9. | I can't seem to keep it out of my mind | 0 | 1 | 2 | 3 | 4 |
| 10. | I keep thinking about how much it hurts | 0 | 1 | 2 | 3 | 4 |
| 11. | I keep thinking about how badly I want the pain to stop | 0 | 1 | 2 | 3 | 4 |
| 12. | There's nothing I can do to reduce the intensity of the pain | 0 | 1 | 2 | 3 | 4 |
| 13. | I wonder whether something serious may happen | 0 | 1 | 2 | 3 | 4 |

Appendix g: Pain Sensitivity Questionnaire

Pain Sensitivity Questionnaire

*This questionnaire contains a series of questions in which you should imagine yourself in certain*
*situations. You should then decide if these situations would be painful for you and if yes, how painful*
*they would be.*

***Let 0 stand for no pain; 1 is an only just noticeable pain arid l0 the most severe pain that you can imagine or consider possible.***

*Please mark the scale with a cross on the number that is most true for you. Keep in mind that there are no "right" or "wrong" answers; only your personal assessment of the situation counts. Please try as much as possible not to allow your fear or aversion of the imagined situations affect your assessment of painfulness.*

1. Imagine you bump your shin badly on a hard edge, for example, on the edge of a glass coffee
   table. How painful would that be for you?
   **0 = not at all painful, 10= most severe pain imaginable**

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

2. Imagine you burn your tongue on a very hot drink.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

3. Imagine your muscles are slightly sore as the result of physical activity.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

4. Imagine you trap your finger in a drawer.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

5. Imagine you take a shower with lukewarm water.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

6. Imagine you have mild sunburn on your shoulders.

0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

7. Imagine you grazed your knee falling off your bicycle.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

8. Imagine you accidentally bite your tongue or cheek badly while eating.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

9. Imagine walking across a cool tiled floor with bare feet.

   0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

10. Imagine you have a minor cut on your finger and inadvertently get lemon juice in the wound.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

11. Imagine you prick your fingertip on the thorn of a rose.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

12. Imagine you stick your bare hands in the snow for a couple of minutes or bring your hands
    in contact with snow for some time, for example, while making snowballs.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

13. Imagine you shake hands with someone who has a normal grip.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

14. Imagine you shake hands with someone who has a very strong grip.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

15. Imagine you pick up a hot pot by inadvertently grabbing its equally hot handles.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

16. Imagine you are wearing sandals and someone with heavy boots steps on your foot.

    0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

17. Imagine you bump your elbow on the edge of a table ("funny bone").

0------1-------2-------3------4-------5-------6-------7-------8-------9-------10

Appendix h: Chronic pain

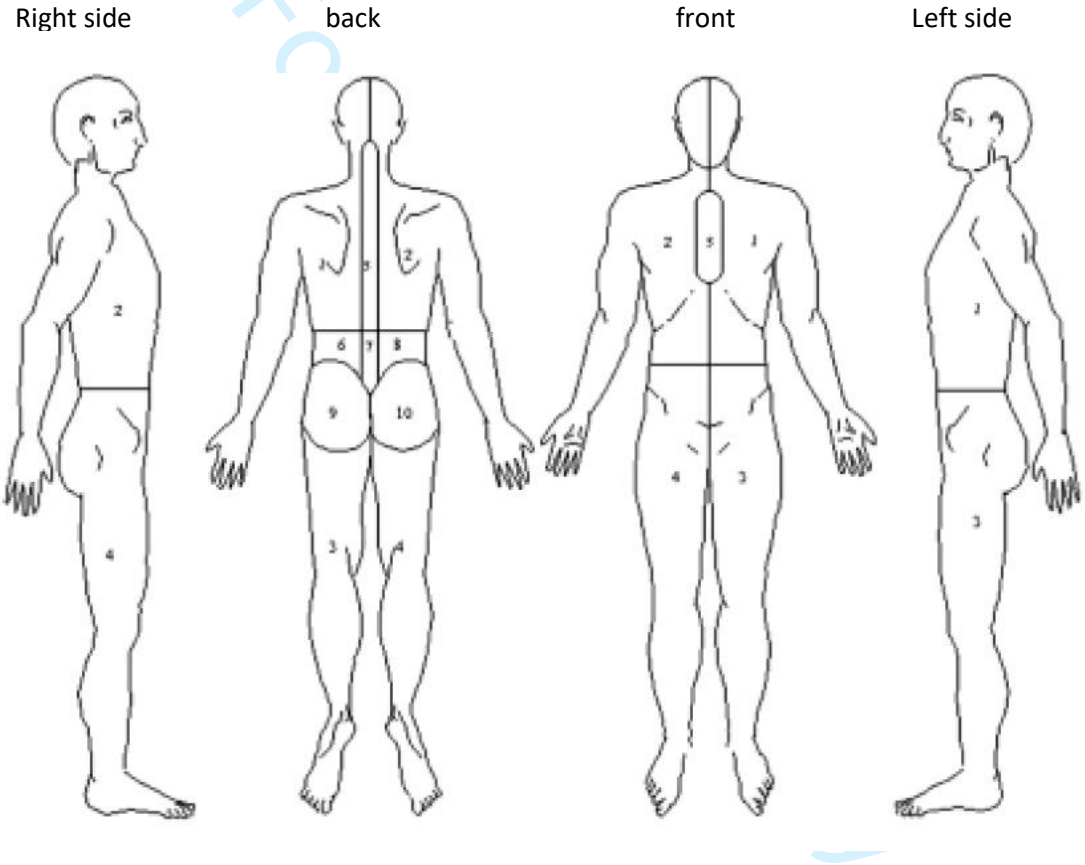
Chronic pain

Did you experience any pain in the last month that lasted for a day or more?

□ Yes, next question

□ No

Can you indicate in the drawings below where you suffer (have suffered) from pain?




Is this the same spot as the spot you are operated on? Yes/no

Does the pain differ from the pain before surgery? Yes/no

How long have you been affected by the above-mentioned pain?

□ Less than three months

□ More than three months

Appendix i: Inventory of depressive symptomatology (self-report) (IDS-SR)

**INVENTORY OF DEPRESSIVE SYMPTOMATOLOGY (SELF-REPORT)**
**(IDS-SR)**

NAME: _____ TODAY'S DATE _____

Please circle the one response to each item that best describes you for the past seven days.

1. Falling Asleep:
   - 0 I never take longer than 30 minutes to fall asleep.
   - 1 I take at least 30 minutes to fall asleep, less than half the time.
   - 2 I take at least 30 minutes to fall asleep, more than half the time.
   - 3 I take more than 60 minutes to fall asleep, more than half the time.

2. Sleep During the Night:
   - 0 I do not wake up at night.
   - 1 I have a restless, light sleep with a few brief awakenings each night.
   - 2 I wake up at least once a night, but I go back to sleep easily.
   - 3 I awaken more than once a night and stay awake for 20 minutes or more, more than half the time.

3. Waking Up Too Early:
   - 0 Most of the time, I awaken no more than 30 minutes before I need to get up.
   - 1 More than half the time, I awaken more than 30 minutes before I need to get up.
   - 2 I almost always awaken at least one hour or so before I need to, but I go back to sleep eventually.
   - 3 I awaken at least one hour before I need to, and can't go back to sleep.

4. Sleeping Too Much:
   - 0 I sleep no longer than 7-8 hours/night, without napping during the day.
   - 1 I sleep no longer than 10 hours in a 24-hour period including naps.
   - 2 I sleep no longer than 12 hours in a 24-hour period including naps.
   - 3 I sleep longer than 12 hours in a 24-hour period including naps.

5. Feeling Sad:
   - 0 I do not feel sad
   - 1 I feel sad less than half the time.
   - 2 I feel sad more than half the time.
   - 3 I feel sad nearly all of the time.

6. Feeling Irritable:
   - 0 I do not feel irritable.
   - 1 I feel irritable less than half the time.
   - 2 I feel irritable more than half the time.
   - 3 I feel extremely irritable nearly all of the time.

7. Feeling Anxious or Tense:
   - 0 I do not feel anxious or tense.
   - 1 I feel anxious (tense) less than half the time.
   - 2 I feel anxious (tense) more than half the time.
   - 3 I feel extremely anxious (tense) nearly all of the time.

8. Response of Your Mood to Good or Desired Events:
   - 0 My mood brightens to a normal level which lasts for several hours when good events occur.
   - 1 My mood brightens but I do not feel like my normal self when good events occur.
   - 2 My mood brightens only somewhat to a rather limited range of desired events.
   - 3 My mood does not brighten at all, even when very good or desired events occur in my life.

9. Mood in Relation to the Time of Day:
   - 0 There is no regular relationship between my mood and the time of day.
   - 1 My mood often relates to the time of day because of environmental events (e.g., being alone, working).
   - 2 In general, my mood is more related to the time of day than to environmental events.
   - 3 My mood is clearly and predictably better or worse at a particular time each day.

   9A. Is your mood typically worse in the morning, afternoon or night? (circle one)

   9B. Is your mood variation attributed to the environment? (yes or no) (circle one)

10. The Quality of Your Mood:
    - 0 The mood (internal feelings) that I experience is very much a normal mood.
    - 1 My mood is sad, but this sadness is pretty much like the sad mood I would feel if someone close to me died or left.
    - 2 My mood is sad, but this sadness has a rather different quality to it than the sadness I would feel if someone close to me died or left.
    - 3 My mood is sad, but this sadness is different from the type of sadness associated with grief or loss.

**Please complete either 11 or 12 (not both)**

11. Decreased Appetite:

0 There is no change in my usual appetite.
1 I eat somewhat less often or lesser amounts of food than usual.
2 I eat much less than usual and only with personal effort.
3 I rarely eat within a 24-hour period, and only with extreme personal effort or when others persuade me to eat.

12. Increased Appetite:

0 There is no change from my usual appetite.
1 I feel a need to eat more frequently than usual.
2 I regularly eat more often and/or greater amounts of food than usual.
3 I feel driven to overeat both at mealtime and between meals.

**Please complete either 13 or 14 (not both)**

13. Decreased Weight (Within the Last Two Weeks):

0 I have not had a change in my weight.
1 I feel as if I've had a slight weight loss.
2 I have lost 2 pounds or more.
3 I have lost 5 pounds or more.

14. Increased Weight (Within the Last Two Weeks):

0 I have not had a change in my weight.
1 I feel as if I've had a slight weight gain.
2 I have gained 2 pounds or more.
3 I have gained 5 pounds or more.

15. Concentration/Decision Making:

0 There is no change in my usual capacity to concentrate or make decisions.
1 I occasionally feel indecisive or find that my attention wanders.
2 Most of the time, I struggle to focus my attention or to make decisions.
3 I cannot concentrate well enough to read or cannot make even minor decisions.

16. View of Myself:

0 I see myself as equally worthwhile and deserving as other people.
1 I am more self-blaming than usual.
2 I largely believe that I cause problems for others.
3 I think almost constantly about major and minor defects in myself.

17. View of My Future:

0 I have an optimistic view of my future.
1 I am occasionally pessimistic about my future, but for the most part I believe things will get better.
2 I'm pretty certain that my immediate future (1-2 months) does not hold much promise of good things for me.
3 I see no hope of anything good happening to me anytime in the future.

18. Thoughts of Death or Suicide:

0 I do not think of suicide or death.
1 I feel that life is empty or wonder if it's worth living.
2 I think of suicide or death several times a week for several minutes.
3 I think of suicide or death several times a day in some detail, or I have made specific plans for suicide or have actually tried to take my life.

19. General Interest:

0 There is no change from usual in how interested I am in other people or activities.
1 I notice that I am less interested in people or activities.
2 I find I have interest in only one or two of my formerly pursued activities.
3 I have virtually no interest in formerly pursued activities.

20. Energy Level:

0 There is no change in my usual level of energy.
1 I get tired more easily than usual.
2 I have to make a big effort to start or finish my usual daily activities (for example, shopping, homework, cooking or going to work).
3 I really cannot carry out most of my usual daily activities because I just don't have the energy.

21. Capacity for Pleasure or Enjoyment (excluding sex):

0 I enjoy pleasurable activities just as much as usual.
1 I do not feel my usual sense of enjoyment from pleasurable activities.
2 I rarely get a feeling of pleasure from any activity.
3 I am unable to get any pleasure or enjoyment from anything.

22. Interest in Sex (Please Rate Interest, not Activity):

0 I'm just as interested in sex as usual.
1 My interest in sex is somewhat less than usual or I do not get the same pleasure from sex as I used to.
2 I have little desire for or rarely derive pleasure from sex.
3 I have absolutely no interest in or derive no pleasure from sex.

23. Feeling slowed down:

0 I think, speak, and move at my usual rate of speed.
1 I find that my thinking is slowed down or my voice sounds dull or flat.
2 It takes me several seconds to respond to most questions and I'm sure my thinking is slowed.
3 I am often unable to respond to questions without extreme effort.

24. Feeling restless:

0 I do not feel restless.
1 I'm often fidgety, wring my hands, or need to shift how I am sitting.
2 I have impulses to move about and am quite restless.
3 At times, I am unable to stay seated and need to pace around.

25. Aches and pains:

0 I don't have any feeling of heaviness in my arms or legs and don't have any aches or pains.
1 Sometimes I get headaches or pains in my stomach, back or joints but these pains are only sometime present and they don't stop me from doing what I need to do.
2 I have these sorts of pains most of the time.
3 These pains are so bad they force me to stop what I am doing.

26. Other bodily symptoms:

0 I don't have any of these symptoms: heart pounding fast, blurred vision, sweating, hot and cold flashes, chest pain, heart turning over in my chest, ringing in my ears, or shaking.
1 I have some of these symptoms but they are mild and are present only sometimes.
2 I have several of these symptoms and they bother me quite a bit.
3 I have several of these symptoms and when they occur I have to stop doing whatever I am doing.

Range 0-84 Score: _____

27. Panic/Phobic symptoms:

0 I have no spells of panic or specific fears (phobia) (such as animals or heights).
1 I have mild panic episodes or fears that do not usually change my behavior or stop me from functioning.
2 I have significant panic episodes or fears that force me to change my behavior but do not stop me from functioning.
3 I have panic episodes at least once a week or severe fears that stop me from carrying on my daily activities.

28. Constipation/diarrhea:

0 There is no change in my usual bowel habits.
1 I have intermittent constipation or diarrhea which is mild.
2 I have diarrhea or constipation most of the time but it does not interfere with my day-to-day functioning.
3 I have constipation or diarrhea for which I take medicine or which interferes with my day-to-day activities.

29. Interpersonal Sensitivity:

0 I have not felt easily rejected, slighted, criticized or hurt by others at all.
1 I have occasionally felt rejected, slighted, criticized or hurt by others.
2 I have often felt rejected, slighted, criticized or hurt by others, but these feelings have had only slight effects on my relationships or work.
3 I have often felt rejected, slighted, criticized or hurt by others and these feelings have impaired my relationships and work.

30. Leaden Paralysis/Physical Energy:

0 I have not experienced the physical sensation of feeling weighted down and without physical energy.
1 I have occasionally experienced periods of feeling physically weighted down and without physical energy, but without a negative effect on work, school, or activity level.
2 I feel physically weighted down (without physical energy) more than half the time.
3 I feel physically weighted down (without physical energy) most of the time, several hours per day, several days per week.

Appendix j: Brief Pain Inventory

Date: _____ /_____ /_____ Time: _____

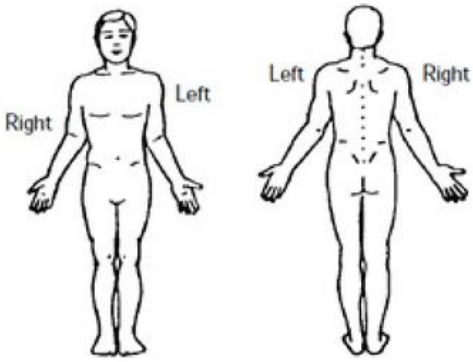
Name: _____ _____ _____

Last First Middle intial

1) Throughout our lives, most of us have had pain from time to time (such as minor headaches, sprains, and toothaches). Have you had pain other than these everyday kinds of pain today?

1. Yes 2. No

2) On the diagram, shade in the areas where you feel pain. Put an X on the area that hurts the most.




3) Please rate your pain by circling the one number that best describes your pain at its **worst** in the past 24 hours.

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| No pain | | | | | | | | | | Pain as bad as you can imagine |

4) Please rate your pain by circling the one number that best describes your pain at its **least** in the past 24 hours.

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| No pain | | | | | | | | | | Pain as bad as you can imagine |

5) Please rate your pain by circling the one number that best describes your pain on **average**.

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| No pain | | | | | | | | | | Pain as bad as you can imagine |

6) Please rate your pain by circling the one number that tells how much pain you have **right now**.

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| No pain | | | | | | | | | | Pain as bad as you can imagine |

7) What treatments or medications are you receiving for your pain?

_____

_____

8) In the past 24 hours, how much **relief** have pain treatments or medications provided? Please circle the one percentage that most shows how much relief you have received.

| 0% | 10 | 20 | 30 | 40 | 50 | 60 | 70 | 80 | 90 | 100% |
|---|---|---|---|---|---|---|---|---|---|---|
| No relief | | | | | | | | | | Complete relief |

9) Circle the one number that describes how, during the past 24 hours, pain has **interfered** with your:

A. General activity

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

B. Mood

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

C. Walking ability

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

D. Normal work (includes both work outside the home and housework)

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

E. Relations with other people

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

F. Sleep

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

G. Enjoyment of life

| 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Does not interfere | | | | | | | | | | Completely interferes |

# Reporting checklist for genetic association study.

Based on the STREGA guidelines.

## Instructions to authors

Complete this checklist by entering the page numbers from your manuscript where readers will find each of the items listed below.

Your article may not currently address all the items on the checklist. Please modify your text to include the missing information. If you are certain that an item does not apply, please write "n/a" and provide a short explanation.

Upload your completed checklist as an extra file when you submit to a journal.

In your methods section, say that you used the STREGAreporting guidelines, and cite them as:

Little J, Higgins JP, Ioannidis JP, Moher D, Gagnon F, von Elm E, Khoury MJ, Cohen B, Davey-Smith G, Grimshaw J, Scheet P, Gwinn M, Williamson RE, Zou GY, Hutchings K, Johnson CY, Tait V, Wiens M, Golding J, van Duijn C, McLaughlin J, Paterson A, Wells G, Fortier I, Freedman M, Zecevic M, King R, Infante-Rivard C, Stewart A, Birkett N; STrengthening the REporting of Genetic Association Studies. STrengthening the REporting of Genetic Association Studies (STREGA): An Extension of the STROBE Statement.

| | | Reporting Item | Page Number |
|---|---|---|---|
| **Title and abstract** | | | |
| Title | #1a | Indicate the study's design with a commonly used term in the title or the abstract | 1 |

| | | | |
|---|---|---|---|
| Abstract | #1b | Provide in the abstract an informative and balanced summary of what was done and what was found | 3 |
| **Background/rationale** | | | |
| | #2 | Explain the scientific background and rationale for the investigation being reported | 6 |
| **Objectives** | | | |
| | #3 | State specific objectives, including any prespecified hypotheses. State if the study is the first report of a genetic association, a replication effort, or both. | 9 |
| **Study design** | | | |
| | #4 | Present key elements of study design early in the paper | 10 |
| **Setting** | | | |
| | #5 | Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection | 10 |
| **Eligibility criteria** | | | |
| | #6a | Cohort study – Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up. Case-control | 11 |

| | | | |
|---|---|---|---|
| | | study – Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls. Cross-sectional study – Give the eligibility criteria, and the sources and methods of selection of participants. Give information on the criteria and methods for selection of subsets of participants from a larger study, when relevant. | |
| | #6b | Cohort study – For matched studies, give matching criteria and number of exposed and unexposed. Case-control study – For matched studies, give matching criteria and the number of controls per case. | n/a, not matched study |
| **Variables** | | | |
| | #7a | Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable | 11, 14-17 |
| | #7b | Clearly define genetic exposures (genetic variants) using a widely-used nomenclature system. Identify variables likely to be associated with population stratification (confounding by ethnic origin). | 18 |
| **Data** | | | |

| | | | |
|---|---|---|---|
| sources/measurement | | | |
| | #8a | For each variable of interest give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group. Give information separately for for exposed and unexposed groups if applicable. | 11, 14-16 |
| | #8b | Describe laboratory methods, including source and storage of DNA, genotyping methods and platforms (including the allele calling algorithm used, and its version), error rates and call rates. State the laboratory / centre where genotyping was done. Describe comparability of laboratory methods if there is more than one group. Specify whether genotypes were assigned using all of the data from the study simultaneously or in smaller batches. | 16 |
| Bias | | | |
| | #9a | Describe any efforts to address potential sources of bias | 22 |
| | #9b | Describe any efforts to address potential sources of bias | 22 |
| Study size | | | |

| | | |
|---|---|---|
| | #10 Explain how the study size was arrived at | 17 |
| **Quantitative variables** | | |
| | #11 Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen, and why. If applicable, describe how effects of treatment were dealt with. | 18 |
| **Statistical methods** | | |
| | #12a Describe all statistical methods, including those used to control for confounding. State software version used and options (or settings) chosen. | 18-19 |
| | #12b Describe any methods used to examine subgroups and interactions | n/a to protocol paper. |
| | #12c Explain how missing data were addressed | n/a to protocol paper. |
| | #12d If applicable, explain how loss to follow-up was addressed | n/a to protocol paper. |
| | #12e Describe any sensitivity analyses | n/a to protocol paper. |
| | #12f State whether Hardy-Weinberg equilibrium was considered and, if so, how. | 18 |
| | #12g Describe any methods used for inferring genotypes or haplotypes | 18 |

| | | | |
|---|---|---|---|
| | #12h | Describe any methods used to assess or address population stratification. | 18 |
| | #12i | Describe any methods used to address multiple comparisons or to control risk of false positive findings. | 18 |
| | #12j | Describe any methods used to address and correct for relatedness among subjects | 18 |
| **Participants** | | | |
| | #13a | Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed. Give information separately for for exposed and unexposed groups if applicable. Report numbers of individuals in whom genotyping was attempted and numbers of individuals in whom genotyping was successful. | n/a to protocol paper. Participant enrollment is not finished yet |
| | #13b | Give reasons for non-participation at each stage | n/a to protocol paper. Participant enrollment is not finished yet |
| | #13c | Consider use of a flow diagram | See figure 1 |

| | | |
|---|---|---|
| **Descriptive data** | | |
| #14a | Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders. Give information separately for exposed and unexposed groups if applicable. Consider giving information by genotype | n/a to protocol paper. Participant enrollment is not finished yet |
| #14b | Indicate number of participants with missing data for each variable of interest | |
| #14c | Cohort study – Summarize follow-up time, e.g. average and total amount. | |
| **Outcome data** | | |
| #15 | Cohort study Report numbers of outcome events or summary measures over time.Give information separately for exposed and unexposed groups if applicable. Report outcomes (phenotypes) for each genotype category over time Case-control study – Report numbers in each exposure category, or summary measures of exposure.Give information separately for cases and controls . Report numbers in each genotype category. Cross-sectional study – Report numbers of outcome events or summary measures. Give information separately for exposed and | n/a to protocol paper. Participant enrollment is not finished yet |

| | | | |
|---|---|---|---|
| | | unexposed groups if applicable. Report outcomes (phenotypes) for each genotype category | |
| **Main results** | | | |
| | #16a | Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included | n/a to protocol paper. Participant enrollment is not finished yet |
| | #16b | Report category boundaries when continuous variables were categorized | |
| | #16c | If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period | |
| | #16d | Report results of any adjustments for multiple comparisons | |
| **Other analyses** | | | |
| | #17a | Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity analyses | n/a to protocol paper. Participant enrollment is not finished yet |
| | #17b | Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity | |

| | | | |
|---|---|---|---|
| | | analyses | |
| | #17c | Report other analyses done—e.g., analyses of subgroups and interactions, and sensitivity analyses | |
| **Key results** | | | |
| | #18 | Summarise key results with reference to study objectives | n/a to protocol paper. Participant enrollment is not finished yet |
| **Limitations** | | | |
| | #19 | Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias. | 21 |
| **Interpretation** | | | |
| | #20 | Give a cautious overall interpretation considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence. | 22 |
| **Generalisability** | | | |
| | #21 | Discuss the generalisability (external validity) of the study results | 22 |

## Funding

| | | |
|---|---|---|
| #22 | Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based | 23 |