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The Swedish chronic Pain Biobank - protocol for a multicenter registry and biomarker project

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Abstract

Introduction: About 20% of the adult population has chronic pain, often associated with psychological distress, sick leave, and poor health. There are large variations in the clinical picture. A biopsychosocial approach is used in investigation and treatment. The concept of personalized medicine, i.e., optimizing medication types and dosages for individual patients based on biomarkers and other patient-related factors, has received increasing attention in different diseases but used less in chronic pain. This cooperative project from all Swedish Universities investigates whether there are changes in protein, inflammation and metabolism patterns in saliva and blood in chronic pain patients and whether the changes correlate with clinical characteristics and rehabilitation outcomes.

Methods and analysis: Patients at multidisciplinary pain centers at University Hospitals in Sweden who have chosen to participate in the Swedish Quality Registry for Pain Rehabilitation and healthy gender- and age-matched individuals will be included in the project. Saliva and plasma samples will be collected in addition to questionnaire data obtained from the Register. From the samples, protein, inflammation, and metabolism patterns are analyzed in relation to, for example, diagnosis, pain characteristics, psychological distress, body weight, pharmacological treatment, and clinical rehabilitation results.

Ethics and Dissemination: The study is approved by the Swedish Ethical Review Authority (Dnr 2021-04929) and will be conducted in accordance with the declaration of Helsinki. The results will be published in open access scientific journals and in popular scientific relevant journals such as those from patient organizations. Data will be also presented in scientific meetings, meeting with healthcare organization and disseminated in different lecturers at the clinic and university.

Keywords: Biomarkers, Chronic pain, Inflammation, Metabolomics, Molecular profile, Proteomics

Strengths and limitations of this study

- This project will play a crucial part in the paradigm shift occurring and focusing on a personalized individual treatment method in chronic pain.

- The research group has broad and adequately high competence for the implementation.
- The participating university clinics have working routines for collecting data and routines for recruiting participants.
- Extensive experience of the clinical, biochemical, and statistical research is available in the research group.
- There is a risk of heterogeneity of the samples as this study is a multicenter study in a large geographical area where the infrastructure for data collection is different.
- Individuals not able to read and write Swedish are excluded which make the results less generalizable to the whole population.

1. Introduction

About 20% of the adult population lives with at least moderate-severe chronic pain; often with concomitant complex psychological distress including depression and anxiety. Pain will also reduce patients' ability to study or work full time, they will show higher levels of sick leave, and show reduced quality of life [2]. There are large variations in the clinical picture in chronic pain patients e.g., regarding psychiatric comorbidities; about 40% of patients managed at specialist level have co-morbid symptoms of depression and/or anxiety [3]. More than 20% of Years lived with disabilities – a measure of non-fatal disabilities - are caused by of pain conditions both in Sweden and globally [38].

Acute tissue damage initiates plastic and reinforcing mechanisms in the pain system peripherally and centrally, which in interaction with psychological and social factors create the experience of pain at a given moment. Chronic pain and in parallel depression constitute a large part of the pain complex, why depression also must be considered when diagnosing the patient. Depression in turn intensifies the pain by pain facilitating pathways. Social factors such as working conditions can be risk factors for the chronification of pain, and when chronic pain has developed return to work will be more difficult (i.e., chronic pain has social consequences). Modern pain care is therefore based on a bio-psycho-social approach to diagnosis and treatment [9; 12].

Chronic pain is not an acute pain that persists over time, but further plastic neurobiological changes occur in interaction with psychological and social factors. Imaging techniques have provided an in-depth understanding of how the brain processes and creates the experience of pain. Chronic pain is associated with chemical changes in structures in the brain that process pain, changes in the physiological interactions between these structures and in the descending control of nociception, and central hyperexcitability (central sensitization) [7; 23].

Chronic widespread pain (CWP) including fibromyalgia (FM) is the extreme of complex pain condition. FM is characterized by altered nociception and is a prototype of a nociplastic pain condition, however, it is well known that patients initially suffering from nociceptive pain can in time develop nociplastic pain conditions [22]. It has been argued that CWP / FM is a typical example of a central pain condition, i.e., peripheral factors

have little or no role. Others believe that peripheral factors initiate and perpetuate the central changes e.g., which is the case in coxarthrosis where central changes are normalized after hip replacement [32].

In various studies, we have shown an increased presence of pain-mediating (e.g., glutamate, serotonin), metabolic (e.g., lactate and pyruvate) and analgesic (e.g., N-acylethanolamines (NAE)) substances in muscle in neck-shoulder pain and in CWP / FM [16]. Other researchers have recently shown changes in the peripheral nociceptors of FM [44]. Proteomic studies are increasingly being performed in the field of pain medicine. Using targeted and untargeted proteomics, our research group has found significant differences in the protein/inflammation pattern in muscles and plasma in CWP / FM [17; 26; 42]. Such changes also correlate with, for example, pain intensity, pain sensitivity and psychological strain [17; 27]. Taken together, this provides support for peripheral factors that significantly contribute to the maintenance of CWP / FM.

In the literature it is suggested that low-grade peripheral or systemic inflammation perpetuates chronic pain and is also involved in psychological / psychiatric conditions such as depression and obesity [11; 39]. Various pro-inflammatory cytokines and chemokines have been studied. However, the literature is not consistent; in a large FM study, we found no changes in “classic” pro-inflammatory cytokines in muscle or plasma [10]. Two recent systematic reviews of peripheral cytokines and chemokines - mainly based on single or few molecules - have not been consistent [1; 25]. Such hypothesis-driven studies have generally focused on a few molecules, while there are only a few exploratory studies of a larger number of molecules (panels) is few. Low-grade inflammation/neuroinflammation has also been studied in neuropathic pain conditions [5; 6; 19].

Preliminary biomarker data available in the literature need to be confirmed in larger studies that reflect the normal flow of chronic pain patients where/in which comparisons are made between pain diagnoses and that include control for gender, pain characteristics, co-morbidity, body weight, and pharmacological therapy. The inflammation pattern according to “classic” inflammation markers seems to be of preliminary importance for the treatment outcome in depression [30] and obesity, as well as for psychological treatment, such as cognitive behavioral therapy (CBT) in chronic

pain [24]. Physical exercise provides anti-inflammatory effects [21; 36]. Interdisciplinary Pain Rehabilitation Programs (IPRP), in which psychological treatment (including CBT) and physical activity and exercise are important components, constitute evidence-based treatment for chronic pain but with small to moderate effect sizes [4; 20; 28; 33]. It has not yet been considered if activated biological mechanisms may impact on the effect of IPRP. It is therefore important to investigate whether IPRP result is related to protein, inflammation, and metabolism patterns.

Aim and hypotheses

The aim of this multicenter project is to investigate whether there are differences in protein, inflammation and metabolism patterns between common chronic pain diagnoses and age- and gender-matched healthy controls from the general Swedish population. Further aims are to characterize and compare the changes of the levels of protein, inflammation, and metabolism in different diagnostic groups of chronic pain patients regarding e.g., gender, diagnosis, pain, pain sensitivity, co-morbidity pattern, body weight, pharmacological therapy, and clinical rehabilitation outcomes.

The hypotheses are:

A) there are unique biomarker signatures in plasma and saliva that distinguish common chronic pain diagnoses from each other and from healthy controls.

B) there are significant correlations between the identified biomarker profiles and pain characteristics e.g., intensity, pain sensitivity and anatomical spread of pain, co-morbidity patterns, body weight and pharmacological therapy, and clinical rehabilitation outcomes.

2. Methods

2.1. Participants

This multicenter project includes patients (> 18 years) with chronic pain who are recruited from the *Swedish Quality Registry for Pain Rehabilitation* (SQRP) and healthy gender- and age-matched controls from university hospital at Linkoping, Stockholm, Uppsala, Umea, Gothenburg, and Lund.

2.2. Patients with chronic pain

The project mainly uses patient-reported data from SQRP (www.ucr.uu.se/nrs/) for the participating university clinics. The project leader (Björn Gerdle) is the research leader for the national research group for SQRP and a member of its steering group.

SQRP is based on *Patient Reported Outcome Measures* (PROM) in the form of a written questionnaire. Validated instruments are used in the survey and collectively capture the various facets of the International Classification of Functional Conditions, Disabilities and Health (ICF). The survey includes background variables (*age, gender, country of origin, education and healthcare consumption, weight, and height*) and pain characteristics, degree of FM-ness, psychological distress, difficulty sleeping, pain coping, fear of movement, consequences of chronic pain concerning activity level and function, physical activity level, as well as health-related quality of life and perceived health (Table 1). The response rate is> 90% in SQRP, which can be interpreted as patients considering it important to be able to share how they experience their situation. The current diagnoses according to ICD-10 are also registered in SQRP.

Table 1. Patient reported outcome measures to be used in the project. NRS = Numeric Rating Scale, MPI = Multidimensional Pain Inventory, ACR = American College of Rheumatology, HADS = Hospital Anxiety and Depression Scale, PCS = Pain Catastrophizing Scale, ISI = insomnia Severity Index, CPAQ = Chronic Pain Acceptance Questionnaire, TSK = Tampa Scale for Kinesiophobia, RAND-36 = modernized version of Short-Form Health Survey-36.

Survey	Instrument	Properties
Pain characteristics		
Intensity	NRS (0-10), MPI part 1	0 = no pain, 10 = worst pain experienced
Anatomical extent	Pain drawing?	Body map front and back side
Frequency	Scale?	0 = never, 1 =, 2 =, 3 =?
Duration		Years
Fibromyalgia		
	ACR 2016 criteria	Widespread pain index (WPI): 0-19 sites

		Symptom severity score (SSS): 0-12 FMness score: WPI + SSS.
Psychological distress		
Depression	HADS (0-21)	7 items, each scored 0-3 according to frequency a summed
Anxiety	HADS (0-21)	7 items, each scored 0-3 according to frequency and summed
	MPI part 1 (0-6)	
	Life control?	2 items scored 0-6
	Affective distress?	3 items scored 0-6
		The mean of each subscale is calculated
Pain catastrophizing	PCS (0-52)	13 items, each scored 0-4 according to frequency and summed
Difficulty sleeping		
	ISI (0-28)	7 items, each scored 0-4 according to frequency and summed
Pain coping		
	CPAQ (0-156?)	20 items, each scored 0-3 according to how often statement is true and summed
Fear of movement		
	TSK	17 items, each scored 0-4 according to agreement with statement and summed
Consequences chronic pain		
Activity level	MPI part 1 and 3 (0-6)	18 items each scored 0-6. The mean for each subscale or the total mean is calculated
Function	MPI part 1 and 3 (0-6)	20 items, each scored 0-6. The mean for each subscale or the total mean is calculated
Health-related quality of life		
	RAND-36 (0-100)	36 items in 8 subcategories scored varying between 0 and 100% (with fixed intervals). The mean for each subscale is calculated
	EQ-5D™	Consists of two parts. The first part is an index obtained from five dimensions items scored 0-5. The second part is self-estimation of today's health according to a 100-point thermometer-like scale (EQ5D-VAS) with defined end points (high values indicate better health and low values indicate worse health)
Physical activity		
Sedentary behavior		Minutes per week
Low intensity		
Moderate intensity		
Vigorous intensity		

The patients who are referred to the clinics respond to the SQRP questionnaire in connection with clinical assessment. Patients participating in IPRP also answer questionnaires immediately before, after and 12 months after rehabilitation.

All patients who respond to SQRP at the relevant university clinics are asked to participate in the project and may submit saliva samples and blood samples (see below).

In this project we will study the following 11 conditions/diagnoses (based on the manual produced within SQRP by, among others, the project leader):

1. Chronic neck-shoulder pain (cervicalgia, trapezius myalgia)
2. Chronic low back pain without sciatica
3. Generalized pain (CWP) according to criteria ACR 2016 criteria.
4. Fibromyalgia according to the ACR 2016 criteria.
5. Neuropathic pain including sciatica
6. Chronic Whiplash Disorders (WAD)
7. Ehlers-Danlos Syndrome (EDS) or Hypermobility Syndrome (HMS)
8. Complex Regional Pain Syndrome (CRPS)
9. Pain post-COVID infection
10. Chronic pain, not elsewhere classified (ICD R52.1, R52.2 and R52.9)

In addition, we will investigate the patterns when several pain diagnoses are present simultaneously.

2.3. Healthy controls

Age- and gender-matched healthy control persons are recruited via advertisements and answer a customized version of the SQRP questionnaire. They donate saliva samples and blood samples. Half of the healthy controls are asked to submit new samples after about 10 weeks.

2.4. Exclusion criteria for patients and controls

Coagulation disorders with predisposition to bleeding, medication with anticoagulants (low-dose aspirin is permitted), hypersensitivity to anesthetic, serious psychiatric disease (investigator's judgement, e.g., psychosis or suicidal ideation), and difficulties in understanding the Swedish language.

2.5. Supplementary survey

In addition to the instruments included in the SQRP (or equivalent adapted for healthy controls), the participants also answer a brief questionnaire in close connection with the tissue sampling. This charts the time of sampling, fasting or not, current medication and health food preparations (e.g., curcumin), pain intensity, and present and previous illnesses.

2.6. Biofluids sampling

Sampling consists of saliva and blood samples (plasma) drawn in the morning and during fasting. Saliva samples are taken with Salivette® (Sarstedt AG & Co, PO Box 1220, D-51582, Nümbrecht, Germany, obtained from VWR, article number: 101093–968). The participants avoid brushing teeth or drinking (water is ok) at least one hour before sampling. They rinse their mouth with water and wait for 15 minutes before placing the swab in the mouth. After 3 minutes, they spit the swab into the salivette tube containing a protease cocktail inhibitor. The salivette will be centrifuged (2 minutes, 1,000 x g) and the supernatant is transferred to a new tube and aliquoted into 200 µl in 0.6 ml eppendorf tubes and stored at -86 ° C until analysis. The total volume of saliva will be recorded.

Blood samples (2 x 8.5 ml) are collected in P100 tubes (article number: 366448, BD Diagnostics System, Frankling Lakes, NJ, USA) according to the manufacture's recommendations. The sample is centrifuged at 2500 g for 20 minutes at room temperature within 2-4 hours. Plasma will be extracted by carefully removing the upper part of the supernatant about 2/3 in fractions to a 10 ml tube on ice. The plasma samples will be aliquoted into 200 µl in 0.6 ml eppendorf tubes and stored at -80 ° C until analysis .

The samples are handled according to previously developed methodology [26; 42].

2.7. Biochemical analyses

The analyzes that will be used in this study are mainly exploratory, i.e., which proteins, metabolites and lipoproteins will be identified cannot be determined in advance. The following analyzes will be performed according to the methodology previously described by us [17; 19; 42].

- Exploratory analyzes using panels for inflammation, cytokines & chemokines and neuroinflammation comprising many proteins (from Olink Bioscience, Uppsala [17] and from Meso Scale Discovery [19].
- Exploratory proteomics analysis using 2D gel electrophoresis and/or shotgun proteomics [26; 43]
- Exploratory Metabolomic and Lipoprotein profile including free fatty acids (FFAs)

- Anti-nociceptive substances, i.e., endocannabinoids, NAE, endorphins, alpha-amylase, and cortisol.
- Metabolites such as lactate, pyruvate, glutamate, and other single pain-mediating molecules such as substance P, bradykinin, serotonin and BDNF.
- Pending analysis, the samples are stored in the biobank facilities with which the participating university clinics are associated.

2.8. Data from SQRP

The surveys in SQRP, including the supplementary questionnaire, will include a careful characterization of the patients and the healthy controls. In the analyzes against the saliva and blood samples, the following variables will initially be used:

- Diagnosis (from manual produced within SQRP)
- Pain intensity
- Widespreadness of pain/Pain localization
- Pain duration
- Psychologic distress (depression, anxiety, pain catastrophizing, fear of movement and insomnia)
- Sex
- Body mass index (BMI)
- Physical activity level
- Pharmacological therapy and health food preparations

2.9. Interdisciplinary multi modal rehabilitation program (IPRP)

The inclusion criteria are decided by the clinical examination and screening questionnaires concerning both anthropometric measures as well as subjective health followed by an inter professional conference between the MD, a psychologist, a physiotherapist, and an occupational therapist – choosing which patients they believe would benefit from the IPRP, in accordance with recommendations from the Swedish Agency for Health Technology Assessments and Assessment of Social Services. The patient should be motivated and have the potential for an active change to be included in IPRP. All patients will participate in a specific individual exercise with a physiotherapist (with both relaxation therapy and strengthening specific regions of the body to unburden), individual- and group therapy led

by a clinical psychologist (cognitive behavioral therapy, education, coping and mindfulness) as well as environmental work changes from an occupational therapist, working with education and return to work strategies [34] in group of six to nine persons, commonly 20 h per week of group-based activities for 6 to 8 weeks [14]. Pain education (including lectures in basic pain science and pain management) will be offered both for patients as well as for their relatives, friends, and colleagues.

2.10. Patient and Public Involvement

Patients and the public were not involved in this research study.

2.11. Statistics

In addition to descriptive statistics, advanced multivariate statistics (MVDA [18]; i.e., advanced principal component analysis (PCA) and OPLS regressions) will be used according to the guidelines presented by Wheelock & Wheelock for omics data [40] using SIMCA®-P + (version17 .0, Sartorius AG, Göttingen Germany). The methods are necessary to manage and take advantage of the intercorrelation pattern (i.e., multicollinearity) between the identified proteins/substances. MVDA enables analyzes where the number of variables is significantly greater than the number of observations (i.e., short and broad data tables), which more traditional multivariate regressions cannot easily handle. The research group has a long experience of the methods and has used these in previous studies and has an established collaboration with statisticians.

2.12 Sample size calculation

To our knowledge, there are no methods for determining the sample size at MVDA. In the literature, for example, proteomics and metabolomics studies for chronic pain have so far been rather small and usually include 20 + 20 participants. The literature points out the need for large cohorts, especially in complex disease states such as chronic pain, and that the results are replicated in new cohorts. Here we make the assessment that at least 200 patients (from both sexes) in each of the 10 diagnostic groups (see above) must be recruited and the same number for replication of the results. In the group with several diagnoses, the heterogeneity is significant, so double the number is deemed necessary. The numbers are required to get a sufficient spread in terms of, for example, psychological load, BMI, and pharmacotherapy and to be able to identify clinically relevant subgroups within a diagnosis. Four hundred healthy controls (even gender

distribution) are deemed necessary; half of these leave new samples after about 10 weeks. For analyzes of the relationships with treatment results (mixed diagnoses), 300 patients are considered necessary.

In summary, we intend to recruit a total of 400 patients in each diagnostic group (1-10) and 800 in mixed diagnostic group for analysis at one time, 300 patients participating in treatment leave samples immediately before the treatment period and are followed up on two occasions, and 400 healthy controls. This means that 4,800 patients and 400 healthy controls, i.e., a total of 5,200 participants, will be involved.

3. Discussion

Sweden has a population of about 10 million people, and the societal costs of approximately 20% of the adult population having moderate-severe chronic pain have been calculated by the Swedish Agency for Health Technology Assessments and Assessment of Social Services in 2003 as SEK 87 billion per year, corresponding to approximatively 1 billion US dollars per million inhabitants and year. Chronic pain is often associated with extensive suffering and poor health. At the same time, it is necessary to state that the effects of, for example, pharmacological treatment are limited; a maximum of 25–30% of the patients report significant and clinically valuable effects. The effect sizes for IPRP are only small to moderate according both to systematic reviews and to registry studies of real-world patients [8; 31].

The importance of developing mechanism-based diagnostics and treatment for chronic pain has been emphasized [37]. Pain clinicians largely lack precision medicine tools that can provide support for treatment choice. This constitutes a significant lack of knowledge about the biological component of the bio-psycho-social model on which modern pain care is expected to rest. This deficiency reasonably helps to explain the relatively small effects of treatment and rehabilitation. Our new cooperative multicenter project from all Swedish Universities combined will play a crucial part in the paradigm shift occurring and focusing on a personalized individual treatment method where we will be using advanced metabolomic as well as proteomic methods. The present project is an example of the paradigm shift that is now taking place in modern clinical medicine in a direction towards precision medicine where *omic* research is a crucial element. The

new research is characterized by having a focus on biological processes and involves developing a clinical medicine which is based not only on anamnesis and clinical examination but that also includes the possibility to measure the actual pathophysiological mechanism at work in each patient (hence, precision medicine). This is crucial for the development of the clinical diagnosis and treatment of chronic pain and other conditions and diseases in which the clinically oriented researchers participating in this project are involved and engaged.

The project is on an international research front and has great potential to contribute to an improved understanding of activated nociceptive and pain mechanisms and thereby better diagnosis and, in the long term, treatment of chronic pain. It will contribute to the development of clinically useful saliva and blood samples that can be used in the investigation and choice of treatment for patients with chronic pain. In this way, the project will be able to contribute to the development of investigation and treatment that is indeed based on the individual patient's activated nociception and pain mechanisms ("personalized medicine", "precision medicine"). The results will also be able to form the basis for new pharmacological development, helping to bridge the gap between clinical pain medicine and animal models (c.f. translation and back-translation).

The feasibility of the project is very high. The research group has broad and adequately high competence for the implementation. The participating university clinics have working routines for collecting data for SQRP; (response rate> 90%) and routines for recruiting healthy participants. Extensive experience of the biochemical and statistical methods that will be used in the project is available. We have access to all necessary equipment partly within our research laboratory Painomics laboratory at Linköping University, through the Faculty of Medicine's Core Facility at Linköping University and the local node in Linköping that is under construction and connected to the Swedish Nuclear Magnetic Resonance spectroscopy (NMR) center in Gothenburg.

In the studies that form the basis of the project and use the same methods, we have found marked differences in the protein/inflammation pattern in muscles and plasma at CWP/FM [26; 42; 43]. In the proteomics studies of musculature and plasma, we have been able to identify proteins that can with great certainty explain the group affiliation (patient or healthy) (R2: 0.81- 0.84) [26; 42]; similar results are obtained with the

inflammation panel [17]. The significant proteins in the proteomics studies reflect activated nociceptive, inflammatory, and various metabolic processes. Similar conclusions are drawn in other existing proteomics studies [18; 29; 41; 42]. Our proteomics/inflammation research has received attention and been highlighted as innovative and internationally leading [13; 35]. A recently published systematic review identified that proteomics research on chronic pain is dominated by researchers from Sweden (mainly this research group) [15].

Overall, this clinically anchored project will be of great importance for the optimization of the clinical investigation and assessment as well as for better and more adapted treatment interventions based on the activated nociceptive mechanisms. This project will fill an important gap in our knowledge of the molecular mechanisms of common chronic pain conditions in humans and will produce significant steps towards a mechanism based and precision pain medicine.

Ethics

The study is approved by the Swedish Ethical Review Authority (Dnr 2021-04929) and will be conducted in accordance with the declaration of Helsinki. All participants will give their informed consent before the experiments and they will repeatedly be informed about their right to interrupt the participation at any time point, without explanation of their actions. Before data are registered in the database it will be blinded. All measurements are performed by registered health care staff. The participants will receive written information with the telephone number of the project responsible physician to be contacted in case of complications. The visits, any complications, and a summary of the study will be recorded in the electronic record. When taking samples and measuring pain sensitivity, participants may experience short periods of (increased) pain. However, this discomfort should not be more extensive than temporary pain that is experienced in everyday situations.

The samples of blood and saliva will be frozen immediately after collection and stored in biobank at; Östergötland, reg nr 1, Örebro biobank, IVO reg nr 454, Biobank Norr, IVO reg nr 472, Uppsala Biobank, IVO reg nr 827, Biobank Väst, IVO reg nr 890, Region Skånes biobank, IVO reg nr 136 and the local biobank at Danderyd hospital AB in Stockholm.

Results will be presented at the group level without the possibility of identifying any single individual. *Overall*, our assessment is that the benefits of this project are considerably greater than the risks.

Authors' contributions

The original scientific idea: BGh, ME, EB, and BG. All authors have contributed to the study design, data collection, analysis methods and writing the protocol.

Competing interests

The authors declare that they have no competing interests

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The Swedish Chronic Pain Biobank - protocol for a multicenter registry and biomarker project

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Abstract

Introduction: About 20% of the adult population have chronic pain, often associated with psychological distress, sick leave, and poor health. There are large variations in the clinical picture. A biopsychosocial approach is used in investigation and treatment. The concept of personalized medicine, i.e., optimizing medication types and dosages for individual patients based on biomarkers and other patient-related factors, has received increasing attention in different diseases but used less in chronic pain. This cooperative project from all Swedish University Hospitals will investigate whether there are changes in inflammation and metabolism patterns in saliva and blood in chronic pain patients and whether the changes correlate with clinical characteristics and rehabilitation outcomes.

Methods and analysis: Patients at multidisciplinary pain centers at University Hospitals in Sweden who have chosen to participate in the Swedish Quality Registry for Pain Rehabilitation and healthy sex- and age-matched individuals will be included in the study. Saliva and blood samples will be collected in addition to questionnaire data obtained from the Register. From the samples, proteins, lipids, metabolites and micro-RNA will be analyzed in relation to, for example, diagnosis, pain characteristics, psychological distress, body weight, pharmacological treatment, and clinical rehabilitation results using advanced multivariate data analysis and bioinformatics.

Ethics and Dissemination: The study is approved by the Swedish Ethical Review Authority (Dnr 2021-04929) and will be conducted in accordance with the declaration of Helsinki. The results will be published in open access scientific journals and in popular scientific relevant journals such as those from patient organizations. Data will be also presented in scientific meetings, meeting with healthcare organization and disseminated in different lecturers at the clinic and university.

Keywords: Biomarkers, Chronic pain, Inflammation, Metabolomics, Molecular profile, Proteomics

Strengths and limitations of this study

- The major strength of this study is that both omics methods and patient reported outcomes measure from the Swedish Quality Registry for Pain Rehabilitation will be used.
- Researchers from different professions with strong research backgrounds in chronic pain, rehabilitation and omics collaborate in this pain precision medicine study.
- There is a risk of heterogeneity of the samples as this study is a multicenter study in a large geographical area where the infrastructure for data collection is different.
- Individuals not able to read and write Swedish are excluded which make the results less generalizable.

1. Introduction

About 20% of the adult population lives with at least moderate-severe chronic pain; often with concomitant complex psychological distress including depression and anxiety. Pain will also reduce patients' ability to study or work full time, they will show higher levels of sick leave, and show reduced quality of life [1]. There are large variations in the clinical picture in chronic pain patients because of the presence of different comorbidities. For example psychiatric comorbidities are common in chronic pain and about 40% of patients managed at specialist level have co-morbid symptoms of depression and/or anxiety [2]. More than 20% of Years lived with disabilities – a measure of non-fatal disabilities - are caused by of pain conditions both in Sweden and globally [3].

Acute tissue damage initiates plastic and reinforcing mechanisms in the pain system peripherally and centrally, which in interaction with psychological and social factors create the experience of pain at a given moment [4]. Chronic pain in combination with depression constitute a large part of the pain complex, why depression also must be considered when diagnosing the patient. Moreover, positive correlations exist between depressive symptoms and pain intensity. Social factors such as working conditions can be risk factors for the chronification of pain, and when chronic pain has developed return to work will be more difficult (i.e., chronic pain has social consequences). Modern pain care is therefore based on a bio-psycho-social approach to diagnosis and treatment[5, 6].

Chronic pain is not an acute pain that persists over time, but further plastic neurobiological changes occur in interaction with psychological and social factors. Imaging techniques have provided an in-depth understanding of how the brain processes and creates the experience of pain. Chronic pain is associated with chemical changes in I) structures in the brain that process pain, II) the physiological interactions between these structures, III) the descending control of nociception, and IV) central hyperexcitability (central sensitization)[7, 8].

Chronic widespread pain (CWP) including fibromyalgia (FM) is the extreme of complex pain condition. FM is characterized by altered nociception and is a prototype of a nociplasic pain condition, however, it is well known that patients initially suffering from

nociceptive pain can in time develop nociplastic pain conditions [9]. It has been argued that CWP / FM is a typical example of a central pain condition, i.e., that peripheral factors have little or no role. Others believe that peripheral factors initiate and perpetuate the central changes in analogy with e.g. coxarthrosis where central changes are normalized after hip replacement [10].

In various studies, we have shown an increased presence of pain-mediating (e.g., glutamate, serotonin), metabolic (e.g., lactate and pyruvate) and analgesic (e.g., N-acylethanolamines (NAE)) substances in muscle in neck-shoulder pain and in CWP / FM [11]. Other researchers have recently shown changes in the peripheral nociceptors of FM [12] and blood biomarkers in chronic pain [13, 14]. Proteomic studies are increasingly being performed in the field of pain medicine. Using targeted and untargeted proteomics, our research group has found significant differences in the protein/inflammation pattern in muscles and plasma in CWP / FM [15-17]. Such changes also correlate with, for example, pain intensity, pain sensitivity and psychological strain [15, 18]. Taken together, this provides support for peripheral factors that significantly contribute to the maintenance of CWP / FM.

In the literature it is suggested that low-grade peripheral or systemic inflammation perpetuates chronic pain and is also involved in psychological / psychiatric conditions such as depression and obesity [19, 20]. Various pro-inflammatory cytokines and chemokines have been studied. However, the literature is not consistent; in a large FM study, we found no changes in “classic” pro-inflammatory cytokines in muscle or plasma [21]. Two recent systematic reviews of peripheral cytokines and chemokines - mainly based on single or few molecules - have not been consistent [22, 23]. Such hypothesis-driven studies have generally focused on a few molecules, while there are only a few exploratory studies of a larger number of molecules (panels). Low-grade inflammation/neuroinflammation has also been studied in neuropathic pain conditions [24-26].

Preliminary biomarker data available in the literature need to be confirmed in larger studies that reflect the normal flow of chronic pain patients where/in which comparisons are made between pain diagnoses and that include control for gender, pain characteristics, co-morbidity, body weight, and pharmacological therapy. The

inflammation pattern according to “classic” inflammation markers seems to be of preliminary importance for the treatment outcome in depression [27] and obesity, as well as for psychological treatment, such as cognitive behavioral therapy (CBT) in chronic pain [28]. Interdisciplinary Pain Rehabilitation Programs (IPRP), in which psychological treatment (including CBT) and physical activity and exercise are important components, constitute evidence-based treatment for chronic pain but with small to moderate effect sizes [29-32]. It has not yet been considered if activated biological mechanisms may impact on the effect of IPRP. It is therefore important to investigate whether IPRP result is related to inflammation, and metabolism patterns. To the best of our knowledge there are no reports on established pain biobank that combine health informatics with biomarkers and bioinformatics to study chronic pain mechanisms in everyday life patients.

Aim and hypotheses

The aim of this multicenter project is to investigate whether there are differences in inflammation and metabolism patterns between common chronic pain diagnoses and to sex- and gender-matched healthy controls from the general Swedish population. Further aims are to characterize and compare the changes of the grade of inflammation, and metabolism in different diagnostic groups of chronic pain patients regarding e.g., sex, diagnosis, pain, pain sensitivity, co-morbidity pattern, body weight, pharmacological therapy, and clinical rehabilitation outcomes.

The hypotheses are:

A) there are unique biomarker signatures in plasma and saliva that distinguish common chronic pain diagnoses from each other and from healthy controls.

B) there are significant correlations between the identified biomarker profiles and pain characteristics e.g., intensity, pain sensitivity and anatomical spread of pain, co-morbidity patterns, body weight and pharmacological therapy, and clinical rehabilitation outcomes.

2. Methods

2.1. Participants

This multicenter project will include patients (> 18 years) with chronic pain who are recruited from the *Swedish Quality Registry for Pain Rehabilitation* (SQRP) and healthy sex- and age-matched controls from university hospital at Linkoping, Stockholm, Uppsala, Umea, Gothenburg, and Lund see figure 1.

2.2. Patients with chronic pain

The project will mainly use patient-reported data from SQRP (www.ucr.uu.se/nrs/) for the participating university clinics. The project leader (Björn Gerdle) is the research leader for the national research group for SQRP and a member of its steering group.

SQRP is based on *Patient Reported Outcome Measures* (PROM) in the form of a written questionnaire. Validated instruments are used in the survey and collectively capture the various facets of the International Classification of Functional Conditions, Disabilities and Health (ICF). The survey includes background variables (*age, sex, country of origin, education and healthcare consumption, weight, height, pain duration and pain extent (according to a list of 36 anatomical regions))* and pain characteristics, degree of FM-ness, psychological distress, difficulty sleeping, pain coping, fear of movement, consequences of living with chronic pain, physical activity level, as well as health-related quality of life and perceived health (Table 1, Appendix 1). The response rate is > 90% in SQRP, which can be interpreted as patients considering it important to be able to share how they experience their situation. The current diagnoses according to ICD-10 are also registered in SQRP.

The patients who refer to the clinics respond to the SQRP questionnaire according to table 1 in connection with clinical assessment. Patients participating in IPRP also answer questionnaires immediately after and 12 months after rehabilitation.

All patients who respond to SQRP at the relevant university clinics will be asked to participate in the study and may submit saliva samples and blood samples (see below). In this project we will study the following 11 conditions/diagnoses (based on the manual produced within SQRP by, among others, the project leader):

1. Chronic neck-shoulder pain (cervicalgia, trapezius myalgia)
2. Chronic low back pain without sciatica
3. Generalized pain (CWP) according to criteria ACR 2016 criteria.
4. Fibromyalgia according to the ACR 2016 criteria.
5. Neuropathic pain including sciatica
6. Chronic Whiplash Disorders (WAD)
7. Ehlers-Danlos Syndrome (EDS) or Hypermobility Syndrome (HMS)
8. Complex Regional Pain Syndrome (CRPS)
9. Pain post-COVID infection
10. Chronic pain, not elsewhere classified (ICD R52.1, R52.2 and R52.9)

In addition, we will investigate the patterns when several pain diagnoses are present simultaneously.

2.3. Healthy controls

Age- and sex-matched healthy control persons will be recruited via advertisements and answer a customized version of the SQRP questionnaire. Saliva and blood samples will be collected. Half of the healthy controls will be asked to submit new samples after about 10 weeks.

2.4. Exclusion criteria for patients and controls

Coagulation disorders with predisposition to bleeding, medication with anticoagulants (low-dose aspirin is permitted), hypersensitivity to anesthetic, serious psychiatric disease (investigator's judgement, e.g., psychosis or suicidal ideation), and difficulties in understanding the Swedish language will be excluded.

2.5. Supplementary survey

In addition to the instruments included in the SQRP (or equivalent adapted for healthy controls), the participants will also answer a brief questionnaire in close connection with the tissue sampling. This charts the time of sampling, fasting or not, current medication and health food preparations (e.g., curcumin), pain intensity, and present and previous illnesses e.g., diabetes.

2.6. Biofluids sampling

Sampling consists of saliva and blood samples drawn in the morning and during fasting. Saliva samples will be taken with Salivette® (Sarstedt AG & Co, PO Box 1220, D-51582, Nümbrecht, Germany, obtained from VWR, article number: 101093–968). The participants ask to avoid brushing teeth or drinking (water is ok) at least one hour before sampling. They rinse their mouth with water and wait for 15 minutes before taking the swab in the mouth. After 3 minutes, they spit the swab into the salivette tube containing a protease cocktail inhibitor. The salivette will be centrifuged (2 minutes, 1,000 x g) and the supernatant is transferred to a new tube and aliquots into 200 µl in 0.6 ml eppendorf tubes and will be stored at -86 ° C until analysis. The swab will be stored at -86°C for future cell extraction. The total volume of saliva will be recorded together with time of sampling, any complication e.g., bleeding, shorter time than 3 minutes due to un-comfortable feelings to have the swab in the mouth.

Blood samples (2 x 8.5 ml) will be collected in P100 tubes (article number: 366448, BD Diagnostics System, Frankling Lakes, NJ, USA) according to the manufacture's recommendations. The sample will be centrifuged at 2500 g for 20 minutes at room temperature within 2-4 hours. Plasma will be extracted by carefully removing the upper part of the supernatant in fractions to a 10 ml tube, and after mixing gently will be aliquoted into 200 µl in 0.6 ml eppendorf tubes and store at -86°C. The cell fraction will be removed to a new tube and store at -86°C. The time for blood sampling and centrifugation, any signs of hemolysis, any complication with sampling will be recorded.

The samples will be handled according to previously developed methodology [16, 17].

2.7. Biochemical analyses

The analyzes that will be used in this study are mainly exploratory omics analysis, i.e., which proteins, metabolites and lipoproteins will be identified cannot be determined in advance. The following analyzes will be performed according to the methodology previously described by our research group [15, 17, 26].

- Exploratory analyzes using panels for inflammation, cytokines & chemokines and neuroinflammation comprising many proteins (from Olink Bioscience, Uppsala [15] and from Meso Scale Discovery [26].

- Exploratory proteomics analysis using 2D gel electrophoresis and/or shotgun proteomics [16, 33]
- Exploratory Metabolomic and Lipoprotein profile including free fatty acids (FFAs)
- Anti-nociceptive substances, i.e., endocannabinoids, NAE, endorphins, alpha-amylase, and cortisol.
- Metabolites such as lactate, pyruvate, glutamate, and other single pain-mediating molecules such as substance P, bradykinin, serotonin and BDNF.
- Pending analysis, the samples are stored in the biobank facilities with which the participating university clinics are associated.

2.8. Data from SQRP

The surveys in SQRP, including the supplementary questionnaire, will include a careful characterization of the patients and the healthy controls. In the correlation analyzes between biomarkers and pain characteristics, the following variables will initially be used:

- Diagnosis (from manual produced within SQRP)
- Pain intensity
- Extent of pain spreading/Pain localization
- Pain duration
- Psychologic distress (depression, anxiety, pain catastrophizing, fear of movement and insomnia)
- Sex
- Body mass index (BMI)
- Physical activity level
- Pharmacological therapy and health food preparations

2.9. Interdisciplinary multi modal rehabilitation program (IPRP)

The inclusion criteria will be decided by the clinical examination and screening questionnaires concerning both anthropometric measures as well as subjective health followed by an inter professional conference between the MD, a psychologist, a physiotherapist, and an occupational therapist. The IPRP team make the decision which patients they believe would benefit from the IPRP, in accordance with recommendations

from the Swedish Agency for Health Technology Assessments and Assessment of Social Services. The patient should be motivated and have the potential for an active change to be included in IPRP. All patients will participate in a specific individual exercise with a physiotherapist (with both relaxation therapy and strengthening specific regions of the body to unburden), individual- and group therapy led by a clinical psychologist (cognitive behavioral therapy, education, coping and mindfulness) as well as environmental work changes from an occupational therapist, working with education and return to work strategies in group of six to nine persons, commonly 20 h per week of group-based activities for 6 to 8 weeks [34, 35]. Pain education (including lectures in basic pain science and pain management) will be offered both for patients as well as for their relatives, friends, and colleagues. For descriptions and content of the Swedish IPRPs see [36-38]

2.10. Statistics

In addition to descriptive statistics, advanced multivariate statistics (MVDA [18]; i.e., advanced principal component analysis (PCA) and OPLS regressions) will be used according to the guidelines presented by Wheelock & Wheelock for omics data [39] using SIMCA®-P + (Sartorius AG, Göttingen Germany). The methods are necessary to manage and take advantage of the intercorrelation pattern (i.e., multicollinearity) between the identified substances. MVDA enables analyzes where the number of variables is significantly greater than the number of observations (i.e., short, and broad data tables), which more traditional multivariate regressions cannot easily handle. The research group has a long experience of the methods and has used these in previous studies and has an established collaboration with statisticians.

2.11 Sample size calculation

There are limited numbers of study that report methods for determining the sample size at MVDA [40]. In the literature, for example, proteomics and metabolomics studies for chronic pain have so far been rather small and usually include 20 + 20 participants. The literature points out the need for large cohorts, especially in complex disease states such as chronic pain, and that the results are replicated in new cohorts. Here we make the assessment that at least 200 patients (from both sexes) in each of the 10 diagnostic groups (see above) must be recruited and the same number for replication of the results. In the group with several diagnoses, the heterogeneity is significant, so double the

number is deemed necessary. The numbers are required to get a sufficient spread in terms of, for example, psychological load, BMI, and pharmacotherapy and to be able to identify clinically relevant subgroups within a diagnosis. Four hundred healthy controls (even sex distribution) are deemed necessary; half of these leave new samples after about 10 weeks. For analyzes of the relationships with treatment results (mixed diagnoses), 300 patients are considered necessary.

In summary, we intend to recruit a total of 400 patients in each diagnostic group (1-10) and 800 in mixed diagnostic group for analysis at one time, 300 patients participating in IPRP treatment will leave samples immediately before the treatment period and are followed up on two occasions, and 400 healthy controls. This means that 4,800 patients and 400 healthy controls, i.e., a total of 5,200 participants, will be involved.

2.12 Patient and Public Involvement

Neither patients nor the public are involved in the design, conduct, reporting, or dissemination plans associated with this research.

3. Discussion

Sweden has a population of about 10 million people, and the societal costs of approximately 20% of the adult population having moderate-severe chronic pain have been calculated by the Swedish Agency for Health Technology Assessments and Assessment of Social Services in 2003 as SEK 87 billion per year, corresponding to approximatively 1 billion US dollars per million inhabitants and year. Chronic pain is often associated with extensive suffering and poor health. At the same time, it is necessary to state that the effects of, for example, pharmacological treatment are limited; a maximum of 25–30% of the patients report significant and clinically valuable effects. The effect sizes for IPRP are only small to moderate according both to systematic reviews and to registry studies of real-world patients [37, 41].

The importance of developing mechanism-based diagnostics and treatment for chronic pain has been emphasized [42]. Pain clinicians largely lack precision medicine tools that can provide support for treatment choice. This constitutes a significant lack of knowledge about the biological component of the bio-psycho-social model on which modern pain care is expected to rest. This deficiency reasonably helps to explain the

relatively small effects of treatment and rehabilitation. Our new cooperative multicenter project from all Swedish Universities combined will play a crucial part in the paradigm shift occurring and focusing on a personalized individual treatment method where we will be using advanced metabolomic as well as proteomic methods. The present project is an example of the paradigm shift that is now taking place in modern clinical medicine in a direction towards precision medicine where *omic* research is a crucial element. The new research is characterized by having a focus on biological processes and involves developing a clinical medicine which is based not only on anamnesis and clinical examination but that also includes the possibility to measure the actual pathophysiological mechanism at work in each patient (hence, precision medicine). This is crucial for the development of the clinical diagnosis and treatment of chronic pain and other conditions and diseases in which the clinically oriented researchers participating in this project are involved and engaged.

The project is on an international research front and has great potential to contribute to an improved understanding of activated nociceptive and pain mechanisms and thereby better diagnosis and, in the long term, treatment of chronic pain. It will contribute to the development of clinically useful saliva and blood samples that can be used in the investigation and choice of treatment for patients with chronic pain. In this way, the project will be able to contribute to the development of investigation and treatment that is indeed based on the individual patient's activated nociception and pain mechanisms ("personalized medicine", "precision medicine"). The results will also be able to form the basis for new pharmacological development, helping to bridge the gap between clinical pain medicine and animal models (c.f. translation and back-translation).

The feasibility of the project is very high. The research group has broad and adequately high competence for the implementation. The participating university clinics have working routines for collecting data for SQRP; (response rate> 90%) and routines for recruiting healthy participants. Extensive experience of the biochemical and statistical methods that will be used in the project is available. We have access to all necessary equipment partly within our research laboratory Painomics laboratory at Linköping University, through the Faculty of Medicine's Core Facility at Linköping University and

the local node in Linköping that is under construction and connected to the Swedish Nuclear Magnetic Resonance spectroscopy (NMR) center in Gothenburg.

In the studies that form the basis of the project and use the same methods, we have found marked differences in the protein/inflammation pattern in muscles and plasma at CWP/FM [16, 17, 33]. In the proteomics studies of musculature and plasma, we have been able to identify proteins that can with great certainty explain the group affiliation (patient or healthy) (R2: 0.81- 0.84) [16, 17]; similar results are obtained with the inflammation panel [15]. The significant proteins in the proteomics studies reflect activated nociceptive, inflammatory, and various metabolic processes. Similar conclusions are drawn in other existing proteomics studies [17, 43-45]. Our proteomics/inflammation research has received attention and been highlighted as innovative and internationally leading [46, 47]. A recently published systematic review identified that proteomics research on chronic pain is dominated by researchers from Sweden (mainly this research group) [48].

Overall, this clinically anchored project will be of great importance for the optimization of the clinical investigation and assessment as well as for better and more adapted treatment interventions based on the activated nociceptive mechanisms. This project will fill an important gap in our knowledge of the molecular mechanisms of common chronic pain conditions in humans and will produce significant steps towards a mechanism based and precision pain medicine.

Ethics

The study is approved by the Swedish Ethical Review Authority (Dnr 2021-04929) and will be conducted in accordance with the declaration of Helsinki. All participants will give their informed consent before the experiments and they will repeatedly be informed about their right to interrupt the participation at any time point, without explanation of their actions. Before data are registered in the database it will be blinded. All measurements are performed by registered health care staff. The participants will receive written information with the telephone number of the project responsible physician to be contacted in case of complications. The visits, any complications, and a summary of the study will be recorded in the electronic record. When taking samples and measuring pain sensitivity, participants may

experience short periods of (increased) pain. However, this discomfort should not be more extensive than temporary pain that is experienced in everyday situations.

The samples of blood and saliva will be frozen immediately after collection and stored in biobank at; Östergötland, reg nr 1, Örebro biobank, IVO reg nr 454, Biobank Norr, IVO reg nr 472, Uppsala Biobank, IVO reg nr 827, Biobank Väst, IVO reg nr 890, Region Skånes biobank, IVO reg nr 136 and the local biobank at Danderyd hospital AB in Stockholm.

Results will be presented at the group level without the possibility of identifying any single individual. *Overall*, our assessment is that the benefits of this project are considerably greater than the risks.

Contributorship statement

BG,BGh and EB were responsible for conception of this research project. All authors contributed to the study design. BGh and ME draft the first version of the manuscript. MR, OG, and BG were responsible for the communication plan with the steering group at the clinic. EK, HW, ÅR, YFL and BG were responsible for the scientific hypothesis. ME and YFL were responsible for ethical application. E B, ML, HG, KU and KR were responsible for the protocol for clinical procedures to recruit patients. BGh, NS and BS were responsible for the protocol for sampling, sample handling and biobanking. PA, RK and MR are responsible for financing plan for the project.

All authors have been involved in editing, review and accepted the final version of the manuscript.

Competing interests

The authors declare no conflict of interest.

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Data sharing statement

The datasets generated during the study will be available from the corresponding author on reasonable request after the final reports have been published.

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Figure legends

Figure 1. Flow chart of the study design outlining numbers of subjects in each chronic pain group and healthy controls.

Tables

Table 1. Patient reported outcome measures to be used in the project. NRS = Numeric Rating Scale, MPI = Multidimensional Pain Inventory Swedish version, HADS = Hospital Anxiety and Depression Scale, PCS = Pain Catastrophizing Scale, ISI = insomnia Severity Index, CPAQ8= Chronic Pain Acceptance Questionnaire based upon 8 items, TSK = Tampa Scale for Kinesiophobia, RAND-36 = modernized version of Short-Form Health Survey-36.

Survey	Instrument/Scales	Properties
Pain characteristics		
Intensity	NRS (0-10)	0 = no pain, 10 = worst pain experienced
	MPI Pain intensity (0-6)	Based on 2 items, each scored 0-6
Interference	MPI-Pain Interference (0-6)	Based on 11 items, each scored 0-6
Fibromyalgia	2016 criteria	Widespread pain index (WPI): 0-19 sites Symptom severity score (SSS): 0-12 FMness score: WPI + SSS.
Psychological distress		
Depression	HADS-Depression (0-21)	Sum of 7 items, each scored 0-3
Anxiety	HADS-Anxiety (0-21)	Sum of 7 items, each scored 0-3
	MPI-Affective distress (0-6)	Based on 3 items each scored 0-6
Pain catastrophizing	PCS (0-52)	Sum of 13 items, each scored 0-4; 3 subscales can be obtained
Difficulty sleeping		
	ISI (0-28)	Sum of 7 items, each scored 0-4
Pain coping		
	CPAQ8 (0-48)	Sum of 8 items, each scored 0-3; 2 subscales can be obtained
Fear of movement		
	TSK (0-68)	17 items, each scored 0-4 according to agreement with statement and summed
Impact of chronic pain		
Control of life	MPI-Life control (0-6)	Based on 4 items, each scored 0-6
Social support	MPI-Social support (0-6)	Based on 2 items, each scored 0-6
Health-related quality of life		
	RAND-36 (0-100)	36 items in 8 subcategories scored varying between 0 and 100% (with fixed intervals). The mean for each subscale is calculated
	EQ-5D™	Consists of two parts. The first part is an index obtained from five dimensions items scored 0-5. The second part is self-estimation of today's health according to a 100-point thermometer-like scale (EQ5D-VAS) with defined end points (high values indicate

	better health and low values indicate worse health)
Physical activity	
Sedentary behavior	Minutes per week
Low intensity	
Moderate intensity	
Vigorous intensity	

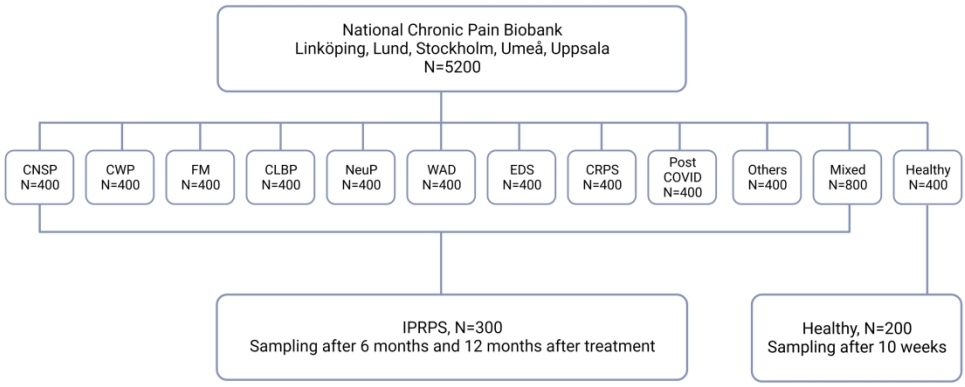

Figure 1. Flow chart of the study design outlining numbers of subjects in each chronic pain group and healthy controls.

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Appendix 1

Table: References to psychometric characteristics for the patient reported outcome measures (cf. Table 1) to be used in the project.

Instrument/Scale	References
Pain intensity (NRS)	1
Fibromyalgia 2016 criteria	2
Psychological distress (HADS)	3 4
Multidimensional Pain Inventory (MPI)	5-7
Pain catastrophizing (PCS)	8 9
Insomnia (ISI)	10 11
Pain coping (CPAQ8)	12-14
Fear of movement (TSK)	15-18
Health-related quality of life (RAND-36)	19-22
Health-related quality of life (EQ-5D™)	23-26
Physical activity	27 28

NRS = Numeric Rating Scale, MPI = Multidimensional Pain Inventory, HADS = Hospital Anxiety and Depression Scale, PCS = Pain Catastrophizing Scale, ISI = insomnia Severity Index, CPAQ = Chronic Pain Acceptance Questionnaire, TSK = Tampa Scale for Kinesiophobia, RAND-36 = modernized version of Short-Form Health Survey-36.

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