Mapping complexity of pain with the Advanced Pain Discovery Platform Awards

UKRI (MRC, BBSRC, ESRC; with funding from the Strategic Priorities Fund), Versus Arthritis, Medical Research Foundation and Eli Lilly launched a call in 2021 to support innovative research grants seeking to increase our understanding of the underlying mechanisms of pain as a part of the APDP. Proposals could seek to explore mechanisms of pain in various disciplines, investigating psychological, biological, cognitive and sociological aspects of pain. Where possible within the scope of the research question, researchers were encouraged to work together across disciplines to facilitate novel research into the underlying mechanisms of pain. Each proposal could focus on a specific pain condition, or a specific aspect of pain, or they could span conditions. Support was available for pre-clinical as well as clinical research projects, with a focus on understanding the mechanisms of pain. All projects had to demonstrate a clear pathway to impact for people with lived experience of chronic pain.

Applications seeking to increase our understanding of the transition from acute to chronic pain, changes in the experience of pain, or the transition to remission were encouraged. Proposals could involve a wide range of methodology across disciplines, and applications involving the use of artificial intelligence, machine learning or other innovative data technologies were especially encouraged. Proposals with a focus on musculoskeletal conditions, both in adults and young people, were also encouraged. Through the involvement of the Medical Research Foundation, a part of the available budget had been ring-fenced for funding of research into mechanisms of pain in children and adolescents, with a particular, but not sole, focus on musculoskeletal conditions in this group. The funding opportunity was intended to support a broad portfolio of research projects ranging from smaller, more focused grants to larger research grants for a maximum of three years and cost of £1 million. Early and mid-career researchers were strongly encouraged to apply. The Panel assessed applications on 1st - 2nd December 2021 and made 12 awards:

Professor Victoria Chapman, University of Nottingham (MR/W02652X/1, 36 months, £627k)

- **MICA: Exploiting specialised pro-resolution molecule mediated analgesia to identify novel targets for the treatment of chronic pain**

Co-investigators:

Professor Ana Valdes	University of Nottingham
Dr Dong-Hyun Kim	University of Nottingham
Dr Federico Dajas-Bailador	University of Nottingham
Professor David Bennett	University of Oxford
Dr Andreas Themistocleous	University of Oxford

Researcher Co-Investigator:

Dr Peter Gowler	University of Nottingham

Patient insight partners:

Lorraine Salt	University of Nottingham Musculoskeletal Patient Advisory Group Member
Jenny Cockshull	University of Nottingham Musculoskeletal Patient Advisory Group Member
Michael Prior	University of Nottingham Musculoskeletal Patient Advisory Group Member
Terry Murphy	University of Nottingham Musculoskeletal Patient Advisory Group Member

Summary: Osteoarthritis (OA) and diabetic neuropathy are often associated with chronic pain which, as highlighted by our PPIE representatives, hugely impacts upon daily life. Not everybody with these diseases experiences chronic pain, and understanding these differences may offer insight into new treatments. A family of natural molecules produced by the body (specialised pro-resolution molecules (SPMs)) have robust analgesic effects in animal models of pain, while healthy volunteers with lower levels of SPMs are more sensitive to pain. Crucially, we showed that people with OA who have lower levels of SPMs experience significantly more pain. The SPMs reduce pain by interfering with multiple signalling pathways, leading to strong analgesic effects. Although SPMs are quickly broken-down into inactive products, they have long-lasting effects - a process likely to be due to the modulation of molecules known to drive pain responses at the gene expression level. We have already identified gene pathways implicated in the effects of the SPMs on OA pain in people.

Aim: To identify the cellular and molecular processes that lead to the powerful and long-lasting analgesia produced by the SPMs and to use this information to identify new therapeutic approaches to improve the treatment of chronic pain.

Team: This new research is guided by our PPIE steering group, and is lead by a team which brings strength in chronic pain mechanisms, genetics of pain and experimental pain. The pharmaceutical company Eli Lilly brings expertise in analysing and integrating large-scale datasets to maximise the benefits of these valuable clinical datasets. The project also supports the career development of researcher co-investigator Dr P Gowler.

Experimental Plan: We will use in-house bioinformatic approaches to analyse our existing and newly acquired data to predict the specific molecules (known as microRNAs) which may regulate changes in gene expression associated with high versus low levels of SPMs and their relationships with chronic pain. First we will collect blood from people with OA pain (and control non-OA group) to undertake a detailed analysis of microRNAs that are predicted to regulate the levels of genes associated with OA pain and levels of a specific stable molecule which is a precursor for many SPMs, known as 17-HDHA. Using existing computational tools we will identify the microRNAs associated with having high versus low levels of 17-HDHA and how this relates to the OA pain experienced. We will validate these findings in patients with a different type of chronic pain (diabetic neuropathic pain with and without pain) to identify commonality and differences between these two types of chronic pain, thus identifying targets that are specific to the diseases, and those shared between them. To focus on potential roles of these pathways in driving pain, computational analysis of existing microRNA datasets from joint tissue and synovial fluid already collected from people with OA (by our collaborator) will identify which microRNAs that we have identified are also present at the site of disease and the source of the pain. Using experimental models and tools we will identify which of the clinically identified microRNAs are altered by 17-HDHA treatment in mice, supporting which pathways are likely candidates for novel therapeutics. Molecular tools specifically developed to manipulate the function of those microRNAs will be used to identify the miRNAs that mediate the effects of the 17-HDHA on excitatory sensory nerve activity that drives pain responses. This approach will prioritise the clinically identified microRNAs to those with the most potential for therapeutic development for people with chronic OA pain. The importance of this outcome was supported by our PPIE representatives, as people living with pain want the development of new treatments which reduce their existing pain and allows them to have a fuller and happier life experience.

Technical summary: Osteoarthritis (OA) is the leading cause of chronic pain. Specialised resolution molecules (SPMs) have robust analgesic effects in models of chronic pain, acting via microRNAs (miRNAs) to shut-down multiple pro-inflammatory signalling pathways to restore tissue homeostasis. Levels of the SPM precursor 17-HDHA are significantly associated with pain thresholds in healthy volunteers and levels of OA pain.

Aim: to identify the miRNAs critical to the analgesic effects of the SPMs to provide new strategies for OA pain treatment. A clinical study will identify miRNAs associated with 17-HDHA levels and OA pain, with validation in people with diabetic neuropathy +/- pain. Bioinformatic analysis will categorise dataset commonality for the two conditions. Using a publicly available synovial fluid dataset we will focus on miRNAs both implicated in the effects of 17-HDHA and locally at the painful joint. The top 30 clinically relevant miRNAs will be mechanistically probed. Levels of 17-HDHA will be augmented in mice in the presence (and absence) of a clinically relevant model of OA pain behaviour, and blood levels of the top 30 clinically relevant miRNAs will be quantified to confirm which miRNAs are modified by 17-HDHA. Using microfluidic chambers for the culture of mouse sensory nerve axons we will quantify the effects of 17-HDHA on calcium (Ca^{2+}) transients in cell bodies after axon terminals are stimulated with PGE2 plus capsaicin (to mimic hyperalgesia). The effects of manipulating (with inhibitors or mimics) 10-15 of the 17-HDHA modifiable miRNAs on evoked Ca^{2+}responses (in the presence and absence of 17-HDHA) will confirm which miRNAs are essential for the inhibitory effects of 17-HDHA on sensory excitability.
The identification of the clinically and functionally relevant miRNAs that underpin SPM-mediated natural analgesia will provide crucial mechanistic understanding which will underpin the development of novel approaches for treating chronic OA pain.

Dr David Andersson, King's College London (MR/W027585/1, 36 months, £631k)

- **Fibromyalgia and refractory pain in rheumatic diseases**

Co-Investigator:
Dr Andreas Goebel University of Liverpool

Researcher Co-Investigators:
Dr Mathilde Israel King's College London
Dr Margot Maurer King's College London

Summary: Fibromyalgia is one of the most common causes of chronic pain worldwide. There is no diagnostic test available, and patients are diagnosed based on how severe and widespread their pain is and whether they have other symptoms, such as fatigue, sleep problems and depression. Treatment of fibromyalgia is focused on exercise and education, which help patients become more active and cope better with pain. Drugs that are used to treat pain in fibromyalgia are effective in some patients, but often cause problematic side effects and regularly become less effective with time. Fibromyalgia has a severe impact on quality of life, but the fact that patients look healthy can make it difficult for them to qualify for benefits and to convince their environment about how they feel. Although fibromyalgia affects more than 1 in 50 people, the cause of disease remains unknown. A better understanding of the cause of fibromyalgia, is likely to dramatically accelerate development of improved treatments and invention of diagnostic tests.
We have discovered that the body's normal defence mechanism, the immune system, is responsible for pain in fibromyalgia patients. Immune cells produce proteins called antibodies, which would normally be used to help us destroy bacteria and other parasites, thereby helping us fight infections, and to become immune to them. Our results show that fibromyalgia patients make antibodies that attack their own bodies, rather than infections. These antibodies stimulate pain-sensing nerves throughout the body, making patients too sensitive to pressure, temperature and thereby experiencing unrelenting pain. In our pilot experiments, we have purified antibodies from fibromyalgia patients (without other health conditions) and healthy volunteers and injected these to mice.
Remarkably, mice that were given patient antibodies developed similar symptoms to the patients that the antibodies were taken from, whereas antibodies from healthy volunteers were without effect.
While about 2% of the general population (mostly women) suffer from fibromyalgia, around 25% of people rheumatic disease suffer from this difficult chronic pain condition. Rheumatic disease can often be well controlled by drugs, but fibromyalgia and the pain it causes throughout the body does typically not improve by treatment. During this project we will determine whether fibromyalgia in rheumatic patients is also caused by antibodies that cause pain. Our results are very likely to lead to development of the first simple diagnostic tests, and this will dramatically reduce the stressful period of time, often a couple of years that it currently takes for patients to be diagnosed. It is also very likely that our work will lead to improved treatment of fibromyalgia, and some of the possible treatments are already used in other conditions caused by antibodies that attack our own bodies.

Technical summary: Fibromyalgia syndrome (FMS) is a common chronic primary pain syndrome with a prevalence exceeding 2% in the general population. FMS is characterized by chronic widespread pain in combination with other neurological symptoms, such as fatigue, sleep disturbances, anxiety, depression, and memory problems. FMS is more common in women than men, and much more prevalent in patients with autoimmune rheumatological conditions. Patients report very poor scores for health- related quality of life, and the available therapies are of limited efficacy.
We have recently discovered that FMS in patients without comorbidities (primary FMS) is caused by autoantibodies and that sensory, anatomical, and motor symptoms can be transferred from patients to mice by administration of IgG. These findings are likely to initiate a paradigm shift in our understanding and management of FMS and have enabled us to embark on detailed cellular and molecular investigations of FMS. Our studies of single afferents in skin-nerve preparations and current-clamp investigations of dorsal root ganglion (DRG) neurons show that FMS IgG produces neuronal hyperexcitability. Transcriptomic analysis of sensory ganglia has identified a single candidate ion channel, and the observed electrophysiological changes in DRG neurons mirror those expected to be caused by dysregulation of the candidate ion channel.
About 20-30% of patients with rheumatoid arthritis, ankylosing spondylitis, and Behcet's disease meet the diagnostic criteria of FMS. While the rheumatic disease in these patients is often well-controlled clinically, FMS is typically refractory to treatment. The proposed studies will determine whether the same

mechanisms that we have identified in primary FMS, are also responsible for comorbid FMS in patients with rheumatic diseases. The results of the proposed studies will
expand our earlier conclusions and may establish FMS as an autoimmune disease in its own right.

Dr Franziska Denk, King's College London (MR/W027518/1, 36 months, £406k)

- **Silencing musculoskeletal pain: can we target spontaneously active neurons?**

Researcher-Co-Investigator:
Dr George Goodwin King's College London

Patient insight partner:
Dr Sara Villa King's College London

Summary: Chronic pain in joints and muscles is one of the leading causes of disability worldwide. The pain killers we have do not work for many people and often have terrible side effects, like addiction in the case of opioids. We therefore urgently need to increase our understanding of what causes chronic muscle and joint pain, to help us develop better treatments.
One particularly promising idea could be to target the nerves that carry sensory information from our body (e.g. touch of the skin) to the spinal cord and brain. The sensory nerve fibres that transmit painful information are meant to be silent unless we injure ourselves. However, in conditions, like osteo- and rheumatoid arthritis, they are known to spontaneously emit signals. This is thought to be an important cause of chronic pain. Indeed, it is believed to be a likely reason for flare-ups and spontaneous pain attacks, which are extremely disruptive to individuals living with arthritis.

Despite this, we understand very little about how these spontaneous signals arise. Most importantly, we do not actually know which type of peripheral nerve fibres (there are many different types) are primarily responsible. Without this information, we cannot develop specific treatments that are aimed just at the malfunctioning nerve fibres.

Our proposal aims to change this. We are working with a recently developed imaging technique that allows us to record spontaneous signals in hundreds of nerve fibres at the same time, rather than having to sample them one-by-one - as was necessary previously. With this technique and other recently developed tools, we can determine exactly which types of nerve fibres develop spontaneous signals in animal models of arthritis. We will then use chemical tools to specifically silence the spontaneous nerve fibres and assess whether this reduces pain in our models.

At the end of our work, we will have fully characterised the peripheral nerve fibres which are responsible for spontaneous pain. We will share this information with drug companies to help their efforts to develop the improved pain killers that millions of individuals so desperately need.

Technical Summary: Musculoskeletal pain conditions are a leading cause of disability and are poorly treated by existing analgesic medications. We know that pain in these conditions can be effectively treated by blocking sensory afferent input in the periphery. This is evidenced, for example, by the acute analgesic effects of local anaesthetic block or by the attenuation of pain following joint replacements in osteoarthritis. However, a non-selective block of all afferent fibres (e.g. via perineural local anaesthetic application) is not viable due to safety concerns. Thus, our proposal will focus on determining which peripheral afferents become sensitised in musculoskeletal pain to facilitate the development of more targeted analgesic treatments. We will focus on spontaneous sensory neuron activity because this has been recorded from nociceptors of individuals living with chronic musculoskeletal pain, as well as in animal models.

Our proposal will use cutting-edge optical imaging technologies, which allow the study of spontaneous firing in afferents in models of musculoskeletal pain. We will identify which types of sensory neurons become spontaneously active using a combination of functional, anatomical and genetic strategies. Once we have identified the major subpopulation of neurons where spontaneous activity resides, we will use a chemogenetic strategy to silence this population and will assess impact on pain behaviours. To understand the relationship between spontaneous activity and rodent behaviour in musculoskeletal pain,

we will optimise the use of Miniscopes, so that we can record activity from DRG neurons in freely behaving awake animals.

Our proposal will reveal which sensory neuron class we should target in order to eliminate spontaneous activity and consequent pain behaviours. This will help refine any analgesic drug development strategies that are currently being developed for the treatment of musculoskeletal pain.

Professor Clare Bryant, University of Cambridge (MR/W027240/1, 36 months, £381k)

- **MICA: Towards targeted treatment for complex regional pain syndrome through determination of the underlying molecular mechanisms**

Patient insight partner
Pauline Pitcher

Summary: Complex regional pain syndrome (CRPS) is a chronic pain condition that can occur after an injury, such as a fracture and results in localised pain and swelling. CRPS spontaneously resolves within a year in 20-30% of cases, but after this time it becomes chronic and rarely improves. Treatment options for CRPS patients, as with many other forms of chronic pain, are very limited, untargeted, and often have significant side effects. It is, therefore, important to understand why CRPS develops and the underlying mechanisms involved in order to try and develop more effective drugs targeting pain and inflammation and improve lives of those living with CRPS.

We can often gain insight into why a disease develops by looking at changes found in genes (mutations), which in turn helps design new drugs, such as pain killers (analgesics). For example, changes in a gene called nerve growth factor, were found in a family unable to feel pain (inherited as a very rare disease), and this led to the discovery of a new class of analgesics that is particularly helpful against arthritic pain. We wanted to see if patients with CRPS had mutations in their genes, compared to non-sufferers, which could alter their susceptibility to the condition. We found that CRPS patients were more likely to have mutations in four genes compared to people without CRPS. Each of these mutations was hypothesised to cause alterations of protein function. We, and others, have shown these four genes are expressed in monocytes and/or macrophages (cells that engulf foreign particles and cause inflammation). Importantly, all four genes are linked to the NOD-, LRR- and pyrin domain-containing protein 3 (NLRP3) inflammatory pathway that senses foreign particles and removes them by causing release of inflammatory molecules and cell death. These data suggest that the NLRP3 inflammasome pathway is likely to be important in the pathology of CRPS.

In this proposal we will investigate how the CRPS-mutated genes affect the activity of the NLRP3 inflammasome. In order to do this, we will introduce the gene mutations into a human monocyte cell line (THP-1 cells). Next, we will determine if these mutations alter the activity of the NLRP3 inflammasome by measuring the production of inflammatory molecules and cell death in THP-1 cells. We will validate the THP-1 model by measuring inflammasome activity in cells from healthy individuals with the CRPS-associated mutations. Our next step will be to determine how these changes in NLRP3 inflammasome activity influence pain neuron activation as a method to model the excessive pain seen in CRPS. Lastly, we will test how NLRP3 modulators affect pain and inflammation in order to translate our findings towards the development of new drugs for CRPS.

We anticipate our research will lead to the identification of a new class of non-addictive pain-killers that will reduce inflammation and pain to stop the chronic stage of CRPS from developing. This would have very significant effects on pain management, inflammation and the depression, morbidity and mortality that are a consequence of prolonged severe pain.

Technical summary: In order to understand the genetic basis for complex regional pain syndrome (CRPS), we recruited two patient cohorts and discovered that four SNPs, each in a different gene, were enriched in CRPS cohorts compared to healthy controls. All four genes are linked to the NOD-, LRR- and pyrin domain-containing protein 3 (NLRP3) inflammasome pathway. We therefore hypothesise that the SNPs identified cause aberrant activation of NLRP3 to promote the development of the chronic inflammatory pain seen in CRPS. In order to study CRPS, we will initially generate a cellular model by the

introduction of each SNP allele into a monocyte cell line THP-1 using CRISPR/Cas9 gene editing (already achieved for one SNP).

Macrophages are highly plastic cells and exist in at least three different states based on environmental cues: naïve M0, pro-inflammatory M1 and anti-inflammatory M2. The transition between M1 and M2 cells is important in wound resolution and healing. We will identify whether the CRPS mutations alter inflammasome activity in macrophages in different activity states by differentiating THP-1 cells harbouring the CRPS SNPs into M0, M1, M2 and M1>M2 states. Inflammasome activity will be determined by measuring pro-inflammatory cytokine secretion (interleukin IL-1B, IL-18) and gasdermin D- driven cell death following activation of NLRP3. These results will be validated in primary macrophages taken from healthy controls with the CRPS-associated SNPs. We will investigate whether the CRPS mutations alter the cross talk between pain and inflammation by measuring the effect of inflammatory mediators released by macrophages on nociceptor sensitisation. We will develop a co-culture system with macrophages and nociceptors differentiated from embryonic stem cells. Nociceptor activity will be measured using electrophysiology. Lastly, we will investigate the effect of inflammasome modulators, provided by NodThera, on the pain and inflammatory responses in the co-culture model.

Dr Annina Schmid, University of Oxford (MR/W027003/1, 36 months, £569k)

- **Factors predicting the transition from acute to persistent pain in people with 'sciatica' - the FORECAST study**

Co-Investigators:

Professor Irene Tracey	University of Oxford
Professor Jeremy Fairbank	University of Oxford
Dr Stuart Clare	University of Oxford
Dr Kathryn Martin	University of Aberdeen
Dr Georgios Baskozos	University of Oxford
Dr Fay Probert	University of Oxford

Patient insight partners:
Christine Price
Claire Robinson

Summary: Sciatica is a term used to describe painful symptoms affecting the hips, buttocks and legs. It is caused by injured or irritated nerves in the lower back. Sciatica pain can feel hot, sharp, or electrical in nature. Sciatica can also cause weakness and pins and needles. Sciatica is a complex condition and the pain may be caused by different mechanisms in different patients. We still do not fully understand the exact mechanisms of sciatica pain.

Sciatica is a very common condition and can have a devastating effect on everyday life. For example, some patients lose their independence and need help with day-to-day tasks such as dressing. Sadly, approximately one in three patients with sciatica develop persistent sciatica pain.
We currently do not understand why some patients develop persistent sciatica pain and why some recover. Previous research has demonstrated that usual clinical findings (e.g., presence of depression or changes on a standard MRI scan) cannot predict persistent sciatica pain. Therefore a different approach is required to identify patients who may develop persistent sciatica pain. This is the goal of the FORECAST study.

Previous studies only included a short clinical examination. Our study is different. We will perform a detailed set of tests (see below). Our ambition is that the detailed tests can predict who will develop persistent sciatica pain. The questions we plan to answer in the FORECAST study are:
1. Can the detailed tests identify patients with similar mechanisms causing their sciatica pain?
2. Which of these detailed tests predict persistent sciatica pain?

The FORECAST study is performed by a team of medical doctors, neuroscientists, statisticians and magnetic resonance imaging (MRI) specialists at Oxford University. The team also includes patient partners. They help us to make sure our study is useful for patients.

We will invite 180 patients with recent onset of sciatica pain (<3 months) to participate in the FORECAST study. At their initial assessment, we will perform the detailed tests. This includes detailed sensory nerve testing (quantitative sensory testing). Quantitative sensory testing evaluates how well patients can feel different stimulations such as cold, warm or pressure. This tells us how well the nerve is working and how sensitive it has become. We will also include a precise set of questionnaires to evaluate the types of pain (e.g., burning or electric shock or achy) and emotional wellbeing. Emotional wellbeing can be affected when a patient has sciatica. For instance, some patients may be anxious or angry, and others may feel lonely and detach themselves from friends and family. We will also take a blood sample to look for signs of inflammation. In some patients we will use specialised MRI scans. These images are much more detailed and specialised than standard MRI scans. They allow us to evaluate the microscopic structure of the small nerves in the back, which is not possible with standard MRI scans. These detailed tests will provide us with a good picture of the starting point of a patient's experience of sciatica.

We will then contact patients again three months and one year later, to ask whether they still have sciatica pain. We will use statistics to identify patients with similar mechanisms causing their sciatica pain. We will also examine which tests predict persistent sciatica pain.

The results of the FORECAST study will help us better understand the complexity of sciatica and who will develop persistent pain. Our findings will also help future research. For example, future studies can examine whether we can prevent or reduce persistent pain by giving more specific treatment to patients who the FORECAST study shows are likely to develop persistent sciatica pain. We hope that the results of the FORECAST study will help reduce suffering and improve quality of life for patients with sciatica.

Technical summary: Sciatica is a common condition (43% lifetime prevalence) and is associated with higher levels of pain, disability, poorer quality of life, and increased use of health resources compared to low back pain alone. Although many patients recover, a third develop persistent sciatica symptoms. Unfortunately, it remains unclear, why some patients develop persistent sciatica as none of the traditionally considered clinical parameters (e.g., symptom severity, routine magnetic resonance imaging, depression) are consistent prognostic factors. Given the failure of traditional variables as prognostic factors, a novel approach is required to understand who may develop persistent pain.

FORECAST includes a prospective prognostic cohort study that aims to:
1. Explore mechanism-based subgroups in patients with sciatica, using deep phenotyping.
2. Investigate whether a mechanism-based approach can identify factors that predict pain persistence in patients with sciatica.

We will evaluate prognostic factors for pain persistence in n=180 patients with acute/subacute sciatica. Prognostic variables will be assessed at baseline and outcome at 3 and 12 months. This will include self-reported sensory and psychosocial profiles, quantitative sensory testing, blood inflammatory markers and advanced neuroimaging. We will use principal component analysis followed by clustering methods to identify subgroups. Univariate associations and machine learning methods optimised for high dimensional small datasets will be used to identify the most powerful predictors and model selection/accuracy.
This project is based on our strong feasibility data and has been shaped by in-depth patient involvement. The results will provide crucial information about the pathophysiological drivers of sciatica symptoms and identify prognostic factors of pain persistence. This will ultimately facilitate patient stratification and streamline management pathways to increase quality of life for patients with sciatica.

Dr Simon Wyn Jones, University of Birmingham (MR/W026961/1, 36 months, £886k)

- **MICA: Synovial fibroblast pain pathotypes: A roadmap to understanding and targeting the complexity of patient-reported joint pain in osteoarthritis**

Co-Investigators:

Mr Edward Davis	Royal Orthopaedic Hospital NHS Fdn Trust
Professor Georgios Gkoutos	University of Birmingham
Professor Victoria Chapman	University of Nottingham
Dr Federico Dajas- Bailador	University of Nottingham
Professor Mark Lindsay	University of Bath

Summary: Background
Osteoarthritis (OA) is highly painful joint disorder and a leading cause of disability. Current pain-relief medications are only minimally effective and are associated with side effects over the long term. One of the challenges in developing a more effective pain-relieving OA drug is understanding of the complex underlying mechanisms of OA joint pain.

Importantly, inflammation of the synovial joint lining tissue (termed synovitis) is associated with increased pain severity in knee OA patients, and we have shown that the cells which reside within the synovial tissue (known as synovial fibroblasts) regulate the inflammatory environment of the joint by secreting factors that promote inflammation.

To investigate the cellular and molecular basis for the relationship between synovitis and OA joint pain, we involved knee OA patients in a study where we mapped the anatomical location of synovitis by MRI and of patient-reported pain by asking patients to mark on a diagram of the knee joint, where they felt most pain and where they felt least pain. We then collected synovial tissue samples from both sites of patient-reported pain and sites of no pain. Significantly, the degree of synovitis was closely associated with the pattern of patient-reported pain, and furthermore that synovial joint lining tissue at the site of patient-reported pain contained distinct populations of synovial fibroblast cells, that promoted inflammation and secreted factors that promoted the growth and survival of nerve cells. Since the synovial joint lining tissue contains numerous nerve endings we believe that the activity of these pain-associated synovial fibroblast cells is a major contributor to OA joint pain. We hypothesise that therapeutics that reduce the activity of these synovial fibroblasts will alleviate joint pain in patients with knee OA.

Aim
The overarching aim of the proposal is to map the relationship between the presence and location of pain-associated synovial fibroblasts with patient-reported pain and synovitis in a larger patient cohort, and then to determine whether modulating the activity of these cells using a novel gene silencing therapeutic reduces sensory nerve function and alleviates pain.

Experimental Plan
To address these aims we will expand our current knee OA patient pain study to 82 patients (41 with early OA disease and 41 with end-stage disease). The anatomical location and degree of synovitis in the joint will be measured by MRI and patient-reported pain severity and pain location captured by questionnaires and completion of the anatomical pain map.
Blood and joint fluid will be collected, and synovial joint lining tissue collected from sites of patient-reported pain and no pain.

The expression and spatial location of pain-associated synovial fibroblast gene signatures will be measured, and their relationship mapped to synovitis, blood and joint fluid biomarkers of inflammation and patient-reported pain determined. Gene expression data of synovial tissue and fibroblast cells from sites of pain and no-pain will be analysed using computational modelling to build networks of the gene pathways that connect the activity of synovial fibroblasts with nerve cells in order to identify new candidate pain gene mediators. We will then design gene silencing therapeutics to target candidate pain genes, and examine the effect of these therapeutics on modulating nerve cell activity and in a mouse model of OA, determine the uptake of the therapeutic to the joint tissues and its effect on reducing joint inflammation and pain.

These experiments will provide the first evidence of whether targeting specific pain-associated synovial fibroblast cells in the joint can alleviate joint inflammation and pain. In summary, this project will advance our understanding of the role of synovial fibroblasts in inflammatory pain and lay the groundwork for the clinical development of a new pain-relieving therapeutic for OA patients.

Technical summary: Current analgesics to treat OA joint pain lack efficacy and are associated with adverse side effects. We have found in knee OA patients that inflammation of the synovial membrane (synovitis) is associated with the pattern of patient-reported pain, and that synovial tissue from sites of pain exhibits a differential phenotype with distinct synovial fibroblast subsets which mediate fibrosis, inflammation and neuronal growth and survival. We hypothesise that the pathotype of these pain-

associated fibroblasts promote neuronal growth and sensitization of joint nociceptors and drive joint pain in knee OA patients.

To address this, we will collect patient-reported pain data (anatomical pain maps and questionnaires), synovitis pathology (MRI), blood, synovial fluid and synovial tissue biopsies from sites of patient-reported pain and sites no pain from n=82 knee OA patients (41 early OA; 41 end-stage OA) undergoing orthopaedic surgery. We will map the synovium expression and spatial location of fibroblast pain pathotype gene signatures and determine the relationship to patient-reported pain and synovitis. RNAseq and proteomic data from OA patient synovium and fibroblasts from pain/no-pain sites will be integrated and computational network models built of the underlying cellular mechanisms between synovial fibroblasts and neurones to identify and validate pathways and candidate targets that mediate nociceptor activity. Antisense oligonucleotides (ASOs) will be designed to silence candidate genes and their efficacy in modulating the fibroblast pain pathotype and reducing the growth and sensory function of neurons determined. Next, the intra-articular delivery of an ASO into the synovial joint tissues and its analgesic efficacy in an experimental model of OA pain will be evaluated. The integration and curation of these multimodal patient datasets and tissue samples will provide new insights into the cellular mechanisms that mediate inflammatory pain.

Professor Anthony Pickering, University of Bristol (MR/W027925/1, 36 months, £662k)

- **SenseCheQ: Community-based sensory testing for early identification of Chemotherapy Induced Peripheral Neuropathy.**

Co-Investigators:

Professor Roger Whittaker	Newcastle University
Professor Anthony O'Neill	Newcastle University
Professor Lesley Colvin	University of Dundee

Researcher-Co-Investigator:

Dr James Dunham	University of Bristol

Patient insight partner:

Mr Alan Young	SenseCheQ PPIE Lead

PPIE co-ordinator:

Dr Gillian Martin	Tayside Clinical Trials Unit

Summary: Chemotherapy is an important treatment for cancer. Unfortunately, it also causes side effects, including chronic pain and changes in the sense of touch. This is due to nerve damage. In up to 1 in 3 people, these changes last longer than 6 months, causing misery, disability and adding to the burden of cancer survivorship.

These changes are vague to begin with, yet early detection is important, as reducing the chemotherapy dose is the only way to reduce the severity of symptoms. Reducing the dose, however, must be balanced against the possible risk of the treatment being less successful. This highlights how important it is to detect these signs early and accurately.

One way of detecting these changes is with careful testing of the skin's senses. This is achieved by asking patients when they feel small changes in temperature or vibrations and then comparing that to readings taken before chemotherapy was started. Such tests are well known in the pain research community where they are called Quantitative Sensory Testing (QST). Frustratingly, QST takes a long time to perform, is expensive and needs the patients to keep coming into clinic during a very vulnerable time in their cancer treatment. These issues mean that this formal QST is not suitable for routine clinical use.

Our team of patients, pain doctors and engineers believe that we can deliver a simple, cheap and effective sense testing kit, that we are calling SenseCheQ. This will enable patients to check the health of their nerves, at home, during their chemotherapy. We will design SenseCHeQ to be sensitive enough to

detect early changes, potentially before the patient notices any symptoms, to enable personalised treatments that will maximise the success of cancer treatment, whilst minimising the risk of chronic pain and loss of sensation.

To deliver SenseCheQ we need to complete four complementary workplans.

Workplan 1 (UK wide) will be led and driven by patient partners to ensure that SenseCHeQ is user friendly and meets the needs of patients. This workplan will also feed into the remaining workplans to ensure that patients remain at the centre of our focus.

Workplan 2 (Newcastle) will engineer solutions, by identifying and integrating off-the-shelf components into suitable 'wearables' and providing power and communications.

Workplan 3 (Bristol) will test these solutions in healthy volunteers. Initially, individual components will be tested alone and compared to commonly used sensory testing equipment. As designs progress, through testing and re-design cycles working closely with WP1 and WP2, this workplan will move to validating early SenseCHeQ versions in models of nerve damage, which will cause temporary numbness or pain, again in healthy volunteers.

Workplan 4 (Dundee) will perform a feasibility study in patients. Workplan 4 will synergise with PAINSTORM, another Versus/MRC advanced pain discovery platform program of work that is seeking to harmonise assessment of pain caused by nerve damage, not just damage caused by chemotherapy. PAINSTORM are funded to perform QST on patients as they move through their chemotherapy. We will ask some of these patients to use SenseCHeQ at home at the same time. We will be most interested in how patients get on with the device - is it easy to use? Robust? We will also compare SenseCHeQ to the QST results to estimate its ability to detect early neuropathy.

If successful, we will apply for further funding to confirm these findings and thus move SenseCHeQ towards a clinically useful tool empowering patients to monitor their own nerve health, at home, minimise their risks of developing chronic pain and numbness and enabling delivery of truly personalised cancer treatment.

Technical summary: Chemotherapy induced peripheral neuropathy (CIPN) affects 30% of patients receiving chemotherapy to treat cancer. CIPN presents with numbness and pain in a glove and stocking distribution. There is no effective preventative or therapeutic treatment, and the only mitigation is chemotherapy dose reduction/switching, which is a difficult clinical decision.

Detection of CIPN relies on patient report and clinical examination which lacks sensitivity in early CIPN. Quantitative Sensory Testing (QST) is the 'gold standard' assessment of sensory function but it is not in routine clinical use for pragmatic reasons. To enable early detection of CIPN we propose to develop an easy-to-use sensory testing device "SenseCheQ" to allow patients to monitor their nerve health at home. This will provide early, accurate assessment of sensory function to detect nerve damage and potentially avoid chronic neuropathic pain.

This will be achieved with iterative and modular device development cycles through collaboration with bio-electronic engineers and patient partners. SenseCheQ will safely deliver automated QST protocols (warmth detection, cool detection and vibration detection), with simplified data capture and be able to transmit that information remotely.

This project will synergise with the APDP PAINSTORM consortium, who will prospectively recruit a cohort of patients due to undergo chemotherapy. This cohort will undergo formal QST during their treatment as well as clinical monitoring. A subgroup of these patients (n=20) will have home QST with the SenseCheQ, within a feasibility study, with measures obtained before treatment starts and at intervals during their chemotherapy. This parallel collection of QST data will enable direct comparison of the device with clinical sensory phenotyping. The project maps the path for delivery of a novel device capable of giving patients and clinicians better and earlier information to optimise and personalise their cancer treatment.

Professor Daniëlle van der Windt Keele University (MR/W026872/1, 36 months, £999k)

- **High Impact Chronic Pain and UK Biobank: presentation, transitions and targets for intervention**

Co-Investigators:

Dr Milisa Blagojevic	Keele University
Professor George Peat	Keele University
Professor Christopher Eccleston	University of Bath
Professor Edmund Keogh	University of Bath
Dr Emma Fisher	University of Bath
Professor Gary Macfarlane	University of Aberdeen
Professor John McBeth	The University of Manchester
Dr Elaine Wainwright	University of Aberdeen
Dr Emma Parry	Keele University

Patient insight partners:

Patient partner group, represented by Robert Taylor - Member Research User Group, Keele University

Summary: Nearly half of UK adults have pain in their muscles or joints lasting longer than three months (chronic pain). While most people manage well, about 25% of people have pain that has far-reaching, negative impacts on their lives, leading to disability, distress, social isolation, and high healthcare needs. Chronic pain presents in many different ways, and it is not clear why some people experience such 'high impact chronic pain' whereas others don't. We also don't know yet why some people improve, whereas others experience ongoing or even increasing impact from their pain.

AIMS OF THE RESEARCH

We aim to find out the possible causes (or 'risk factors') of high impact chronic pain. This will inform the design or choice between selfcare or treatment options that may more effectively address people's individual needs. Early care or support that target specific risk factors may also help prevent the onset of high impact chronic pain.

Specifically, we plan to use existing data from a large, national study of older adults (UK Biobank) to

1. Identify groups of people with distinct patterns of high impact chronic pain who may have different outcomes and different care needs
2. Understand the reasons why high impact chronic pain affects some more than others, and why this can change over time, focusing on the influence of:
    a. psychological factors, including mood, attention, memory, ability to problem-solve
    b. trauma or traumatic events that may occur at different times in people's lives (e.g., fracture, bereavement, sudden illness)
3. Identify selfcare or treatment options that can reduce the influence of these risk factors for people with chronic pain.

DESIGN AND METHODS

We will use data from UK Biobank, which includes health information from 500,000 people in the UK, who were aged 40-69 years when data collection started in 2006-2010. A questonnaire on the nature and impact of chronic pain was completed by over 173,000 participants in 2019. A second pain questionnaire will be sent to participants in 2023.

The study team will work together with patient partners on the following three workpackages:

(1) We will use state-of-the art statistical and artificial intelligence methods to identify subgroups with distinct patterns of chronic pain, and describe its impact on daily activity, mood, and healthcare use.

(2) We will use data from questionnaires and healthcare records collected over a period of more than 15 years, to investigate the role of factors that may explain changes over time in the impact of chronic pain, including (a) mood, attention, memory, problem-solving, and (b) physical or psychological trauma.

(3) Based on the results from (2) we will identify factors that can be effectively targeted by selfcare or treatment. We will then estimate how much benefit could be achieved if such interventions would successfully be offered to people with chronic pain.

PATIENT AND PUBLIC INVOLVEMENT

Research questions have been informed by patient partners, who shared their life stories, and suggested factors that may explain the development of chronic pain, and how impact can change over time. They will continue to be involved in defining and prioritising factors for analysis; reviewing interventions; interpreting results; formulating key messages and dissemination of findings.

COMMUNICATING FINDINGS
We will work together with patient partners and clinical advisors to
- Publish results in scientific journals and during (international) pain conferences
- Share our findings with other research teams and the PPIE network in APDP, and with UK Biobank
- Engage with our existing networks, including Versus Arthritis, Public Health England, NHS partners, research funders
- Design infographics, visual animations, or YouTube videos to communicate key findings in a way that is suitable for a wide audience
- Hold webinars to discuss findings and implications

Technical summary: BACKGROUND: Chronic pain (CP), lasting more than 3 months, represents a major global burden in terms of years lived with disability and economic impact due to healthcare use and work absenteeism. Approximately 25% of those with CP (10% of the adult UK population) experience far-reaching impact from CP, including widespread disability and distress, social exclusion, and high healthcare needs, recognised as High Impact Chronic Pain (HICP).

AIMS AND OBJECTIVES: We propose an interdisciplinary programme of research to inform prevention, selfcare and treatment options for HICP. Specific objectives are to
1. Use clustering methods to identify and describe phenotypes of HICP, and validate these in other datasets
2. Use causal inference methods to investigate the role of (a) cognitive and affective factors, and (b) trauma or traumatic events in explaining transitions of HICP
3. Identify potential targets for prevention and treatment, and estimate expected benefit from individual or population level intervention

DESIGN AND METHODS: Working with patient partners we will use data from UK Biobank to investigate changes in the impact of chronic musculoskeletal pain over time. The presence of CP was assessed in all UKB participants at inclusion in 2006-2010. A detailed CP survey was launched in 2019 (completed by >173,000 individuals) and will be repeated in 2023. Additional data on risk factors, mediating and moderating factors will be extracted from existing surveys (e.g. lifestyle, work cogntive function, mental health) and linked healthcare records (trauma, sudden illness, comorbidity). Social determinants (e.g. age, gender, ethnicity, deprivation), will be taken into account in all analyses.

DISSEMINATION: Open access publications, conference presentations, lay summaries, and infographics will be co- created with patient partners and clinical advisors, and shared within the research community, public health/clinical networks, and the wider public.

Professor Stuart Bevan, King's College London (MR/W027623/1, 36 months, £853k)
- **Pain Mechanisms in long-Covid**

Co-Investigators:

Dr Michael Lee	University of Cambridge
Dr David Andersson	King's College London

Summary: The COVID-19 pandemic has affected hundreds of millions of people and while many individuals recover fully about 1 in 10 suffer from long term symptoms known as long-COVID. We know already that some patients still experience symptoms after more than one year. Muscle and joint pain and fatigue are common symptoms of long-COVID along with breathing problems and loss of smell. Some patients also suffer from abnormal sensations (pins and needles and burning pain) which could be due to damage to some sensory nerves. The mechanisms underlying pain and abnormal sensations in

long-COVID are unknown and there are no effective therapies. Also there is currently no laboratory method to diagnose long-COVID. Our research combines patient and lab-based studies to understand why long-COVID patients suffer from pain To do this, we will characterise the qualities, intensities and location of pain in long-COVID patients, test whether there is damage to pain-sensing nerves, and measure the impact of pain and fatigue on daily life.

The pain and fatigue seen in long-COVID resembles the symptoms found in patients with fibromyalgia, and we have very recently shown that the body's normal defence mechanism, the immune system, is responsible for pain in fibromyalgia patients. Immune cells produce antibodies which normally help us to fight infections. Our results showed that fibromyalgia patients produce antibodies that attack their own bodies and stimulate pain sensing nerves. We will therefore test to see whether antibodies are also responsible for pain and fatigue in long-COVID. To do this we will purify antibodies from people with long-COVID, inject them into mice and measure if the mice develop symptoms of pain and fatigue. We will also examine if long-COVID antibodies obtained from people who have sensory nerve damage, targets the similar sensory nerves in the mice. We will find out at a molecular level what part of the nerves are being targeted and begin to understand how the antibodies have their effects. Our work will improve management of long-COVID and potentially provide a laboratory method for diagnosing this condition.

Technical summary: About 10% of patients infected with COVID-19 have persistent symptoms (long-COVID) that can last for over a year. These include musculoskeletal pain and fatigue as well as paresthesias (tingling, burning pain) and symptoms of dysautonomia (e.g. breathlessness, chest pain). Our proposal will investigate if small fibre neuropathy (SFN) and autommunity are responsible for these neurological symptoms. We will recruit patients who have A) pain and fatigue, B) fatigue but no pain, C) symptoms resolved after COVID, and D) asymptomatic with no COVID infection. Validated questionnaires will be used to characterise the location, quality and intensity of pain and to identify patients whose pain and symptoms suggest (SFN). A diagnosis of SFN will be ascertained by quantitative sensory testing and by histological analysis of intraepidermal nerve fibre density (IENFD) in skin biopsy samples.

In addition, we will assess the presence of dysautonomia by measuring cardiovascular parameters in the laboratory at baseline and under provocative testing. We will assess the impact of pain on physical activity during daily life using wearable sensors which capture movement, posture and autonomic data. Blood will be collected from patients for antibody studies. IgG from patient blood will be isolated and administered to mice by intraperitoneal injection. Mice will be tested for their sensitivity to mechanical and thermal stimulation before and then daily after antibody administration. Effects on spontaneous activity will also be assessed as this may be affected by pain/fatigue. The presence of SFN in antibody treated mice will be assessed by measurements of skin IENFD. Fatigue will be assessed by performance on treadmill and rotarod tests. These experiments will show if we can passively transfer symptoms of pain from patients to mice. Immunohistochemistry and Western blotting will be used to identify the cellular binding sites and likely epitopes for patient IgG.

Dr Ben Seymour, University of Oxford (MR/W027593/1, 36 months, £989k)

- **The Role of Learning in Chronic Musculoskeletal Pain**

Co-Investigators:

Dr Flavia Mancini	University of Cambridge
Professor Timothy Denison	University of Oxford
Dr Katja Wiech	University of Oxford
Dr Anushka Soni	University of Oxford

Researcher-Co-Investigator:

Dr Suyi Zhang	University of Oxford

Patient insight partners:

Sally Ainsworth
Nicola Warrick

Summary: Aims and objectives:
Despite its prevalence, we still don't know why some people get chronic pain, and others do not. One influential idea is that the processes in the brain that normally allow us to adapt to an injury and recover from it, are used excessively, meaning that pain is exaggerated and prolonged beyond what is necessary. This 'maladaptive brain learning' hypothesis, in its various forms, is a popular model of chronic musculoskeletal pain. However, evidence is currently limited by the lack of sufficient tools required to measure and quantify learning. The project addresses this by implementing a novel set of experimental tools based on a basic science understanding of how learning works in the brain. We will use these tools to study outcomes in two complementary longitudinal studies: i) recent onset lower back pain and ii) fibromyalgia patients embarking on a multidisciplinary treatment (MDT) program. These tools will also be disseminated openly across the APDP and further afield.

Over the last year, we have established a unique partnership with people with lived experience of pain, physiotherapists, clinicians and engineers at Oxford and Cambridge, to create a core toolkit for pain-related learning evaluation. This is a set of tablet games that people play at home. People can also use their tablet camera to record specific physio exercises which we use to reconstruct and quantify the impact of pain on movement. By using a patient-led design at the outset, the tools are user-friendly, engaging, and maximise accessibility. This provides the means to test the maladaptive learning hypothesis in a broad range of clinical cohorts.

Data to be collected:
We will pursue 3 integrated workstreams:
i) Development and dissemination of an open data analysis platform to accompany the experimental toolkit.
ii) Data collection in a cohort of 140 patients with recent onset (acute) lower back pain, presenting via NHS GP services, to predict outcomes at 12 months
iii) Data collection in a cohort of 80 patients with fibromyalgia undergoing an NHS MDT program.
Both clinical cohorts aim to look for the ability of learning metrics to predict clinical outcomes, and validate the findings with neuroimaging. All data will be made open via the ADPD datahub.

Potential benefits:
The main scientific outcome will be the identification and characterisation of how learning correlates with chronic pain outcomes. This is important because such mechanisms directly imply treatment targets, which can be realised using the non-pharmaceutical interventions (cognitive and physical rehabilitation). This therefore provides a springboard for treatment innovation across the APDP and partners.
These benefits are facilitated by the patient-led design of the tools. Practically, they are easy-to-implement, require minimal expertise, and come with open data analysis pipelines and collaborative support if required. This builds a UK-based network that will capitalise on the innovation and expertise across the whole APDP. Furthermore, the data generated by this infrastructure opens up new opportunities for bioinformatics and related applications (e.g. in clinical stratification and outcome prediction).

Legacy and sustainability.
The very nature of data collected, being based on a common set of tools, will seed a database that will grow over time. This is self-sustaining because the statistical 'power' of the database increases as more data is added, permitting comparative analyses of individual datasets to a wide-range of other conditions. The tools also provide a technological backbone that can be developed and refined over time, as new insights, tools and techniques become available. In effect once the systems are in place, they grow organically over time. This is likely to be realised in new therapies, potentially as early as 5 years, given its potential to be integrated with existing technology-based treatment methodologies.

Technical summary: We address whether maladaptive learning systems contribute to the maintenance of chronic pain. This has been difficult to answer because learning comprises a set of complex, interacting processes, and hence difficult to evaluate and quantify. Based on computational models of learning, we have designed a suite of tasks and analysis tools that probe domain- general value-based and sensorimotor learning. These are implemented online as a set of tablet-based computer games that can be applied easily and widely in observational or longitudinal clinical studies in domestic settings.

We will study whether learning metrics can predict clinical outcomes in: i) a longitudinal study of patients with recent onset low back pain, and ii) fibromyalgia patients undergoing an NHS MDT program. We will measure neural changes (using resting-state fMRI) and physical outcomes using a novel video-based tool to quantify movement during physiotherapy exercises.

The tools, analysis pipeline and data will all be made free and openly available to the research community. Within the APDP open Datahub, data from our and other partners can be pooled across various conditions, allowing further comprehensive characterisation of how learning is linked to chronic pain.

We adopt a systems engineering approach to the development of our tools, in which patients have and will continue to have a fundamental role in the design process itself, necessary to ensure the usability and relevance of the framework to the lived experience of pain. This builds the joint vision of researchers and patients to develop an open platform for integrated multidisciplinary evaluation and treatment.

The proposed project will have a lasting impact: building a self-sustaining data generating infrastructure focused on learning mechanisms; providing a technological platform to support innovation in non-pharmacological treatment; and building a UK pain neuroscience and engineering community focused on chronic pain

Professor Katy Vincent, University of Oxford (MR/W02697X/1, 36 months, £976k)

- **Understanding the Role of Adolescent Dysmenorrhoea as a risk factor for the transition to chronic Pain (RoAdPain)**

Co-Investigators:

Professor Krina Zondervan	University of Oxford
Professor Mina Fazel	University of Oxford
Dr Gemma Sharp	University of Bristol
Dr Sharon Dixon	University of Oxford
Ms Emma Cox	Endometriosis UK

Researcher-Co-Investigators:

Dr Kate Stein	University of Oxford
Dr MaryAnn Noonan	University of Oxford

Summary: Chronic pain is defined as pain that lasts for more than 3 months. It is really common, affecting up to 30% of people worldwide with impacts on all areas of life. Chronic pain is difficult to treat once it has developed. Therefore, understanding which people might be at risk of developing chronic pain and protecting them from it starting, would be a really positive step forward.

We know that women are more likely to develop almost all types of chronic pain than men. We start to see a sex difference in chronic pain after puberty, suggesting that changes happening at this time may be contributing to this increased risk.
One important change that happens at this time is periods starting. Despite periods often being very painful, period pain has traditionally been dismissed as "normal" and something girls must learn to live with. However, in adult women with period pain we see many differences across a range of body systems when compared to women without period pain. These include increased sensitivity to pain; increased sensitivity of the bladder, bowel and womb; altered brain structure and function; and altered stress responses. Similar changes to those seen with period pain can be seen with other chronic pain. We don't know whether these changes are caused by repeated or continuous pain or if they are part of the reason why chronic pain develops, or a combination of both.

Our project has 3 aims:
1. To understand whether period pain during adolescence increases the risk of developing chronic pain as a young woman.
2. To see whether the differences in other body systems described above in adult women with period pain are also seen in girls in the first few years of having periods.

3. To find out whether there are any factors present in childhood that increase the risk of period pain developing in the first few years of having periods.

We will use two different approaches to answer these questions. For questions 1 and 3, we will use data collected in the Avon Longitudinal Study of Parents and Children (ALSPAC). ALSPAC is a large research resource which has been collecting data from children born in the early 1990s and their parents for almost 30 years. This dataset includes:

- detailed assessments of chronic pain at the ages of 17 and 26 years
- questions about periods every year between the ages of 8 and 17 years
- a wide variety of childhood assessments
- information about the mothers health and wellbeing
- genetic information.

For objective 2 we will recruit a group of adolescent girls, including those with and without period pain, 1, 3 and 5 years after starting their periods. With their agreement, we will undertake tests to understand more about their body systems and pain. All the tests we will use have been previously used in children and adolescents and do not involve significant pain.

We hope this work will reduce the risk of both adolescent girls and adult women suffering with period pain and other chronic pain conditions. We will use what we learn about the long term risks of period pain, including how long it takes for experiencing period pain to increase your risk of chronic pain, to make sure period pain is taken seriously and to produce advice and guidance for those with period pain, health professionals, policy makers and educators. A better understanding of the risk factors for developing period pain when periods start will let us identify girls at risk of early-onset period pain, ensuring they are educated and empowered to seek treatment early. Our findings on how period pain leads to chronic pain will be developed into novel strategies to prevent chronic pain, including future work with other researchers and drug companies.

The research team come from a wide variety of backgrounds and have strong links with key stakeholders and industry partners. We will use these existing networks to ensure our findings are widely publicised (including to the public) and developed to maximise benefits.

Technical summary: Chronic pain affects up to 30% of people worldwide with significantly reduced quality of life and associated socioeconomic cost. Once established it is difficult to treat and therefore preventative strategies are urgently required. Being a woman is a risk factor for chronic pain and this sex difference emerges after puberty. Periods, which start during puberty, are themselves painful for many women. Alterations in a variety of pain-relevant body systems have been demonstrated in adults with dysmenorrhoea, including sensitivity to noxious stimuli, visceral sensitivity, brain structure and function and activity of both the hypothalamic-pituitary-adrenal axis and the autonomic nervous system. We hypothesise that the experience of dysmenorrhoea during adolescence is a risk factor for the development of chronic pain, with the potential to act through a variety of pathways. We will address this in two ways. Epidemiological analyses will use the ALSPAC data, a resource that has been collecting prospective data from children born in the early 1990s and their parents for almost 30 years, to determine whether adolescent dysmenorrhoea is an independent risk factor for chronic pain in young women. We will additionally explore whether factors present in childhood increase the risk of dysmenorrhoea early in menstrual life.

Alongside this work, an experimental study using psychophysical and neuroimaging assessments will determine whether dysfunction in pain-relevant systems can be demonstrated in adolescent girls in the early years of menstruation. We anticipate results with clear and direct implications for adolescent menstrual health and for drug target discovery of relevance to chronic pain in women. We will use findings from this project to highlight the importance of dysmenorrhoea and optimise its timely management. Ultimately we hope that this work will reduce the risk of both adolescent girls and adult women suffering with dysmenorrhoea and other chronic pain conditions.

Professor Ana Valdes University of Nottingham (MR/W026813/1, 36 months, £355k)

- **Molecular signatures of endocannabinoid induced pain relief in humans: lifestyle interventions, systemic and localised changes.**

Co-Investigators:

Dr Moira Taylor	University of Nottingham
Dr Stefan Kluzek	University of Nottingham
Dr Cristina Menni	King's College London

Researcher-Co-Investigator:

Dr Amrita Vijay	University of Nottingham

Patient insight partners:

Mrs Lorna Weir
Mr Terry Murphy

Summary: Endocannabinoids are molecules made by our bodies which are similar to some of the substances in marijuana. They regulate appetite, mood, sleep, muscle strength, inflammation and they also are involved in how strongly we feel and respond to pain. There have been efforts to generate new drugs that are similar to "endocannabinoids" to treat various diseases but because they are involved in so many functions these compounds end up having unforeseen side effects.

The microbes in our guts have been implicated in the levels of pain and inflammation that people experience. And physical exercises are well known to reduce the levels of pain experienced by people with chronic conditions such as arthritis.

We propose to understand (identify) the molecules that are involved in all of these related processes linked to pain by asking people to take fibre and or do exercise for six weeks, both of which increase endocannabinoid levels

We will measure levels of pain and pain sensitivity, peoples' mood, and take bloods to measure endocannabinoids, gut microbes, substances produced by microbes that alter inflammation, inflammatory substances, other pain related substances

The results of this study will help people with chronic pain in the following ways:

1. Understand the mechanisms by which lifestyle interventions like diet and exercise work and whether they can be added to each other to improve pain.
2. To measure how much of the effects of changes in endocannabinoids on pain reduction is linked to changes in mood or to changes in inflammation or to different levels of substances directly related to pain transmission and which genes in our bodies are involved
3. Understand if there are molecular factors that result in improved or reduced endocannabinoid changes and their effects on pain
4. Help develop new drugs to treat pain by understanding which mechanisms are modulated by the various receptors of drugs that target the endocannabinoid system

Technical summary: Modulation of endocannabinoids (ECB) in animal models has strong antinociceptive properties and use of cannabis appears effective in reducing neuropathic pain symptoms in humans. However, the pleiotropic effects of ECB receptors and the various feedback loops in the system make the prediction of undesirable central and peripheral effects of such compounds challenging, hindering the deployment of ECB targeting compounds to market. Endocannabinoid tone can also be modulated non-pharmacologically. Both physical exercise and alteration of the gut microbiome via dietary supplementation modify the endocannabinoid tone.

Strong clinical evidence that exercise interventions are effective in achieving clinically meaningful musculoskeletal pain relief. Observational and interventional evidence suggest that the gut microbiome is involved in pain intensity, progression and sensitivity for various types of pain.

We hypothesize (based on solid preliminary data) that the mechanisms by which these non-pharmacological interventions relieve pain are mostly mediated by modulation of the endocannabinoid system resulting in meaningful anti-inflammatory and antinociceptive effects

Using a factorial design of exercise and microbiome modulation of short chain fatty acids. We will perform extensive molecular phenotyping and characterisation of pain responses to these interventions and apply artificial intelligence/machine learning algorithms as we have done for other complex molecular traits. This will allow us to identify the key molecular mediators involved in effective responses to lifestyle interventions.

We will validate the pathways involved using independent cohorts such as TwinsUK, StepUp OA, IBEAT-OA with existing data on pain sensitivity and molecular markers.

Finally we will assess the direct relevance of ECs in the joint by assessing levels of ECs in synovial fluid from severe knee OA patients and their change with exercise/ gut microbiome modulating interventions