2024

Understanding pain in persons with endometriosis, evaluating the role of the hypothalamus and brainstem

https://hdl.handle.net/2144/51348

BOSTON UNIVERSITY

ARAM V. CHOBANIAN & EDWARD AVEDISIAN SCHOOL OF MEDICINE

Thesis

UNDERSTANDING PAIN IN PERSONS WITH ENDOMETRIOSIS, EVALUATING THE ROLE OF THE HYPOTHALAMUS AND BRAINSTEM

by

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B.S., University at Buffalo, 2022

Submitted in partial fulfillment of the

requirements for the degree of

Master of Science

2024

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DEDICATION

I would like to dedicate this work to my mom and my grandparents, Appachan and Ammachi.

ACKNOWLEDGMENTS

I am sincerely grateful to my PI, Dr. Holmes, for his invaluable advice and mentorship, continuous support throughout every stage of this thesis project, and fueling my interest in neuroimaging and pain research. I would like to thank my professor and mentor, Dr. Ratner for all her extra efforts in taking the time to ensure my understanding of different concepts, offering words of encouragement, and her enthusiasm that has instilled in me a desire to keep on learning and growing as a student.

I want to thank my lab mates, Amanda Dynak, Madison Dudek, Cameron Talbert, Angela Brillantes, John Lee, Emily Tomchak, Clara Moon, and Priya Patel for their friendship, moral support, and thoughtful insights and suggestions. I am so grateful I got to work alongside all of them. I want to thank my friends Victoria Rust and Kasey Joy for keeping my spirits up throughout this writing period, making me laugh, our late-night work sessions, and for their endless support throughout this entire program.

Finally, I would like to give a special thanks to my mom and my grandparents, Appachan and Ammachi, for their prayers, countless sacrifices, words of encouragement, and unwavering support and faith in me.

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ABSTRACT

Endometriosis is a chronic gynecological disorder associated with severe pain and impacts the quality of life for over 190 million girls and women of reproductive age (Zondervan et al., 2020). Prolonged pain exposure is associated with altered central processing of pain signals which is believed to largely implicate brain stem structures. To date, limitations in terms of technologies have prevented accurate segmentation of brain stem structures to evaluate clinical conditions. In the proposed investigation, we use a novel method of brain stem and hypothalamic segmentation to understand the impact of endometriosis and the relevancy of findings toward pain behaviors. This cross-sectional study applied structural neuroimaging and pain measures scales to understand endometriosis associated in a cohort with surgically confirmed endometriosis (n=43) and a cohort of healthy controls (n=25). Participants with endometriosis had significantly higher anxiety, depression, pain interference, and pain catastrophizing than healthy controls. Group differences were observed in several brain stem structures wherein the endometriosis cohort showed higher levels of brain stem volumes than healthy controls. Correlation analysis highlighted brain-behavioral relationships including the pain catastrophizing scale. Together, findings from this investigation provide insight into the

centralized impact of endometriosis and highlight relevant targets for immunological and neurological treatment options.

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LIST OF ABBREVIATIONS

ACC	Anterior Cingulate Cortex
ACS	American Community Survey
AVP	Arginine Vasopressin
BCH	Boston Children’s Hospital
BWH	Brigham and Women’s Hospital
CPP	Chronic Pelvic Pain
CPU	Central Processing Unit
CRF	Corticotropin-Releasing Factor
CRP	Coordinated Response Protocol
ETCV	Estimated Total Cranial Volume
FA	Flip Angle
FoV	Field of View
GAD	Generalized Anxiety Disorder
GPU	Graphics Processing Unit
GRAPPA	Generalized Autocalibrating Partially Parallel Acquisition
HPA	Hypothalamic Pituitary Adrenal
IASP	International Association for the Study of Pain
IRB	Institutional Review Board
LC-RVM	Locus Coeruleus-Rostral Ventromedial Medulla
ME/CFS	Encephalomyelitis/Chronic Fatigue Syndrome
MPRAGE	Magnetization-Prepared Rapid Acquisition with Gradient Echo

MRI	Magnetic Resonance Imaging
PACU	Post-Anesthesia Care Unit
PAG	Periaqueductal Gray
PCS	Pain Catastrophizing Scale
PCS-C	Pain Catastrophizing Scale-Child
PFC	Pre-Frontal Cortex
PROMIS	Patient Reported Outcomes Measurement Information System
PVN	Paraventricular Nucleus
RVM	Rostral Ventromedial Medulla
SCP	Superior Cerebellar Peduncle
SNR	Signal-to-Noise Ratio
SON	Supraoptic Nucleus
SPSS	Statistical Package for the Social Sciences
TAC	Trigeminal Autonomic Cephalalgias
TE	Echo Time
TNC	Trigeminal Nucleus Caudalis
TR	Repetition Time
VLM	Ventrolateral Medulla

INTRODUCTION

Background

Endometriosis is a chronic disease in which abnormal tissue growing outside the lining of the uterus causes severe pain to girls and women of reproductive age, and in some cases prior to the onset of menarche (Reese et al., 1996). During each menstruation cycle, the lining of the uterus, also known as the endometrium, sheds its superficial layer as a menstrual period (Maybin & Critchley, 2015). However, in cases of endometriosis, instead of shedding, a buildup of abnormal tissue and inflammation can lead to scarring, in the form of adhesions, fibrosis, or painful cysts in the pelvic region or abdominal cavity (Abd El-Kader et al., 2019). Endometriosis is the leading cause of infertility and is associated with life impacting symptoms, including chronic pelvic pain during menses, urination, bowel movements, sexual intercourse, as well as fatigue, depression, and anxiety (Verkauf, 1987; Saunders, 2021).

Endometriosis affects roughly 10-15% of women of reproductive age in the United States (Mehedintu et al., 2014). However, patients receive diagnostic delays of around seven to nine years and sixty percent of women affected by endometriosis are undiagnosed (Ghai et al., 2020; Morassutto et al., 2016). The current gold standard in diagnosis for endometriosis is laparoscopy (Hsu, 2010). This procedure takes a looks inside the pelvic cavity to check for signs of endometrial lesions. However, it can be painful, expensive, time consuming, and poorly accessed in lower socioeconomic demographics. Transvaginal gynecological ultrasounds and Magnetic Resonance Imaging (MRI) scans are also used as diagnostic tools, but there are limitations as it may lack adequate resolution to identify lesions (Hsu, 2010). All these factors can result in a delay in diagnosis, as well as, imprecise symptoms, deficiency in provider awareness of the condition, and need for noninvasive diagnostic tools. This makes it difficult to manage treatment and places a financial burden on both patients and the health care system.

Endometriosis is a debilitating condition which decreases the quality of life for individuals who experience chronic pain, making it difficult to go back to school or work (Nnoaham et al., 2011). The economic burden of endometriosis due to loss of work, decreased productivity, and disability is $4.7 billion in 2022 (Simoens et al., 2007). The annual healthcare costs in the United States associated with endometriosis is estimated to be from $78 billion each year due to the costs of surgeries, hospitalization, MRI scans, physician visits, and medication (Soliman et al., 2016). The number of endometriosis-related hospital visits in Australia increased up to 43% this past year, resulting in 40,500 hospitalizations in 2022 (Australian Institute of Health and Welfare, 2023). In the United States, the Nationwide Emergency Department Sample reported that there were 12,351 hospital visits related to endometriosis among patients ages 21 to 50 (Manuel et al., 2019). The prevalence of the condition, the economic burden, and impact on quality of life confirms that endometriosis is an under-researched and under-diagnosed condition.

Since there is currently no cure, the primary concern for individuals with endometriosis experiencing chronic pain is seeking pain management or minimization of symptoms. Current treatment methods include laparoscopic surgery, nerve injections, psychotherapy, hormone therapy, and NSAID medications (Al Kadri et al., 2009; Brown et al., 2017; Somigliana et al., 2009; Khodaverdi et al., 2021; Donatti et al., 2022).

However, many of these procedures have adverse reactions, and the endometriosis symptoms can re-occur. Even removal of the endometrioma may not cure the pain, due to a phenomenon known as central sensitization.

Classification of Pain

Pain is one of the most recurring reasons as to why individuals seek medical attention (Schappert et al., 2001-2002), also due to its relation to a variety of other conditions. Classification systems are implemented to aid with diagnosis, exercise precision medicine, and tailor treatments based on the needs and characteristics of the patient. The International Association for the Study of Pain has three main classifications for pain: nociceptive, neuropathic, and nociplastic pain. Clinical studies have shown that individuals with chronic pain can experience one and/or the other, a term referred to as "mixed pain" (Coxon et al., 2021; Freynhagen et al., 2019).

Nociceptive Pain

Pain is defined as an uncomfortable sensory and psychological occurrence, as a result of real or possible damage to tissues whereas, nociception is the neural action involved in the encoding of harmful stimuli. Nociceptive pain is derived from real or threatened noxious (harmful) stimuli to non-neural tissue. By changing the sensory response, it alters one's perception of pain by producing pain hypersensitivity, also referred as hyperalgesia, to non-inflamed tissues (Latremoliere & Woolf, 2009). The somatosensory nervous system, including the pain signaling pathways, has normal activity in nociceptive pain. When a noxious stimulus is encoded by a nociceptor, high blood pressure or a motor withdrawal reflex could occur. The nociceptive flexion reflex involves neural pathways acting on a stimulus before it has arrived at the brain, activating a proper withdrawal response by contracting the muscle that results in reflex muscle movement.

Neuropathic Pain

Neuropathic pain is defined by the International Association for the Study of Pain (IASP) as pain that results from different lesions, revealed during a diagnostic test, or diseases of the somatosensory nervous system. Central sensitization contributes to neuropathic pain and causes the nervous system to mistake innocuous (harmless) stimuli as noxious. The onset of neuropathic pain is influenced by several factors, including metabolic disorders, (e.g. diabetic neuropathy), viruses (e.g. postherpetic neuralgia), leprosy, spinal cord injury, autoimmune disorders (e.g. multiple sclerosis), and stroke (Colloca et al., 2017). Peripheral neuropathy can make modifications to the electrical properties of sensory nerves, which can result in alterations in inhibitory signaling, impairing the descending control pathway (Colloca et al., 2017). The cause of peripheral disorders that lead to neuropathic pain is associated with certain nerve fibers. For instance, in endometriosis, endometrial implants cause an inflammatory reaction which may stimulate nerves and cause severe pain. This sensation of pain is contributed by myelinated A delta, unmyelinated sensory C, cholinergic, and adrenergic fibers, activated by a noxious stimulus (Tokushige et al., 2007). These nerve fibers were found in higher quantity in women affected by endometriosis in the functional and basal layers of the endometrium (Medina & Lebovic, 2009).

Nociplastic Pain

Nociplastic pain is defined as persistent pain that is caused by altered nociception, regardless of the presence of damaged tissue, causing the peripheral nociceptors to become activated. It can also be demonstrated as a lesion or disease impairing the somatosensory system. Such alteration of neural processing pathways can lead to central sensitization. Central sensitization is in which the nervous system undergoes alterations, which can amplify pain sensitivity and affect the way the nervous system processes sensory input (Harte et al., 2018). Nociplastic pain can appear in many chronic pain disorders, including headache, fibromyalgia, lower back pain, and chronic pelvic pain (Raja et al., 2020).

Acute and Chronic Pain

Pain can further be divided on the basis of intensity, characteristics, and duration: acute and chronic pain. Acute pain, caused by an illness or injury, acts as a protective response to noxious stimuli. It occurs suddenly, and last no more than three months. Under normal conditions, acute pain typically passes as the noxious stimuli weakens and the underlying condition has been treated or recovered. However, when the pain becomes persistent and intense, it can progress to chronic pain. This occurrence is also caused by the alteration of receptors, ion channels, and functional connections involved in the normal pain response (Yang et al., 2019). Chronic pain can have overlapping phenotypes among nociceptive, neuropathic, and nociplastic pain (Maixner et al., 2016; Fitzcharles et al., 2021). It is associated with neuroplasticity which reinforces pain pathways, even in the absence of injury or illness, resulting in hyperalgesia, consistent heightened levels of pain. This consistent state of reactivity can also impact an individual’s cognition, and result in poor memory, anxiety, difficulty in concentration (Yunus, 2007).

Neuroimaging

Several brain imaging studies have demonstrated that the hypothalamus and brain stem are active during chronic pain (Denuelle et al., 2007; Weiller et al., 1995). However, the use of neuroimaging to recognize patterns in structural abnormalities in the brain of individuals with endometriosis has not been well studied. This can be due to a shortage of automated segmentation tools (Billot et al., 2020). While manual segmentation is the most accurate technique for studying the brain, it is arduous, as it requires two to three hours to complete a scan (Billot et al., 2020). Manual segmentation runs into issues with scalability, as it scans at most with 1mm resolution, and has difficulties with delineation which affects its ability to be reproducible, increasing its liability to intra and inter-rater variability (Billot et al., 2020). Magnetic resonance imaging (MRI) is the best procedure in which to examine the brain due to its superior soft tissue contrast (Billot et al., 2020). MRI can also provide pre-operative brain mapping by delineating the extent of the disease, which can expand the scope of resection, decreasing time in surgery, and potentially reducing the size of a craniotomy (Petrella et al., 2006).

MRI

An MRI is a non-invasive imaging technology that uses a large magnet and radio waves generated by a computer to create high resolution images of the internal structures and organs of the body. In a closed-bore MRI, the magnet surrounds the individual, while in an open MRI, the magnets are positioned at the top and bottom, allowing the sides to be open. Open MRIs are more comfortable for individuals who are claustrophobic, however closed MRIs are more accurate in diagnosing medical conditions due to higher imaging quality (Matsui et al., 2019). Cross sectional images are produced using the magnetic field found in the MRI and the hydrogen atoms found in an individual’s body, which align to create a magnetic vector, directed towards the axis of the MRI scanner (Berger, 2002). The difference in the amount of hydrogen atoms reflects on the amount of water in tissues, influencing the MRI signal and tissue contrast, and may also account for changes in structural brain volumes across different neuropsychiatric conditions (Bansal et al., 2013). Slice selection, phase encoding, and frequency encoding are used to locate where the protons are positioned in the patient, in order to generate an image. In an MRI, the image matrix is composed of pixels, and the number of pixels necessary to create an image is dependent by the number of frequency encodings and phase encoding steps for a particular field of view (FoV). The size of the pixels also affects the resolution, as smaller pixel size leads to a higher resolution (Van Geuns et al., 1999). The frequency of the wobbling or precession around the direction of magnetic field can be given by the Larmor equation, $F = \gamma B_0 / 2\pi$, in which F is the frequency of precession, B_0 is the strength of the magnetic field, and γ is the gyromagnetic ratio of the nucleus (Van Geuns et al., 1999). Radio frequency coils serve to excite proton spins in the body and to receive signal reception generated by tissue in the MRI (Gruber et al., 2018). The amount of time it takes for a signal to return to its external magnetic field is known as longitudinal relaxation time, T1 (Van Geuns et al., 1999). MRIs are the most frequently used imaging test for diagnosis or monitoring of treatment as ionizing radiation is not utilized, unlike X-rays (Van Geuns et al., 1999). The MRI strength can vary from less than 1 to 3 T (Grover et al., 2015). Advantages to higher field strengths include improved signal to noise ratio (SNR), resulting in shorter imaging times and enhanced spectral, spatial, and temporal resolution (Di Costanzo et al., 2003).

Prior Research Findings using MRI

Previous neuroimaging studies have examined structural and functional changes in brain regions related with pain perception and modulation in individuals with endometriosis. A study using voxel-based morphometry to investigate structural alterations in individuals with chronic pelvic pain (CPP) showed that individuals with CPP and endometriosis (symptomatic) appeared to have decreased gray matter volume in the right posterior insula, right putamen, left thalamus, and left cingulate gyrus (As-Sanie et al., 2012). Asymptomatic individuals (without CPP) with endometriosis showed increases in the PAG volume; this could clarify why some individuals with endometriosis experience hardly any pain, even with significant morbidity (As-Sanie et al., 2012; Brawn et al., 2014). Individuals with endometriosis displayed increased resting state functional connectivity between the anterior insula and medial prefrontal cortex, which was correlated with increasing pain intensity, depression, and anxiety symptoms (As-Sanie et al., 2016). Furthermore, participants with endometriosis associated pain demonstrated significant decreased functional connectivity between the right anterior insula and the right cerebellum, which was positively correlated with pain intensity (Szabo et al., 2022). Decreased functional connectivity between the right anterior insula and the left middle frontal gyrus was also observed in women with endometriosis associated pain compared to healthy volunteers (Szabo et al., 2022).

Cortical and Subcortical Structural Alterations

Previous research has shown that nociplastic alterations in the prefrontal and subcortical areas of the brain have resulted in the persistence of nociceptive signals, due to these regions being active above average, leading to the chronification of pain (Thompson, 2019). The prefrontal cortex, anterior cingulate cortex, and amygdala are functionally connected in the processing of pain (Stevens et al., 2011). The prefrontal cortex plays a critical role in executive functioning, regulating emotions, and pain modulation (Ong et al., 2019). The persistence of nociceptive signals allows for aversive learning, which is regulated by the prefrontal cortex (Kummer et al., 2020). During chronic pain, the prefrontal cortex experiences alterations in neurotransmitters and inflammation, resulting in alterations in functional connectivity and structural changes (Ong et al., 2019). Grey matter volume has been shown to decrease in the medial prefrontal cortex, affecting cognitive and emotional functioning (Gustin et al., 2014). The anterior cingulate cortex, connected to the PFC, is activated by noxious stimuli in pain processing. Activation of the prefrontal cortex and the ACC is correlated with increased activity of the periaqueductal gray (PAG) (Peyron, 2014).

Ascending nociceptive input reaches the medial prefrontal cortex and its cortical projections extend into the ACC and input on PAG activity, which is involved in the descending inhibitory pain pathway (An et al., 1998; Kummer et al., 2020). Chronic pain brings about an increased input upon medial prefrontal cortex GABAergic interneurons causing the descending pain modulation to weaken, exacerbating the painful state (Kummer et al., 2020). The medial prefrontal cortex transmits excitatory projections to the amygdala. Prior research has shown that patients with chronic pain were seen to have abnormal amygdala functional connectivity, correlating with an increase in pain intensity and pain catastrophizing (Jiang et al., 2016).

Hypothalamus and Brainstem

The central extended amygdala has major connections with the lateral hypothalamus and the brain stem, specifically the dorsolateral pons and medulla (Alheid, 2009). This thesis study investigated changes in structural segments of the brainstem and hypothalamus brain regions. The hypothalamic regions that were examined were the anterior-inferior, anterior-superior, posterior, inferior tubular, superior tubular, whole left, and whole right region. The hypothalamus contains several nuclei that control neuroendocrine function. The paraventricular nucleus, ventromedial nucleus, supraoptic nuclei, dorsomedial nuclei, as well as the lateral hypothalamus are involved in nociceptive processing and transmission of descending projections (Bernard, 2007). The paraventricular nucleus (PVN) is involved in the inhibition of spinal nociception (Condés-Lara et al., 2015). The PVN and the supraoptic nucleus (SON) synthesizes oxytocin, which contains analgesic properties, and may project to the spinal cord and cortical regions (Li et al., 2021). The ventromedial and dorsomedial nucleus projects extensively to the PAG and receive nociceptive input from the parabrachial region (Bernard, 2007). The lateral hypothalamus has been shown to be involved in the regulating neuronal activity in the periaqueductal gray region (Behbehani et al., 1988). It is also the major site for orexin expressing neurons, whose receptors are distributed through the pain circuitry including the PAG, involved in pain modulation (Peyron et al., 1998; Marcus et al., 2001). In several studies, the role of the orexinergic system has shown to produce antinociceptive effects on pain, specifically by OX1 receptors (Jeong et al., 2009). Corticotropin is a neurohormone that is released in response to stressful conditions, such as painful conditions. However, chronic pain can result in cortical dysfunction, resulting in extensive inflammation, pain, and psychological stress, increasing anxiety and depression (Heim et al., 2000). These comorbid mood disorders and intensity of symptoms are caused by modifications in the functioning of the hypothalamic pituitary adrenal axis (HPA), regulated by corticotropin-releasing factor (CRF) receptors (Heim et al., 1998).

The brainstem is a prominent region for pain processing, nociception, and production of analgesic and hyperalgesic responses, one of the factors being, due to its involvement in descending pain control. The results of the investigations presented in this thesis examined four different brainstem structures: medulla oblongata, pons, midbrain, and superior cerebellar peduncle (SCP). Several brainstem nuclei play a prominent role in pain processing, including the periaqueductal gray (PAG) and nucleus cuneiformis in the midbrain, raphe nuclei, parabrachial nucleus, and locus coeruleus in the pons, and the rostral ventromedial medulla (RVM), ventrolateral medulla (VLM), and dorsal reticular nucleus in the medulla oblongata (Napadow et al., 2019). The involvement of the SCP has been demonstrated in individuals with migraine; those with lower heat pain thresholds were shown have a smaller medulla and cerebellar peduncles, which indicates hypersensitivity to pain (Chong et al., 2016). In addition, patients with a cerebellar infarction appeared to have reduced activation of endogenous pain inhibitory mechanisms and increased pain perception (Ruscheweyh et al., 2014).

Stress has been known to exacerbate symptoms of chronic pain. To compensate, the brainstem pain modulating circulatory will create an analgesic response as a way to suppress the pain. However, stress can also induce hyperalgesia (i.e., heightened sensitivity to pain). Pain signals can prompt the descending control pathway, to modulate the pain intensity through neuronal inhibition. The PAG, dorsal reticular nucleus, and the parabrachial region will process the nociceptive information and relay it to the RVM, which will extend those signals to the spinal dorsal horn, which will activate the endogenous analgesia system to suppress the pain (Gauriau et al., 2002; Leite-Almeida et al., 2006). When noradrenergic neurons in the locus coeruleus are activated by gabapentin to release noradrenaline, it can result in analgesic effects (Pertovaara, 2006). However, the locus coeruleus has also been shown to induce hyperalgesia and anxiety related disorders through the activation of the LC-RVM circuit in mice with visceral inflammation (Kong et al., 2023).

Alterations in the neurons of in the rostral ventromedial medulla is known to be a factor in pain persistence. Stimulation of the VLM and RVM has been found to produce analgesic effects, inhibiting the nociceptive action of dorsal horn neurons (Fong et al., 1986; Gebhart, 2004). The nucleus cuneiformis is found ventrolateral to the PAG, and interacts with the RVM, involved in pain modulation (Haghparast et al., 2007). The nucleus cuneiformis and the raphe nuclei also contains a vast number of GABA_A

receptors (Nagai et al.,1985), which when stimulated induces antinociception effects, and can alleviate chronic pain (Mahmoudi & Zarrindast, 2002). Research has also shown that removing or altering the y2 subunit of GABA$_A$ receptors resulted in depressive-like behaviors (Smith & Rudolph, 2012). The a5-subunit, containing GABA$_A$ receptors, regulate tonic inhibition and are shown to be involved in the preservation of long-lasting secondary hypersensitivity (Bravo-Hernández et al., 2016).

Self-Report Metrics

Since pain is subjective for each individual, the most valid and relevant measurement to understand pain is through self-report metrics (Katz & Melzack, 1999). Laboratory and imaging findings may only partially address a patient's health outcome. Patient self-reports provide a comprehensive approach to administering quality care through the utilization of assessment tools to collect information regarding their health, as well as measure the effectiveness of a treatment. Patients may only report and remember their most recent or worst experience of pain, also known as "peak-end phenomenon" (Stone et al., 2000). Fluctuations in chronic pain can go unreported during medical appointments, and a more holistic assessment should be conducted to understand the underlying cause and improve patient health outcomes. An inadequate pain assessment could cause health care providers to overestimate or underestimate a patient's pain condition, affecting the course of treatment received (Leigheb et al., 2017). Since individuals' verbal and behavioral responses to express pain may vary, offering a diverse set of assessment tools is important to provide an inclusive method to evaluate pain

(Hadjistavropoulos & Craig, 2002). Several self-report methods used for pain measurement include Likert scales, facial distress scales, and verbal scales.

Likert Scales

Likert scales is a rating scale used to measure subjects' level of agreement, opinions, feelings, behaviors, or attitudes, and can be used to assess pain-related measures (Sullivan et al., 2013). Providing an accessible, easy to fill out, and comprehensive assessment has shown to increase general subject response, and due to these characteristics, Likert scales have been more favorable than visual analogue scale (Guyatt et al., 1987). The construct of this scale comprises a statement or question being asked, and a 5- or 7-point ordinal scale is typically provided for subjects to respond (Sullivan et al., 2013). The more options presented do signify more detailed responses, however, a study conducted by Borgers and colleagues have shown that in Likert scales there was an increase in stability of responses up to six options, while more than seven indicated to reduce scale reliability and increase the number of non-responses (Borgers et al., 2004). Likert scales are more susceptible to response bias, subjects may choose the more socially desirable option, central tendency bias, participants may choose most of their answers in the middle of the rating scale, and acquiescence bias, participants may have a strong stand toward one view, which can all possibly affect the validity of the assessment (Westland, 2022). Certain options may also be interpreted differently, for instance, participants may have difficulty choosing between "likely" and "somewhat likely". In the current study, Patient Reported Outcomes Measurement Information

System (PROMIS) and Pain Catastrophizing Scale (PCS) items were scored on Likert scales.

Facial Distress Scales

Facial scales use a visual representation to measure pain intensity in the context of facial expressions, allowing individuals to choose a face that best portrays their pain experience. It is especially helpful in pediatric populations, where children may struggle to articulate their pain due to limited expressive language. Health care providers may also experience more ease discussing pain with patients, as minimum cognitive demands are necessary for this scale. Faces Pain Scale and Wong-Baker pain scales are examples of facial distress scales that implement numbers with faces to better assist individuals with understanding the level of pain they may be undergoing (Bieri et al., 1990; Khin et al., 2014). One limitation in facial distress scales may be addressing fluctuations in pain, improvements or declines, especially when values may be in increments of two, but they may only experience a one-point difference (Adeboye et al., 2021).

Verbal Scales

Verbal pain rating scales are comprised of a series of verbal descriptions placed in order from least to most intense pain level, and individuals will select the word that best fits their pain intensity. Options may range from “no pain at all” to “worst pain possible” and consist of a series of numbers attached to the pain experience (Katz & Melzack, 1999). Verbal pain scales demonstrated a higher response rate for individuals who are cognitively impaired, compared to numeric rating and visual analogue scales (Closs et al., 2004). Post-Anesthesia Care Unit (PACU) patients showed an increased response rate to verbal rating scales compared to numeric rating scales, which could be attributed to their level of consciousness following surgery (Lee et al., 2021). However, verbal scales should not be the sole assessment tool as it depends on an individual's ability to interpret the words, and it may be more difficult for those with limited vocabulary (Briggs & Closs, 1999).

Study Rationalization

Endometriosis is a highly prevalent condition, and due to its impact on quality of life and economic hardship, it places a burden on both patients and the health care system. Limitations in technologies have prevented accurate segmentation of brainstem and hypothalamus structures to assess clinical conditions. Magnetic Resonance Imaging (MRI) has long been used as a neuroimaging technique in identifying alterations in brain regions. It will be useful in understanding the structural regions associated with pain in individuals with endometriosis, and other conditions related to chronic pain. Pain measure scales such as PROMIS, used to measure anxiety, depression, and pain interference, and PCS will be used to assess cognitions around pain. It is hypothesized that there would be significant differences in the volumes of the brainstem and hypothalamus of individuals with endometriosis compared to healthy volunteers, and these differences observed in neuroimaging findings would be correlated with pain reporting. Additionally, findings from this study can provide insight into the impact of endometriosis and highlight relevant targets for immunological and neurological treatment options.

Aims and Objectives

The first aim of this thesis study is using psychological testing to investigate group differences in pain reporting in endometriosis and healthy cohorts. The second aim is to use MRI to investigate volumetric differences in structural segments of the brainstem and hypothalamus regions. The final aim of this study is to use correlation analysis to understand how pain reporting correlates with brainstem and hypothalamus volumes.

METHODS

This thesis study obtained its study design and patient selection from grant-funded, Boston Children’s Hospital Institutional Review Board (IRB) approved research led by Dr. Christine Sieberg and Dr. Scott Holmes.

Study Design Overview

The study focused on a sample of 68 adolescents and women, ages 12 to 44 who had surgically confirmed endometriosis. The proposed age range was chosen to include individuals from the onset of menarche to the onset of menopause. This study aims to produce generalizable results reflective of adults and women in the United States who have endometriosis. They were recruited from Boston Children’s Hospital (BCH), Brigham and Women’s Hospital (BWH), and from the general public in the Boston area. A series of assessments, including the Pain Catastrophizing Scale (PCS) and Pain Reported Outcomes Measurement Information System (PROMIS), which measured anxiety, depression, and pain interference, were used to evaluate cognitions associated with pain. The endometriosis cohort consisted of women who reported a visual analogue pain score of over 3 out of 10 during the time of enrollment. After the completion of questionnaires, participants who were eligible went to Boston Children’s Hospital to undergo magnetic resonance imaging (MRI). They were requested to lie still on a flat surface in the scanner for up to an hour per scan, to capture brain measurements, in order to detect structural alterations. The regions of interest investigated using neuroimaging were the hypothalamus and brainstem.

Patient Selection & Inclusion/Exclusion Criteria

Before approaching participants, doctors involved in the study provided a list of potentially eligible participants, who were then contacted to inquire about their interest in taking part in the study. Patients from the Department of Adolescent Medicine at Boston Children's Hospital and Department of Gynecology at Brigham and Women's Hospital were presented with the opportunity to take part in the study during their clinical visit, as well as members from the community. Participants were made aware that the study would take approximately two to three hours. The healthy cohort consisted of individuals who were without pain and have not been diagnosed with endometriosis. The endometriosis cohort included individuals who were interested in the research study and had surgical confirmation of endometriosis. The research study strived to have a diverse cohort to heighten generalizability that reflected the population in the Boston area. As stated in the most recent American Community Survey (ACS), minorities make up around 51% of the Boston population. Therefore, members involved in the research study ensured that all participants that meet the eligibility criteria were included in this study.

Before participants were recruited, the IRB approved the protocol used in research to enroll human subjects to the study. The following outlined are the inclusion and exclusion criteria for the cross-sectional study cohort.

Inclusion criteria

i. Ability to speak sufficient English to complete surveys and questionnaires.
ii. Patients 12-44 years old with surgically confirmed endometriosis.

iii. Individuals 12-44 years old who will represent the healthy cohort from the general community and will complete a health screening form.

Exclusion criteria

i. Inability to speak sufficient English to complete surveys and questionnaires.
ii. History of severe cognitive impairment (e.g., intellectual disability, severe head injury, other neurological disorders).
iii. Medical history of asthma, schizophrenia, psychosis, bipolar disorder, personality disorder, and other significant disorders.
iv. Patients with co-morbid medical and/or pain conditions, which could potentially confound the data (e.g., cancer, sickle-cell disease, juvenile idiopathic arthritis).
v. Individuals who currently use opioid analgesics.
vi. Metallic implants (due to safety concerns, as well as distortion of images in MRI)
vii. Pregnant (confirmed with negative urine pregnancy test during the study visit)
viii. Claustrophobic
ix. Weight >350 lbs (due to limitation of the MRI table)
x. History of hysterectomy or oophorectomy (unless having been done prior to the start of the study) and menopause were excluded to minimize hormonal changes.
xi. Individuals who have reached menopause.

The participants in the endometriosis and healthy cohort were chosen in accordance with the inclusion/exclusion criteria of the study to account for confounding variables and provide reliable, reproducible results. The thesis study consisted of 25 participants in the healthy cohort and 43 participants in the endometriosis cohort. There were three pediatric participants, under eighteen years of age. Two participants identified as non-binary or gender fluid.

Recruitment Methods

Participants in the healthy controls cohort were recruited through flyers, which were displayed around Boston Children's Hospital, the Longwood Medical Area, and the surrounding Boston community. Several doctors collaborating with the study recruited patients with endometriosis from clinical visits. Consent and assent forms were reviewed by members of the research team. Consent forms were provided to participants of legal age and parents with patients under the age of 18, and assent forms were completed by adolescents and young adults.

At the start of the visit, a urine drug test was given to participants to confirm the absence of drugs, including barbiturates, benzodiazepines, amphetamine, barbiturates cocaine, phencyclidine, and opioids. If participants tested positive, they were no longer eligible to continue with the study. Participants who were eligible underwent MRI, which took approximately one hour to complete, and the entire study took approximately 2-3 hours. Participants received reimbursements through Clincards for their time and involvement in the study. They received up to $200 after completing their scan and series of questionnaires. If they partially completed the study, they were given $100 for completion of 1-1.5 hours of the study visit, $50 for completion of less than an hour, and no compensation for completing the consent form but not participating in study tasks. Transportation costs were reimbursed up to $50. Participants received $5 for every referral that completed the study. Data collected for the purpose of this thesis study only included participants who completed the full study visit.

Data collection

For participants to be involved in the study, they needed to agree to the informed consent, and individuals under 18 had to complete the assent form. This document disclosed the purpose of the study, procedures (MRI/questionnaires), retrieval of medical records, a confidentiality agreement, potential risks and benefits, and their rights as participants in the study. After completing consent and assent forms, participants were asked to fill out several self-report questionnaires that asked for their demographic information, endometriosis diagnosis, existence or non-existence of pain, menstruation, and psychiatric and neurological disorders. The psychological study data was collected using electronic measures through REDCap, a secure data capture tool for research studies. Participant chose to fill out the questionnaires through their own personal device or by iPad, which was provided by the lab. They were sent a password protected link, in compliance with Coordinated Response Protocol (CRP), by members of the study team, verifying their authenticity and email address, and through that link were taken to a secure site that stored their responses.

Description of Study Questionnaires

Participants were asked to fill out a series of questionnaires which took approximately thirty minutes. This included PCS and PROMIS, which measured anxiety, depression, and pain interference, and were used to evaluate cognitions associated with pain. They were asked to rate their pain (current, worst, and average) in the past two weeks and during their menstrual cycle using a 10-point Likert scale (0 = no pain at all, 10 = worst pain imaginable). They also had to list their current medications prescribed for chronic pain, which included their drug class, dosage, and detriment. A Health Screening Form was administered to assess a participant's overall health.

PROMIS

Patient Reported Outcomes Measurement Information System (PROMIS) was funded by the National Institute of Health to gather reports of patients' physical, mental, and social health conditions, including their symptoms, effects of treatment, perceptions, and emotions (Bevans et al., 2014); allowing clinicians to better understand their patients and incorporate their perspective when tailoring a treatment plan. Previous patient-reported outcomes had patients complete several, long questionnaires that were not relevant to all patients (Calvert et al., 2019). PROMIS addresses these issues by applying efficiency when measuring items relevant of physical function, pain interference, depression, anxiety, fatigue, social health, and sleep disturbance (Amtmann et al, 2010; Pilkonis et al., 2011; Buysse et al., 2010; Rose at al., 2014; Lai et al., 2011). Pain interference, defined by PROMIS, is the measure in which pain impedes engagement with physical, emotional, social, cognitive, and recreational activities (Amtmann et al., 2010). In the current study, pain interference was measured using a 5-point scale which asked participants to specify to what extent their pain had hindered their daily functioning and quality of life. Standardized scores for Pain interference followed PROMIS guidelines, indicating that a score below 55 was characterized as "mild" pain interference, and a score between 70 and greater than 80 indicated "severe" pain interference for adults and greater than 65 indicated “severe” pain interference for adolescents (Hassett et al., 2020; Miró et al., 2023). The PROMIS Anxiety and Depression measures assessed how frequently participants experienced symptoms of anxiety and depression in the past 7 days, from 8 items listed on a 5-point scale (Pilkonis et al., 2011; Pilkonis et al., 2014).

Standardized scores for Anxiety and Depression followed PROMIS guidelines, indicating that a score below 55 was characterized as “mild” symptoms, and a score between 70 and greater than 80 indicated “severe” symptoms for adults and greater than 65 indicated “severe” symptoms for adolescents (Hassett et al., 2020; Miró et al., 2023). PROMIS measures are well suitable for research as they have been validated by several studies (Van Balen et al., 2021; Pecorelli et al., 2023; McMullen et al., 2022).

PCS

Pain catastrophizing is defined as the propensity to overemphasize feelings towards a painful situation, experiencing helpless, and heightened pain intensity, outside of the norm. Pain Catastrophizing Scale (PCS) and PCS-C, for adolescents, has been developed to evaluate the characteristics associated with catastrophizing: rumination, magnification, and helplessness, which result from noxious stimuli (Sullivan et al., 1995). PCS uses a 5-point scale to asks participants to reflect on previous painful occurrences and to indicate to what extent they experienced those thoughts and feelings outlined by the 13 items listed on the scale. For both adult and adolescent questionnaires, the total scores ranged from 0 to 52, indicating the level of catastrophizing. Those scoring below

15 indicated low catastrophizing, and those scoring above 24 indicated high catastrophizing (Sullivan et al., 1995). Research conducted by Chen and colleagues found that catastrophizing thoughts may possibly lead to the chronification of depression and pain (Chen et al., 2022). Catastrophizing not only results in heightened intensity of pain and emotional distress, but also increases the persistence of pain.

Data Analysis

Statistical analysis for PROMIS and PCS self-report pain measures and volumetric differences were analyzed using the Statistical Package for the Social Sciences (SPSS) Version 28.0.1.0. Descriptive statistics were used to observe the distribution and variation in the way pain is presented among both cohorts. It provided a statistical summary of the number of participants, mean, standard deviation, minimum and maximum values. Group differences were analyzed using parametric tests, such as an independent sample t-test to measure continuous variables. Parametric tests are used when the sample means are normally distributed. To control for confounding variables, the regional brain volumes were divided by the estimated total intracranial volume. The confidence interval chosen for the analysis was 95%, a significance level of $p<0.05$. Cohen’s effect size and behavioral correlations using Pearson Correlation will be used for group comparison. PROMIS measures and PCS will be used as covariates and predictors of interests in group level multivariable regression analysis, incorporating both imaging and non-imaging data.

MRI Acquisition

A 3T Siemens Prisma MRI scanner was used to visualize and produce images of the subcortical regions of the hypothalamus and brainstem. A 32-channel head coil was used to maximize spatial resolution signal-to-noise ratio (SNR). T1-weighted Magnetization-prepared rapid acquisition with gradient echo (MPRAGE) was utilized as it provides fine anatomic detail, high SNR, and clear gray-white matter tissue contrast, which allows for high level delineation (Nelson et al., 2008). The parameters used in the research study included an echo time (TE) of 2.19 ms, repetition time (TR) of 2400 ms, field of view (FOV) of 240 mm, voxel size of 0.8 *0.8*0.8 mm3, flip angel (FA) of 8°, slice thickness of 0.8mm, and GRAPPA with an acceleration factor of 2.

Freesurfer

FreeSurfer utilizes automated brain segmentation methods to label subcortical structures in 3D T1 weighted scans of around 1mm isotropic resolution (Iglesias et al., 2015). In this research study, it was used to measure for subcortical thickness and volume through segmentation of five subregions of the hypothalamus: anterior inferior, anterior superior, posterior, inferior tubular, and superior tubular, and four regions of the brainstem: medulla oblongata, pons, midbrain, and superior cerebellar peduncle. Freeview is a visualization tool under Freesurfer, used to view multiple output volumes (brainmask.mgz and wm.mgz), the surfaces (rh.white and lh.white), and the subcortical segmentation (aseg.mgz). Freesurfer addresses issues associated with partial voluming effect sighted in volumetric images, when more than one type of tissue from distinct anatomical regions are comprised within a voxel, affecting the accuracy of structural characteristics, such as grey matter volume and thickness. (González et al., 2002). The software tools applied in Freesurfer is used create boundaries between white matter and cortical gray matter, in addition to the pial surface, which can be used to measure cortical thickness, volume, surface area, and curvature. T1-weighted images are generally used to outline brain structures (Misaki et al., 2015). Once the skull is stripped from the volume, it will be segmented into the cortex, white matter, and subcortical structures. The distance from the white surface to the pial surface is the gray matter thickness. The processing speed by which it computes segmentations in the hypothalamus is very miniscule, around 10 seconds with a CPU and lower than a second with GPU (Billot et al., 2020). The processing steps included motion correction and taking an average of several volumetric T1 weighted images (Reuter et al. 2010), skull stripping (Segonne et al., 2004), performing Talairach transformation, segmentation of the structures of the subcortical white matter and deep grey matter, (Fischl et al., 2002; Fischl et al., 2004a) intensity normalization (Sled et al., 1998), tessellation of the boundary between the gray and white matter, automated topology correction (Fischl et al., 2001; Segonne et al., 2007), surface deformation (Dale et al., 1999; Dale & Sereno, 1993; Fischl & Dale, 2000), and registration of the subjects brain to a spherical atlas (Fischl et al., 1999b). The morphometric procedures performed from Freesurfer have been shown to produce high test-retest reliability (Dickerson et al., 2008). After calculating the subcortical volumes and standardizing to the whole brain volume, statistically significant differences between the endometriosis and healthy cohort were analyzed using a t-test, shown to have high test-retest reliability (Dickerson et al., 2008).

RESULTS

Behavioral findings

The first objective of this study was to examine the differences in pain reporting in patients with endometriosis and healthy participants. This was assessed in the form of questionnaires, PROMIS and PCS, completed by participants during their study visit. There was a total of 68 participants, ranging from 12-44 years old, 43 patients with endometriosis (M age 27.69 ± 8.40) and 25 healthy volunteers (M age 26.90 ± 7.18). Table 1 shows descriptive data, including age at study visit, race/ethnicity, level of education completed, as well as pain measures: anxiety, depression, pain interference, and PCS scores. Education of participants in the healthy control cohort ranged from currently in high school to earning a Master's degree. In the endometriosis cohort, participants' academic background ranged from currently in school to earning a PhD. When compared to healthy controls, individuals with endometriosis reported higher levels of anxiety, depression, pain interference, and PCS scores.

Table 1. Demographic & Behavioral Characteristics of Participants. This table provides a summary of the demographic characteristics, in which 25 participants were healthy volunteers and 43 were patients with endometriosis (N=68). Demographic characteristics included age at study visit, race/ethnicity, level of education completed, as well as behavioral pain measures: anxiety, depression, pain interference, and PCS.

Age at study visit	**Healthy Controls (n=25)**	**Endometriosis (n=43)**
Mean; SD; Range (12-44)	26.90; 7.18; 29	27.69; 8.40; 30
Race and ethnicity		
White, not of Hispanic Origin	14	35
White of Hispanic Origin	1	1
Black/African American, not of Hispanic Origin	0	2
Black/African American of Hispanic Origin	1	1
Asian, or Pacific Islander	5	2
Native American	0	0
Other/Multiple	4	2
Level of education completed		
High school	7	9
Undergraduate	17	28
N/A	1	6
Anxiety T-Score		
Mean; SD; Range	51.56; 9.63; 37.5	58.55; 7.80; 35.9
Depression T-Score		
Mean; SD; Range	46.58; 8.37; 34.4	55.28; 8.50; 37.6
Pain Interference T-Score		
Mean; SD; Range	41.18; 2.45;12	57.29; 7.17; 30.3
PCS T-Score		
Mean; SD; Range	18.88; 9.66; 33	31.46; 10.29; 40

Table 2. Independent Samples T-Test of Brainstem Regions. This table shows the level of significance for volumetric differences in the brainstem of healthy controls and patients with endometriosis. An independent samples t- test was conducted (n=68). Equal variances not assumed. *p<0.05 is significant.

Brain Regions	**T-score**	**df**	**Two-sided p**
SCP	2.690	69.825	**0.009***
pons	2.282	72.784	**0.025***
midbrain	2.002	69.262	**0.049***
medulla oblongata	2.036	72.489	**0.045***
whole brainstem	2.257	71.876	**0.027***

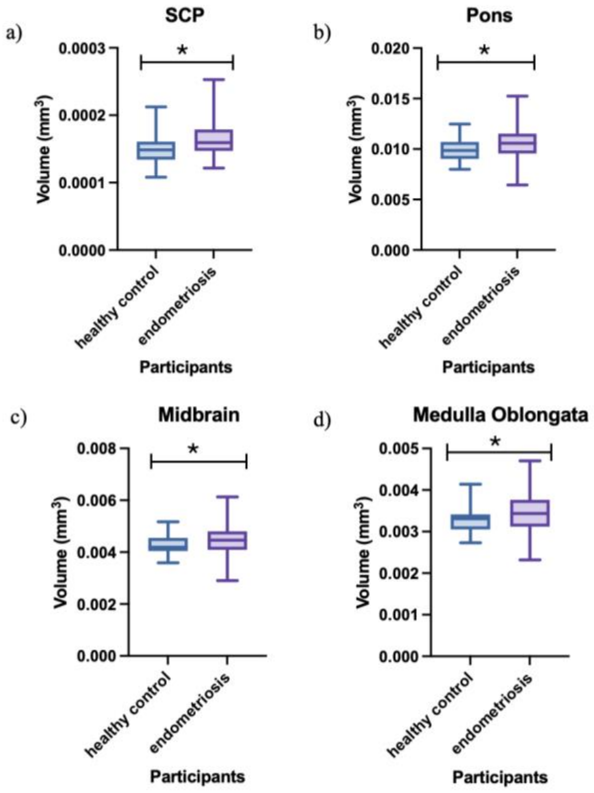

Figure 1. Whisker Plot of Brainstem Volumes in Healthy Control vs Patients with Endometriosis. Whisker plot indicating distribution of volume in segments of brainstem structures of the a) SCP, b) pons, c) midbrain, d) medulla oblongata in healthy controls and patients with endometriosis. Error bars represent the standard error of the mean. *p<0.05 is significant.

Structural findings

The second objective of this study was to use neuroimaging to investigate volumetric group differences in structural segments of the brainstem and hypothalamus regions. Freesurfer was used for statistical analysis of neuroimaging data to quantify and compare the subcortical volumes in both cohorts. After controlling for possible confounding variables, by dividing subcortical volumes by estimated total intracranial volume, an independent samples t-tests was administered. Table 2 revealed significant volumetric differences in the SCP (t (69.825) = 2.690, p = 0.009), pons (t (72.784) = 2.282, p = 0.025), medulla oblongata (t (72.489) = 2.036, p = 0.045), midbrain (t (69.262) = 2.002, p = 0.049), and whole brainstem (t (71.876) = 2.257, p = 0.027).

The endometriosis cohort was found to have larger SCP, pons, medulla oblongata, midbrain, and whole brainstem volumes compared to healthy controls, as demonstrated in Table 10 and 11. No significant volumetric differences were observed in the hypothalamic regions as shown in Table 3: left anterior inferior (t (60.592) = -0.765, p = 0.447), left anterior superior (t (46.105) = -0.284, p = 0.778), left posterior (t (52.488) = 0.237, p = 0.814), left tubular inferior (t (41.160) = -0.267, p = 0.791), left tubular superior (t (50.420) = 0.313, p = 0.756), right anterior inferior (t (71.637) = -1.346, p = 0.182), right anterior superior (t (53.075) = -0.253, p = 0.801), right posterior (t (46.321) = 1.359, p = 0.181), right tubular inferior (t (43.989) = 0.115, p = 0.909), right tubular superior (t (51.851) = 0.540, p = 0.591), whole left (t (43.880) = -0.009, p = 0.993), and whole right (t (44.253) = 0.583, p = 0.563).

Table 3. Independent Samples T-Test of Hypothalamus Regions. This table shows the level of significance for volumetric differences in the hypothalamus of healthy controls and patients with endometriosis. An independent samples t-test was conducted (n=68). Equal variances not assumed. *p<0.05 is significant.

Brain Regions	**T-score**	**df**	**Two-sided p**
Left anterior inferior	-0.765	60.592	.447
Left anterior superior	-0.284	46.105	.778
Left posterior	0.237	52.488	.814
Left tubular inferior	-0.267	41.160	.791
Left tubular superior	0.313	50.420	.756
Right anterior inferior	-1.346	71.637	.182
Right anterior superior	-0.253	53.075	.801
Right posterior	1.359	46.321	.181
Right tubular inferior	0.115	43.989	.909
Right tubular superior	0.540	51.851	.591
Whole left	-0.009	43.880	.993
Whole right	0.583	44.253	.563

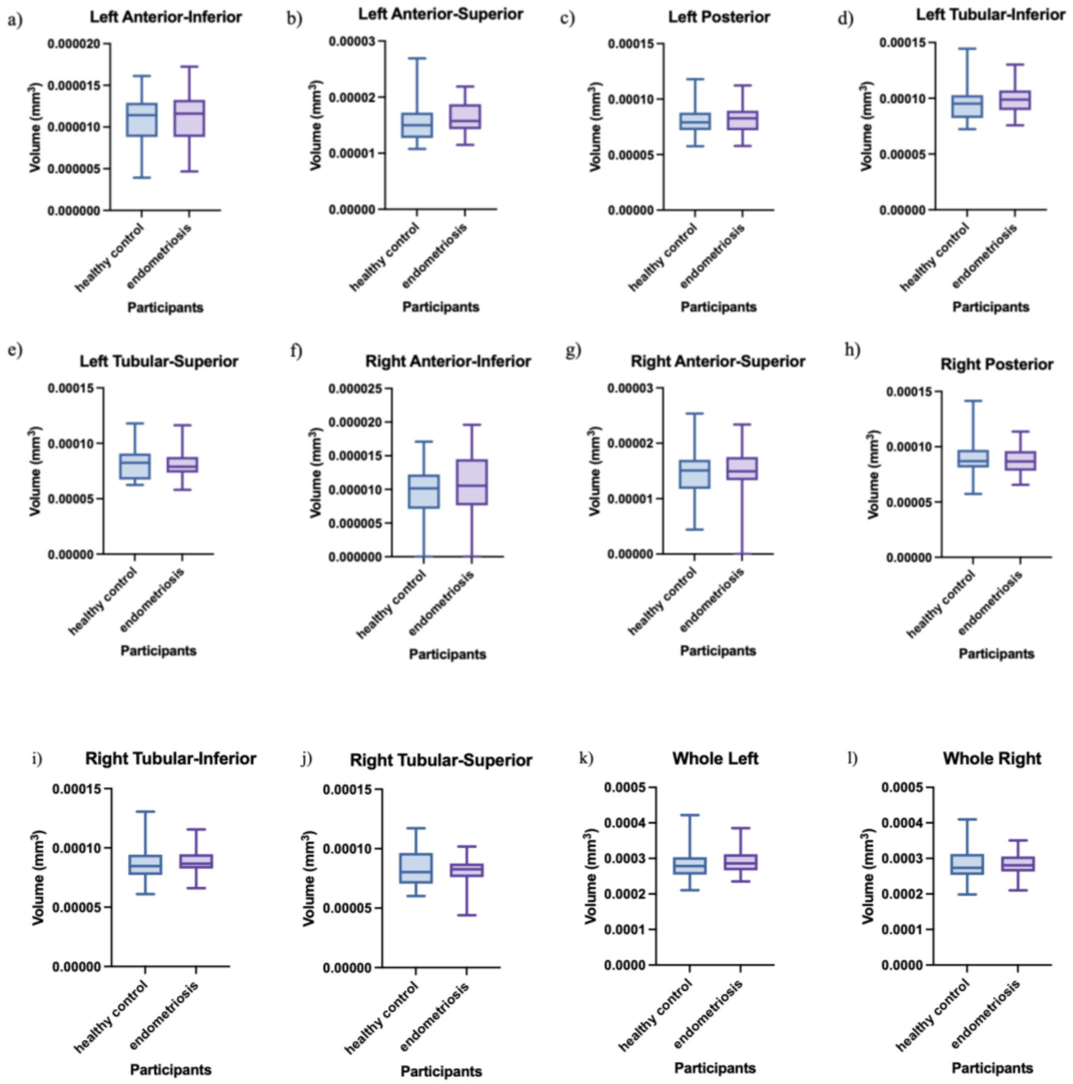

Figure 2. Whisker Plot of Hypothalamus Volumes in Healthy Controls vs Patients with Endometriosis. Whisker plot of indicating distribution of volume in segments of hypothalamus structures of the a) left anterior-inferior, b) left anterior-superior, c) left posterior, d) left tubular-inferior, e) left tubular-superior, f) right anterior inferior, g) right anterior superior, h) right posterior, i) right tubular-inferior, j) right tubular-superior, k) whole left, l) whole right for healthy controls and patients with endometriosis. Error bars represent the standard error of the mean. *p<0.05 is significant.

Correlation findings

The third objective of this thesis study was to use correlation analysis to investigate the relationship between pain metrics and brainstem and hypothalamus volumes. As shown on Table 4 and 5, when correlating pain metrics with each other, there were significant correlations between anxiety and depression, anxiety and pain interference, and depression and pain interference for endometriosis and healthy controls. There was a significant correlation with PCS and anxiety, PCS and depression, and PCS and pain interference for the endometriosis cohort, but not for the healthy controls. As seen on Table 6, there were no significant correlations between pain metrics and brainstem volumes in healthy controls. When comparing the brainstem volumes of individuals with endometriosis to pain metrics, PCS was significantly negatively correlated with pons volume, and approaching significance with medulla, midbrain, and whole brainstem volumes, as shown on Table 7.

When comparing the hypothalamus volumes of healthy individuals with pain metrics as demonstrated on Table 8, there were significant negative correlations observed in the left anterior inferior volume with depression, right posterior volume with depression, and an approaching significance with depression and smaller right anterior superior volume and smaller whole right volume with depression. When comparing anxiety scores in healthy individuals with hypothalamic volumes, a significant negative correlation was found with the right posterior volume. Pain interference scores were significantly negatively correlated with the whole right volume, specifically the right anterior superior volume, right posterior volume, right tubular inferior volume, and approaching significance with smaller whole left volume, specifically smaller left anterior inferior volume and left posterior volume in healthy participants. No significant correlations were observed in PCS and hypothalamic volumes in healthy controls. As shown on Table 9, pain interference scores approached significance with smaller left anterior superior volume in patients with endometriosis. Depression scores approached significance with larger whole left hypothalamic volume and were significantly positively correlated with larger left tubular superior volume in patients with endometriosis. PCS scores approached significance with larger left anterior inferior hypothalamic volume. There were no significant correlations observed in anxiety with hypothalamic volumes in individuals with endometriosis.

Table 4. Pearson Correlation Coefficient between Pain Measures in Healthy Controls. This table shows correlations between anxiety, depression, pain interference, and pain catastrophizing (PCS) scores. Correlation is significant at * p<0.05 and ** p<0.01. (–) signifies healthy controls hit baseline scores for pain interference and PCS scores. (~) signifies pain metric correlated with itself not measured for significance.

Pain Metric		**Anxiety t-score**	**Depression t-score**	**Pain Interference t-score**	**PCS t-score**
Anxiety	Pearson Correlation	1	**.809****	**.505****	0.056
	Sig.(2-tailed)	~	0.000	0.010	0.796
Depression	Pearson Correlation	**.809****	1	**.661****	-0.057
	Sig.(2-tailed)	0.000	~	0.000	0.790
Pain Interference	Pearson Correlation	**.505***	**.661****	1	-
	Sig. 2-tailed)	0.010	0.000	~	-
PCS	Pearson Correlation	0.056	-0.057	-	1
	Sig.(2-tailed)	0.796	0.790	-	~

Table 5. Pearson Correlation Coefficient between Pain Measures in Patients with Endometriosis. This table shows correlations between anxiety, depression, pain interference, and pain catastrophizing (PCS) scores. Correlation is significant at * p<0.05 and ** p<0.01. (–) signifies healthy controls hit baseline scores for pain interference and PCS scores. (~) pain metric correlated with itself not measured for significance.

Pain Metric		**Anxiety t-score**	**Depression t-score**	**Pain Interference t-score**	**PCS t-score**
Anxiety	Pearson Correlation	1	**.728****	**.453****	**.675****
	Sig. (2-tailed)	~	0.000	0.002	0.000
Depression	Pearson Correlation	**.728****	1	**.531****	**.664****
	Sig. (2-tailed)	0.000	~	0.000	0.000
Pain Interference	Pearson Correlation	**.453****	**.531****	1	**.501****
	Sig. (2-tailed)	0.002	0.000	~	0.002
PCS	Pearson Correlation	**.675****	**.664****	**.501****	1
	Sig. (2-tailed)	0.000	0.000	0.002	~

Table 6. Pearson Correlation Coefficient between Brainstem Volumes and Pain Measures in Healthy Controls. This table shows correlations between pain measures of anxiety, depression, pain interference, and pain catastrophizing (PCS) with brainstem regions of medulla, pons, SCP, midbrain, and whole brainstem. Correlation is significant at * p<0.05 and ** p<0.01.

Brain regions		Anxiety t-score	Depression t-score	Pain Interference t- score	PCS t- score
medulla	Pearson Correlation	0.097	0.222	-0.006	0.072
	Sig. (2-tailed)	0.644	0.286	0.978	0.739
pons	Pearson Correlation	0.123	0.284	0.126	-0.112
	Sig. (2-tailed)	0.560	0.169	0.549	0.602
SCP	Pearson Correlation	0.238	0.281	-0.003	-0.007
	Sig. (2-tailed)	0.252	0.174	0.989	0.976
midbrain	Pearson Correlation	0.021	0.127	-0.055	-0.021
	Sig. (2-tailed)	0.922	0.545	0.795	0.923
whole brainstem	Pearson Correlation	0.104	0.254	0.069	-0.064
	Sig. (2-tailed)	0.620	0.220	0.744	0.767

Table 7. Pearson Correlation Coefficient between Brainstem Volumes and Pain Measures in Patients with Endometriosis. This table shows correlations between pain measures of anxiety, depression, pain interference, and pain catastrophizing (PCS) with brainstem regions of medulla, pons, SCP, midbrain, and whole brainstem. Correlation is significant at * p<0.05 and ** p<0.01.

Brain Regions		**Anxiety t-score**	**Depression t- score**	**Pain Interference t- score**	**PCS t-score**
medulla	Pearson Correlation	-0.075	-0.135	-0.163	-0.282
	Sig. (2-tailed)	0.634	0.388	0.295	0.091
pons	Pearson Correlation	-0.051	-0.103	-0.130	**-0.324***
	Sig. (2-tailed)	0.744	0.511	0.405	0.050
SCP	Pearson Correlation	-0.107	-0.154	-0.048	-0.255
	Sig. (2-tailed)	0.496	0.324	0.760	0.127
midbrain	Pearson Correlation	-0.030	-0.063	-0.119	-0.288
	Sig. (2-tailed)	0.849	0.689	0.447	0.084
whole brainstem	Pearson Correlation	-0.052	-0.102	-0.135	-0.315
	Sig. (2-tailed)	0.741	0.516	0.388	0.058

Table 8. Pearson Correlation Coefficient between Hypothalamus Volumes and Pain Measures in Healthy Controls. This table shows correlations between pain measures of anxiety, depression, pain interference, and pain catastrophizing (PCS) scores with hypothalamus regions of left anterior inferior, left anterior superior, left posterior, left tubular inferior, left tubular superior, right anterior inferior, right anterior superior, right posterior, right tubular inferior, right tubular superior, whole left, and whole right in healthy controls. Correlation is significant at * p<0.05 and ** p<0.01.

Brain Regions		**Anxiety t-score**	**Depression t-score**	**Pain Interference t-score**	**PCS t-score**
Left anterior inferior	Pearson Correlation	-0.248	**-0.406***	-0.382	0.211
	Sig. (2-tailed)	0.231	0.044	0.060	0.323
Left anterior superior	Pearson Correlation	0.028	-0.083	-0.291	0.293
	Sig. (2-tailed)	0.896	0.693	0.158	0.165
Left posterior	Pearson Correlation	-0.313	-0.271	-0.370	-0.295
	Sig. (2-tailed)	0.128	0.189	0.069	0.161
Left tubular inferior	Pearson Correlation	-0.007	0.001	-0.189	-0.059
	Sig. (2-tailed)	0.972	0.998	0.365	0.785
Left tubular superior	Pearson Correlation	0.262	0.088	-0.221	0.020
	Sig. (2-tailed)	0.205	0.675	0.288	0.927
Right anterior inferior	Pearson Correlation	-0.133	-0.267	-0.103	0.274
	Sig. (2-tailed)	0.526	0.197	0.623	0.195
Right anterior superior	Pearson Correlation	-0.173	-0.347	**-0.473***	0.286
	Sig. (2-tailed)	0.407	0.089	0.017	0.176

Right posterior	Pearson Correlation	**-0.414***	**-0.479***	**-0.557****	-0.256
	Sig. (2-tailed)	0.040	0.015	0.004	0.227
Right tubular inferior	Pearson Correlation	-0.063	-0.095	**-0.417***	0.127
	Sig. (2-tailed)	0.763	0.652	0.038	0.555
Right tubular superior	Pearson Correlation	0.072	-0.030	-0.088	0.079
	Sig. (2-tailed)	0.733	0.887	0.677	0.715
Whole left	Pearson Correlation	-0.045	-0.116	-0.371	-0.077
	Sig. (2-tailed)	0.830	0.581	0.068	0.721
Whole right	Pearson Correlation	-0.226	-0.358	**-0.545****	0.087
	Sig. (2-tailed)	0.278	0.079	0.005	0.686

Table 9. Pearson Correlation Coefficient between Hypothalamus Volumes and Pain Measures in Patients with Endometriosis. This table shows correlations between pain measures of anxiety, depression, pain interference, and pain catastrophizing (PCS) with hypothalamus regions of left anterior inferior, left anterior superior, left posterior, left tubular inferior, left tubular superior, right anterior inferior, right anterior superior, right posterior, right tubular inferior, right tubular superior, whole left, and whole right in patients with endometriosis. Correlation is significant at * p<0.05 and ** p<0.01.

Brain Regions		**Anxiety t-score**	**Depression t-score**	**Pain Interference t-score**	**PCS t-score**
Left anterior inferior	Pearson Correlation	0.036	0.063	0.178	0.300
	Sig. (2-tailed)	0.820	0.686	0.252	0.071
Left anterior superior	Pearson Correlation	0.110	0.119	-0.297	0.228
	Sig. (2-tailed)	0.484	0.449	0.053	0.175
Left posterior	Pearson Correlation	0.226	0.126	-0.228	0.168
	Sig. (2-tailed)	0.145	0.422	0.142	0.321
Left tubular inferior	Pearson Correlation	0.120	0.151	-0.002	0.020
	Sig. (2-tailed)	0.443	0.332	0.988	0.907
Left tubular superior	Pearson Correlation	0.198	**0.329***	0.112	0.166
	Sig. (2-tailed)	0.204	0.031	0.474	0.326
Right anterior inferior	Pearson Correlation	-0.160	-0.053	-0.040	0.204
	Sig. (2-tailed)	0.304	0.737	0.799	0.225
Right anterior superior	Pearson Correlation	-0.104	0.016	-0.102	0.107
	Sig. (2-tailed)	0.508	0.919	0.515	0.529

Right posterior	Pearson Correlation	0.221	0.091	-0.178	0.010
	Sig. (2-tailed)	0.154	0.562	0.254	0.951
Right tubular inferior	Pearson Correlation	0.161	0.243	0.119	0.020
	Sig. (2-tailed)	0.302	0.117	0.446	0.907
Right tubular superior	Pearson Correlation	-0.187	-0.035	-0.167	-0.152
	Sig. (2-tailed)	0.229	0.821	0.286	0.369
Whole left	Pearson Correlation	0.254	0.282	-0.066	0.209
	Sig. (2-tailed)	0.101	0.067	0.674	0.214
Whole right	Pearson Correlation	0.045	0.103	-0.136	0.006
	Sig. (2-tailed)	0.777	0.513	0.386	0.971

DISCUSSION

This thesis study investigated group differences in pain reporting and the structural changes in the hypothalamus and brainstem in patients with endometriosis and healthy controls, as a way to examine the correlation between structural changes and psychological functioning. The findings from this study demonstrated significant structural brain changes that may be associated with self-report pain measures and provided insight into the centralized impact of endometriosis.

Behavioral findings

Individuals in the endometriosis cohort were found to have greater pain interference, pain catastrophizing, anxiety, and depression scores compared to healthy controls. These findings align with several studies that have reported that endometriosis significantly reduces one's quality of life and increases anxiety, depression, and stress (Pope et al., 2015; Friedl et al., 2015; Chen et al., 2016). Psychiatric responses to endometriosis could also be a result of the pain itself, along with infertility, difficulties with intercourse, and fatigue, which occur consequently due to the condition (Ramezanzadeh et al., 2014; Saunders, 2021). Individuals who have experienced symptoms for a longer period of time are more likely to encounter higher levels of perceived stress (Lazzeri et al., 2015) suggesting the greater impact of chronic pain on the perception of stress compared to acute pain. Increases in symptoms could also be due to delays in diagnosis which causes prolonged pain left untreated, exacerbating the risks of psychiatric comorbidity. Higher pain interference scores were observed in the endometriosis cohort, corresponding to the difficulties for many adolescents and women to go back to school or work and partake in activities of daily living, negatively affecting their quality of life.

Structural findings

There were significant volumetric group differences found in the SCP, pons, midbrain, and medulla oblongata when comparing healthy controls with participants with endometriosis, with the endometriosis cohort having higher volumes in all the mentioned brain stem segments. These observations expand upon previous research looking at structural brain changes associated with endometriosis. Specifically, prior research indicated that individuals with chronic pelvic pain (CPP) and endometriosis presented with decreased gray matter volume in the right posterior insula, right putamen, left thalamus, and the left cingulate gyrus, and asymptomatic individuals displayed increases in PAG volume and the right prefrontal cortex (As-Sanie et al., 2012). The findings from the current study also suggest that the observed structural changes in the brainstem correlate with patient responses to measures of pain. The pons contains several nuclei involved in pain mechanisms, including the parabrachial nucleus which relays nociceptive information to the RVM (Gauriau et al., 2002), and the locus coeruleus, shown to induce hyperalgesia through the activation of the LC-RVM circuit (Kong et al., 2023). In the midbrain, the PAG and the nucleus cuneiformis interact with the RVM, which is located in the medulla oblongata, and involved in pain modulation Haghparast et al., 2007).

The involvement of the SCP, pons, midbrain, and medulla oblongata has been shown in individuals with migraine; Chong and colleagues found that migraine patients who experienced severe symptoms of allodynia had smaller medulla, midbrain, and cerebellar peduncle volume. (Chong et al., 2016). Migraineurs demonstrated morphological inward deformations in the ventral side of the pons and midbrain, in addition to the outward deformations in the lateral side of the pons and medulla (Chong et al., 2016). Patients with fibromyalgia, a chronic pain disorder characterized by musculoskeletal pain, displayed structural alterations in the medulla and reduced gray matter volumes in the pons (Fallon et al., 2013). These correlations may provide insight to alterations in circuitry in central pain mechanisms.

When comparing brainstem volumes of individuals with encephalomyelitis/chronic fatigue syndrome (ME/CFS) to healthy controls, Thapaliya and colleagues found that the ME/CFS group had larger volumes in pons and whole brainstem, which could be caused by cerebral edema. Positive correlations were observed between the pons, SCP, and the whole brain stem volumes with pain scale scores. Several studies have demonstrated the development of cerebral edema caused by pro-inflammatory responses (Ting-Ting et al., 2020; Thapaliya et al., 2023). In endometriosis, the continuous activation of sensory nerve fibers can result in the recruitment of mast cells and release of proinflammatory cytokines, and interleukins (Velho et al., 2021). These inflammatory responses increase hyperpermeability of the blood-brain barrier (BBB) resulting in its breakdown and accumulation of fluid into the brain parenchyma, (i.e., edema) (Stamatovic et al., 2006; Michinaga & Koyama, 2015). Mast cells have been seen in prior studies in abundance in endometriotic lesions and proinflammatory cytokines were identified in high amounts in the peritoneal fluid of patients with endometriosis (Vallvé-Juanico et al., 2019). Increased levels of proinflammatory cytokines correlate with central hyperexcitability due to repeated electrical stimulations and alterations in pain response (Maddern et al., 2020). These findings could explain the larger brainstem volumes observed in the endometriosis cohort, as inflammation could have a significant impact on formation of edema in the brain.

The hypothalamus has major connections with the amygdala, thalamus, and the brainstem (Kuner & Flor, 2016; Alheid, 2009), which are involved in pain processing. Nociceptive information from the hypothalamus projects into the PAG and RVM. Furthermore, the hypothalamic-pituitary-adrenal (HPA) axis, activated through the secretion of the corticotropin releasing hormone and arginine vasopressin synthesized in the paraventricular nucleus of the hypothalamus, plays a vital role in the relationship between stress and pain (Heim et al., 1998; Chrousos & Gold, 1992). Based on these findings, it could be hypothesized that structural alterations in the HPA and specifically in the hypothalamus would be associated with chronic pain in patients with endometriosis. However, there were no significant group differences observed in the hypothalamus regions for patients with endometriosis and healthy controls. This could perhaps be due to variability in pain intensity, age, and a limited number of participants.

Correlation findings

Pearson correlation coefficient was used to investigate the relationship between the different pain metrics scores and regional brain volumes. When correlating pain metrics with each other, there were significant correlations between anxiety and depression, anxiety and pain interference, and depression and pain interference for endometriosis and healthy controls. Specifically, there were significant correlations with PCS and anxiety, PCS and depression, and PCS and pain interference for the endometriosis cohort, but not for the healthy controls, as they may not experience as intense levels of pain to have significant PCS scores. Anxiety, depression, and pain interference can amplify the severity of pain, and cause individuals to be hyperalert, which would explain the higher pain catastrophizing scores. These findings align with several studies which indicate that anxiety and depression are commonly co-occurring among patients with endometriosis (Pope et al., 2015; Smorgick et al., 2013).

Brainstem-pain metrics correlations

When comparing the brainstem regions to pain metrics for the endometriosis cohort, there were significant negative correlations with PCS and pons. The locus coeruleus in the pons is heavily involved in modulation of many cognitive processes, including regulating attention, perception, and arousal (Sara, 2009). Alterations in the neural circuits of the locus coeruleus due to excess stress can result in pain chronification (Taylor & Westlund, 2017), which could elucidate why individuals with endometriosis perceive more intense feelings of pain. In this study, the pons volume was larger in the endometriosis cohort, however PCS scores were correlated to smaller pons volume within this cohort. Ongoing pain can cause the activity of the locus coeruleus to gradually decrease over time (Suárez-Pereira et al., 2022), which may be reflected by decreases in pons volume compared to PCS scores observed in endometriosis participants. These findings correspond with previous research in patients with fibromyalgia that displayed reduced gray matter volumes in the pons (Fallon et al., 2013). The pons also has a number of nerve fibers passing through the medulla oblongata and midbrain. The PAG in the midbrain sends its descending projections to the RVM in the medulla, which are heavily involved in stress-induced hyperalgesia and allodynia (Pagliusi & Gomes, 2023) Chong and colleagues found that migraine patients have smaller medulla and midbrain in correlation with severe symptoms of allodynia, and those with lower heat pain thresholds, more sensitive to pain, had smaller medulla oblongata volume (Chong et al., 2016). This study aligns with our findings which indicate that PCS scores were approaching significance in the medulla oblongata and the midbrain, demonstrating that higher pain catastrophizing scores were correlated with smaller medulla oblongata and midbrain volumes.

Hypothalamus-pain metrics correlations

Circadian behavioral functions are regulated by oscillations produced by the suprachiasmatic nucleus (Bumgarner et al., 2021). Disruptions in circadian rhythm are associated with inflammation and disorder of the endocrine function, which can result in reduced pain thresholds, increasing pain sensitivity (Bumgarner et al., 2021; Kundermann et al., 2004). Alterations in sleep patterns have also been known to be linked to the development of depression. Sleep-wake disturbances were reported in individuals with chronic pain, including fibromyalgia (Korszun, 2000). In a study comparing hypothalamus volumes in different migraine patients, chronic migraine patients had lower hypothalamus volumes, specifically in the anterior hypothalamus compared to healthy control (Chen et al., 2019). These volumetric changes were correlated with headache frequency and alterations in the functional connectivity with different brain regions (Chen et al., 2019). These observations and prior literature indicating the occurrence of depression due to alterations in the function of the suprachiasmatic nucleus found in the anterior inferior hypothalamus, align with our study findings which showed that in healthy controls higher depression scores were significantly correlated with smaller left anterior inferior hypothalamus volumes. Higher pain interference scores which could be related to depression and pain intensity were approaching significance with smaller left anterior inferior hypothalamus volumes in healthy controls. In patients with endometriosis, we found that higher PCS scores were approaching significance with larger left anterior inferior volume of the hypothalamus. Similar findings were observed in a study conducted by Arkink and colleague, which found that the anterior hypothalamus appeared to be enlarged in episodic and chronic cluster headache (Arkink et al., 2017).

The paraventricular nucleus of the anterior-superior hypothalamus is vital in intervening in the regulation of nociceptive input. It synthesizes oxytocin (Li et al., 2021), inhibiting spinal nociception (Condés-Lara et al., 2015), and is involved in the relationship between stress and pain by releasing corticotropin releasing hormone to activate HPA axis activity. In accordance with previous study results which found lower anterior hypothalamus volumes in chronic migraine patients (Chen et al., 2019), our findings indicated that higher pain interference scores were approaching significance with smaller left anterior superior volume in the endometriosis cohort. In healthy controls, higher pain interference scores were significantly correlated with smaller right anterior superior volume of the hypothalamus, and higher depression scores were approaching significance with smaller right anterior superior volume. These findings correspond with prior literature which explains that the medial preoptic area of the anterior-superior hypothalamus produces prostaglandin E_2 (PGE_2) which magnifies inflammatory and neuropathic pain by activating the RVM neurons to promote nociception and produce hyperalgesia (Heinricher et al., 2004).

The lateral hypothalamus is the major site for orexin expressing neurons, which have been shown to be involved in pain modulation, and these OX1 receptors are distributed throughout the pain circuitry, including the PAG region (Behbehani et al., 1988; Peyron et al., 1998; Marcus et al., 2001; Jeong et al., 2009). The posterior hypothalamus has appeared to be activated in trigeminal autonomic cephalalgies (TACs), and inhibition of PH activity produced anti-nociceptive effects on trigeminal neuropathic pain signal transmission by modulation of the PAG and trigeminal nucleus caudalis (TNC) (Islam et al., 2023). In a study comparing hypothalamus volumes in different migraine patients, episodic migraine patients had lower hypothalamus volumes, specifically in the posterior hypothalamus compared to healthy control (Chen et al., 2019). These volumetric changes were associated with headache frequency (Chen et al., 2019). Our findings indicated that higher anxiety, depression, and pain interference scores significantly correlated with smaller right posterior volume of the hypothalamus in healthy controls, and higher pain interference scores were approaching significance with smaller left posterior volume. This corresponds with prior literature indicating decreased hypothalamic volumes in generalized anxiety disorder (GAD) patients. Since depression and pain interference have shown to be co-existing with anxiety, this explains why similar trends of decreased volume in the posterior hypothalamus occur.

The supraoptic nuclei of the inferior tubular hypothalamic region and the PVN of the anterior-superior hypothalamus synthesize and secrete oxytocin and arginine vasopressin (AVP), influencing nociception (Landgraf & Neumann, 2004). The parabrachial region projects nociceptive information to the ventromedial nucleus of the inferior tubular region, which projects extensively to the PAG. The ventromedial nucleus of the hypothalamus is responsible for generating an innate affective reaction, such as vocalization, in response to a painful stimulus (e.g., tail shock in a rodent model) (Borszcz, 2006). Stimulation of the ventromedial nucleus of the hypothalamus induces analgesia, which may be mediated by periaqueductal output (Bernard, 2007). The arcuate nucleus of the inferior tubular region contains insulin receptors which when activated promote secretion of endogenous opioids, and inhibition of the nucleus has been shown to increase pain sensitivity (Khaleghzadeh-Ahangar et al., 2020). Based on the involvement of the right tubular inferior hypothalamus in pain processing, our results in adjacent hypothalamic regions, may explain our findings that higher pain interference scores were correlated with smaller right tubular inferior volume of the hypothalamus in healthy controls.

Within the hypothalamus, there was variability in regional volumes that differentially correlated with depression scores in control subjects and subjects with endometriosis. Specifically, the volume of the right posterior hypothalamic region was negatively correlated with depression in control subjects. By contrast, the volume of the left superior tubular hypothalamic region was positively correlated with depression in the endometriosis participants. These findings are consistent with emerging evidence suggesting that the response to stress is associated with structural and functional asymmetry in the hypothalamus in patients with mood disorders (Schindler et al., 2019). This regional variability in hypothalamic volumes and depression scores may reflect differences in the etiology of depression in these two groups. Further research is needed to elucidate these relationships.

Limitations

The study implemented cross sectional data to evaluate volumetric and behavioral group differences in endometriosis and healthy controls. However, longitudinal studies could provide valuable insight into disease progression over time. In statistics, power computes the extent to which a study is able to detect true differences or associations within the study population (Fox & Mathers, 1997). G Power was used to compute effect sizes and statistical power (Faul et al., 2007). The study contained a sample of 25 healthy controls and 43 endometriosis participants, with a statistical power of 0.48, which reflected structural differences in brainstem volumes between endometriosis and healthy controls. The participants in the study were predominately white, not reflective of the incidence of endometriosis across all racial/ethnic groups. A sufficiently powered study with a similar number of healthy controls to endometriosis participants would better differentiate the effects of pain on psychiatric measures and subcortical volumes between cohorts. A more diverse and larger sample size would produce more reliable and generalizable results. To receive an endometriosis diagnosis, individuals would have to undergo an invasive procedure, and if they are not experiencing symptoms, they may not have a reason to. Therefore, there is a possibility that the healthy control group could have included individuals who are asymptomatic with endometriosis. However, in the study, participants in the endometriosis cohort had surgically diagnosed endometriosis, and reported severe symptoms prior to surgery, and participants in the healthy control cohort would be excluded from the study if they reported experiencing any life-impacting chronic pain, including chronic pelvic pain.

Conclusions

Our findings identified significant differences in pain reporting and brain stem volumes in patients with endometriosis and healthy controls and indicated a relationship between brainstem and hypothalamus volumes and pain reporting. Future research should seek to expand on these findings and determine how these structural changes correlate with different treatment modalities with the expectation that this additional information may help guide improvements in treatment from an evidence-based approach. In addition, longitudinal analysis could be implemented to understand age-related regional brain and behavioral changes, as well as differences in presentation reflected by duration of the disease. Finally, there is an under-representation of racial and ethnic minority populations in research studies on endometriosis stemming from biases and barriers to gynecological care, resulting in disparities in endometriosis diagnosis, and therefore future studies should examine how endometriosis is presented in non-White populations.

APPENDIX

Table 10. Summary Statistics of Brainstem Volumes in Healthy Controls. This table shows the mean, standard deviation, minimum, and maximum values of brainstem regions: SCP, pons, midbrain, medulla oblongata, and whole brainstem in healthy controls.

Brain Regions	Mean	SD	Min	Max
SCP	1.51×10^{-4}	2.45×10^{-5}	1.08×10^{-4}	2.12×10^{-4}
pons	1.00×10^{-2}	1.20×10^{-3}	7.99×10^{-3}	1.25×10^{-2}
midbrain	4.29×10^{-3}	3.95×10^{-4}	3.59×10^{-3}	5.17×10^{-3}
medulla oblongata	3.30×10^{-3}	3.32×10^{-4}	2.73×10^{-3}	4.14×10^{-3}
whole brainstem	1.78×10^{-2}	1.86×10^{-3}	1.44×10^{-2}	2.13×10^{-2}

Table 11. Summary Statistics of Brainstem Volumes in Patients with Endometriosis. This table shows the mean, standard deviation, minimum, and maximum values of brainstem regions: SCP, pons, midbrain, medulla oblongata, and whole brainstem in patients with endometriosis.

Brain Regions	Mean	SD	Min	Max
SCP	1.67×10^{-4}	2.91×10^{-5}	1.21×10^{-4}	2.52×10^{-4}
pons	1.07×10^{-2}	1.86×10^{-3}	6.44×10^{-3}	1.52×10^{-2}
midbrain	4.50×10^{-3}	7.14×10^{-4}	2.90×10^{-3}	6.13×10^{-3}
medulla oblongata	3.47×10^{-3}	5.13×10^{-4}	2.32×10^{-3}	4.70×10^{-3}
whole brainstem	1.88×10^{-2}	3.05×10^{-3}	1.18×10^{-2}	2.60×10^{-2}

Table 12. Summary Statistics of Hypothalamus Volumes in Healthy Controls. This table shows the mean, standard deviation, minimum and maximum values of hypothalamus regions: left anterior inferior, left anterior superior, left posterior, left tubular inferior, left tubular superior, right anterior inferior, right anterior superior, right posterior, right tubular inferior, right tubular superior, whole left, and whole right in healthy controls.

Brain Regions	**Mean**	**SD**	**Min**	**Max**
Left anterior inferior	1.06×10^{-5}	3.08×10^{-6}	3.92×10^{-6}	1.61×10^{-5}
Left anterior superior	1.58×10^{-5}	4.02×10^{-6}	1.07×10^{-5}	2.69×10^{-5}
Left posterior	8.21×10^{-5}	1.51×10^{-5}	5.75×10^{-5}	1.18×10^{-4}
Left tubular inferior	9.83×10^{-5}	2.04×10^{-5}	7.24×10^{-5}	1.44×10^{-4}
Left tubular superior	8.18×10^{-5}	1.43×10^{-5}	6.25×10^{-5}	1.18×10^{-4}
Right anterior inferior	9.80×10^{-6}	3.09×10^{-6}	3.37×10^{-6}	1.71×10^{-5}
Right anterior superior	1.46×10^{-5}	4.92×10^{-6}	4.41×10^{-6}	2.54×10^{-5}
Right posterior	9.14×10^{-5}	1.79×10^{-5}	5.73×10^{-5}	1.41×10^{-4}
Right tubular inferior	8.87×10^{-5}	1.61×10^{-5}	6.11×10^{-5}	1.31×10^{-4}
Right tubular superior	8.30×10^{-5}	1.42×10^{-5}	6.02×10^{-5}	1.17×10^{-4}
Whole left	2.89×10^{-4}	5.17×10^{-5}	2.11×10^{-4}	4.22×10^{-4}
Whole right	2.87×10^{-4}	4.96×10^{-5}	1.98×10^{-4}	4.10×10^{-4}

Table 13. Summary Statistics of Hypothalamus Volumes in Patients with Endometriosis. This table shows the mean, standard deviation, minimum and maximum values of hypothalamus regions: the left anterior inferior, left anterior superior, left posterior, left tubular inferior, left tubular superior, right anterior inferior, right anterior superior, right posterior, right tubular inferior, right tubular superior, whole left, and whole right region in patients with endometriosis.

Brain Regions	**Mean**	**SD**	**Min**	**Max**
Left anterior inferior	7.91×10^{-5}	9.26×10^{-6}	5.80×10^{-5}	1.01×10^{-4}
Left anterior superior	1.61×10^{-5}	2.60×10^{-6}	1.15×10^{-5}	2.19×10^{-5}
Left posterior	8.23×10^{-5}	1.19×10^{-5}	5.78×10^{-5}	1.12×10^{-4}
Left tubular inferior	9.93×10^{-5}	1.17×10^{-5}	7.58×10^{-5}	1.30×10^{-4}
Left tubular superior	8.05×10^{-5}	1.10×10^{-5}	5.80×10^{-5}	1.16×10^{-4}
Right anterior inferior	1.12×10^{-5}	4.02×10^{-6}	3.75×10^{-6}	1.96×10^{-5}
Right anterior superior	1.53×10^{-5}	3.08×10^{-6}	8.74×10^{-6}	2.34×10^{-5}
Right posterior	8.78×10^{-5}	1.23×10^{-5}	6.56×10^{-5}	1.14×10^{-4}
Right tubular inferior	8.81×10^{-5}	1.03×10^{-5}	6.61×10^{-5}	1.16×10^{-4}
Right tubular superior	8.13×10^{-5}	1.05×10^{-5}	4.41×10^{-5}	1.02×10^{-4}
Whole left	2.89×10^{-4}	3.19×10^{-5}	2.35×10^{-4}	3.85×10^{-4}
Whole right	2.83×10^{-4}	3.05×10^{-5}	2.10×10^{-4}	3.51×10^{-4}

BIBLIOGRAPHY

Abd El-Kader, A. I., Gonied, A. S., Lotfy Mohamed, M., & Lotfy Mohamed, S. (2019). Impact of Endometriosis-Related Adhesions on Quality of Life among Infertile Women. *International Journal of Fertility & Sterility*, *13*(1), 72–76. https://doi.org/10.22074/ijfs.2019.5572

Adeboye, A., Hart, R., Senapathi, S. H., Ali, N., Holman, L., & Thomas, H. W. (2021). Assessment of Functional Pain Score by Comparing to Traditional Pain Scores. *Cureus*, *13*(8), e16847. https://doi.org/10.7759/cureus.16847

Alheid, G.F. (2009). Extended Amygdala. In: Binder, M.D., Hirokawa, N., Windhorst, U. (eds.) *Encyclopedia of Neuroscience*. Springer, Berlin, Heidelberg. https://doi.org/10.1007/978-3-540-29678-2_3221

Al Kadri, H., Hassan, S., Al-Fozan, H. M., & Hajeer, A. (2009). Hormone therapy for endometriosis and surgical menopause. *The Cochrane Database of Systematic Reviews*, (1), CD005997. https://doi.org/10.1002/14651858.CD005997.pub2

Amtmann, D., Cook, K. F., Jensen, M. P., Chen, W. H., Choi, S., Revicki, D., Cella, D., Rothrock, N., Keefe, F., Callahan, L., & Lai, J. S. (2010). Development of a PROMIS item bank to measure pain interference. *Pain*, *150*(1), 173–182. https://doi.org/10.1016/j.pain.2010.04.025

An, X., Bandler, R., Ongür, D., & Price, J. L. (1998). Prefrontal cortical projections to longitudinal columns in the midbrain periaqueductal gray in macaque monkeys. *The Journal of Comparative Neurology*, *401*(4), 455–479.

As-Sanie, S., Harris, R. E., Harte, S. E., Tu, F. F., Neshewat, G., & Clauw, D. J. (2013). Increased pressure pain sensitivity in women with chronic pelvic pain. *Obstetrics and Gynecology*, *122*(5), 1047–1055. https://doi.org/10.1097/AOG.0b013e3182a7e1f5

As-Sanie, S., Harris, R. E., Napadow, V., Kim, J., Neshewat, G., Kairys, A., Williams, D., Clauw, D. J., & Schmidt-Wilcke, T. (2012). Changes in regional gray matter volume in women with chronic pelvic pain: a voxel-based morphometry study. *Pain*, *153*(5), 1006–1014. https://doi.org/10.1016/j.pain.2012.01.032

As-Sanie, S., Kim, J., Schmidt-Wilcke, T., Sundgren, P. C., Clauw, D. J., Napadow, V., & Harris, R. E. (2016). Functional Connectivity is Associated With Altered Brain Chemistry in Women With Endometriosis-Associated Chronic Pelvic Pain. *The Journal of Pain*, *17*(1), 1–13. https://doi.org/10.1016/j.jpain.2015.09.008

Australian Institute of Health and Welfare. (2023). *Treatment and management: Hospitalizations.* Australian Institute of Health and Welfare.

https://www.aihw.gov.au/reports/chronic-disease/endometriosis-in-australia/contents/treatment-management/hospitalisations

Bansal, R., Hao, X., Liu, F., Xu, D., Liu, J., & Peterson, B. S. (2013). The effects of changing water content, relaxation times, and tissue contrast on tissue segmentation and measures of cortical anatomy in MR images. *Magnetic Resonance Imaging*, *31*(10), 1709–1730. https://doi.org/10.1016/j.mri.2013.07.017

Bazot, M., Lafont, C., Rouzier, R., Roseau, G., Thomassin-Naggara, I., & Daraï, E. (2009). Diagnostic accuracy of physical examination, transvaginal sonography, rectal endoscopic sonography, and magnetic resonance imaging to diagnose deep infiltrating endometriosis. *Fertility and Sterility*, *92*(6), 1825–1833. https://doi.org/10.1016/j.fertnstert.2008.09.005

Berger A. (2002). Magnetic resonance imaging. *BMJ: British Medical Journal (Clinical research ed.)*, *324*(7328), 35. https://doi.org/10.1136/bmj.324.7328.35

Bernard, JF. (2007). Hypothalamus and Nociceptive Pathways. In: Schmidt, R., Willis, W. (eds) *Encyclopedia of Pain*. Springer, Berlin, Heidelberg. https://doi.org/10.1007/978-3-540-29805-2_1847

Bevans, M., Ross, A., & Cella, D. (2014). Patient-Reported Outcomes Measurement Information System (PROMIS): efficient, standardized tools to measure self-reported health and quality of life. *Nursing Outlook*, *62*(5), 339–345. https://doi.org/10.1016/j.outlook.2014.05.009

Bieri, D., Reeve, R. A., Champion, D. G., Addicoat, L., & Ziegler, J. B. (1990). The Faces Pain Scale for the self-assessment of the severity of pain experienced by children: development, initial validation, and preliminary investigation for ratio scale properties. *Pain*, *41*(2), 139–150. https://doi.org/10.1016/0304-3959(90)90018-9

Billot, B., Bocchetta, M., Todd, E., Dalca, A. V., Rohrer, J. D., & Iglesias, J. E. (2020). Automated segmentation of the hypothalamus and associated subunits in brain MRI. *NeuroImage*, *223*, 117287. https://doi.org/10.1016/j.neuroimage.2020.117287

Borgers, N., Sikkel, D. & Hox, J. (2004). Response Effects in Surveys on Children and Adolescents: The Effect of Number of Response Options, Negative Wording, and Neutral Mid-Point. *Quality & Quantity* 38, 17–33. https://doi.org/10.1023/B:QUQU.0000013236.29205.a6

Borszcz G. S. (2006). Contribution of the ventromedial hypothalamus to generation of the affective dimension of pain. *Pain*, *123*(1–2), 155–168. https://doi.org/10.1016/j.pain.2006.02.026

Bravo-Hernández, M., Corleto, J. A., Barragán-Iglesias, P., González-Ramírez, R., Pineda-Farias, J. B., Felix, R., Calcutt, N. A., Delgado-Lezama, R., Marsala, M., & Granados-Soto, V. (2016). The α5 subunit containing GABAA receptors contribute to chronic pain. *Pain*, *157*(3), 613–626. https://doi.org/10.1097/j.pain.0000000000000410

Brawn, J., Morotti, M., Zondervan, K. T., Becker, C. M., & Vincent, K. (2014). Central changes associated with chronic pelvic pain and endometriosis. *Human Reproduction Update*, *20*(5), 737–747. https://doi.org/10.1093/humupd/dmu025

Briggs, M., & Closs, J. S. (1999). A descriptive study of the use of visual analogue scales and verbal rating scales for the assessment of postoperative pain in orthopedic patients. *Journal of Pain and Symptom Management*, *18*(6), 438–446. https://doi.org/10.1016/s0885-3924(99)00092-5

Brigham and Women's Hospital (n.d.), *Endometriosis pain management for adult women.* Brigham and Women's Hospital. Accessed January 27, 2024. https://www.brighamandwomens.org/obgyn/infertility-reproductive-surgery/endometriosis/endometriosis-pain-management-for-adult-women#:~:text=Management%20of%20chronic%20pelvic%20pain,their%20pain%20due%20to%20endometriosis.

Brown, J., Crawford, T. J., Allen, C., Hopewell, S., & Prentice, A. (2017). Nonsteroidal anti-inflammatory drugs for pain in women with endometriosis. *The Cochrane Database of Systematic Reviews*, *1*(1), CD004753. https://doi.org/10.1002/14651858.CD004753.pub4

Bumgarner, J. R., Walker, W. H., 2nd, & Nelson, R. J. (2021). Circadian rhythms and pain. *Neuroscience and Biobehavioral Reviews*, *129*, 296–306. https://doi.org/10.1016/j.neubiorev.2021.08.004

Buysse, D. J., Yu, L., Moul, D. E., Germain, A., Stover, A., Dodds, N. E., Johnston, K. L., Shablesky-Cade, M. A., & Pilkonis, P. A. (2010). Development and validation of patient-reported outcome measures for sleep disturbance and sleep-related impairments. *Sleep*, *33*(6), 781–792. https://doi.org/10.1093/sleep/33.6.781

Calvert, M., Kyte, D., Price, G., Valderas, J. M., & Hjollund, N. H. (2019). Maximising the impact of patient reported outcome assessment for patients and society. *BMJ: British Medical Journal (Clinical research ed.)*, *364*, k5267. https://doi.org/10.1136/bmj.k5267

Chen, L. C., Hsu, J. W., Huang, K. L., Bai, Y. M., Su, T. P., Li, C. T., Yang, A. C., Chang, W. H., Chen, T. J., Tsai, S. J., & Chen, M. H. (2016). Risk of developing major depression and anxiety disorders among women with endometriosis: A longitudinal follow-up study. *Journal of Affective Disorders*, *190*, 282–285. https://doi.org/10.1016/j.jad.2015.10.030

Chen, Y., Ju, P., Xia, Q., Cheng, P., Gao, J., Zhang, L., Gao, H., Cheng, X., Yu, T., Yan, J., Wang, Q., Zhu, C., & Zhang, X. (2022). Potential Role of Pain Catastrophic Thinking in Comorbidity Patients of Depression and Chronic Pain. *Frontiers in Psychiatry*, *13*, 839173. https://doi.org/10.3389/fpsyt.2022.839173

Chen, Z., Chen, X., Liu, M., Ma, L., & Yu, S. (2019). Volume of Hypothalamus as a Diagnostic Biomarker of Chronic Migraine. *Frontiers in Neurology*, *10*, 606. https://doi.org/10.3389/fneur.2019.00606

Chong, C. D., Plasencia, J. D., Frakes, D. H., & Schwedt, T. J. (2016). Structural alterations of the brainstem in migraine. *NeuroImage. Clinical*, *13*, 223–227. https://doi.org/10.1016/j.nicl.2016.10.023

Chrousos, G. P., & Gold, P. W. (1992). The concepts of stress and stress system disorders. Overview of physical and behavioral homeostasis. *The Journal of the American Medical Association*, *267*(9), 1244–1252.

Closs, S. J., Barr, B., Briggs, M., Cash, K., & Seers, K. (2004). A comparison of five pain assessment scales for nursing home residents with varying degrees of cognitive impairment. *Journal of Pain and Symptom Management*, *27*(3), 196–205. https://doi.org/10.1016/j.jpainsymman.2003.12.010

Colloca, L., Ludman, T., Bouhassira, D., Baron, R., Dickenson, A. H., Yarnitsky, D., Freeman, R., Truini, A., Attal, N., Finnerup, N. B., Eccleston, C., Kalso, E., Bennett, D. L., Dworkin, R. H., & Raja, S. N. (2017). Neuropathic pain. *Nature Reviews. Disease Primers*, *3*, 17002. https://doi.org/10.1038/nrdp.2017.2

Condés-Lara, M., Martínez-Lorenzana, G., Rubio-Beltrán, E., Rodríguez-Jiménez, J., Rojas-Piloni, G., & González-Hernández, A. (2015). Hypothalamic paraventricular nucleus stimulation enhances c-Fos expression in spinal and supraspinal structures related to pain modulation. *Neuroscience Research*, *98*, 59–63. https://doi.org/10.1016/j.neures.2015.04.004

Coxon, L., Wiech, K., & Vincent, K. (2021). Is There a Neuropathic-Like Component to Endometriosis-Associated Pain? Results From a Large Cohort Questionnaire Study. *Frontiers in Pain Research (Lausanne, Switzerland)*, *2*, 743812. https://doi.org/10.3389/fpain.2021.743812

Dale, A. M., Fischl, B., & Sereno, M. I. (1999). Cortical surface-based analysis. I. Segmentation and surface reconstruction. *NeuroImage*, *9*(2), 179–194. https://doi.org/10.1006/nimg.1998.0395

Dale, A. M., & Sereno, M. I. (1993). Improved Localizadon of Cortical Activity by Combining EEG and MEG with MRI Cortical Surface Reconstruction: A Linear Approach. *Journal of Cognitive Neuroscience*, *5*(2), 162–176. https://doi.org/10.1162/jocn.1993.5.2.162

Defrère, S., Lousse, J. C., González-Ramos, R., Colette, S., Donnez, J., & Van Langendonckt, A. (2008). Potential involvement of iron in the pathogenesis of peritoneal endometriosis. *Molecular Human Reproduction*, *14*(7), 377–385. https://doi.org/10.1093/molehr/gan033

Denuelle, M., Fabre, N., Payoux, P., Chollet, F., & Geraud, G. (2007). Hypothalamic activation in spontaneous migraine attacks. *Headache*, *47*(10), 1418–1426. https://doi.org/10.1111/j.1526-4610.2007.00776.x

Di Costanzo, A., Trojsi, F., Tosetti, M., Giannatempo, G. M., Nemore, F., Piccirillo, M., Bonavita, S., Tedeschi, G., & Scarabino, T. (2003). High-field proton MRS of human brain. *European Journal of Radiology*, *48*(2), 146–153. https://doi.org/10.1016/j.ejrad.2003.08.009

Dickerson, B. C., Fenstermacher, E., Salat, D. H., Wolk, D. A., Maguire, R. P., Desikan, R., Pacheco, J., Quinn, B. T., Van der Kouwe, A., Greve, D. N., Blacker, D., Albert, M. S., Killiany, R. J., & Fischl, B. (2008). Detection of cortical thickness correlates of cognitive performance: Reliability across MRI scan sessions, scanners, and field strengths. *NeuroImage*, *39*(1), 10–18. https://doi.org/10.1016/j.neuroimage.2007.08.042

Donatti, L., Malvezzi, H., Azevedo, B. C., Baracat, E. C., & Podgaec, S. (2022). Cognitive Behavioral Therapy in Endometriosis, Psychological Based Intervention: A Systematic Review. Intervenção psicológica baseada na terapia cognitivo-comportamental na endometriose: Uma revisão sistemática. *Revista brasileira de ginecologia e obstetricia*, *44*(3), 295–303. https://doi.org/10.1055/s-0042-1742406

Fallon, N., Alghamdi, J., Chiu, Y., Sluming, V., Nurmikko, T., & Stancak, A. (2013). Structural alterations in brainstem of fibromyalgia syndrome patients correlate with sensitivity to mechanical pressure. *NeuroImage. Clinical*, *3*, 163–170. https://doi.org/10.1016/j.nicl.2013.07.011

Faul, F., Erdfelder, E., Lang, A. G., & Buchner, A. (2007). G*Power 3: a flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behavior Research Methods*, *39*(2), 175–191. https://doi.org/10.3758/bf03193146

Faul, F., Erdfelder, E., Buchner, A., & Lang, A.G. (2009). Statistical power analyses using G*Power 3.1: Tests for correlation and regression analyses. *Behavior Research Methods*, *41*, 1149–1160. https://doi.org/10.3758/BRM.41.4.1149

Ferraro, S., Nigri, A., Bruzzone, M. G., Medina Carrion, J. P., Fedeli, D., Demichelis, G., Chiapparini, L., Ciullo, G., Gonzalez, A. A., Proietti Cecchini, A., Giani, L., Becker, B., & Leone, M. (2024). Involvement of the ipsilateral-to-the-pain anterior-superior hypothalamic subunit in chronic cluster headache. *The Journal of Headache and Pain*, *25*(1), 7. https://doi.org/10.1186/s10194-023-01711-0

Fischl, B., & Dale, A. M. (2000). Measuring the thickness of the human cerebral cortex from magnetic resonance images. *Proceedings of the National Academy of Sciences of the United States of America*, *97*(20), 11050–11055. https://doi.org/10.1073/pnas.200033797

Fischl, B., Liu, A., & Dale, A. M. (2001). Automated manifold surgery: constructing geometrically accurate and topologically correct models of the human cerebral cortex. *IEEE Transactions on Medical Imaging*, *20*(1), 70–80. https://doi.org/10.1109/42.906426

Fischl, B., Salat, D. H., Busa, E., Albert, M., Dieterich, M., Haselgrove, C., van der Kouwe, A., Killiany, R., Kennedy, D., Klaveness, S., Montillo, A., Makris, N., Rosen, B., & Dale, A. M. (2002). Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain. *Neuron*, *33*(3), 341–355. https://doi.org/10.1016/s0896-6273(02)00569-x

Fischl, B., Salat, D. H., van der Kouwe, A. J., Makris, N., Ségonne, F., Quinn, B. T., & Dale, A. M. (2004). Sequence-independent segmentation of magnetic resonance images. *NeuroImage*, *23 Suppl 1*, S69–S84. https://doi.org/10.1016/j.neuroimage.2004.07.016

Fischl, B., Sereno, M. I., Tootell, R. B., & Dale, A. M. (1999). High-resolution intersubject averaging and a coordinate system for the cortical surface. *Human Brain Mapping*, *8*(4), 272–284. https://doi.org/10.1002/(sici)1097-0193(1999)8:4<272::aid-hbm10>3.0.co;2-4

Fitzcharles, M. A., Cohen, S. P., Clauw, D. J., Littlejohn, G., Usui, C., & Häuser, W. (2021). Nociplastic pain: towards an understanding of prevalent pain conditions. *Lancet*, *397*(10289), 2098–2110. https://doi.org/10.1016/S0140-6736(21)00392-5

Foong, F. W., & Duggan, A. W. (1986). Brain-stem areas tonically inhibiting dorsal horn neurones: studies with microinjection of the GABA analogue piperidine-4-

sulphonic acid. *Pain*, *27*(3), 361–371. https://doi.org/10.1016/0304-3959(86)90160-0

Fox, N., & Mathers, N. (1997). Empowering research: statistical power in general practice research. *Family Practice*, *14*(4), 324–329. https://doi.org/10.1093/fampra/14.4.324

Freynhagen, R., Parada, H. A., Calderon-Ospina, C. A., Chen, J., Rakhmawati Emril, D., Fernández-Villacorta, F. J., Franco, H., Ho, K. Y., Lara-Solares, A., Li, C. C., Mimenza Alvarado, A., Nimmaanrat, S., Dolma Santos, M., & Ciampi de Andrade, D. (2019). Current understanding of the mixed pain concept: a brief narrative review. *Current Medical Research and Opinion*, *35*(6), 1011–1018. https://doi.org/10.1080/03007995.2018.1552042

Friedl, F., Riedl, D., Fessler, S., Wildt, L., Walter, M., Richter, R., Schüßler, G., & Böttcher, B. (2015). Impact of endometriosis on quality of life, anxiety, and depression: an Austrian perspective. *Archives of Gynecology and Obstetrics*, *292*(6), 1393–1399. https://doi.org/10.1007/s00404-015-3789-8

Gauriau, C., & Bernard, J. F. (2002). Pain pathways and parabrachial circuits in the rat. *Experimental Physiology*, *87*(2), 251–258. https://doi.org/10.1113/eph8702357

Gebhart G. F. (2004). Descending modulation of pain. *Neuroscience and Biobehavioral Reviews*, *27*(8), 729–737. https://doi.org/10.1016/j.neubiorev.2003.11.008

Geiger, J., Bley, T., Uhl, M., Frydrychowicz, A., Langer, M., & Markl, M. (2010). Diagnostic value of T2-weighted imaging for the detection of superficial cranial artery inflammation in giant cell arteritis. *Journal of Magnetic Resonance Imaging: JMRI*, *31*(2), 470–474. https://doi.org/10.1002/jmri.22047

Ghai, V., Jan, H., Shakir, F., Haines, P., & Kent, A. (2020). Diagnostic delay for superficial and deep endometriosis in the United Kingdom. *Journal of Obstetrics and Gynaecology*, *40*(1), 83–89. https://doi.org/10.1080/01443615.2019.1603217

González Ballester, M. A., Zisserman, A. P., & Brady, M. (2002). Estimation of the partial volume effect in MRI. *Medical Image Analysis*, *6*(4), 389–405. https://doi.org/10.1016/s1361-8415(02)00061-0

Grover, V. P., Tognarelli, J. M., Crossey, M. M., Cox, I. J., Taylor-Robinson, S. D., & McPhail, M. J. (2015). Magnetic Resonance Imaging: Principles and Techniques: Lessons for Clinicians. *Journal of Clinical and Experimental Hepatology*, *5*(3), 246–255. https://doi.org/10.1016/j.jceh.2015.08.001

Gruber, B., Froeling, M., Leiner, T., & Klomp, D. W. J. (2018). RF coils: A practical guide for nonphysicists. *Journal of Magnetic Resonance Imaging: JMRI*, *48*(3), 590–604. https://doi.org/10.1002/jmri.26187

Gustin, S. M., McKay, J. G., Petersen, E. T., Peck, C. C., Murray, G. M., & Henderson, L. A. (2014). Subtle alterations in brain anatomy may change an individual's personality in chronic pain. *PLoS One*, *9*(10), e109664. https://doi.org/10.1371/journal.pone.0109664

Guyatt, G. H., Townsend, M., Berman, L. B., & Keller, J. L. (1987). A comparison of Likert and visual analogue scales for measuring change in function. *Journal of Chronic Diseases*, *40*(12), 1129–1133. https://doi.org/10.1016/0021-9681(87)90080-4

Hadjistavropoulos, T., & Craig, K. D. (2002). A theoretical framework for understanding self-report and observational measures of pain: a communications model. *Behaviour Research and Therapy*, *40*(5), 551–570. https://doi.org/10.1016/s0005-7967(01)00072-9

Haghparast, A., Soltani-Hekmat, A., Khani, A., & Komaki, A. (2007). Role of glutamatergic receptors located in the nucleus raphe magnus on antinociceptive effect of morphine microinjected into the nucleus cuneiformis of rat. *Neuroscience Letters*, *427*(1), 44–49. https://doi.org/10.1016/j.neulet.2007.09.003

Harte SE, Harris RE, Clauw DJ (2018).The neurobiology of central sensitization. *Journal of Applied Behavioral Research, 23,* e12137. https://doi.org/10.1111/jabr.12137

Hassett, A.L., Whibley, D., Kratz, A. and Williams, D.A. (2020), Measures for the Assessment of Pain in Adults. *Arthritis Care and Research, 72,* 342–357. https://doi.org/10.1002/acr.24222

Heim, C., Ehlert, U., Hanker, J. P., & Hellhammer, D. H. (1998). Abuse-related posttraumatic stress disorder and alterations of the hypothalamic-pituitary-adrenal axis in women with chronic pelvic pain. *Psychosomatic Medicine*, *60*(3), 309–318. https://doi.org/10.1097/00006842-199805000-00017

Heim, C., Ehlert, U., & Hellhammer, D. H. (2000). The potential role of hypocortisolism in the pathophysiology of stress-related bodily disorders. *Psychoneuroendocrinology*, *25*(1), 1–35. https://doi.org/10.1016/s0306-4530(99)00035-9

Heinricher, M. M., Neubert, M. J., Martenson, M. E., & Gonçalves, L. (2004). Prostaglandin E2 in the medial preoptic area produces hyperalgesia and activates pain-modulating circuitry in the rostral ventromedial medulla. *Neuroscience*, *128*(2), 389–398. https://doi.org/10.1016/j.neuroscience.2004.06.050

Hsu, A. L., Khachikyan, I., & Stratton, P. (2010). Invasive and noninvasive methods for the diagnosis of endometriosis. *Clinical Obstetrics and Gynecology*, *53*(2), 413–419. https://doi.org/10.1097/GRF.0b013e3181db7ce8

Iglesias, J. E., Van Leemput, K., Bhatt, P., Casillas, C., Dutt, S., Schuff, N., Truran-Sacrey, D., Boxer, A., Fischl, B., & Alzheimer's Disease Neuroimaging Initiative (2015). Bayesian segmentation of brainstem structures in MRI. *NeuroImage*, *113*, 184–195. https://doi.org/10.1016/j.neuroimage.2015.02.065

International Association for the Study of Pain (2011). *Terminology*. International Association for the Study of Pain. Accessed on 25 February 2024. https://www.iasp-pain.org/resources/terminology/

Islam, J., Kc, E., So, K. H., Kim, S., Kim, H. K., Park, Y. Y., & Park, Y. S. (2023). Modulation of trigeminal neuropathic pain by optogenetic inhibition of posterior hypothalamus in CCI-ION rat. *Scientific Reports*, *13*(1), 489. https://doi.org/10.1038/s41598-023-27610-7

Jeong, Y., & Holden, J. E. (2009). The role of spinal orexin-1 receptors in posterior hypothalamic modulation of neuropathic pain. *Neuroscience*, *159*(4), 1414–1421. https://doi.org/10.1016/j.neuroscience.2009.02.006

Jiang, Y., Oathes, D., Hush, J., Darnall, B., Charvat, M., Mackey, S., & Etkin, A. (2016). Perturbed connectivity of the amygdala and its subregions with the central executive and default mode networks in chronic pain. *Pain*, *157*(9), 1970–1978. https://doi.org/10.1097/j.pain.0000000000000606

Katz, J., & Melzack, R. (1999). Measurement of pain. *The Surgical Clinics of North America*, *79*(2), 231–252. https://doi.org/10.1016/s0039-6109(05)70381-9

Khaleghzadeh-Ahangar, H., Sadeghimahalli, F., Khandan, S., Shahabi, S., & Moghadamnia, A. (2020). Insulin within the Arcuate Nucleus Has Paradoxical Effects on Nociception in Healthy and Diabetic Rats. *Basic and Clinical Neuroscience*, *11*(6), 727–736. https://doi.org/10.32598/bcn.11.6.1983.1

Khin Hla, T., Hegarty, M., Russell, P., Drake-Brockman, T. F., Ramgolam, A., & von Ungern-Sternberg, B. S. (2014). Perception of pediatric pain: a comparison of postoperative pain assessments between child, parent, nurse, and independent observer. *Paediatric Anaesthesia*, *24*(11), 1127–1131. https://doi.org/10.1111/pan.12484

Khodaverdi, S., Alebouyeh, M. R., Sadegi, K., Mehdizadehkashi, A., Kaveh, M., Entezari, S. R., Mirzaei, H., Khaledi, M., & Khodaverdi, M. (2021). Superior hypogastric plexus block as an effective treatment method for endometriosis-related chronic pelvic pain: an open-label pilot clinical trial. *Journal of Obstetrics and Gynaecology*, *41*(6), 966–971. https://doi.org/10.1080/01443615.2020.1820468

Kong, D., Zhang, Y., Gao, P., Pan, C., Deng, H., Xu, S., Tang, D., Xiao, J., Jiao, Y., Yu, W., & Wen, D. (2023). The locus coeruleus input to the rostral ventromedial medulla mediates stress-induced colorectal visceral pain. *Acta Neuropathologica Communications*, *11*(1), 65. https://doi.org/10.1186/s40478-023-01537-6

Korszun A. (2000). Sleep and circadian rhythm disorders in fibromyalgia. *Current Rheumatology Reports*, *2*(2), 124–130. https://doi.org/10.1007/s11926-000-0052-4

Kummer, K. K., Mitrić, M., Kalpachidou, T., & Kress, M. (2020). The Medial Prefrontal Cortex as a Central Hub for Mental Comorbidities Associated with Chronic Pain. *International Journal of Molecular Sciences*, *21*(10), 3440. https://doi.org/10.3390/ijms21103440

Kundermann, B., Spernal, J., Huber, M. T., Krieg, J. C., & Lautenbacher, S. (2004). Sleep deprivation affects thermal pain thresholds but not somatosensory thresholds in healthy volunteers. *Psychosomatic Medicine*, *66*(6), 932–937. https://doi.org/10.1097/01.psy.0000145912.24553.c0

Kuner, R., & Flor, H. (2016). Structural plasticity and reorganisation in chronic pain. *Nature Reviews. Neuroscience*, *18*(1), 20–30. https://doi.org/10.1038/nrn.2016.162

Laganà, A. S., Vitale, S. G., Trovato, M. A., Palmara, V. I., Rapisarda, A. M., Granese, R., Sturlese, E., De Dominici, R., Alecci, S., Padula, F., Chiofalo, B., Grasso, R., Cignini, P., D'Amico, P., & Triolo, O. (2016). Full-Thickness Excision versus Shaving by Laparoscopy for Intestinal Deep Infiltrating Endometriosis: Rationale and Potential Treatment Options. *BioMed Research International*, *2016*, 3617179. https://doi.org/10.1155/2016/3617179

Lai, J. S., Cella, D., Choi, S., Junghaenel, D. U., Christodoulou, C., Gershon, R., & Stone, A. (2011). How item banks and their application can influence measurement practice in rehabilitation medicine: a PROMIS fatigue item bank example. *Archives of Physical Medicine and Rehabilitation*, *92*(10 Suppl), S20–S27. https://doi.org/10.1016/j.apmr.2010.08.033

Landgraf, R., & Neumann, I. D. (2004). Vasopressin and oxytocin release within the brain: a dynamic concept of multiple and variable modes of neuropeptide communication. *Frontiers in Neuroendocrinology*, *25*(3–4), 150–176. https://doi.org/10.1016/j.yfrne.2004.05.001

Latremoliere, A., & Woolf, C. J. (2009). Central sensitization: a generator of pain hypersensitivity by central neural plasticity. *The Journal of Pain*, *10*(9), 895–926. https://doi.org/10.1016/j.jpain.2009.06.012

Lazzeri, L., Orlandini, C., Vannuccini, S., Pinzauti, S., Tosti, C., Zupi, E., Nappi, R. E., & Petraglia, F. (2015). Endometriosis and perceived stress: impact of surgical and medical treatment. *Gynecologic and Obstetric Investigation*, *79*(4), 229–233. https://doi.org/10.1159/000368776

Lee, H. J., Cho, Y., Joo, H., Jeon, J. Y., Jang, Y. E., & Kim, J. T. (2021). Comparative study of verbal rating scale and numerical rating scale to assess postoperative pain intensity in the post anesthesia care unit: A prospective observational cohort study. *Medicine*, *100*(6), e24314. https://doi.org/10.1097/MD.0000000000024314

Leigheb, M., Sabbatini, M., Baldrighi, M., Hasenboehler, E. A., Briacca, L., Grassi, F., Cannas, M., Avanzi, G., & Castello, L. M. (2017). Prospective analysis of pain and pain management in an emergency department. *Acta Bio-medica: Atenei Parmensis*, *88*(4S), 19–30. https://doi.org/10.23750/abm.v88i4-S.6790

Leite-Almeida, H., Valle-Fernandes, A., & Almeida, A. (2006). Brain projections from the medullary dorsal reticular nucleus: an anterograde and retrograde tracing study in the rat. *Neuroscience*, *140*(2), 577–595. https://doi.org/10.1016/j.neuroscience.2006.02.022

Li, X. H., Matsuura, T., Xue, M., Chen, Q. Y., Liu, R. H., Lu, J. S., Shi, W., Fan, K., Zhou, Z., Miao, Z., Yang, J., Wei, S., Wei, F., Chen, T., & Zhuo, M. (2021). Oxytocin in the anterior cingulate cortex attenuates neuropathic pain and emotional anxiety by inhibiting presynaptic long-term potentiation. *Cell Reports*, *36*(3), 109411. https://doi.org/10.1016/j.celrep.2021.109411

Maddern, J., Grundy, L., Castro, J., & Brierley, S. M. (2020). Pain in Endometriosis. *Frontiers in Cellular Neuroscience*, *14*, 590823. https://doi.org/10.3389/fncel.2020.590823

Mahmoudi, M., & Zarrindast, M. R. (2002). Effect of intracerebroventricular injection of GABA receptor agents on morphine-induced antinociception in the formalin test. *Journal of Psychopharmacology*, *16*(1), 85–91. https://doi.org/10.1177/026988110201600108

Maixner, W., Fillingim, R. B., Williams, D. A., Smith, S. B., & Slade, G. D. (2016). Overlapping Chronic Pain Conditions: Implications for Diagnosis and

Classification. *The Journal of Pain*, *17*(9 Suppl), T93–T107. https://doi.org/10.1016/j.jpain.2016.06.002

Mansour, A. R., Farmer, M. A., Baliki, M. N., & Apkarian, A. V. (2014). Chronic pain: the role of learning and brain plasticity. *Restorative Neurology and Neuroscience*, *32*(1), 129–139. https://doi.org/10.3233/RNN-139003

Manuel E, Vogt E, Jiang C, Caldwell M, and Marsh E. (2019). Analysis of National Emergency Department Utilization for Endometriosis by Race and Socioeconomic Status. *Reproductive Sciences*, *30,* 51A. https://scholarlycommons.henryford.com/administration_mtgabstracts/12/

Marcus, J. N., Aschkenasi, C. J., Lee, C. E., Chemelli, R. M., Saper, C. B., Yanagisawa, M., & Elmquist, J. K. (2001). Differential expression of orexin receptors 1 and 2 in the rat brain. *The Journal of Comparative Neurology*, *435*(1), 6–25. https://doi.org/10.1002/cne.1190

Matsui, Y., Sakurai, J., Hiraki, T., Okamoto, S., Iguchi, T., Tomita, K., Uka, M., Gobara, H., & Kanazawa, S. (2019). MRI-guided percutaneous needle biopsy with 1.2T open MRI: study protocol for a prospective feasibility study (SCIRO-1701). *Nagoya Journal of Medical Science*, *81*(3), 463–468. https://doi.org/10.18999/nagjms.81.3.463

Maybin, J. A., & Critchley, H. O. (2015). Menstrual physiology: implications for endometrial pathology and beyond. *Human Reproduction Update*, *21*(6), 748–761. https://doi.org/10.1093/humupd/dmv038

McMullen, K., Bamer, A., Ryan, C. M., Schneider, J. C., Gibran, N., Stewart, B. T., Mroz, T., Wolf, S., & Amtmann, D. (2022). Validation of PROMIS-29 domain scores among adult burn survivors: A National Institute on Disability, Independent Living, and Rehabilitation Research Burn Model System Study. *The Journal of Trauma and Acute Care Surgery*, *92*(1), 213–222. https://doi.org/10.1097/TA.0000000000003365

Medina, M. G., & Lebovic, D. I. (2009). Endometriosis-associated nerve fibers and pain. *Acta Obstetricia et Gynecologica Scandinavica*, *88*(9), 968–975. https://doi.org/10.1080/00016340903176826

Mehedintu, C., Plotogea, M. N., Ionescu, S., & Antonovici, M. (2014). Endometriosis still a challenge. *Journal of Medicine and Life*, *7*(3), 349–357.

Michinaga, S., & Koyama, Y. (2015). Pathogenesis of brain edema and investigation into anti-edema drugs. *International Journal of Molecular Sciences*, *16*(5), 9949–9975. https://doi.org/10.3390/ijms16059949

Miró, J., Roman-Juan, J., Sánchez-Rodríguez, E., Solé, E., Castarlenas, E., & Jensen, M. P. (2023). Chronic Pain and High Impact Chronic Pain in Children and Adolescents: A Cross-Sectional Study. *The Journal of Pain*, *24*(5), 812–823. https://doi.org/10.1016/j.jpain.2022.12.007

Misaki, M., Savitz, J., Zotev, V., Phillips, R., Yuan, H., Young, K. D., Drevets, W. C., & Bodurka, J. (2015). Contrast enhancement by combining T1- and T2-weighted structural brain MR Images. *Magnetic Resonance in Medicine*, *74*(6), 1609–1620. https://doi.org/10.1002/mrm.25560

Morassutto, C., Monasta, L., Ricci, G., Barbone, F., & Ronfani, L. (2016). Incidence and Estimated Prevalence of Endometriosis and Adenomyosis in Northeast Italy: A Data Linkage Study. *PLoS One*, *11*(4), e0154227. https://doi.org/10.1371/journal.pone.0154227

Nagai, T., Maeda, T., Imai, H., McGeer, P. L., & McGeer, E. G. (1985). Distribution of GABA-T-intensive neurons in the rat hindbrain. *The Journal of Comparative Neurology*, *231*(2), 260–269. https://doi.org/10.1002/cne.902310213

Napadow, V., Sclocco, R., & Henderson, L. A. (2019). Brainstem neuroimaging of nociception and pain circuitries. *Pain Reports*, *4*(4), e745. https://doi.org/10.1097/PR9.0000000000000745

Nelson, F., Poonawalla, A., Hou, P., Wolinsky, J. S., & Narayana, P. A. (2008). 3D MPRAGE improves classification of cortical lesions in multiple sclerosis. *Multiple Sclerosis*, *14*(9), 1214–1219. https://doi.org/10.1177/1352458508094644

Nnoaham, K. E., Hummelshoj, L., Webster, P., d'Hooghe, T., de Cicco Nardone, F., de Cicco Nardone, C., Jenkinson, C., Kennedy, S. H., Zondervan, K. T., & World Endometriosis Research Foundation Global Study of Women's Health consortium (2011). Impact of endometriosis on quality of life and work productivity: a multicenter study across ten countries. *Fertility and Sterility*, *96*(2), 366–373.e8. https://doi.org/10.1016/j.fertnstert.2011.05.090

Norman G. (2010). Likert scales, levels of measurement and the "laws" of statistics. *Advances in Health Sciences Education: Theory and Practice*, *15*(5), 625–632. https://doi.org/10.1007/s10459-010-9222-y

Ong, W. Y., Stohler, C. S., & Herr, D. R. (2019). Role of the Prefrontal Cortex in Pain Processing. *Molecular Neurobiology*, *56*(2), 1137–1166. https://doi.org/10.1007/s12035-018-1130-9

Pagliusi, M., Jr, & Gomes, F. V. (2023). The Role of The Rostral Ventromedial Medulla in Stress Responses. *Brain Sciences*, *13*(5), 776. https://doi.org/10.3390/brainsci13050776

Pecorelli, N., Guarneri, G., Vallorani, A., Limongi, C., Licinio, A. W., Di Salvo, F., Tamburrino, D., Partelli, S., Crippa, S., & Falconi, M. (2023). Validation of the PROMIS-29 Questionnaire as a Measure of Recovery After Pancreatic Surgery. *Annals of Surgery*, *278*(5), 732–739. https://doi.org/10.1097/SLA.0000000000006020

Pertovaara A. (2006). Noradrenergic pain modulation. *Progress in Neurobiology*, *80*(2), 53–83. https://doi.org/10.1016/j.pneurobio.2006.08.001

Petrella, J. R., Shah, L. M., Harris, K. M., Friedman, A. H., George, T. M., Sampson, J. H., Pekala, J. S., & Voyvodic, J. T. (2006). Preoperative functional MR imaging localization of language and motor areas: effect on therapeutic decision making in patients with potentially resectable brain tumors. *Radiology*, *240*(3), 793–802. https://doi.org/10.1148/radiol.2403051153

Peyron, C., Tighe, D. K., van den Pol, A. N., de Lecea, L., Heller, H. C., Sutcliffe, J. G., & Kilduff, T. S. (1998). Neurons containing hypocretin (orexin) project to multiple neuronal systems. *The Journal of Neuroscience*, *18*(23), 9996–10015. https://doi.org/10.1523/JNEUROSCI.18-23-09996.1998

Peyron R. (2014). Imagerie de la douleur [Functional imaging of pain]. *Biologie aujourd'hui*, *208*(1), 5–12. https://doi.org/10.1051/jbio/2014003

Pilkonis, P. A., Choi, S. W., Reise, S. P., Stover, A. M., Riley, W. T., Cella, D., & PROMIS Cooperative Group (2011). Item banks for measuring emotional distress from the Patient-Reported Outcomes Measurement Information System (PROMIS®): depression, anxiety, and anger. *Assessment*, *18*(3), 263–283. https://doi.org/10.1177/1073191111411667

Pilkonis, P. A., Yu, L., Dodds, N. E., Johnston, K. L., Maihoefer, C. C., & Lawrence, S. M. (2014). Validation of the depression item bank from the Patient-Reported Outcomes Measurement Information System (PROMIS) in a three-month observational study. *Journal of Psychiatric Research*, *56*, 112–119. https://doi.org/10.1016/j.jpsychires.2014.05.010

Pope, C. J., Sharma, V., Sharma, S., & Mazmanian, D. (2015). A Systematic Review of the Association Between Psychiatric Disturbances and Endometriosis. *Journal of obstetrics and gynaecology Canada: JOGC = Journal d'obstetrique et gynecologie du Canada : JOGC*, *37*(11), 1006–1015. https://doi.org/10.1016/s1701-2163(16)30050-0

Raja, S. N., Carr, D. B., Cohen, M., Finnerup, N. B., Flor, H., Gibson, S., Keefe, F. J., Mogil, J. S., Ringkamp, M., Sluka, K. A., Song, X. J., Stevens, B., Sullivan, M. D., Tutelman, P. R., Ushida, T., & Vader, K. (2020). The revised International Association for the Study of Pain definition of pain: concepts, challenges, and compromises. *Pain*, *161*(9), 1976–1982. https://doi.org/10.1097/j.pain.0000000000001939

Ramezanzadeh, F., Aghssa, M. M., Abedinia, N., Zayeri, F., Khanafshar, N., Shariat, M., & Jafarabadi, M. (2004). A survey of relationship between anxiety, depression and duration of infertility. *BMC Women's Health*, *4*(1), 9. https://doi.org/10.1186/1472-6874-4-9

Reese, K. A., Reddy, S., & Rock, J. A. (1996). Endometriosis in an adolescent population: the Emory experience. *Journal of Pediatric and Adolescent Gynecology*, *9*(3), 125–128. https://doi.org/10.1016/s1083-3188(96)70021-9

Reuter, M., Rosas, H. D., & Fischl, B. (2010). Highly accurate inverse consistent registration: a robust approach. *NeuroImage*, *53*(4), 1181–1196. https://doi.org/10.1016/j.neuroimage.2010.07.020

Rose, M., Bjorner, J. B., Gandek, B., Bruce, B., Fries, J. F., & Ware, J. E., Jr (2014). The PROMIS Physical Function item bank was calibrated to a standardized metric and shown to improve measurement efficiency. *Journal of Clinical Epidemiology*, *67*(5), 516–526. https://doi.org/10.1016/j.jclinepi.2013.10.024

Ruscheweyh, R., Kühnel, M., Filippopulos, F., Blum, B., Eggert, T., & Straube, A. (2014). Altered experimental pain perception after cerebellar infarction. *Pain*, *155*(7), 1303–1312. https://doi.org/10.1016/j.pain.2014.04.006

Sara S. J. (2009). The locus coeruleus and noradrenergic modulation of cognition. *Nature reviews. Neuroscience*, *10*(3), 211–223. https://doi.org/10.1038/nrn2573

Saunders, P. T. K., & Horne, A. W. (2021). Endometriosis: Etiology, pathobiology, and therapeutic prospects. *Cell*, *184*(11), 2807–2824. https://doi.org/10.1016/j.cell.2021.04.041

Schappert, S. M., & Burt, C. W. (2006). Ambulatory care visits to physician offices, hospital outpatient departments, and emergency departments: United States, 2001–02. *Vital and health statistics. Series 13, Data from the National Health Survey*, (159), 1–66.

Schindler, S., Schmidt, L., Stroske, M., Storch, M., Anwander, A., Trampel, R., Strauß, M., Hegerl, U., Geyer, S., & Schönknecht, P. (2019). Hypothalamus enlargement in mood disorders. *Acta Psychiatrica Scandinavica*, *139*(1), 56–67. https://doi.org/10.1111/acps.12958

Ségonne, F., Dale, A. M., Busa, E., Glessner, M., Salat, D., Hahn, H. K., & Fischl, B. (2004). A hybrid approach to the skull stripping problem in MRI. *NeuroImage*, *22*(3), 1060–1075. https://doi.org/10.1016/j.neuroimage.2004.03.032

Ségonne, F., Pacheco, J., & Fischl, B. (2007). Geometrically accurate topology-correction of cortical surfaces using nonseparating loops. *IEEE Transactions on Medical Imaging*, *26*(4), 518–529. https://doi.org/10.1109/TMI.2006.887364

Simoens, S., Hummelshoj, L., & D'Hooghe, T. (2007). Endometriosis: cost estimates and methodological perspective. *Human Reproduction Update*, *13*(4), 395–404. https://doi.org/10.1093/humupd/dmm010

Simons, L. E., Sieberg, C. B., Carpino, E., Logan, D., & Berde, C. (2011). The Fear of Pain Questionnaire (FOPQ): assessment of pain-related fear among children and adolescents with chronic pain. *The Journal of Pain*, *12*(6), 677–686. https://doi.org/10.1016/j.jpain.2010.12.008

Sled, J. G., Zijdenbos, A. P., & Evans, A. C. (1998). A nonparametric method for automatic correction of intensity nonuniformity in MRI data. *IEEE Transactions on Medical Imaging*, *17*(1), 87–97. https://doi.org/10.1109/42.668698

Smith, K. S., & Rudolph, U. (2012). Anxiety and depression: mouse genetics and pharmacological approaches to the role of GABA(A) receptor subtypes. *Neuropharmacology*, *62*(1), 54–62. https://doi.org/10.1016/j.neuropharm.2011.07.026

Smorgick, N., Marsh, C. A., As-Sanie, S., Smith, Y. R., & Quint, E. H. (2013). Prevalence of pain syndromes, mood conditions, and asthma in adolescents and young women with endometriosis. *Journal of Pediatric and Adolescent Gynecology*, *26*(3), 171–175. https://doi.org/10.1016/j.jpag.2012.12.006

Soliman, A. M., Yang, H., Du, E. X., Kelley, C., & Winkel, C. (2016). The direct and indirect costs associated with endometriosis: a systematic literature review. *Human Reproduction*, *31*(4), 712–722. https://doi.org/10.1093/humrep/dev335

Somigliana, E., Vigano, P., Barbara, G., & Vercellini, P. (2009). Treatment of endometriosis-related pain: options and outcomes. *Frontiers in Bioscience (Elite edition)*, *1*(2), 455–465. https://doi.org/10.2741/e41

Stamatovic, S. M., Dimitrijevic, O. B., Keep, R. F., & Andjelkovic, A. V. (2006). Inflammation and brain edema: new insights into the role of chemokines and their receptors. *Acta Neurochirurgica. Supplement*, *96*, 444–450. https://doi.org/10.1007/3-211-30714-1_91

Stevens, F. L., Hurley, R. A., & Taber, K. H. (2011). Anterior cingulate cortex: unique role in cognition and emotion. *The Journal of Neuropsychiatry and Clinical Neurosciences*, *23*(2), 121–125. https://doi.org/10.1176/jnp.23.2.jnp121

Stone, A. A., Broderick, J. E., Kaell, A. T., DelesPaul, P. A., & Porter, L. E. (2000). Does the peak-end phenomenon observed in laboratory pain studies apply to real-world pain in rheumatoid arthritics? *The Journal of Pain*, *1*(3), 212–217. https://doi.org/10.1054/jpai.2000.7568

Suárez-Pereira, I., Llorca-Torralba, M., Bravo, L., Camarena-Delgado, C., Soriano-Mas, C., & Berrocoso, E. (2022). The Role of the Locus Coeruleus in Pain and Associated Stress-Related Disorders. *Biological Psychiatry*, *91*(9), 786–797. https://doi.org/10.1016/j.biopsych.2021.11.023

Sullivan, G. M., & Artino, A. R., Jr (2013). Analyzing and interpreting data from Likert-type scales. *Journal of Graduate Medical Education*, *5*(4), 541–542. https://doi.org/10.4300/JGME-5-4-18

Sullivan, M. J. L., Bishop, S. R., & Pivik, J. (1995). The Pain Catastrophizing Scale: Development and validation. *Psychological Assessment, 7*(4), 524–532. https://doi.org/10.1037/1040-3590.7.4.524

Szabo, E., Timmers, I., Borsook, D., Simons, L. E., & Sieberg, C. B. (2022). Altered anterior insula functional connectivity in adolescent and young women with endometriosis-associated pain: Pilot resting-state fMRI study. *European Journal of Paediatric Neurology: EJPN*, *41*, 80–90. https://doi.org/10.1016/j.ejpn.2022.10.004

Taylor, B. K., & Westlund, K. N. (2017). The noradrenergic locus coeruleus as a chronic pain generator. *Journal of Neuroscience Research*, *95*(6), 1336–1346. https://doi.org/10.1002/jnr.23956

Thapaliya, K., Marshall-Gradisnik, S., Barth, M., Eaton-Fitch, N., & Barnden, L. (2023). Brainstem volume changes in myalgic encephalomyelitis/chronic fatigue syndrome and long COVID patients. *Frontiers in Neuroscience*, *17*, 1125208. https://doi.org/10.3389/fnins.2023.1125208

Thompson, J. M., & Neugebauer, V. (2019). Cortico-limbic pain mechanisms. *Neuroscience Letters*, *702*, 15–23. https://doi.org/10.1016/j.neulet.2018.11.037

Tokushige, N., Markham, R., Russell, P., & Fraser, I. S. (2007). Different types of small nerve fibers in eutopic endometrium and myometrium in women with endometriosis. *Fertility and Sterility*, *88*(4), 795–803. https://doi.org/10.1016/j.fertnstert.2006.12.078

Vallvé-Juanico, J., Houshdaran, S., & Giudice, L. C. (2019). The endometrial immune environment of women with endometriosis. *Human Reproduction Update*, *25*(5), 564–591. https://doi.org/10.1093/humupd/dmz018

Van Balen, E. C., Haverman, L., Hassan, S., Taal, E. M., Smit, C., Driessens, M. H., Beckers, E. A. M., Coppens, M., Eikenboom, J., Hooimeijer, H. L., Leebeek, F. W. G., van Vulpen, L. F. D., Schols, S. E. M., Terwee, C. B., Rosendaal, F. R., van der Bom, J. G., & Gouw, S. C. (2021). Validation of PROMIS Profile-29 in adults with hemophilia in the Netherlands. *Journal of Thrombosis and Haemostasis: JTH*, *19*(11), 2687–2701. https://doi.org/10.1111/jth.15454

Van Geuns, R. J., Wielopolski, P. A., de Bruin, H. G., Rensing, B. J., van Ooijen, P. M., Hulshoff, M., Oudkerk, M., & de Feyter, P. J. (1999). Basic principles of magnetic resonance imaging. *Progress in Cardiovascular Diseases*, *42*(2), 149–156. https://doi.org/10.1016/s0033-0620(99)70014-9

Velho, R. V., Taube, E., Sehouli, J., & Mechsner, S. (2021). Neurogenic Inflammation in the Context of Endometriosis-What Do We Know? *International Journal of Molecular Sciences*, *22*(23), 13102. https://doi.org/10.3390/ijms222313102

Verkauf B. S. (1987). Incidence, symptoms, and signs of endometriosis in fertile and infertile women. *The Journal of the Florida Medical Association*, *74*(9), 671–675.

Wei, Y., Liang, Y., Lin, H., Dai, Y., Shuzhong, Y. (2020). Autonomic nervous system and inflammation interaction in endometriosis-associated pain. *Journal of Neuroinflammation 17*, 80. https://doi.org/10.1186/s12974-020-01752-1

Weiller, C., May, A., Limmroth, V., Jüptner, M., Kaube, H., Schayck, R. V., Coenen, H. H., & Diener, H. C. (1995). Brain stem activation in spontaneous human migraine attacks. *Nature Medicine*, *1*(7), 658–660. https://doi.org/10.1038/nm0795-658

Westland J. C. (2022). Information loss and bias in likert survey responses. *PLoS One*, *17*(7), e0271949. https://doi.org/10.1371/journal.pone.0271949

Yang, S., & Chang, M. C. (2019). Chronic Pain: Structural and Functional Changes in Brain Structures and Associated Negative Affective States. *International Journal of Molecular Sciences*, *20*(13), 3130. https://doi.org/10.3390/ijms20133130

Yunus M. B. (2007). Role of central sensitization in symptoms beyond muscle pain, and the evaluation of a patient with widespread pain. *Best Practice & Research. Clinical Rheumatology*, *21*(3), 481–497. https://doi.org/10.1016/j.berh.2007.03.006

Zondervan, K. T., Becker, C. M., & Missmer, S. A. (2020). Endometriosis. *The New England Journal of Medicine*, *382*(13), 1244–1256. https://doi.org/10.1056/NEJMra1810764

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