



Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.



Symptomatic but not Asymptomatic COVID-19 Impairs Conditioned Pain Modulation in Young Adults

Jessica A. Peterson,^{*,†,‡} Michael G. Bembien,[‡] Rebecca D. Larson,[‡] Hugo Pereira,[‡] H. Michael Crowson,^{†,§} and Christopher D. Black[‡]

^{*}Pain Research and Intervention Center of Excellence (PRICE) at the University of Florida, Gainesville, Florida, [†]Department of Community Dentistry and Behavioral Science, the University of Florida, Gainesville, Florida, [‡]Department of Health and Exercise Science, University of Oklahoma, Norman, Oklahoma, [§]Department of Educational Psychology, University of Oklahoma, Norman, Oklahoma

Abstract: Pain is a common symptom reported in COVID-19 patients. Impaired endogenous pain-modulatory mechanisms such as conditioned pain modulation (CPM), and exercise-induced hypoalgesia (EIH) have been found in chronic pain conditions but is often overlooked in acute conditions that evoke painful symptoms, such as COVID-19. The purpose was to compare pressure-pain sensitivity, CPM, and EIH function among individuals who previously had COVID-19, both symptomatically and asymptotically, and a healthy control group. Pressure pain thresholds of 59 participants were assessed in the forearm and leg using a pressure algometer before and after 1) submersion of their dominant foot in cold water (2°C) for 1min; and 2) isometric knee extension performed to task-failure at 25% of their maximal contraction. The CPM response was attenuated in individuals who were infected with symptomatic COVID-19 (N = 26) compared to asymptomatic COVID-19 (N = 13) in arm (-1.0% ± 20.3 vs 33.3% ± 26.2; $P < .001$) and leg (12.8% ± 22.0 vs 33.8% ± 28.2; $P = .014$) and compared to controls (N = 20) in arm only (-1.0% ± 26.2 vs 23.4% ± 28.2; $P = .004$). The EIH response was not different between groups. CPM was impaired in individuals who had symptomatic COVID-19, which may have long-term implications on pain modulation.

Perspective: This study reveals that CPM was impaired in individuals who had symptomatic COVID-19 during the first wave of COVID-19, pre vaccine. These findings present a preliminary motive to study the long-term implications of COVID-19 and its effects on pain modulation.

© Published by Elsevier Inc. on behalf of United States Association for the Study of Pain, Inc.

Key words: Conditioned pain modulation, Exercise induced hyperalgesia, Pressure pain thresholds, COVID-19.

Introduction

Novel coronavirus disease 2019 (COVID-19) has infected more than 532 million individuals and lead to 6.3 million deaths since it was first identified in December of 2019,⁸⁵ making it one of, if not the

most significant public health issues of the past century. The majority of COVID-19 infections result in mild-to-moderate symptoms/disease with perhaps one-third being “asymptomatic”.⁴⁵ Although the risk of mortality and developing severe disease and mortality is low (<1.5%),^{79,85} a recent study suggests an excess mortality rate of approximately 3 times the reported number of COVID related deaths,⁷⁹ suggesting the impact of the pandemic has been significantly greater than indicated by reported death rates. The long-term consequences of infection remain somewhat unclear especially in those who were asymptotically infected. Persistence of symptoms for longer than 60 days, often termed “long-COVID” has been reported in roughly 30%⁷² of those reporting a symptomatic infection, but scant data exist on possible residual effects in individuals who

Received March 21, 2022; Revised June 7, 2022; Accepted June 15, 2022.
Support/grant: Funding for this study was provided by the Department of Health and Exercise Science Helen Riddle Dissertation Award and the College of Arts and Sciences Robberson Research Award to Jessica Peterson.

Conflicts of interest statement: The authors report no conflicts of interest.

Address reprint requests to Jessica Peterson, PhD, The University of Florida, Pain Research & Intervention Center of Excellence, 1329 SW 16th Street, Suite 5180, Gainesville, FL, 32610. E-mail: jpeterson@dental.ufl.edu

1526-5900/\$36.00

© Published by Elsevier Inc. on behalf of United States Association for the Study of Pain, Inc.

<https://doi.org/10.1016/j.jpain.2022.06.010>

were asymptomatic and/or do not present with long-COVID.

Pain is a common, acute symptom of active coronavirus infection (COVID-19). Myalgia and headache have been reported as the most common pain symptoms with sore throat, abdominal pain, and chest pain experienced less frequently.²⁸ Acute pain during COVID-19 infection likely originates from inflammation consequent to the infection.^{69,89} There is evidence that more severe acute symptoms are associated with a larger inflammatory response. Neuroinflammation and the presence of pro-inflammatory cytokines and chemokines in the peripheral and central nervous system can produce functional or structural abnormalities of neurons, that over time can develop into chronic pain³⁰ and has been previously observed following other infections with other viruses such as parvovirus, hepatitis B and C, HIV, etc.⁸² In addition to inflammatory mediated changes in neuron function, SARS-CoV-2 may also directly affect the nociceptive system via its action on the angiotensin converting enzyme 2 (ACE2) which has been shown to be a receptor for the SARS-CoV-2 spike protein.²⁹ ACE2 expressing sensory neurons synapse with neurons in the central nervous system (CNS) which can lead to pain symptomology.^{46,48}

Impairments in pain modulation, often assessed via conditioned pain modulation (CPM) or exercise-induced hypoalgesia (EIH), have been demonstrated in a host of chronic pain conditions including chronic fatigue syndrome (CFS),^{47,78} and fibromyalgia (FM).^{11,33,34,36,67,68,86} Impaired endogenous pain modulation has been shown to be predictive of the development of chronic pain^{38,86} and is impaired in those with chronic pain [see³ for review]. Therefore, a better understanding of the effects of COVID-19 infection on pain modulation could provide insight into the potential future risk of chronic pain in the hundreds of millions of individuals previously infected with COVID-19, who did not experience long lasting symptoms. As such, this study sought to compare pain modulatory function assessed via CPM and EIH in a group of individuals who had been previously infected by COVID-19, who did not present with reported symptoms of long-COVID. Individuals reporting both symptomatic and asymptomatic infections were included and compared to a control group who had not been infected. It was hypothesized that individuals who had been infected with COVID-19 would have an attenuated CPM and EIH response compared to controls.

Methods

Sample

A total of 64 individuals were recruited for this study; 4 were excluded as they had received the COVID-19 vaccine while enrolled in the study and had tested positive for antibodies, and 1 was excluded since they had tested positive for pregnancy. A total of 59 participants were included in the analysis; 26 in the symptomatic group (61% female), 13 in the asymptomatic group (66%

female), and 20 controls (44% female). A sample of 18 participants per group was determined to be sufficient to detect a moderate, but clinically relevant effect (Cohens d of 0.50 SD) using a three group x 2 assessment site mixed model (between-within) analysis of variance (ANOVA; for interaction) via an *a priori* power analysis assuming a correlation between repeated measures of .90. With 13 participants in the asymptomatic group we were powered ($\beta = .80$) to detect an effect of .59 SD at α level of .05. All participants self-reported that they were free of any musculoskeletal injuries, diagnosed chronic pain conditions, and other diseases known to affect sensory processing at the time of testing. Participants who currently had an active case of COVID-19 or those who had a COVID-19 vaccine were also excluded from the study. After providing informed consent, participants who had previously tested positive for COVID-19 (past 12 months) were assigned to 1 of 2 COVID-19 groups (see below). Those who had not tested positive had a polymerase chain reaction (PCR) and antibody test performed in order to confirm they were not currently infected and to determine if they might have been asymptomatic positive. Participants placed into the following 3 groups: 1) symptomatic COVID-19 infection, 2) asymptomatic COVID-19 infection, and 3) non-infection healthy control. The following criteria were used for group assignment: for the symptomatic COVID-19 group participants self-reported their symptoms that they had experienced when they had an active COVID-19 infection from a list modified from the Center of Disease Control (CDC) and provided evidence of a previous positive PCR or antibody ELISA test indicating infection. For the asymptomatic group, participants self-reported no symptoms and provided evidence of a previous positive PCR and/or positive antibody test or self-reported no symptoms, but had a positive antibody test. For the control group they self-reported no previous symptoms and were negative on the PCR and antibody test administered immediately prior to testing. The participants provided written informed consent prior to the experiment, and all testing procedures were approved by the University of Oklahoma institutional review board and complied with the Declaration of Helsinki.

Experimental Protocol

Participants were required to visit the Sensory and Muscle Function laboratory located within the department of Health and Exercise Science for 3 testing visits during the months of March through May 2021. All instruments and procedures were approved by the University of Oklahoma ethics committee and complied with the Declaration of Helsinki. Participants completed three testing visits that occurred within a 7-day period. On the first visit, written and verbal informed consent was completed. Following consent, a physical activity readiness questionnaire (PAR-Q), menstrual and drug history questionnaire and a COVID-19 symptom review survey were completed. Following consent and preliminary questionnaires, the participants completed a battery of psychological/pain questionnaires: 1) short form

of the profile of mood states (POMS),²⁷ 2) pain catastrophizing scale (PCS),⁷⁰ 3) and pain attitudes questionnaire-revised (PAQ-R).⁸⁸ POMS data was collected as the pandemic has been shown to influence mood⁵⁵ and PCS and PAQ were collected as pain catastrophizing and pain attitudes are both associated with CPM and EIH.^{43,75,81} The International Physical Activity Questionnaire (IPAQ) was also administered to assess self-reported physical activity.¹² Physical activity amount has been shown to influence pain modulatory function.^{50,51,65,76} Participants were familiarized with the procedure for pressure pain threshold (PPT) testing. PPT assessment involved determining the minimum amount of applied pressure (force) required to evoke "pain." Progressively increasing amounts of pressure (30kPa's per second) were applied to the muscle belly of the vastus lateralis of their dominant leg and ipsilateral muscle belly of the brachioradialis (arm) using an electronic AlgoMed pressure algometer (Medoc Ltd., Ramat Yishai, Israel) interfaced with Medoc Algomed software (Medoc Ltd., Ramat Yishai, Israel). Participants were instructed to press a handheld button when they deemed the applied pressure to first "hurt." Three assessments were performed on each muscle in an alternating fashion, with a 10-second interval between each PPT. The three measurements on each muscle were averaged. The arm and leg were chosen as local and distal sites for the two pain modulatory tests.

During visit two, participants had their height and weight measured which was used to calculate body mass index (BMI). CPM was assessed using a cold pressor test (CPT) where the participants first submerged their dominant foot into room temperature water for 1 minute. Following the room temperature water, ten minutes of quiet rest was completed, and PPT's using the same method from the familiarization were measured three times in the dominant leg, and three times in the ipsilateral arm in an alternating fashion. Averages were taken of the PPT's in each limb. Participants then submerged their dominant foot into ice water (2-3°C) for 1 minute. PPT's were then reassessed immediately after the cold water submersion. An additional 15 minutes of quiet rest was then provided and PPTs were reassessed a final time. The neutral cool water bath was used to control for potential distraction associated with water immersion⁴⁹ and this method has been used previously.^{13,42,44}

During visit 3, EIH was measured using a maximal isometric leg extension exercise that involved three maximal voluntary contractions (MVC), followed by a time-to-task failure (TTF) procedure of contacting the knee extensors at 25% of the highest MVC. PPT's were assessed in the leg and ipsilateral arm before the MVC's and PPT's were measured again immediately following the TTF bout, and 15minutes post. An isometric dynamometer (KinCom; Biopac, Goleta, CA) was used to measure MVC's. The participants were seated with their dominant knee at 110 degrees (full leg extension being 180 degrees) for both the TTF and MVC protocols. The ankle of each leg was secured against the end of an immobile lever arm. The force signal was digitalized

with Biopac MP-150 converter. The signals were instantly displayed to participants via acknowledge software and recorded for analysis. Initially, three MVC's were performed by kicking against the locked isokinetic dynamometer as forcefully as possible for a 3s bout. Three minutes of rest were provided between attempts. The highest value was taken as their MVC and used to calculate the target force for the time to task failure exercise bout. Participants were instructed to hold 25% of their MVC until volitional exhaustion or when the participant could no longer produce the force required. Verbal encouragement was provided to the participants. The pre PPT values during visit 2 and 3 served as baseline pain sensitivity measures for PPT's as the familiarization process during visit one.⁶

Statistical Analysis

The data collected during the experiment was analyzed using SPSS 26. Alpha was set at <.05 for statistical significance. Averages of 3 trials were taken for the PPT's in both the arm and the leg measures. Day-to-day-reliability was measured using single measures intraclass correlations coefficients (ICC's) to assess consistency. In order to account for differences in baseline PPT among participants, CPM and EIH are reported as the percent change from pre-values for the immediately post and 15-minute post assessments.

One-way ANOVAs were performed on normally distributed data to assess differences among the 3 groups on participant characteristics (height, weight, age), self-reported physical activity, mood pain attitudes, pain catastrophizing, and PPT's and were presented as means and standard deviations. Non-parametric tests, such as Kruskal-Wallis test, were performed on non-normally distributed questionnaire results (self-reported physical activity and mood) and were presented as medians. A 3 group (symptomatic, asymptomatic, and control) x 2 sex (males and females) ANOVA was performed to assess group and sex differences in PPT's. A 3 group (symptomatic, asymptomatic, and control) x 2 time points (immediately post and 15-minute post) mixed model ANOVA was performed to assess differences in the CPM and EIH responses. A Bonferroni adjustment was used to identify differences in the variables over the time points.

Effect sizes were calculated as partial eta squared (η_p^2) statistic for ANOVA analysis and Cohen's d statistic as the differences in means divided by the pooled standard deviation of the means. Effects of $\sim .01$ were judged to be small, $\sim .06$ were judged to be moderate, and $\geq .14$ were judged to be large when computing η_p^2 and effects of $\sim .20$ were judged to be small, $\sim .50$ were judged to be moderate, and $\geq .80$ were judged to be large when computing d.

Results

Participant Characteristics

Mean values and group differences for participant characteristics can be found in [Table 1](#). The control

Table 1. Group Descriptive for Symptomatic, Asymptomatic, and Control Groups (means \pm SD's)

	SYMPTOMATIC COVID-19 (N=26)	ASYMPTOMATIC COVID-19 (N=13)	CONTROL (N=20)	P-VALUE	PARTIAL ETA SQUARED
Age	21.6 \pm 2.5	23.2 \pm 3.2	24.3 \pm 4.8 ^{*,†}	.04	.11
Height (cm)	172.6 \pm 8.6	170.6 \pm 9.2	175.6 \pm 9.4	.31	.04
Weight (kg)	75.6 \pm 10.32	69.3 \pm 16.8	76.3 \pm 24.6	.33	.04
BMI	25.5 \pm 4.1	23.5 \pm 4.2	24.6 \pm 3.6	.34	.04
PPT leg (kPa)	588.9 \pm 205.6	438.5 \pm 166.6	629.2 \pm 334.1	.11	.07
PPT arm (kPa)	384.7 \pm 141.3	320.1 \pm 138.9	478.4 \pm 284.6	.08	.08
TTF (seconds)	190.9 \pm 103.2	177.4 \pm 49.0	181.4 \pm 81.8	.87	.00
CPM pain rating	5.1 \pm 1.7	4.5 \pm 1.5	5.7 \pm 1.7	.16	.07
IPAQ Walking (MET/mins)	3286.7 \pm 5711.6	2923.3 \pm 3171.4	2077.3 \pm 2089.6	.65	.02
IPAQ Moderate (MET/mins)	1769.6 \pm 1949.9	2432.3 \pm 3170.4	1285.8 \pm 1375.6	.33	.04
IPAQ Vigorous (MET/mins)	3639.7 \pm 5001.8	2450.0 \pm 2742.2	2280.8 \pm 2452.6	.45	.03
IPAQ Time Sitting (hours)	44.0 \pm 17.4	46.3 \pm 19.6	50.4 \pm 18.7	.51	.02
IPAQ Total (MET/mins)	8696.0 \pm 11013.0	7805.3 \pm 8595.5	5643.8 \pm 8832.2	.52	.02
PCS	10.9 \pm 5.7	10.6 \pm 6.5	15.6 \pm 10.3	.09	.08
PAQ SF	16.6 \pm 4.6	15.7 \pm 3.2	15.8 \pm 3.4	.70	.01
PAQ SC	12.0 \pm 3.5	12.4 \pm 3.2	13.4 \pm 2.7	.34	.04
PAQ SS	13.7 \pm 4.4	14.1 \pm 2.4	14.1 \pm 3.7	.93	.00
PAQ CSD	16.4 \pm 5.2	13.2 \pm 4.0	17.8 \pm 4.3 ^{*,^}	.03	.12
PAQ CR	13.6 \pm 3.7	13.1 \pm 3.2	14.0 \pm 3.0	.80	.01
POMS Tension	6.1 \pm 4.0	5.2 \pm 3.4	4.7 \pm 4.7	.48	.03
POMS Anger	3.2 \pm 2.9	3.6 \pm 3.0	2.8 \pm 4.4	.78	.01
POMS Fatigue	7.8 \pm 4.9	7.4 \pm 5.5	6.5 \pm 3.9	.65	.02
POMS Vigor	8.1 \pm 4.1	9.4 \pm 2.4	9.4 \pm 4.4	.45	.03
POMS Confusion	4.3 \pm 3.4	3.8 \pm 3.4	3.6 \pm 2.4	.68	.01
POMS Depression	4.4 \pm 4.2	4.2 \pm 3.8	3.3 \pm 3.6	.61	.02
POMS TMD	17.8 \pm 15.7	14.9 \pm 15.9	11.2 \pm 16.3	.40	.03

*difference between symptomatic vs asymptomatic,

†difference between symptomatic and control,

^difference between asymptomatic and control. BMI, Body mass index, BF%, body fat percentage, PPT, pressure pain threshold, CPM, Conditioned Pain Modulation, TTF, Time to task failure, IPAQ, international physical activity questionnaire, PCS, pain catastrophizing scale, PAQ, Pain attitudes questionnaire, SF, stoic fortitude, SC, stoic concealment, SS, stoic superiority, CSD, cautious self-doubt, CR, cautious reluctance, TMD, total mood disturbance

group was older than the asymptomatic COVID-19 group ($P = .042$ for ANOVA; $\eta_p^2 = .107$). No differences were found among the groups in all dimensions of the IPAQ, PCS, and POMS. Furthermore, no differences were found in the PAQ with the exception of 1 dimension; cautious self-doubt ($P = .034$; $\eta_p^2 = .122$), which demonstrated differences between asymptomatic and control groups ($P = .027$).

COVID-19 Symptoms

Fever was reported in 60% of participants ($N = 15$) who had previously experienced symptoms of COVID-19. For respiratory symptoms, cough was experienced in 60% ($N = 15$), hemoptysis in 4% ($N = 1$), congested nose in 80% ($N = 20$), phlegm in 68% ($N = 17$), and 72% experienced some degree of shortness of breath ($N = 18$). Every symptomatic individual reported experiencing at least 1 neurological symptom. Fatigue and headaches were seen in 84% of symptomatic COVID-19 participants ($N = 21$), myalgia in 72% ($N = 18$), anosmia in 60% ($N = 15$), ageusia in 56% ($N = 14$), sore throat in 48% ($N = 12$), 44% reported difficulties in concentration ($N = 11$) and jaw/facial pain was reported in 12% ($N = 3$). Gastrointestinal manifestations of COVID-19 were less common ($N = 14$; 56%), with 44% experiencing diarrhea ($N = 11$) and 28% experiencing nausea ($N = 7$).

Pressure Pain Sensitivity

No differences were found in baseline PPT's among groups in the leg ($P = .111$; $\eta_p^2 = .07$), or the arm ($P = .082$ for ANOVA; $\eta_p^2 = .08$) (Table 1). PPT's across all three visits were consistent and demonstrated high reliability in both leg (ICC = .965) and arm (ICC = .940).

Pain Modulation

A significant group \times time interaction was observed for CPM in both the leg ($P = .008$) and the arm ($P = .001$). There were no group differences observed at 15-minute post ice bath in the leg ($P = .15$) or arm ($P = .70$). Bonferroni adjustment indicated that the magnitude of CPM differed between the symptomatic and asymptomatic groups in the leg (12.8% \pm 22.0 vs 33.8% \pm 28.2; $P = .014$), but not between the symptomatic and control (12.8% \pm 22.0 vs 18.1% \pm 14.1; $P = .678$), or the asymptomatic and control groups (33.8% \pm 28.2 vs 18.1% \pm 14.1; $P = .107$) (Fig 1). In the arm significant differences were observed between symptomatic and asymptomatic groups (-1.0% \pm 20.3 vs 33.3% \pm 26.2; $P < .001$), between the symptomatic and control group (-1.0% \pm 26.2 vs 23.4% \pm 28.2; $P = 0.004$), but not between the asymptomatic and control groups (33.3% \pm 26.2 vs 23.4% \pm 28.2; $P = .504$) (Fig 2).

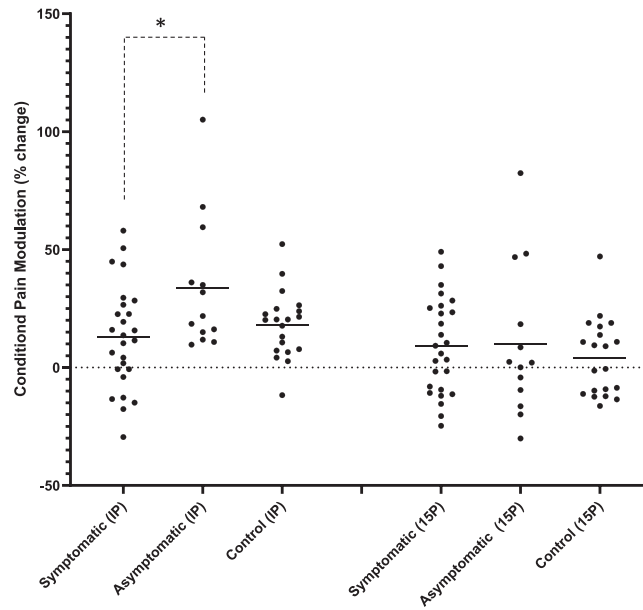


Figure 1. Group differences in Condition Pain Modulation between symptomatic COVID-19, asymptomatic COVID-19, and control groups in the leg. *denotes significant difference between symptomatic and asymptomatic. IP = immediately post, 15P = 15minutes post.

The group*time interaction for EIH in the leg was not significant ($P = .94$), but there was a main effect for time (36.1% vs 20.8% for immediately post and 15-minute post; $P < .001$) (Fig 3). No differences were observed in the arm ($P = .82$ for the interaction, and $P = .39$ for time main effect) (Fig 4).

Discussion

This study compared pressure pain sensitivity and pain modulatory function in individuals who have been previously infected by COVID-19, both symptomatically and

asymptomatically, to a control group that did not have COVID-19. The primary findings of this study were 1) pressure pain sensitivity did not differ among groups, 2) the magnitude of EIH was not different between groups, and 3) the CPM response in symptomatic COVID-19 was significantly lower than the asymptomatic and control groups in the arm. The magnitude of the CPM response in the arm was significantly lower in the symptomatic group than asymptomatic group and control group whereby the symptomatic COVID-19 group showed a lack of CPM response.

Pain sensitivity in both the upper and lower extremity in previously symptomatic COVID-19, asymptomatic

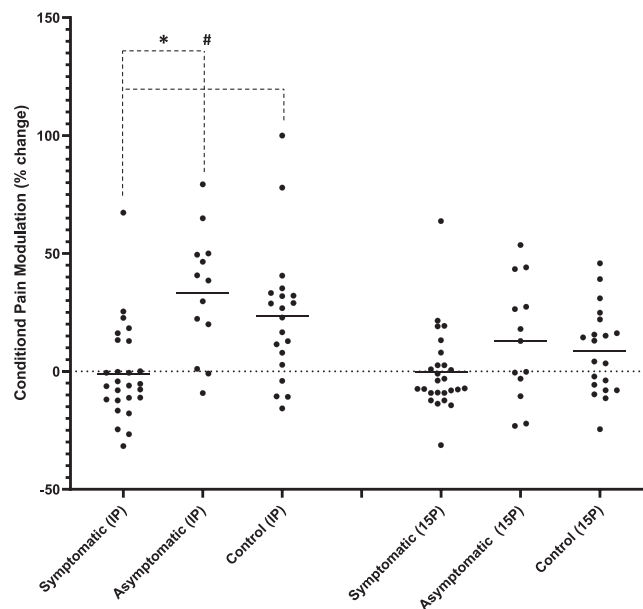


Figure 2. Group differences in Condition Pain Modulation between symptomatic COVID-19, asymptomatic COVID-19, and control groups in the arm. *denotes significant difference between symptomatic and asymptomatic. # denotes significant difference between symptomatic and control group. IP = immediately post, 15P = 15minutes post.

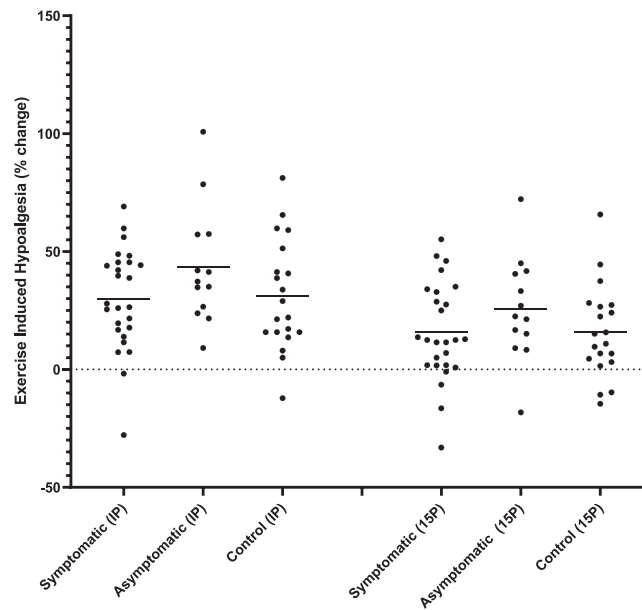


Figure 3. Group differences in Exercise Induced Hypoalgesia between symptomatic COVID-19, asymptomatic COVID-19, and control groups in the leg. IP = immediately post, 15P = 15minutes post.

COVID-19 and control groups showed no statistical difference. While we did expect to find a difference between groups, previous exposure to SARS-CoV-2 appeared to not have an effect on pressure pain sensitivity. Our group was homogeneous and did not differ in mood,⁷ catastrophizing,¹⁶ and physical activity¹⁷ which have all been reported as determinants that may affect pain sensitivity. Participants that were tested did not have an active COVID-19 infection and it would be interesting to investigate whether an active COVID-19 case attenuates pressure pain responses.

Individuals who had symptomatic COVID-19 seemed to have a dysfunctional CPM response in a distal location;

the forearm compared to the other 2 groups. The mean arm CPM response was -1% in the symptomatic group compared to the control group and the asymptomatic group whose pain thresholds increased following the cold pressor test by 33% and 26%, respectively. This magnitude is similar to the mean magnitude of CPM evoked (29%) in studies using healthy controls in a recent meta-analysis review⁶¹. The included studies in the analysis used the CPT,^{15,25,37,60,64,68,74,80} heat,^{25,39,57,68} ischemia,^{9,20,21,62} and chemical stimuli^{2,23,26,71} as the conditioning stimulus, with the most common being the CPT⁶¹ similar to that used in the present study. Our finding of an attenuated response in the symptomatic group is similar to those who

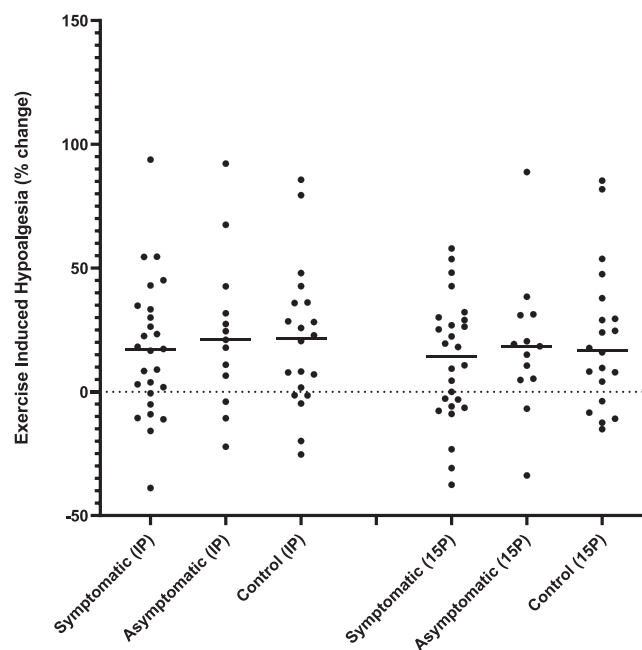


Figure 4. Group differences in Exercise Induced Hypoalgesia between symptomatic COVID-19, asymptomatic COVID-19, and control groups in the arm. IP = immediately post, 15P = 15minutes post.

have been diagnosed with chronic pain conditions such as fibromyalgia,^{34,53,58,59} headache/migraine,^{10,14,54,63,73} arthritis,^{1,35,41} irritable bowel syndrome^{83,84} and myalgia⁴⁰ whereby the CPM response magnitude was significantly lower compared to the control groups. Impaired CPM predicts chronic post-operative pain when assessed in pain-free individuals prior to surgery⁸⁷ and has potential utility as a biomarker for chronic pain risk. Pro-inflammatory cytokines can sensitize or activate nociceptors and transmit painful stimuli to the brain.⁸⁹ The surface expressed angiotensin converting enzyme 2 (ACE2) has been found to be main receptor for uptake of SARS-CoV-2 spike protein.²⁹ Evidence indicates that ACE2 expressing sensory neurons synapse with spinal and brainstem central nervous system (CNS) neurons leading to neurological effects, including headache and nerve pain^{46,48}(17, 18). Additionally activation of ACE2 receptors lead to an inflammatory response and a viral infection may cause localized and systematic inflammation. Inflammation is a key proponent of pain and impaired pain modulation, as such, could be a reason as to why pain modulation is impaired in those who have had symptomatic COVID-19 and may be experiencing lingering inflammation.

Despite our findings of differences in CPM among groups, we found no differences in the EIH response in individuals who have had symptomatic COVID-19 and those who did not. CPM and EIH have been shown to correlate,^{42,77} however in younger adults, this relationship has not been consistently observed.⁵⁶ Our sample, other than COVID-19 infection status, was relatively homogenous, especially in regards to age and PA, which are thought to play a role in EIH. Endogenous opioids,³² hemodynamics,²² pro-inflammatory cytokines,³¹ and distraction¹⁹ have also been proposed as possible mechanisms underlying EIH. Our findings seem to indicate that COVID-19 infection does not seem to alter this pathway (s). This further highlights existing evidence¹⁸ that CPM and EIH do not necessarily modulate pain sensitivity using the same shared pathways. Furthermore, regular physical activity across all three groups may attribute to the groups not having an impaired EIH response following COVID-19 infection as exercise elicits an anti-inflammatory mechanism²⁴ that may sub serve as protection from the pro-inflammatory effects of COVID-19. Similar to other pain conditions, there is broad inter-individual variability in both EIH and CPM but it is generally accepted that multiple factors affect pain modulation (see^{52,66} for review) and could be contributing to the high levels of variability of CPM and EIH response within our sample.

This study had several limitations to note. Firstly, individuals who previously had asymptomatic COVID-19 may not have had antibodies present. SARS-CoV-2 antibodies remain stable for at least 5 months after an infection with the virus,⁸ so participants who were in the control group may have had asymptomatic COVID-19 prior to the 5 months. Second, we used recall for symptoms that the participants had when they had an active case of COVID-19. We did not assess whether or not they had long COVID-19, nor did we collect detailed information on current health status or current symptoms. At the time of testing (spring 2021), data on long-

COVID was emerging and was not fully established, therefore additional studies assessing long COVID-19 symptoms and their impact on modulatory function is warranted. While we collected data on the painfulness of the CPM protocol, we did not ask the participants to rate the pain of the EIH protocol. This would have been helpful to evaluate the endogenous pain-inhibitory effects of the exercise, as EIH magnitude can be influenced by the painfulness of the exercise. Time of day was not controlled for during PPT testing. It has been suggested that time of day is a contributing factor to pain sensitivity; however, inconsistent findings have been found at which time participants were most sensitive to pain at its peak.^{4,5} It is possible that this study was underpowered, however, since we did find a result with CPM and not the other two tested variables (EIH and pain sensitivity); it is possible that what we found regarding CPM is significant and concerning.

Despite these limitations, the current study adds COVID-19 infection to a growing list of conditions characterized by impaired pain modulation. The results from the study indicate CPM function was impaired in individuals who had symptomatic COVID-19. No differences between groups were found in pressure pain sensitivity, and no differences between symptomatic group and the other two groups were found in EIH. CPM seemed to be effected by symptomatic COVID-19 with the EIH response remaining unaffected; the exact mechanisms of CPM and EIH need further exploration especially regarding conditions that involve acute systemic inflammation. Impaired CPM response could be an indication on whether or not COVID-19 exposure may have long-term implications on pain modulation and increased risk of chronic pain development. This study used younger adults who were not admitted into the ICU; additional research in older adults and those who had more severe symptoms of COVID-19 is warranted. Those who were admitted to the ICU with more severe symptoms may have different or more profound implications to pain modulatory function and this is very important to address due to the association between chronic pain and impaired pain modulation.

This study and its findings add to the growing body of literature that there are residual consequences of COVID-19. It is unknown whether CPM remains dysregulated long term and this should be addressed in future research studies, both cross sectional and longitudinal. CPM was impaired in individuals who had symptomatic COVID-19, which may have long-term implications on pain modulation. These findings are concerning in that normally healthy, young adults who have demonstrated only mild to moderate symptoms of COVID-19 have an impaired CPM response, as such, pain modulatory function should be examined in symptomatic COVID-19 older adults and in those vulnerable populations who are at risk for chronic pain development.

Acknowledgments

The authors would like to thank the participants for their time.

References

- Arendt-Nielsen L, Nie H, Laursen MB, Laursen BS, Madeleine P, Simonsen OH, Graven-Nielsen T: Sensitization in patients with painful knee osteoarthritis. *Pain* 149:573-581, 2010
- Arendt-Nielsen L, Sluka KA, Nie HL: Experimental muscle pain impairs descending inhibition. *Pain* 140:465-471, 2008
- Arendt-Nielsen L, Morlion B, Perrot S, Dahan A, Dickenson A, Kress H, Wells C, Bouhassira D, Mohr Drewes A: Assessment and manifestation of central sensitisation across different chronic pain conditions. *Eur J Pain* 22:216-241, 2018
- Aviram J, Pud D, Shochat T: Pain sensitivity in healthy young men is modified by time-of-day. *Sleep Med* 14:e265, 2013
- Aviram J, Shochat T, Pud D: Pain perception in healthy young men is modified by time-of-day and is modality dependent. *Pain Med* 16:1137-1144, 2015
- Black CD, Pickowitz KE: Day-to-day reliability of pressure pain threshold and pain ratings in college-aged men. *Int J Rehabil Res* 38:213-218, 2015
- Boettger MK, Schwier C, Bär K-J: Sad mood increases pain sensitivity upon thermal grill illusion stimulation: Implications for central pain processing. *PAIN®*. 152:123-130, 2011
- Buss LF, Prete CA, Abraham CM, Mendrone A, Salomon T, de Almeida-Neto C, França RF, Belotti MC, Carvalho MP, Costa AG: Three-quarters attack rate of SARS-CoV-2 in the Brazilian Amazon during a largely unmitigated epidemic. *Science* 371:288-292, 2021
- Campbell CM, France CR, Robinson ME, Logan HL, Geffken GR, Fillingim RB: Ethnic differences in diffuse noxious inhibitory controls. *J Pain* 9:759-766, 2008
- Cathcart S, Winefield AH, Lushington K, Rolan P: Noxious inhibition of temporal summation is impaired in chronic tension-type headache. *Headache: J Head and Face Pain* 50:403-412, 2010
- Chalaye P, Goffaux P, Bourgault P, Lafrenaye S, Devroede G, Watier A, Marchand S: Comparing pain modulation and autonomic responses in fibromyalgia and irritable bowel syndrome patients. *Clin J Pain* 28:519-526, 2012
- Craig CL, Marshall AL, Sjostrom M, Bauman AE, Booth ML, Ainsworth BE, Pratt M, Ekelund U, Yngve A, Sallis JF, Oja P: International physical activity questionnaire: 12-country reliability and validity. *Med Sci Sport Exer* 35:1381-1395, 2003
- Dailey DL, Rakel BA, Vance CG, Liebano RE, Amrit AS, Bush HM, Lee KS, Lee JE, Sluka KA: Transcutaneous electrical nerve stimulation reduces pain, fatigue and hyperalgesia while restoring central inhibition in primary fibromyalgia. *PAIN®* 154:2554-2562, 2013
- Drummond PD, Knudsen L: Central pain modulation and scalp tenderness in frequent episodic tension-type headache. *Headache: J Head Face Pain* 51:375-383, 2011
- Edwards RR, Fillingim RB, Ness TJ: Age-related differences in endogenous pain modulation: A comparison of diffuse noxious inhibitory controls in healthy older and younger adults. *Pain* 101:155-165, 2003
- Edwards RR, Haythornthwaite JA, Sullivan MJ, Fillingim RB: Catastrophizing as a mediator of sex differences in pain: Differential effects for daily pain versus laboratory-induced pain. *Pain* 111:335-341, 2004
- Ellingson LD, Colbert LH, Cook DB: Physical activity is related to pain sensitivity in healthy women. *Med Sci Sports Exercise* 44:1401-1406, 2012
- Ellingson LD, Koltyn KF, Kim JS, Cook DB: Does exercise induce hypoalgesia through conditioned pain modulation? *Psychophysiology* 51:267-276, 2014
- Fillingim RB, Roth DL, Haley WE: The effects of distraction on the perception of exercise-induced symptoms. *J Psychosom Res* 33:241-248, 1989
- France CR, Suchowiecki S: A comparison of diffuse noxious inhibitory controls in men and women. *Pain* 81:77-84, 1999
- Fujii K, Motohashi K, Umino M: Heterotopic ischemic pain attenuates somatosensory evoked potentials induced by electrical tooth stimulation: Diffuse noxious inhibitory controls in the trigeminal nerve territory. *Eur J Pain* 10:495-504, 2006
- Fuller A, Robinson M: A test of exercise analgesia using signal detection theory and a within-subjects design. *Percept Mot Skills* 76:1299-1310, 1993
- Ge H-Y, Madeleine P, Arendt-Nielsen L: Sex differences in temporal characteristics of descending inhibitory control: An evaluation using repeated bilateral experimental induction of muscle pain. *Pain* 110:72-78, 2004
- Gleeson M, Bishop NC, Stensel DJ, Lindley MR, Mastana SS, Nimmo MA: The anti-inflammatory effects of exercise: Mechanisms and implications for the prevention and treatment of disease. *Nat Rev Immunol* 11:607-615, 2011
- Granot M, Weissman-Fogel I, Crispel Y, Pud D, Granovsky Y, Sprecher E, Yarnitsky D: Determinants of endogenous analgesia magnitude in a diffuse noxious inhibitory control (DNIC) paradigm: Do conditioning stimulus painfulness, gender and personality variables matter? *Pain* 136:142-149, 2008
- Graven-Nielsen T, Babenko V, Svensson P, Arendt-Nielsen L: Experimentally induced muscle pain induces hypoalgesia in heterotopic deep tissues, but not in homotopic deep tissues. *Brain Res* 787:203-210, 1998
- Grove JR, Prapavessis H: Preliminary evidence for the reliability and validity of an abbreviated profile of mood states. *Int J Sport Psychol* 23:93-109, 1992
- Guan W-j, Ni Z-y, Hu Y, Liang W-h, Ou C-q, He J-x, Liu L, Shan H, Lei C-l, Hui DS: Clinical characteristics of coronavirus disease 2019 in China. *N Engl J Med* 382:1708-1720, 2020
- Hoffmann M, Kleine-Weber H, Schroeder S, Krüger N, Herrler T, Erichsen S, Schiergens TS, Herrler G, Wu N-H, Nitsche A: SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor. *Cell*, 2020
- Ji R-R, Nackley A, Huh Y, Terrando N, Maixner W: Neuroinflammation and central sensitization in chronic and widespread pain. *Anesthesiology* 129:343-366, 2018
- Kami K, Tajima F, Senba E: Exercise-induced hypoalgesia: Potential mechanisms in animal models of neuropathic pain. *Anat Sci Int* 92:79-90, 2017

32. Koltyn KF, Brellenthin AG, Cook DB, Sehgal N, Hillard C: Mechanisms of exercise-induced hypoalgesia. *J Pain* 15:1294-1304, 2014
33. Kosek E, Ekholm J, Hansson P: Modulation of pressure pain thresholds during and following isometric contraction in patients with fibromyalgia and in healthy controls. *Pain* 64:415-423, 1996
34. Kosek E, Hansson P: Modulatory influence on somatosensory perception from vibration and heterotopic noxious conditioning stimulation (HNCS) in fibromyalgia patients and healthy subjects. *Pain* 70:41-51, 1997
35. Kosek E, Ordeberg G: Lack of pressure pain modulation by heterotopic noxious conditioning stimulation in patients with painful osteoarthritis before, but not following, surgical pain relief. *Pain* 88:69-78, 2000
36. Lannersten L, Kosek E: Dysfunction of endogenous pain inhibition during exercise with painful muscles in patients with shoulder myalgia and fibromyalgia. *Pain* 151:77-86, 2010
37. Larivière M, Goffaux P, Marchand S, Julien N: Changes in pain perception and descending inhibitory controls start at middle age in healthy adults. *Clin J Pain* 23:506-510, 2007
38. Larsen DB, Laursen M, Edwards RR, Simonsen O, Arendt-Nielsen L, Petersen KK: The combination of preoperative pain, conditioned pain modulation, and pain catastrophizing predicts postoperative pain 12 Months after total knee arthroplasty. *Pain Med* 22:1583-1590, 2021
39. Lautenbacher S, Rollman G: Possible deficiencies of pain modulation in fibromyalgia. *Clin J Pain* 13:189-196, 1997
40. Leffler A-S, Hansson P, Kosek E: Somatosensory perception in a remote pain-free area and function of diffuse noxious inhibitory controls (DNIC) in patients suffering from long-term trapezius myalgia. *Eur J Pain* 6:149-159, 2002
41. Leffler A-S, Kosek E, Lerndal T, Nordmark B, Hansson P: Somatosensory perception and function of diffuse noxious inhibitory controls (DNIC) in patients suffering from rheumatoid arthritis. *Eur J Pain* 6:161-176, 2002
42. Lemley KJ, Hunter SK, Bement MK: Conditioned pain modulation predicts exercise-induced hypoalgesia in healthy adults. *Med Sci Sports Exerc* 47:176-184, 2015
43. Lemley KJ, Hunter SK, Bement MKH: Conditioned pain modulation predicts exercise-induced hypoalgesia in healthy adults. *Med Sci Sports Exercise* 47:176-184, 2015
44. Liebano RE, Vance C, Rakel BA, Lee JE, Cooper NA, Marchand S, Walsh DM, Sluka KA: Transcutaneous electrical nerve stimulation and conditioned pain modulation influence the perception of pain in humans. *Eur J Pain* 17:1539-1546, 2013
45. Ma Q, Liu J, Liu Q, Kang L, Liu R, Jing W, Wu Y, Liu M: Global Percentage of Asymptomatic SARS-CoV-2 Infections Among the Tested Population and Individuals With Confirmed COVID-19 Diagnosis: A Systematic Review and Meta-analysis. *JAMA Netw Open* 4:e2137257, 2021
46. Mao L, Jin H, Wang M, Hu Y, Chen S, He Q, Chang J, Hong C, Zhou Y, Wang D: Neurologic manifestations of hospitalized patients with coronavirus disease 2019 in Wuhan, China. *JAMA Neurol* 77:683-690, 2020
47. Meeus M, Nijs J, Van de Wauwer N, Toeback L, Truijten S: Diffuse noxious inhibitory control is delayed in chronic fatigue syndrome: An experimental study. *Pain* 139:439-448, 2008
48. Montalvan V, Lee J, Bueso T, De Toledo J, Rivas K: Neurological manifestations of COVID-19 and other coronavirus infections: A systematic review. *Clin Neurol Neurosurg* 194:105921, 2020
49. Moont R, Pud D, Sprecher E, Sharvit G, Yarnitsky D: 'Pain inhibits pain' mechanisms: Is pain modulation simply due to distraction? *Pain* 150:113-120, 2010
50. Naugle KM, Ohlman T, Naugle KE, Riley ZA, Keith NR: Physical activity behavior predicts endogenous pain modulation in older adults. *Pain* 158:383-390, 2017
51. Naugle KM, Riley JL 3rd: Self-reported physical activity predicts pain inhibitory and facilitatory function. *Med Sci Sports Exerc* 46:622-629, 2014
52. Nir R-R, Yarnitsky D: Conditioned pain modulation. *Curr Opin Support Palliat Care* 9:131-137, 2015
53. Normand E, Potvin S, Gaumond I, Cloutier G, Corbin J-F, Marchand S: Pain inhibition is deficient in chronic widespread pain but normal in major depressive disorder. *J Clin Psychiatry* 72:219-224, 2010
54. Perrotta A, Serrao M, Sandrini G, Burstein R, Sances G, Rossi P, Bartolo M, Pierelli F, Nappi G: Sensitisation of spinal cord pain processing in medication overuse headache involves supraspinal pain control. *Cephalalgia* 30:272-284, 2010
55. Peterson JA, Chesbro G, Larson R, Larson D, Black CD: Short-term analysis (8 weeks) of social distancing and isolation on mental health and physical activity behavior during COVID-19. *Front Psychol* 12:738, 2021
56. Peterson JA, Schubert DJ, Campbell J, Bembem MG, Black CD: Endogenous pain inhibitory function: endurance-trained athletes vs active controls. *Pain Med* 20:1822-1830, 2019
57. Pielsticker A, Haag G, Zaudig M, Lautenbacher S: Impairment of pain inhibition in chronic tension-type headache. *Pain* 118:215-223, 2005
58. Potvin S, Larouche A, Normand E, de Souza JB, Gaumond I, Grignon S, Marchand S: DRD3 Ser9Gly polymorphism is related to thermal pain perception and modulation in chronic widespread pain patients and healthy controls. *J Pain* 10:969-975, 2009
59. Potvin S, Larouche A, Normand E, de Souza JB, Gaumond I, Marchand S, Grignon S: No relationship between the ins del polymorphism of the serotonin transporter promoter and pain perception in fibromyalgia patients and healthy controls. *Eur J Pain* 14:742-746, 2010
60. Potvin S, Stip E, Tempier A, Pampoulova T, Bentaleb LA, Lalonde P, Lipp O, Goffaux P, Marchand S: Pain perception in schizophrenia: No changes in diffuse noxious inhibitory controls (DNIC) but a lack of pain sensitization. *J Psychiatr Res* 42:1010-1016, 2008
61. Pud D, Granovsky Y, Yarnitsky D: The methodology of experimentally induced diffuse noxious inhibitory control (DNIC)-like effect in humans. *Pain* 144:16-19, 2009

62. Reinert A, Treede R-D, Bromm B: The pain inhibiting pain effect: An electrophysiological study in humans. *Brain Res* 862:103-110, 2000
63. Sandrini G, Rossi P, Milanov I, Serrao M, Cecchini A, Nappi G: Abnormal modulatory influence of diffuse noxious inhibitory controls in migraine and chronic tension-type headache patients. *Cephalalgia* 26:782-789, 2006
64. Serrao M, Rossi P, Sandrini G, Parisi L, Amabile GA, Nappi G, Pierelli F: Effects of diffuse noxious inhibitory controls on temporal summation of the RIII reflex in humans. *Pain* 112:353-360, 2004
65. Shiro Y, Ikemoto T, Terasawa Y, Arai Y-CP, Hayashi K, Ushida T, Matsubara T: Physical activity may be associated with conditioned pain modulation in women but not men among healthy individuals. *Pain Res Manag* 2017, 2017
66. Sluka KA, Law LF, Bement MH: Exercise-induced pain and analgesia? Underlying mechanisms and clinical translation. *Pain* 159:S91, 2018
67. Staud R, Robinson ME, Price DD: Isometric exercise has opposite effects on central pain mechanisms in fibromyalgia patients compared to normal controls. *Pain* 118:176-184, 2005
68. Staud R, Robinson ME, Vierck CJ Jr, Price DD: Diffuse noxious inhibitory controls (DNIC) attenuate temporal summation of second pain in normal males but not in normal females or fibromyalgia patients. *Pain* 101:167-174, 2003
69. Su S, Cui H, Wang T, Shen X, Ma C: Pain: A potential new label of COVID-19. *Brain Behav Immun* 87:159, 2020
70. Sullivan M, Bishop S, Pivik J: The pain catastrophizing scale: Development and validation. *Psych Assess* 7:524-532, 1995
71. Svensson P, Hashikawa CH, Casey KL: Site-and modality-specific modulation of experimental muscle pain in humans. *Brain Res* 851:32-38, 1999
72. Taquet M, Dercon Q, Luciano S, Geddes JR, Husain M, Harrison PJ: Incidence, co-occurrence, and evolution of long-COVID features: A 6-month retrospective cohort study of 273,618 survivors of COVID-19. *PLoS Med* 18:e1003773, 2021
73. Tommaso Md, Sardaro M, Pecoraro C, fruscolo OD, Serpino C, Lamberti P, Livrea P: Effects of the remote C fibres stimulation induced by capsaicin on the blink reflex in chronic migraine. *Cephalalgia* 27:881-890, 2007
74. Tousignant-Laflamme Y, Pagé S, Goffaux P, Marchand S: An experimental model to measure excitatory and inhibitory pain mechanisms in humans. *Brain Res* 1230:73-79, 2008
75. Traxler J, Hanssen MM, Lautenbacher S, Ottawa F, Peters ML: General versus pain-specific cognitions: Pain catastrophizing but not optimism influences conditioned pain modulation. *Eur J Pain* 23:150-159, 2019
76. Umeda M, Newcomb LW, Ellingson LD, Koltyn KF: Examination of the dose-response relationship between pain perception and blood pressure elevations induced by isometric exercise in men and women. *Biol Psychol* 85:90-96, 2010
77. Vaegter HB, Handberg G, Graven-Nielsen T: Similarities between exercise-induced hypoalgesia and conditioned pain modulation in humans. *Pain* 155:158-167, 2014
78. Van Oosterwijck J, Nijs J, Meeus M, Lefever I, Huybrechts L, Lambrecht L, Paul L: Pain inhibition and postexertional malaise in myalgic encephalomyelitis/chronic fatigue syndrome: An experimental study. *J Intern Med* 268:265-278, 2010
79. Wang H, Paulson KR, Pease SA, Watson S, Comfort H, Zheng P, Aravkin AY, Bisignano C, Barber RM, Alam T: Estimating excess mortality due to the COVID-19 pandemic: A systematic analysis of COVID-19-related mortality, 2020 –21. *Lancet North Am Ed* 399:1513-1536, 2022
80. Washington LL, Gibson SJ, Helme RD: Age-related differences in the endogenous analgesic response to repeated cold water immersion in human volunteers. *Pain* 89:89-96, 2000
81. Weissman-Fogel I, Sprecher E, Pud D: Effects of catastrophizing on pain perception and pain modulation. *Exp Brain Res* 186:79-85, 2008
82. Widyadharma IPE, Dewi PR, Wijayanti IAS, Utami DKI: Pain related viral infections: A literature review. *Egypt J Neurol Psychiatr Neurosurg* 56:105, 2020
83. Wilder-Smith C, Schindler D, Lovblad K, Redmond S, Nirikko A: Brain functional magnetic resonance imaging of rectal pain and activation of endogenous inhibitory mechanisms in irritable bowel syndrome patient subgroups and healthy controls. *Gut* 53:1595-1601, 2004
84. Wilder-Smith CH, Robert-Yap J: Abnormal endogenous pain modulation and somatic and visceral hypersensitivity in female patients with irritable bowel syndrome. *World J Gastroenterol* 13:3699, 2007
85. World-Health-Organization: WHO Coronavirus (COVID-19) Dashboard. 2022 Available at: <https://covid19.who.int/> 2022
86. Yarnitsky D: Conditioned pain modulation (the diffuse noxious inhibitory control-like effect): Its relevance for acute and chronic pain states. *Curr Opin Anaesthesiol* 23:611-615, 2010
87. Yarnitsky D, Crispel Y, Eisenberg E, Granovsky Y, Ben-Nun A, Sprecher E, Best L-A, Granot M: Prediction of chronic post-operative pain: Pre-operative DNIC testing identifies patients at risk. *Pain* 138:22-28, 2008
88. Yong HH, Gibson SJ, Horne DJ, Helme RD: Development of a pain attitudes questionnaire to assess stoicism and cautiousness for possible age differences. *J Gerontol B Psychol Sci Soc Sci* 56:P279-P284, 2001
89. Zhang X, Tan Y, Ling Y, Lu G, Liu F, Yi Z, Jia X, Wu M, Shi B, Xu S: Viral and host factors related to the clinical outcome of COVID-19. *Nature* 583:437-440, 2020