

Cortical differences in diverticular disease and correlation with symptom reports

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KEY MESSAGES:

- There is an increasing body of evidence that patients with chronic gastrointestinal disease have brain structural abnormalities in areas linked to the pain network.
- Magnetic resonance imaging, whole brain volumetry, cortical thickness analysis and voxel based morphometry were used to investigate structural brain differences in Diverticular Disease (DD) patients for the first time. In particular differences between High Somatization DD (HSDD) and Low Somatization DD (LSDD) patients (as characterized using the Patient Health Questionnaire 12, PHQ12-SS) were investigated.
- The structural brain differences highlighted in this work suggest that these patient groups differ in terms of pathophysiology. Increased understanding will help direct pharmacological and psychological interventions in this widespread disease.

Abstract

Background

Recent studies have shown that the brain of patients with gastrointestinal disease differ both structurally and functionally from that of controls. Highly somatizing diverticular disease (HSDD) patients were also shown to differ from low somatizing (LSDD) patients functionally. This study aimed to investigate how they differed structurally.

Methods

Four diseases subgroups were studied in a cross-sectional design: 20 patients with asymptomatic diverticular disease (ADD), 18 LSDD, 16 HSDD, and 18 with irritable bowel syndrome. We divided DD patients into LSDD and HSDD using a cutoff of 6 on the Patient Health Questionnaire 12 Somatic Symptom (PHQ12-SS) scale. All patients underwent a 1-mm isotropic structural brain MRI scan and were assessed for somatization, hospital anxiety, depression, and pain catastrophizing. Whole brain volumetry, cortical thickness analysis and voxel-based morphometry were carried out using Freesurfer and SPM.

Key Results

We observed decreases in grey matter density in the left and right dorso-lateral prefrontal cortex (dlPFC), and in the mid-cingulate and motor cortex, and increases in the left (19, 20) and right (19, 38) Brodmann Areas. The average cortical thickness differed overall across groups ($P=0.002$) and regionally: HSDD>ADD in the posterior cingulate cortex ($P=0.03$), HSDD>LSDD in the dlPFC ($P=0.03$) and in the ventro-lateral PFC ($P<0.001$). The thickness of the anterior cingulate cortex and of the mid-prefrontal cortex were also found to correlate with Pain Catastrophizing (Spearman's $\rho=0.24$, $P=0.043$ uncorrected and Spearman's $\rho=0.25$, $P=0.03$ uncorrected).

Conclusion & Inferences

This is the first study of structural grey matter abnormalities in diverticular disease patients. The data shows brain differences in the pain network.

Keywords

Magnetic resonance imaging; brain; cortex; diverticular disease; pain; cortical thickness analysis; MRI; voxel-based morphometry; grey matter; catastrophizing

Abbreviations:

anterior cingulate cortex (ACC), asymptomatic diverticular disease (ADD), anterior insula (aINS), amygdala (AMYG), Contact Heat- Evoked Potential Stimulator (CHEPS), descending noxious inhibitory control (DNIC), echo-planar imaging (EPI), functional magnetic resonance imaging (fMRI), general linear model (GLM), Hospital Anxiety and Depression Score (HAD), haemodynamic response function (HRF), high somatization score symptomatic diverticular disease (HSDD), irritable bowel syndrome (IBS), insula (INS), lateral (lat), low somatization score symptomatic diverticular disease (LSDD), mid cingulate cortex (MCC), medial pre-frontal cortex (mPFC), pain catastrophizing score (PCS), posterior insula (pINS), Patient Health Questionnaire 12 Somatic Symptom (PHQ12-SS), Patient Health Questionnaire 15 (PHQ15), pre-frontal cortices (PFC), random effects (RFX), symptomatic diverticular disease (SDD), Sir Peter Mansfield Magnetic Resonance Centre (SPMMRC), thalamus (THAL), visual analogue score (VAS).

Introduction

Diverticular disease (DD) of the colon, characterized by mucosal herniation, affects predominantly those over 65 years old and is associated with considerable morbidity. The incidence of DD and its complications are increasing¹⁻³ however our understanding of DD is still incomplete. Diverticulosis without symptoms is termed asymptomatic DD (ADD). A significant minority of patients have recurrent episodes of pain (symptomatic DD, SDD). Chronic pain symptoms can be present for prolonged periods of time resulting in an associated reduction in quality of life and increased cost to the health service^{4,5}.

Visceral pain pathways involve the enteric nervous plexus, signaling to a variety of regions in the brain via afferent tracts through the spine, and back via descending nociceptive inhibitory control mechanisms within the brain^{6,7}. Key cortical brain regions involved in the response to pain include the anterior cingulate cortex (ACC), amygdala (AMYG), hypothalamus (HpTH), the posterior, mid and anterior insula (INS) locus coeruleus (LC), periaqueductal grey (PAG), prefrontal cortex (PFC) including both dorsolateral and orbitofrontal areas, rostroventral medulla (RVM), primary and secondary somatosensory cortices (SI and SII) and the thalamus (Thal). There are widespread connections between these areas which are often shown to be activated in brain imaging studies of emotion processing touching upon affective, emotional and somatosensory aspects of pain. The descending nociceptive inhibitory control (DNIC) network and the PFC⁸ produce a fronto-limbic regulatory network⁹⁻¹².

Recent work has shown that SDD patients have visceral hypersensitivity to rectal barostat distension^{13, 14} which also occurs in irritable bowel syndrome (IBS). In another study using rectal barostat distension, SDD patients had a significantly lower pain threshold than ADD and healthy volunteers¹⁵. In that study mucosal biopsies revealed elevation in RNA expression of tachykinins and galanin receptors (GALR1 and NK1R), TNF-alpha and IL-6 in the SDD group, suggesting that the development of painful DD is associated with these neurochemical changes and low level chronic inflammation¹⁵. Those patients with SDD have also been shown to report higher levels of somatization as measured by the Patient Health Questionnaire 12 Somatic Symptom scale (PHQ12-SS) compared to those with asymptomatic disease¹⁶. This suggests that both central (psychological) and peripheral factors such as prior inflammation and changes in the enteric nervous system play a role in symptom reporting in this group¹⁷. Therefore it is possible to stratify these symptomatic patients by levels of somatization as it may be the mechanisms of pain perception are different between these two groups. Brain activation imaging studies of visceral pain¹⁸ tend to consider acute stimuli to different

parts of the gastrointestinal tract and reveal a consistent network response including posterior and anterior INS and ACC, S1, regions of the PFC and Thal, some of which have direct anatomical connections as studied with diffusion tensor imaging (DTI) ¹⁹. A recent study of anticipation of somatic pain in DD showed brain activation differences between high and low somatizing DD patients ²⁰. In addition to the increasing number of imaging studies showing alterations of brain function in gastrointestinal diseases there is increasing evidence that structural changes can also occur in the brain. Studies in IBS showed grey matter density and cortical thickness differences in the anterior cingulate cortex (ACC), mid-cingulate cortex (MCC), medial prefrontal cortex, posterior parietal cortex, orbitofrontal cortex, Thal, (para)hippocampus, secondary somatosensory cortex SII, and correlations between the thickness of both the insula (INS) and dorsolateral prefrontal cortex (dlPFC) with clinical scores ^{21, 22}. Some of these affected areas correspond well with brain areas found to have structural alterations in chronically painful conditions such as ACC, MCC, INS, SII in chronic pancreatitis ²³; ACC, dlPFC, prefrontal cortex, motor cortex, medial frontal gyrus in fibromyalgia ^{24, 25}; Thal, , MCC, anterior INS and dlPFC in chronic pain ^{26, 27}.

Recent studies have shown structural changes in similar areas also in other gastrointestinal disease groups such as Crohn's disease and functional dyspepsia patients ²⁸⁻³⁰. However, it is not known if grey and white matter changes occur in symptomatic DD and particularly if differences may relate to low (LSDD) or high somatization DD (HSDD). The primary aim of this cross-sectional study was to test the hypothesis that HSDD patients differed from LSDD patients in a selected number of brain regions commonly identified by brain imaging studies related to the pain network: dlPFC, MCC, motor cortex, frontal superior orbital, SII, INS (anterior, posterior, frontal operculum), middle frontal gyrus, temporal pole, ACC, orbitofrontal, hippocampus, parahippocampus, posterior parietal and Thal. In particular, we hypothesized that HSDD would show increased cortical thickness in regional areas of the prefrontal cortex such as dlPFC compared to LSDD. Secondly this study aimed to explore correlations between standard clinical scores of anxiety, depression and pain catastrophizing and structural brain imaging findings.

Materials and Methods

Subjects

Seventy-two participants (24 males, 48 females) between 21 and 75 years of age were recruited from the gastrointestinal medicine and surgery clinics and a database of interested patients held

at the Nottingham Digestive Diseases Centre (NDDC) NIHR Biomedical Research Unit. All participants were screened by a medical doctor or a research nurse using a structured telephone interview before the study day to confirm the gastrointestinal diagnosis and check for inclusion and exclusion criteria (Table 1). We aimed to recruit and study separate groups of patients with diverticulosis. These were subdivided according to whether they reported recurrent abdominal pain into those with no pain, labelled asymptomatic (ADD), and those with recurrent pain, labelled “symptomatic diverticular disease” (SDD). These SDD were again subdivided according to the level of somatization as assessed by the PHQ12-SS into low somatization diverticular disease (LSDD) with PHQ12-SS <7 and high somatization diverticular disease (HSDD) patients with PHQ12-SS >6. We also aimed to recruit a group of patients with a gastrointestinal disease of different origin but with similar characteristics. Therefore we recruited a group of irritable bowel syndrome (IBS) patients. The symptomatic DD patients exhibited a range of bowel habits hence the IBS patients were selected using the recurrent abdominal pain/discomfort Rome III criteria without specifying a particular bowel habit. No analysis was taken of the specific location of the pain region in the abdomen. All study participants had structural bowel imaging as part of their hospital diagnosis, either with flexible sigmoidoscopy or colonoscopy, CT, or barium enema. Patients with current acute diverticulitis were not included in the study. Patients with prior episodes of acute diverticulitis were included in the study as this is a known risk factor for the development of symptomatic disease and therefore excluding these patients would introduce a potential selection bias. Patients with prior inflammatory bowel disease were also excluded. The final disease subgroups selected had a similar number of participants: 20 patients with ADD, 18 patients with LSDD, 16 patients with HSDD and 18 patients with IBS. The study was approved by the Nottingham Regional Ethics Committee 1 (09/H0403/43) and written informed consent was obtained for all participants. The patients were recruited from the local gastroenterology clinics endoscopy lists and colorectal surgical clinics. ADD patients were recruited from those with no symptoms but one or more colonic diverticulum identified on endoscopy, barium enema or CT scan. Symptomatic DD were included if any diverticula were present. A total of 426 potential participants were identified and contacted. Respondents were initially screened by structured questionnaire over the phone, leading to the enrollment of the 72 patients studied here. Recruitment started in February 2010 and completed in September 2011.

Symptoms questionnaires

Psychological factors can be present in symptomatic DD ¹⁷. The patients completed a number of validated questionnaires to assess their levels of anxiety, depression and somatization of symptoms. The hospital anxiety and depression scores (HAD)³¹ and pain catastrophizing score (PCS)³² were used. Somatization was assessed using the Patient Health Questionnaire 12 Somatic Symptom scale (PHQ12-SS) ^{16, 33}, an adaptation of the standard PHQ15 without the assessment of the gastrointestinal symptoms. Patients are considered to have abnormal values if they score more than 6 on the scale ¹⁶. The DD patients were therefore divided into low somatization (LSDD) and high somatization (HSDD) if they had a PHQ12-SS up to 6 (LSDD) or 7 and higher (HSDD).

MRI Protocol

MRI was performed on a Philips 3T Achieva MRI scanner at the Sir Peter Mansfield Imaging Centre, using 8-channel receive head coil. A three-dimensional, gradient-echo, T₁-weighted (MPRAGE) sequence was used to acquire sagittal anatomical images of the whole brain, acquired and reconstructed at 1 mm³ spatial resolution. The sequence employed a repetition time (TR) of 8.2 ms, echo time (TE) of 3.8 ms, 8° flip angle, TI of 960 ms, linear phase encoding order with data acquired with a 256×256 matrix.

MR image quality control

The MRI scans were first inspected by AP and a record was made of those artifacts that could affect image processing, such as movement noise, susceptibility effects or Gibbs artifacts. The results of all volumetric, cortical thickness and voxel based morphometry analyses were quality controlled by AP: scans inadequately processed were either re-analyzed or excluded.

Whole Brain Volumetry and Cortical Thickness Analysis

Freesurfer ³⁴ was used to estimate the global grey and white matter volume, as well as global and regional cortical thickness averages. For each patient, the standard, recommended FreeSurfer pipeline was run. Briefly, scans were first corrected for intensity inhomogeneity and skull stripped before being projected to Talairach space, where grey matter, white matter and a variety of structures were segmented to form tissue maps. These were then triangulated, optimized for the location of the tissue boundaries, corrected for topological anomalies and used

to calculate local, regional and global thickness measurements, which were then averaged across the hemispheres.

Voxel Based Morphometry (VBM)

Statistical Parametric Mapping (SPM) software package version 12 was used to assess local morphological differences between low and high somatization patients in terms of grey matter density. The VBM analysis was conducted using the high-precision DARTEL approach recommended by Ashburner³⁵. Scans were first manually corrected for position and orientation. Grey and white matter tissues maps were then generated and imported into the DARTEL tool, in order to create an unbiased, cross group template, to which all scans were precisely registered. The group-specific template was then registered to the SPM's standard template in Montreal Neurological Institute (MNI) space, which made it possible to project all grey matter maps onto the MNI space, whilst ensuring excellent correspondence across patients. Finally, the maps were smoothed with a Gaussian kernel of 8mm full width half maximum. A between-subject t-test was carried out between LSDD and HSDD patients on the modulated smoothed maps, with age, gender, BMI and the total cranial volumes obtained above as covariates, in accordance with the literature. As an extra quality control process, tissue volumes obtained with VBM (by integrating over the modulated tissue maps) were compared to those obtained with Freesurfer.

Statistical analysis

All statistical analyses were performed in SPSS (IBM SPSS Statistics, Version 22.0. Armonk, NY).

We used one-way, independent-sample ANOVA and ANCOVA analyses to compare demographic characteristics as well as volumetric and thickness measurements at the global and regional level across patient groups. We focused on those regions identified *a priori* from the literature, as discussed in the Introduction, i.e. various sub-structures of the cingulate gyrus (ACC, anterior, posterior and dorsal MCC, PCC), prefrontal cortex (dlPFC, MPFC, vlPFC), and the insula. We systematically ran all post-hoc pair-wise comparisons, which were corrected for multiple comparisons with the conservative Tukey HSD test.

Since clinical characteristics deviated substantially from a normal distribution they were compared across groups using non-parametric independent-samples Kruskal-Wallis tests.

We ran all *post-hoc* pair-wise comparisons using Dunn's tests, corrected for multiple comparisons with the 5% False Discovery Rate. Finally, we used Spearman's ρ to correlate thickness measurements and clinical characteristics.

In terms of VBM, a between-subject t-test was carried out between LSDD and HSDD patients on the modulated smoothed maps, with age, gender, BMI and the total cranial volumes as covariates, in accordance with the literature. The statistical comparisons were corrected for multiple comparison using first the standard Random Field Theory (RFT) cluster approach³⁶ and then a more liberal approach (clusters larger than 20 voxels with uncorrected threshold at the voxel level of 0.001) where no cluster survived the RFT threshold.

We used $\alpha=0.05$ as the threshold for significance and reported 95% confidence intervals and η^2 effect sizes where appropriate. The statistical review of the study was performed by a biomedical statistician.

Results

All volumetric and thickness analyses were successful therefore no data were discarded.

Clinical Characteristics

Demographics and clinical information for each patient group is provided in Table 2. One-way, independent-sample ANOVAs showed that patients differed in terms of age ($F=12.4$, $P<0.001$, partial $\eta^2=0.35$), with IBS patients being significantly younger than others (IBS<ADD: $P<0.001$, $CI_{95\%}=[-24.58,-9.00]$; IBS<HSDD: $P=0.03$, $CI_{95\%}=[-17.06,-0.58]$; IBS<LSDD: $P<0.001$, $CI_{95\%}=[-22.38, -6.40]$).

Independent-Samples Kruskal-Wallis tests showed statistical differences across patient groups in terms of Somatic Symptom Severity ($\chi^2=33.18$, $P<0.001$, $\eta^2=0.47$), Depression ($\chi^2=12.6$, $P=0.006$, $\eta^2=0.18$), and Anxiety ($\chi^2=8.66$, $P=0.03$, $\eta^2=0.12$), but much less so in terms of Pain Catastrophizing ($\chi^2=3.21$, $P=0.36$, $\eta^2=0.05$). In the first two cases, patients with HSDD had higher means than patients with LSDD with the *post-hoc* Dunn's P values at <0.001 and 0.03, respectively. We also found the following remarkable differences: IBS>ADD ($P<0.001$), IBS>LSDD ($P=0.006$), and ADD<HSDD ($P<0.001$) in terms of Somatic Symptom Severity; and ADD<HSDD ($P=0.006$) in terms of Depression.

Whole Brain Results

The correlation between tissue volumes assessed with Freesurfer and SPM were high: $r=0.82$ ($P<0.001$) for grey matter and $r=0.94$ ($P<0.001$) for white matter.

The one-way ANCOVA showed a substantial effect of age on the total amount of grey matter ($P<0.001$, partial $\eta^2=0.15$), and on the total amount of white matter ($P=0.008$, partial $\eta^2=0.11$), and a reduced effect on average thickness ($P=0.054$, partial $\eta^2=0.06$), as can be qualitatively observed on Figure 1. No tissue volume differences across groups passed the significance threshold when adjusting for age, and no interaction either. However, mean cortical thickness differed substantially across groups ($F=5.733$, $P=0.002$, $\eta^2=0.21$). Post-hoc comparisons showed IBS>ADD ($P=0.002$, $CI_{95\%}=[0.03,0.15]$) and IBS>LSDD ($P=0.01$, $CI_{95\%}=[0.01,0.14]$). Using age as a covariate of no interest abolished those differences.

Region of Interest Results

We first report the differences across groups in terms of regional thickness averages for those specific structures we identified in the introduction.

In the *cingulate cortex*, there were differences in (a) the pmCC ($F=2.756$, $P=0.049$, $\eta^2=0.11$), though no *post-hoc* comparisons survived correction for multiple comparisons, and (b) the PCC ($F=3.304$, $P=0.02$, $\eta^2=0.13$) with *post-hoc* comparisons showing that HSDD>ADD ($P=0.03$, $CI_{95\%}=[0.01,0.22]$).

In the *prefrontal cortex*, differences were detected in (a) the dlPFC ($F=4.56$, $p=0.006$, $\eta^2=0.17$) with *post-hoc* comparisons showing that HSDD>LSDD ($P=0.03$, $CI_{95\%}=[0.01,0.16]$) and (b) the vlPFC ($F=4.232$, $P<0.001$, $\eta^2=0.16$) with post-hoc comparisons showing that IBS>ADD ($P=0.01$, $CI_{95\%}=[0.02,0.18]$) and IBS>LSDD ($P=0.03$, $CI_{95\%}=[0.01,0.17]$).

We also saw differences in the *insula* ($F=2.855$, $P=0.04$, $\eta^2=0.11$) with *post-hoc* comparisons showing that IBS>LSDD ($P=0.037$, $CI_{95\%}=[0.01,0.24]$).

No statistically significant differences across groups were found for the following (sub-) structures: ACC, aMCC, dPCC and mPFC (see Table 3). However, the thickness of the ACC and the mPFC both correlated with Pain Catastrophizing score (Spearman's $\rho=0.24$, $P=0.043$ uncorrected and Spearman's $\rho=0.25$, $P=0.031$ uncorrected, respectively).

In terms of voxel based morphometry, no cluster survived the random field theory threshold when age, gender, BMI and total cranial volume were used as covariates. However, there were a number of clusters larger than 20 voxels with uncorrected threshold at the voxel level 0.001 that showed a difference in grey matter density. We report here results for the LSDD versus

HSDD contrast since it is the focus of our study; please refer to supplementary table S1 for the complete set of VBM results.

We observed areas with increased gray matter density for HSDD patients w.r.t. LSDD patients in the **cingulate cortex** (MCC), in the **pre-frontal cortex** (dlPFC), and in the frontal, somatosensory and motor cortices. Areas with decreased density were observed in the frontal, occipital and temporal cortices (see Figure 2).

Discussion

This is the first study of structural brain grey matter abnormalities in SDD. The data support our hypothesis of structural brain differences in HSDD patients compared to LSDD and in parts of the hypothesized network response, including MCC, dlPFC, INS, motor and frontal superior orbital cortices. The current focus on the pathophysiology of symptomatic diverticular disease has been on the role of prior inflammation in the form of acute diverticulitis and ongoing low grade inflammation¹⁵. However recognition of the role of alterations in central processing both in the anticipation and response to painful stimuli in this group of patients is providing new insights into the pathophysiological mechanisms responsible for symptom reporting²⁰.

In IBS patients the INS and MCC were shown to have more stress-induced activation during rectal stimulation and reduced modulation of INS activity during relaxation compared to healthy volunteers³⁷. This suggests that anxiety and depression may play a role in altered pain processing. Circuit interaction may be present in DD and be amenable to pharmacological or psychological intervention³⁸. Patients with SDD are more likely to report higher levels of anxiety and depression on the Hospital Anxiety and Depression Scale¹⁷. During rectal stimulation, IBS patients also show modulation of INS, MCC, VL-PFC, but reduced modulation of INS³⁷. Indeed these areas were highlighted in our recent study of anticipation of thermal pain in DD²⁰. This suggested that the SDD group and the IBS group may have greater emotional awareness of the painful stimuli and that this may influence stimulus perception possibly via increased MCC and aINS connectivity within a “salience network”³⁹.

The prefrontal cortex is involved in high level appraisal of anticipated painful events⁴⁰ and emotional awareness⁴¹, expectation and anticipation of pain⁴². In anorexia nervosa, greater activation of the DL-PFC and cingulate was found compared to healthy women undergoing anticipated painful heat stimuli⁴³. Fibromyalgia patients showed increased activation of the PAG, posterior parietal cortex and DL-PFC during anticipation of pain⁴⁴. In our functional

MRI study greater DL-PFC deactivation was seen in response to anticipation of pain in the ADD and the LSDD patients compared to HSDD and IBS²⁰.

In patients with chronic pain the cortical thickness^{45,46} is altered in various areas, and some abnormalities can be reversed by analgesia or symptom improvement. Grey matter changes have also been reported in other pain matrix regions in chronic pain conditions such as the amygdale, hippocampus, post central and superior frontal gyri, INS, prefrontal and ACC^{22,24,47-49}. In fibromyalgia these changes also correlate with disease duration and age⁵⁰. In IBS changes in grey matter thickness have also been reported, with the hippocampus having thickened grey matter while the mid-cingulate cortex was thinned. The insular regions also showed altered thickness, with a reduction for IBS patients with a short duration of symptoms and increased in those who had long term pain²¹. White matter changes have also been detected in thalamo-cortical tracts and insular regions^{47,49,51}. However similarities and differences in the regions affected have been seen between different conditions⁵²⁻⁵⁴. There is also suggestion that effective treatment may reverse these changes in some chronic pain²⁶.

In our patients the ACC and mPFC sub-structures significantly correlated with Pain Catastrophizing scores. Pain catastrophizing is a 'negative cognitive-affective response to anticipated or actual pain'⁵⁵. Catastrophizing, may be linked with activity in the cerebellum and mPFC (anticipation), dorsolateral PFC and dACC (attention) and lentiform nuclei⁵⁶. It can be associated with anxiety and depression hence assessing possible correlation of cortical structures with these scores is valuable. In our recent study of anticipation of pain in DD the HSDD and IBS groups showed areas of correlated activity with PCS scores²⁰. It is worth noting that despite having more days with pain the changes in HSDD and IBS patient' cerebral cortex were remarkably similar, suggesting that anxiety and catastrophizing drive the changes more than just pain sensation.

It was not entirely surprising that whole brain volumetry showed an effect of age on cortical thickness. The limited sample size and the comparisons made only between patient groups, not against a healthy control group, meant that small whole brain volumetry differences did not survive the correction for multiple comparisons. Hypothesis driven regional volumetry and VBM allowed more precise assessment of differences between our groups. Another limitation of this study was that the IBS patients are younger than the DD groups which was expected considering the typical IBS phenotype. There was also an imbalance in gender between ADD and the other groups.

Conclusions

In conclusion, whole brain volumetry, cortical thickness analysis and voxel based morphometry are effective tools to investigate structural brain differences in DD and in particular differences between high somatization DD (HSDD) and low somatization DD (LSDD) patients. The structural brain differences highlighted in this work suggest that these patient groups have differences in pathophysiology. Increased understanding of DD is important since DD is the 5th most costly gastrointestinal condition in the USA after gastro-oesophageal reflux disease, gallbladder disease, colorectal cancer and peptic ulcer disease⁵⁷. Previous therapies have been dominated by a surgical approach directed towards the peripheral causation of symptoms. However our findings suggest directing pharmacological and psychological interventions aimed at altering pain processing may help to reduce the cost and burden of DD, which is likely to increase further as the population age^{3, 58, 59}.

Acknowledgments

The Authors would like to thank all the staff at the NIHR Nottingham Digestive Diseases Biomedical Research Unit for their help with this study. This study was funded by the Wellcome Trust Research Training Fellowship awarded to J Smith (WT086609MA). There are no conflicts of interest competing interests declared for any of the authors.

JKS, DJH, STF, PAG, RCS, and LM designed the research.

JG and JKS recruited the patients.

JKS collected the data. AP analyzed the data.

AP, DJH and LM wrote the manuscript draft.

All authors revised the final manuscript.

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Tables

Table 1: Inclusion and Exclusion criteria

Inclusion criteria	
Participants must have either	<ul style="list-style-type: none">• Symptomatic diverticular disease with short-lived recurrent abdominal pain on 3 or more days a month and at least one or more colonic diverticulum identified on endoscopy, barium enema, or CT scan• Asymptomatic diverticular disease, with no abdominal pain and at least one or more colonic diverticulum identified on endoscopy, barium enema or CT scan• Irritable bowel syndrome, which has been diagnosed by a gastroenterologist at the hospital using ROME II or III criteria
Age	18–85 years
Handedness	Right
Informed consent	Yes
Exclusion criteria	
General	<ul style="list-style-type: none">• Pregnant or lactating women• Severe co-morbidity; for example, heart failure, respiratory failure, alcoholism, or drug dependence• Participation in any other study on Nottingham University campus in the last 3 months• No restrictions on the use of HRT, contraceptives medications, or timing of menstrual cycle with the study day were imposed
Metallic implants or objects	<ul style="list-style-type: none">• Cardiac pacemaker• Implanted cardiac defibrillator• Metallic heart valves• Aneurysm clips

	<ul style="list-style-type: none"> • Carotid artery vascular clamp • Neurostimulator • Insulin or infusion pump or implanted drug infusion device • Non-removable cochlear, otologic, or ear implant • Shot or shrapnel inside the body • Metallic fragments in the eye
Medications	<ul style="list-style-type: none"> • Inability to stop NSAIDs (non-steroidal anti-inflammatory agents), antibiotics or immunosuppressant drugs or taking anti-epileptic, gabapentin, long-term opiates, or antipsychotic medications • Participants taking ondansetron were included in the study, but the medication was not taken until after the study • No exclusions for patients taking antihypertensive medications, diuretics, alcohol, or caffeine prior to the study
Inflammatory conditions	Presence of other gastrointestinal conditions such as ulcerative colitis, Crohn's disease and Celiac disease, malignancy, cirrhosis, current hematological malignancy, untreated peptic ulcer disease, Polymyalgia rheumatic
Abdominal surgery	Previous abdominal surgery (other than appendectomy, hysterectomy, cholecystectomy and sterilization, hernia repair)
Neurological conditions	Previous diagnosis of neurological conditions, for example, stroke, cerebral malignancy, essential tremor, Parkinson's disease and Parkinson plus syndromes, motor neuron disease, dementia, storage disorders, Wilsons disease etc. Peripheral neuropathy (e.g., diabetic, alcohol, stroke)
Other	Claustrophobia, broken skin

Table 2: Patients demographics and clinical scores (mean±SD)

	Patient group			
	ADD	LSDD	HSDD	IBS
<i>n</i> =	20	18	16	18
Gender	10M, 10F	6M, 12F	4M, 12F	4M, 14F
Age (years)	64±7	61±8	56±10	47±11
BMI (kg/m ²)	27±4	29±6	29±4	26±4
PHQ12-SS	3±3	4±2	8±2	7±4
Pain catastro- phizing	11±10	10±8	16±14	16±10
HAD-Anxiety	5±3	6±3	9±4	9±5
HAD-Depres- sion	3±2	4±4	6±3	5±4
Previous diver- ticulitis	10%	39%	31%	0%
Bowel fre- quency (/day)	1.8±0.7	1.7±0.8	2±2	2±2
Days per month of abdominal pain	0.5±0.7	12±11	18±11	11±9

ADD: asymptomatic diverticular disease

LSDD: low somatization diverticular disease

HSDD: high somatization diverticular disease

IBS: irritable bowel syndrome

Table 3: Cortical thickness results comparing all groups for the selected (sub-) structures (see Introduction)

Cortex	(Sub-) structure of interest	Contrast (p<0.05)
prefrontal	dIPFC	HSDD>LSDD (P=0.03)
	vlPFC	IBS>ADD (P=0.01) & IBS>LSDD (P=0.03)
	mPFC	no group difference
cingulate	ACC	no group difference
	aMCC	no group difference
	pMCC	group difference only
	PCC	HSDD > ADD (P=0.02)
	dPCC	no group difference
insular	insula	IBS>LSDD (P=0.037)

Table 4: Voxel-based morphometry (VBM) results comparing HSDD and LSDD

Contrast	Cortex	Region	MNI coordinates (x, y, z)	Peak T
			(45,12,43)	4.09
	prefrontal	L & R middle frontal gyrus, dlPFC	(33,39,42)	4.07
			(42,23,55)	3.83
			(-40,35,42)	3.67
HSDD >	frontal	R superior frontal gyrus	(6,35,63)	4.06
LSDD	cingulate	L & R median cingulate gy- rus (MCC)	(12,39,31)	3.75
			(10,23,45)	3.74
			(-14,15,37)	3.57
	somatosensory	R pre/post-central gyrus	(32,-27,75)	3.47
	motor	L supplementary motor area	(-10,-1,60)	3.86
	frontal	L middle frontal gyrus	(-51,-58,-5)	4.43
			(51,-76,3)	4.12
	occipital	L & R middle occipital	(-52,-84,9)	3.91
LSDD >			(-48,-78,12)	3.35
HSDD			(66,-3,-30)	4.04
	temporal	L & R middle and inferior temporal	(-60,-25,-17)	3.92
			(48,6,-32)	3.79
			(-60,-30,-30)	3.64

LSDD: low somatization diverticular disease

HSDD: high somatization diverticular disease

MNI: Montreal Neurological Institute

dlPFC: dorso-lateral pre-frontal cortex

MCC: median cingulate cortex

L: Left, R: Right

Region names standardized according to the Online Brain Atlas Reconciliation Tool (OBART)

Figure Legends

Figure 1. Average thickness, grey and white matter volumes as a function of age. Average thickness, gray and white matter volumes as a function of age, for each of the four patient groups. ADD: asymptomatic diverticular disease, LSDD: low somatization diverticular disease, HSDD: high somatization diverticular disease, IBS: irritable bowel syndrome.

Figure 2. Voxel-Based Morphometry (VBM) results. Clusters (red) larger than 20 voxels with uncorrected threshold at the voxel level 0.001 for (a) HSDD > LSDD, and (b) LSDD > HSDD. LSDD: low somatization diverticular disease, HSDD: high somatization diverticular disease.