

# 1 **Supporting Information**

## 2 **S1 QST Protocols**

3 The cold pressor tests are utilized to measure time exposed to cold to first perceived pain and  
4 perceived intolerable pain. Participants submerged a whole hand into an ice-bath cooled to 2°C.  
5 If this was intolerable, fingers up to the knuckles were submerged. Participants indicated when  
6 they first felt pain and to remove their hand when the pain became intolerable. Times from  
7 submersion to perception of pain was recorded as CPT. Time from submersion to pain  
8 intolerance was recorded as CIT. If a participant was able to keep their hand submerged for 2  
9 min, they removed their hand to prevent nerve damage. Shorter submersion times indicated  
10 higher pain sensitivity and lower pain tolerance.

11 HPT and HPST were used to measure heat pain sensitivity and tolerance respectively.  
12 Participants rested their arm on a table in front of them while in a seated position. Following  
13 inspection for possible confounding factors (e.g. cuts, bruises), a 25 x 50 mm probe connected  
14 to a Modular Sensory Analyzer (Somedic) was secured onto the volar surface of the forearm [1].  
15 To measure HPT, the probe was slowly heated from an adaptation temperature of 32°C at a  
16 rate of 0.5°C/s until the participant stopped the test at the onset of perceived pain by pressing a  
17 button. Pain onset temperature (HPT) was recorded, and the probe temperature returned to  
18 32°C. Probe temperature did not exceed 50°C to prevent thermal burning. A lower HPT  
19 indicates higher pain sensitivity. All participants were given standardized instructions and a  
20 practice run of the HPT experiment prior to testing.

21 To measure HPST, the probe was removed from the starting arm and placed at the same  
22 position on the opposite arm. From 32°C the probe was heated at a rate of 1°C/s until the  
23 participant pressed a button to indicate change in stimulus perception from painful to  
24 unbearable. The temperature at this point was logged as HPST, and the probe was quickly

25 returned to the adaptation temperature. If the test was not stopped before reaching 50°C, the  
26 probe temperature was automatically returned to 32°C to prevent thermal burning. A higher  
27 HPST indicates higher pain tolerance. All participants were given standardized instructions.  
28 There was no practice run when assessing HPST.

29 MDT was measured using a standardized set of modified von Frey hairs (Optihair2-Set,  
30 Marstock Nervtest, Germany) to assess sensitivity to mechanical force. Filaments were applied  
31 to the forearm. If participants were able to detect the first presentation, a 32 mN von Frey hair,  
32 lighter force hairs were applied. The force of the last von Frey filament the participant could  
33 sense upon application was recorded as MDT. Lower MDT scores reflect higher mechanical  
34 force sensitivity. MPT was measured using custom-made weighted pinprick mechanical  
35 stimulators to assess pinprick hypoalgesia, a decreased sensitivity to painful stimuli. Pinpricks  
36 were applied to the participant's forearm in ascending force order from 8 mN. MPT identifies the  
37 first force at which a blunt pinprick is perceived as sharp. A higher MPT indicates higher  
38 hypoalgesia. Both MDT and MPT tests were repeated a total of five times, and the final scores  
39 for each test were the geometric mean values of the trials.

40 The thermal burn protocol has previously been described and includes a number of measures of  
41 sensitization following a thermal burn, including: pain during burn induction, skin flare extent,  
42 punctate hyperalgesia, and thermal hyperalgesia [2]. A 32 mm<sup>2</sup> probe connected to a servo-  
43 controlled Peltier device (TSA-II, Medoc, Israel) was secured onto the volar surface of the right  
44 forearm with a fabric-covered band, approximately equidistant between the elbow and wrist. A  
45 mild thermal burn injury was induced by heating the probe from 32°C to 45°C at a rate of  
46 0.5°C/s and then maintained at 45°C for 330 s. Pain intensity at 0 s, 120 s, and 210 s were  
47 recorded on a 100 mm visual analogue scale (VAS) measured to 1 mm accuracy. 0 was defined  
48 as 'no pain' and 100 as 'the worst pain you can imagine.' These pain ratings were added and  
49 recorded to provide a total rating of pain intensity during the burn protocol (out of 300).

50 At the end of the thermal burn procedure, the burn site was marked on the skin according to an  
51 acetate template. Skin flare (reddening of unheated skin due to the primary burn site) was  
52 measured at each spoke to the nearest 0.5 cm and used to calculate a total area of flare extent.  
53 Larger flare area reflected greater inflammation in response to thermal burn. Punctate  
54 hyperalgesia, an increased sensation of pain following a perpendicular application of a  
55 mechanical force to the skin, was assessed using a 128 mN pinprick stimulus. The stimulus was  
56 applied to the outermost point of each spoke to serve as a reference point for normal sensation  
57 for the participant; the stimulus was then applied to each dot along the spoke inward towards  
58 the primary burn area. If the stimulus was perceived to be too painful, a smaller pinprick  
59 stimulus (i.e., 64 mN) was used. The pressure was maintained for approximately 1 s at each  
60 point and stopped once the participant reported a change in sensation from 'a prodding  
61 sensation' to a 'sharp pricking.' The distance of this point from the outermost point of the spoke  
62 was recorded and repeated across all spokes to calculate an area of increased pain sensation  
63 (punctate hyperalgesia). 15 min after the thermal burn induction, the time of peak response,  
64 flare extent and punctate hyperalgesia were measured and calculated a second time using the  
65 same technique. The average of the initial and secondary measurements of flare extent and  
66 punctate hyperalgesia were used in analysis.

67 Thermal hyperalgesia, an increased sensation of pain following the application of a thermal  
68 stimulus to the skin, was assessed by re-measuring HPT on the same arm as first  
69 measurement. Thermal hyperalgesia was calculated as the change in HPT before and after  
70 thermal burn. A negative value indicated increased pain sensitivity at a lower temperature.  
71 For all sensory tests, participants had their eyes closed to prevent visual clues from influencing  
72 pain perception.

73 1. Vehof J, Kozareva D, Hysi PG, Harris J, Nessa A, Williams FK, et al. Relationship  
74 Between Dry Eye Symptoms and Pain Sensitivity. *JAMA Ophthalmology*. 2013;131(10):1304-8.

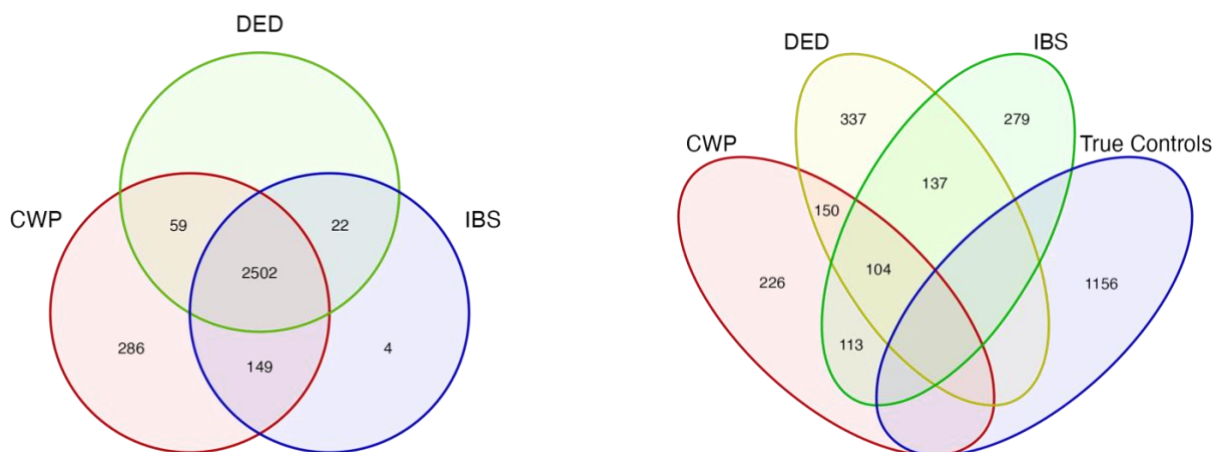
75 2. Norbury TA, MacGregor AJ, Urwin J, Spector TD, McMahon SB. Heritability of  
76 responses to painful stimuli in women: a classical twin study. *Brain*. 2007;130(11):3041-9.

77

78 **S1 Fig. Venn diagram of QST analytical populations by CPS**

79 (A) By CPS questionnaire completion

(B) By CPS diagnosis



80

81 CPSs, chronic pain syndrome; CWP, chronic widespread pain; IBS, irritable bowel syndrome;

82 DED, dry eye disease

83 (A) Venn diagram includes all unique participants represented in Tables 1-3 (N=3022). Each

84 ellipse represents a CPS questionnaire completed by participants with quantitative sensory

85 testing (QST) data and serves as an analytical group. Overlap indicates completion of multiple

86 CPS questionnaires by participants. These participants were included in multiple analytical

87 groups. (B) Venn diagram depicts a subset of participants represented in S1A Fig who

88 completed all CPS questionnaires (N=2502). Each ellipse represents participants with a

89 diagnosis of the indicated CPS or participants with no CPS (true controls). Overlap indicates

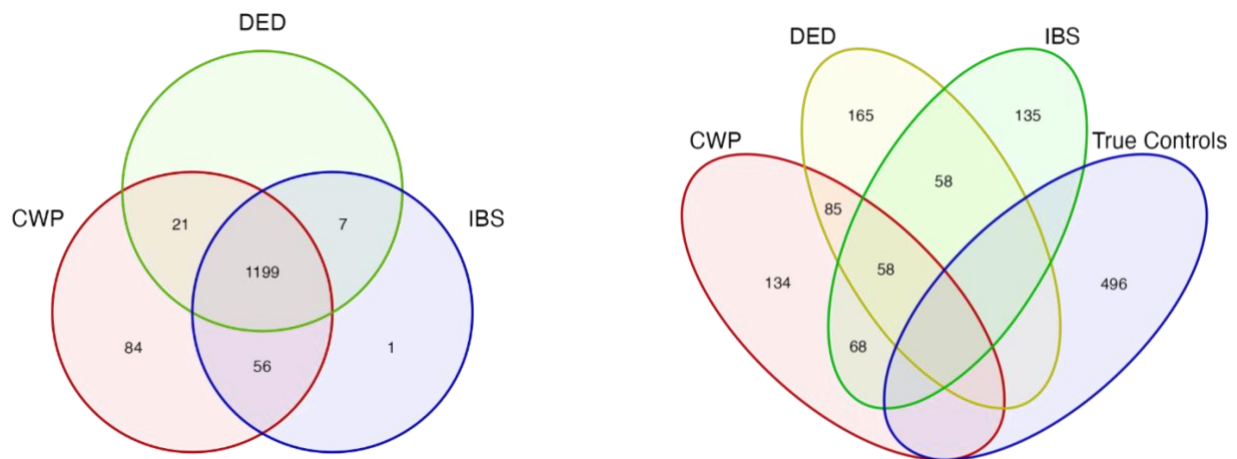
90 multiple CPS diagnoses.

91

92 **S2 Fig. Venn diagram of participants with inflammatory marker data by CPS**

93 (A) By CPS Questionnaire Completion

(B) By CPS Diagnosis



94

95 CPS, chronic pain syndromes; CWP, chronic widespread pain; IBS, irritable bowel syndrome;

96 DED, dry eye disease

97 (A) Venn diagram includes all unique participants represented in S1, S3, S5 Tables (N=1368).

98 Each ellipse represents a CPS questionnaire completed by participants with quantitative

99 sensory testing (QST) and inflammatory marker data and serves as an analytical group. Overlap

100 indicates completion of multiple CPS questionnaires by participants. These participants were

101 included in multiple analytical groups. (B) Venn diagram depicts a subset of participants

102 represented in S2A Fig who completed all CPS questionnaires (N=1199). Each ellipse

103 represents participants with a diagnosis of the indicated CPS or participants with no CPS (true

104 controls). Overlap indicates multiple CPS diagnoses.

105 **S3 Fig. Flowchart of study populations**

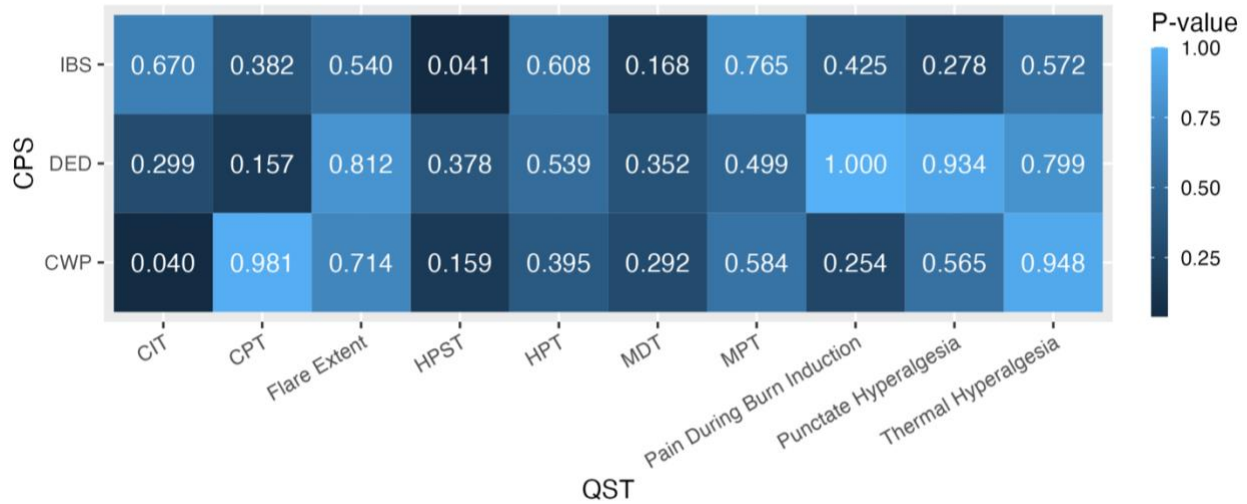


106

107 QST, quantitative sensory testing; CWP, chronic widespread pain; DED, dry eye disease; IBS,

108 irritable bowel syndrome.

109 **S4 Fig. Heatmap of p-values from Mann-Whitney U tests comparing QST scores in**  
 110 **participants with a CPS and true controls.**



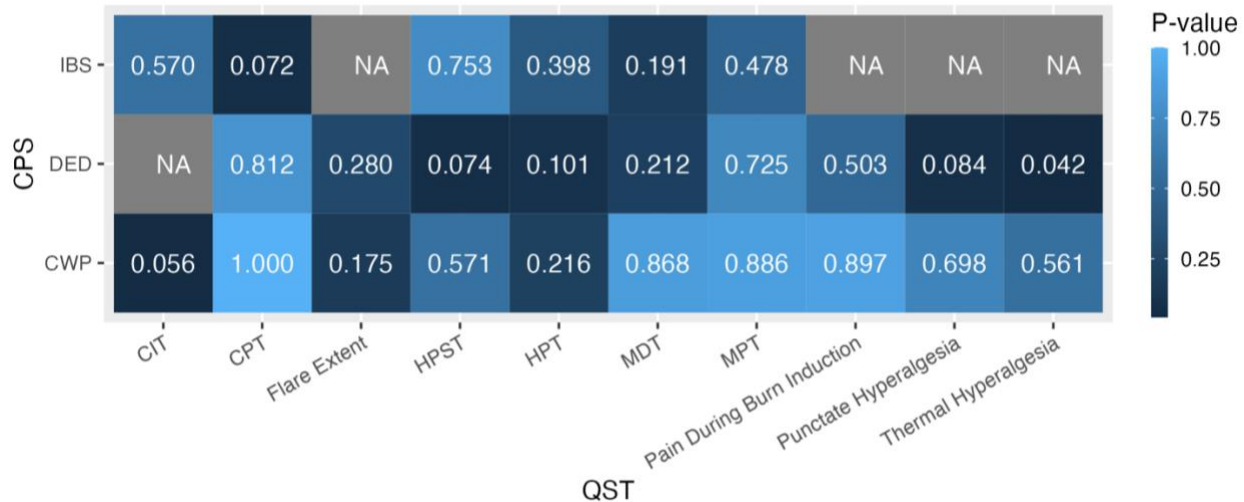
111

112 QST, quantitative sensory testing; CPS, chronic pain syndromes; CWP, chronic widespread  
 113 pain; DED, dry eye disease; IBS, irritable bowel syndrome; CIT, cold intolerable threshold; CPT,  
 114 cold pain threshold; HPST, heat pain supra threshold; HPT, heat pain threshold; MDT,  
 115 mechanical detection threshold; MPT, mechanical pain threshold.

116 Each cell represents the p-value of an individual Mann-Whitney U test for the corresponding  
 117 QST in the relevant CPS questionnaire population (i.e., p-value for Mann-Whitney U test  
 118 comparing CIT scores in participants with CWP and true controls (participants without any CPS)  
 119 = 0.040). Bonferroni-corrected p-value cutoff = 0.005.



120 **S5 Fig. Heatmap of p-values from Mann-Whitney U tests comparing QST scores in**  
 121 **participants with prevalent CPS and incident CPS.**



122  
 123 QST, quantitative sensory testing; CPS, chronic pain syndromes; CWP, chronic widespread  
 124 pain; DED, dry eye disease; IBS, irritable bowel syndrome; CIT, cold intolerable threshold; CPT,  
 125 cold pain threshold; HPST, heat pain supra threshold; HPT, heat pain threshold; MDT,  
 126 mechanical detection threshold; MPT, mechanical pain threshold; NA, not available.

127 Each cell represents the p-value of an individual Mann-Whitney U test for the corresponding  
 128 QST in the relevant CPS questionnaire population (i.e., p-value for Mann-Whitney U test  
 129 comparing CIT scores in participants with prevalent CWP at QST visit and participants with  
 130 incident CWP = 0.056). Cells marked NA did not have enough participants with the CPS of  
 131 interest at date of QST for appropriate comparison. Bonferroni-corrected p-value cutoff = 0.005.

132

133 **S1 Table. Characteristics of female QST participants with CWP data and inflammatory**  
 134 **markers collected within two years of QST visit**

	<b>Total<sup>1</sup> (N=1342)</b>	<b>Participants without CWP (N=973)</b>	<b>Participants with CWP (N=369)</b>
<b>Zygoty</b>			
Dizygotic (%)	782 (58.3)	568 (58.4)	214 (58.0)
Monozygotic (%)	552 (41.1)	398 (40.9)	154 (41.7)
Missing (%)	8 (0.6)	7 (0.7)	1 (0.3)
<b>Age (years)</b>			
Mean (SD)	60.7 (9.5)	60.1 (9.9)	62.3 (8.2)
<b>BMI Category</b>			
Underweight (%)	15 (1.1)	10 (1.0)	5 (1.4)
Healthy (%)	556 (41.4)	430 (44.2)	126 (34.1)
Overweight (%)	513 (38.2)	369 (37.9)	144 (39.0)
Obese (%)	258 (19.2)	164 (16.9)	94 (25.5)

135 CWP, chronic widespread pain; QST, quantitative sensory testing; SD, standard deviation; BMI,  
 136 body mass index.

137 <sup>1</sup> Total female participants in TwinsUK who have completed a CWP questionnaire and at least  
 138 one QST modality with inflammatory marker samples collected within 2 years of QST visit date.

139 **S2 Table. Characteristics of female twin pairs discordant for CWP diagnosis with**  
 140 **inflammatory markers collected on date of QST visit**

	<b>Total<sup>1</sup> (N=234)</b>	<b>Participants without CWP (N=117)</b>	<b>Participants with CWP (N=117)</b>
<b>Zygoty</b>			
Dizygotic (%)	172 (73.5)	86 (73.5)	86 (73.5)
Monozygotic (%)	60 (25.6)	30 (25.6)	30 (25.6)
Missing (%)	2 (0.9)	1 (0.9)	1 (0.9)
<b>Age (years)</b>			
Mean (SD)	62.0 (9.3)	62.0 (9.3)	62.0 (9.3)
<b>BMI Category</b>			
Underweight (%)	2 (0.9)	1 (0.9)	1 (0.9)
Healthy (%)	99 (42.3)	52 (44.4)	47 (40.2)
Overweight (%)	80 (34.2)	41 (35.0)	39 (33.3)
Obese (%)	53 (22.6)	23 (19.7)	30 (25.6)

141 CWP, chronic widespread pain; QST, quantitative sensory testing; SD, standard deviation; BMI,  
 142 body mass index.

143 <sup>1</sup> Total female twins in TwinsUK discordant with their twin for CWP diagnosis with inflammatory  
 144 marker samples collected on date of QST visit.

145 **S3 Table. Characteristics of female QST participants with DED data and inflammatory**  
 146 **markers collected within two years of QST visit**

	<b>Total<sup>1</sup> (N=1211)</b>	<b>Participants without DED (N=843)</b>	<b>Participants with DED (N=368)</b>
<b>Zygoty</b>			
Dizygotic (%)	703 (58.1)	505 (59.9)	198 (53.8)
Monozygotic (%)	502 (41.5)	332 (39.4)	170 (46.2)
Missing (%)	6 (0.5)	6 (0.7)	0 (0.0)
<b>Age (years)</b>			
Mean (SD)	60.9 (9.2)	60.5 (9.7)	61.8 (7.8)
<b>BMI Category</b>			
Underweight (%)	12 (1.0)	10 (1.2)	2 (0.5)
Healthy (%)	508 (41.9)	349 (41.4)	159 (43.2)
Overweight (%)	464 (38.3)	328 (38.9)	136 (37.0)
Obese (%)	227 (18.7)	156 (18.5)	71 (19.3)

147 DED, dry eye disease; QST, quantitative sensory testing; SD, standard deviation; BMI, body  
 148 mass index.

149 <sup>1</sup> Total female participants in TwinsUK who have completed a DED questionnaire and at least  
 150 one QST modality with inflammatory marker samples collected within 2 years of QST visit date.

151 **S4 Table. Characteristics of female twin pairs discordant for DED diagnosis with**  
 152 **inflammatory markers collected on date of QST visit**

	<b>Total<sup>1</sup> (N=258)</b>	<b>Participants without DED (N=129)</b>	<b>Participants with DED (N=129)</b>
<b>Zygoty</b>			
Dizygotic (%)	164 (63.6)	82 (63.6)	82 (63.6)
Monozygotic (%)	94 (36.4)	47 (36.4)	47 (36.4)
<b>Age (years)</b>			
Mean (SD)	62.2 (8.2)	62.2 (8.2)	62.3 (8.3)
<b>BMI Category</b>			
Underweight (%)	0 (0.0)	0 (0.0)	0 (0.0)
Healthy (%)	99 (38.4)	49 (38.0)	50 (38.8)
Overweight (%)	111 (43.0)	58 (45.0)	53 (41.1)
Obese (%)	48 (18.6)	22 (17.1)	26 (20.2)

153 DED, dry eye disease; QST, quantitative sensory testing; SD, standard deviation; BMI, body  
 154 mass index.

155 <sup>1</sup> Total female twins in TwinsUK discordant with their twin for DED diagnosis with inflammatory  
 156 marker samples collected on date of QST visit.

157 **S5 Table. Characteristics of female QST participants with IBS data and inflammatory**  
 158 **markers collected within two years of QST visit**

	<b>Total<sup>1</sup></b> <b>(N=1248)</b>	<b>Participants</b> <b>without IBS</b> <b>(N=911)</b>	<b>Participants with</b> <b>IBS</b> <b>(N=337)</b>
<b>Zygoty</b>			
Dizygotic (%)	722 (57.9)	532 (58.4)	190 (56.4)
Monozygotic (%)	521 (41.7)	374 (41.1)	147 (43.6)
Missing (%)	5 (0.4)	5 (0.5)	0 (0.0)
<b>Age (years)</b>			
Mean (SD)	60.7 (9.3)	61.1 (9.0)	59.5 (9.9)
<b>BMI Category</b>			
Underweight (%)	12 (1.0)	7 (0.8)	5 (1.5)
Healthy (%)	521 (41.7)	384 (42.2)	137 (40.7)
Overweight (%)	477 (38.2)	347 (38.1)	130 (38.6)
Obese (%)	238 (19.1)	173 (19.0)	65 (19.3)

159 IBS, irritable bowel syndrome; QST, quantitative sensory testing; SD, standard deviation; BMI,  
 160 body mass index.

161 <sup>1</sup> Total female participants in TwinsUK who have completed an IBS questionnaire and at least  
 162 one QST modality with inflammatory marker samples collected within 2 years of QST visit date.

163 **S6 Table. Characteristics of female twin pairs discordant for IBS diagnosis with**  
 164 **inflammatory markers collected on date of QST visit**

	<b>Total<sup>1</sup> (N=250)</b>	<b>Participants without IBS (N=125)</b>	<b>Participants with IBS (N=125)</b>
<b>Zygoty</b>			
Dizygotic (%)	156 (62.4)	78 (62.4)	78 (62.4)
Monozygotic (%)	94 (37.6)	47 (37.6)	47 (37.6)
<b>Age (years)</b>			
Mean (SD)	61.6 (8.8)	61.6 (8.8)	61.7 (8.8)
<b>BMI Category</b>			
Underweight (%)	3 (1.2)	1 (0.8)	2 (1.6)
Healthy (%)	104 (41.6)	49 (39.2)	55 (44.0)
Overweight (%)	81 (32.4)	42 (33.6)	39 (31.2)
Obese (%)	62 (24.8)	33 (26.4)	29 (23.2)

165 IBS, irritable bowel syndrome; QST, quantitative sensory testing; SD, standard deviation; BMI,  
 166 body mass index.

167 <sup>1</sup> Total female twins in TwinsUK discordant with their twin for IBS diagnosis with inflammatory  
 168 marker samples collected on date of QST visit.

169 **S7 Table. Detectable effect sizes for Mann-Whitney U tests with 80% (1-β) power**

<b>QST</b>	<b>CWP</b>	<b>DED</b>	<b>IBS</b>
CIT	0.859	0.915	1.001
CPT	0.734	0.736	0.848
Flare extent	0.823	0.802	0.939
HPST	0.186	0.178	0.186
HPT	0.164	0.163	0.165
MDT	0.432	0.423	0.456
MPT	0.429	0.421	0.455
Pain During Burn Induction	0.823	0.802	0.939
Punctate Hyperalgesia	0.832	0.810	0.941
Thermal Hyperalgesia	0.823	0.802	0.939

170 QST, quantitative sensory testing; CWP, chronic widespread pain; DED, dry eye disease; IBS,  
 171 irritable bowel syndrome; CIT, cold intolerable threshold; CPT, cold pain threshold; HPST, heat  
 172 pain supra threshold; HPT, heat pain threshold; MDT, mechanical detection threshold; MPT,  
 173 mechanical pain threshold.

174 Each cell represents the detectable effect size of an individual two-tailed Mann-Whitney U test  
 175 for the corresponding QST in the relevant CPS questionnaire population (i.e., detectable effect  
 176 size for Mann-Whitney U test comparing CIT scores in participants with CWP and participants  
 177 without CWP = 0.859) at 80% (1-β) power and  $\alpha = 0.005$ . Effect sizes were determined using  
 178 G\*Power 3.1 [45]. Analytic samples in which Mann-Whitney U comparison groups did not meet  
 179 the unequal variances assumption are shaded red.



180 **S8 Table. QST mixed effects logistic regression analyses**

181 (A) CIT Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
CIT (scaled)	0.507 (0.220, 1.168)	0.111	0.912 (0.487, 1.705)	0.772	1.276 (0.564, 2.886)	0.559

182

183 (B) CIT Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
CIT (scaled)	0.505 (0.202, 1.263)	0.144	1.034 (0.474, 2.253)	0.933	1.317 (0.555, 3.128)	0.532
Age (scaled)	1.482 (0.353, 6.222)	0.591	2.266 (0.495, 10.37)	0.292	1.251 (0.297, 5.280)	0.760
BMI: overweight	0.921 (0.155, 5.516)	0.931	0.097 (0.010, 0.966)	0.047	0.989 (0.151, 6.493)	0.991
BMI: obese	2.020 (0.236, 17.32)	0.521	0.204 (0.021, 1.967)	0.169	1.080 (0.141, 8.259)	0.941

184

185 (C) CPT Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
CPT (scaled)	1.143 (0.668, 1.956)	0.626	0.718 (0.389, 1.328)	0.291	0.803 (0.408, 1.582)	0.526

186

187 (D) CPT Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
CPT (scaled)	1.109 (0.632, 1.945)	0.718	0.848 (0.443, 1.625)	0.620	0.771 (0.375, 1.587)	0.481
Age (scaled)	1.190 (0.453, 3.125)	0.724	1.932 (0.598, 6.246)	0.271	1.348 (0.405, 4.485)	0.626
BMI: overweight	1.394 (0.398, 4.880)	0.604	0.225 (0.051, 1.004)	0.051	1.786 (0.350, 9.108)	0.485
BMI: obese	0.829 (0.193, 3.556)	0.801	0.216 (0.038, 1.242)	0.086	1.298 (0.219, 7.705)	0.774

188

189 (E) Flare Extent Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Flare Extent (scaled)	0.811 (0.419, 1.570)	0.534	0.996 (0.484, 2.049)	0.991	0.733 (0.299, 1.801)	0.499

190

191 (F) Flare Extent Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Flare Extent (scaled)	0.839 (0.417, 1.687)	0.623	0.996 (0.456, 2.179)	0.993	0.788 (0.305, 2.035)	0.622
Age (scaled)	0.983 (0.291, 3.321)	0.978	1.646 (0.373, 7.268)	0.511	1.737 (0.328, 9.206)	0.516
BMI: overweight	2.898 (0.711, 11.81)	0.138	0.513 (0.103, 2.544)	0.414	1.692 (0.224, 12.76)	0.610
BMI: obese	1.275 (0.241, 6.758)	0.775	0.198 (0.021, 1.904)	0.161	1.079 (0.106, 10.98)	0.949

192

193 (G) HPST Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
HPST (scaled)	0.965 (0.845, 1.102)	0.597	0.993 (0.892, 1.106)	0.902	0.863 (0.769, 0.969)	0.013

194

195 (H) HPST Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
HPST (scaled)	0.933 (0.818, 1.063)	0.298	0.979 (0.879, 1.090)	0.693	0.858 (0.764, 0.964)	0.010
Age (scaled)	1.898 (1.564, 2.303)	<0.001	1.275 (1.106, 1.469)	<0.001	1.066 (0.915, 1.242)	0.412
BMI: overweight	1.232 (0.920, 1.650)	0.161	0.707 (0.558, 0.897)	0.004	0.758 (0.582, 0.986)	0.039
BMI: obese	1.894 (1.327, 2.705)	<0.001	0.681 (0.504, 0.919)	0.012	0.816 (0.588, 1.132)	0.223

196

197

198 (I) HPT Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
HPT (scaled)	1.039 (0.922, 1.170)	0.531	0.972 (0.880, 1.073)	0.570	0.939 (0.846, 1.043)	0.243

199

200 (J) HPT Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
HPT (scaled)	0.986 (0.877, 1.109)	0.820	0.948 (0.858, 1.047)	0.293	0.945 (0.850, 1.051)	0.295
Age (scaled)	1.871 (1.606, 2.179)	<0.001	1.380 (1.225, 1.555)	<0.001	0.928 (0.824, 1.045)	0.220
BMI: overweight	1.253 (0.961, 1.634)	0.095	0.726 (0.582, 0.906)	0.005	0.803 (0.632, 1.022)	0.075
BMI: obese	2.270 (1.658, 3.108)	<0.001	0.728 (0.552, 0.959)	0.024	1.001 (0.747, 1.341)	0.994

201

202 (K) MDT Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
MDT (scaled)	1.445 (0.989, 2.110)	0.057	0.625 (0.319, 1.226)	0.171	1.029 (0.734, 1.440)	0.870

203

204 (L) MDT Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
MDT (scaled)	1.407 (0.963, 2.057)	0.078	0.663 (0.342, 1.286)	0.224	1.003 (0.704, 1.430)	0.985
Age (scaled)	1.636 (0.945, 2.835)	0.079	1.273 (0.823, 1.970)	0.278	1.017 (0.569, 1.818)	0.954
BMI: overweight	0.900 (0.425, 1.904)	0.782	0.534 (0.280, 1.018)	0.057	2.007 (0.861, 4.678)	0.107
BMI: obese	0.976 (0.392, 2.429)	0.959	0.573 (0.256, 1.283)	0.175	1.322 (0.475, 3.683)	0.593

205

206 (M) MPT Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
MPT (scaled)	1.349 (0.982, 1.852)	0.064	0.904 (0.683, 1.197)	0.483	1.165 (0.832, 1.632)	0.374

207

208 (N) MPT Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
MPT (scaled)	1.321 (0.958, 1.821)	0.090	0.927 (0.692, 1.241)	0.609	1.115 (0.790, 1.574)	0.535
Age (scaled)	1.612 (0.946, 2.744)	0.079	1.279 (0.832, 1.965)	0.262	1.010 (0.565, 1.803)	0.974
BMI: overweight	0.935 (0.448, 1.953)	0.858	0.527 (0.278, 0.999)	0.050	1.952 (0.836, 4.560)	0.122
BMI: obese	1.127 (0.468, 2.716)	0.790	0.525 (0.240, 1.148)	0.107	1.352 (0.491, 3.722)	0.559

209

210 (O) Pain During Burn Induction Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Pain During Burn Induction (scaled)	0.589 (0.298, 1.166)	0.129	1.098 (0.562, 2.147)	0.784	0.728 (0.348, 1.522)	0.399

211

212 (P) Pain During Burn Induction Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Pain During Burn Induction (scaled)	0.613 (0.321, 1.171)	0.138	1.121 (0.557, 2.259)	0.749	0.712 (0.341, 1.488)	0.366
Age (scaled)	1.106 (0.343, 3.572)	0.866	1.629 (0.393, 6.742)	0.501	2.059 (0.416, 10.180)	0.376
BMI: overweight	2.934 (0.699, 12.310)	0.141	0.520 (0.106, 2.536)	0.418	1.601 (0.224, 11.440)	0.639
BMI: obese	1.389 (0.252, 7.657)	0.706	0.196 (0.021, 1.853)	0.155	1.217 (0.126, 11.760)	0.865

213



214 (Q) Punctate Hyperalgesia Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Punctate Hyperalgesia (scaled)	0.831 (0.450, 1.535)	0.554	1.399 (0.613, 3.191)	0.425	0.518 (0.170, 1.576)	0.247

215

216 (R) Punctate Hyperalgesia Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Punctate Hyperalgesia (scaled)	0.883 (0.482, 1.617)	0.687	1.383 (0.560, 3.418)	0.482	0.551 (0.183, 1.657)	0.289
Age (scaled)	1.028 (0.338, 3.129)	0.961	1.792 (0.305, 10.530)	0.519	1.885 (0.388, 9.161)	0.432
BMI: overweight	2.561 (0.636, 10.320)	0.186	0.310 (0.037, 2.615)	0.282	1.428 (0.192, 10.630)	0.728
BMI: obese	1.251 (0.251, 6.240)	0.784	0.135 (0.008, 2.393)	0.172	0.988 (0.107, 9.105)	0.992

217

218 (S) Thermal Hyperalgesia Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Thermal Hyperalgesia (scaled)	0.948 (0.539, 1.670)	0.854	1.033 (0.537, 1.987)	0.923	0.725 (0.339, 1.550)	0.406

219

220 (T) Thermal Hyperalgesia Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
Thermal Hyperalgesia (scaled)	0.867 (0.484, 1.555)	0.633	0.910 (0.439, 1.887)	0.800	0.631 (0.239, 1.663)	0.351
Age (scaled)	1.109 (0.357, 3.444)	0.859	1.666 (0.398, 6.978)	0.485	1.994 (0.277, 14.360)	0.493
BMI: overweight	3.039 (0.746, 12.370)	0.121	0.522 (0.106, 2.578)	0.425	2.442 (0.169, 35.29)	0.512
BMI: obese	1.197 (0.217, 6.591)	0.326	0.187 (0.018, 1.887)	0.155	0.815 (0.049, 13.660)	0.887

221

222 CPS, chronic pain syndromes; CWP, chronic widespread pain; DED, dry eye disease; IBS,  
 223 irritable bowel syndrome; OR, odds ratio; CI, confidence interval; QST, quantitative sensory  
 224 testing; CIT, cold intolerable threshold; CPT, cold pain threshold; HPST, heat pain supra  
 225 threshold; HPT, heat pain threshold; MDT, mechanical detection threshold; MPT, mechanical  
 226 pain threshold.

227 Univariate model: adjusted for Family ID (random effect); no fixed effects

228 Multivariate model: adjusted for Family ID (random effect), age (scaled) and BMI category

229 (nominal) (fixed effects)

230 **S9 Table. Conditional logistic regressions of inflammatory marker level on CPS**

231 **diagnosis in discordant twin pairs**

232 (A) Twin pairs discordant for CWP

233

Inflammatory Marker Level (NPX)	Univariate Model <sup>1</sup>		Multivariate Model <sup>2</sup>	
	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-6	1.10 (0.75, 1.62)	0.615	1.00 (0.67, 1.49)	0.985
IL-8	1.74 (0.97, 3.11)	0.064	1.66 (0.93, 2.97)	0.085
IL-10	0.86 (0.50, 1.51)	0.607	0.90 (0.51, 1.59)	0.723
MCP-1	1.32 (0.72, 2.41)	0.373	1.28 (0.69, 2.35)	0.435
TNF	0.85 (0.46, 1.58)	0.607	0.77 (0.40, 1.46)	0.417

234

235

236

(B) Twin pairs discordant for DED

Inflammatory Marker Level (NPX)	Univariate Model <sup>1</sup>		Multivariate Model <sup>2</sup>	
	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-6	0.80 (0.53, 1.22)	0.306	0.81 (0.54, 1.22)	0.315
IL-8	0.94 (0.57, 1.53)	0.791	0.95 (0.58, 1.55)	0.833
IL-10	0.73 (0.42, 1.25)	0.252	0.74 (0.44, 1.27)	0.280
MCP-1	1.05 (0.58, 1.92)	0.862	1.08 (0.60, 1.94)	0.808
TNF	0.83 (0.53, 1.31)	0.426	0.84 (0.54, 1.32)	0.459

237

238 (C) Twin pairs discordant for IBS  
239

Inflammatory Marker Level (NPX)	Univariate Model <sup>1</sup>		Multivariate Model <sup>2</sup>	
	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-6	0.76 (0.48, 1.21)	0.252	0.84 (0.51, 1.36)	0.472
IL-8	1.26 (0.77, 2.07)	0.361	1.29 (0.78, 2.14)	0.326
IL-10	1.09 (0.63, 1.86)	0.761	1.13 (0.66, 1.95)	0.652
MCP-1	1.17 (0.60, 2.29)	0.652	1.21 (0.61, 2.38)	0.589
TNF	1.06 (0.61, 1.83)	0.832	1.10 (0.63, 1.91)	0.735

240 CPS, chronic pain syndromes; CWP, chronic widespread pain; DED, dry eye disease; IBS,  
241 irritable bowel syndrome; NPX, Normalized Protein eXpression; OR, odds ratio; CI, confidence  
242 interval; IL-6, interleukin-6; IL-8, interleukin-8; IL-10, interleukin-10; MCP-1, monocyte  
243 chemoattractant protein-1; TNF, tumor necrosis factor.

244 <sup>1</sup> Strata: Family ID; Covariates: none

245 <sup>2</sup> Strata: Family ID; Covariates: BMI category (nominal)

246

247 **S10 Table. Inflammatory marker mixed effects logistic regression analyses**

248 (A) IL-6 Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-6	1.307 (1.026, 1.664)	0.02996	1.109 (0.898, 1.369)	0.33646	0.937 (0.743, 1.181)	0.57955

249

250 (B) IL-6 Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-6	1.062 (0.818, 1.377)	0.65217	1.067 (0.847, 1.343)	0.58416	0.983 (0.767, 1.261)	0.89275
Age (scaled)	1.367 (1.104, 1.692)	0.00415	1.167 (0.983, 1.385)	0.07861	0.836 (0.698, 1.002)	0.05244
BMI: overweight	1.561 (1.045, 2.332)	0.02953	0.930 (0.669, 1.293)	0.66595	0.955 (0.669, 1.365)	0.80234
BMI: obese	2.458 (1.471, 4.107)	6E-04	1.022 (0.667, 1.566)	0.92165	0.979 (0.619, 1.548)	0.92652

251

252 (C) IL-8 Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-8	1.283 (0.995, 1.654)	0.05483	1.027 (0.821, 1.285)	0.81548	1.201 (0.951, 1.516)	0.12401

253

254 (D) IL-8 Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-8	1.169 (0.902, 1.515)	0.23672	0.967 (0.766, 1.219)	0.77464	1.291 (1.017, 1.638)	0.03555
Age (scaled)	1.356 (1.108, 1.660)	0.00307	1.239 (1.044, 1.471)	0.01412	0.778 (0.654, 0.926)	0.00466
BMI: overweight	1.484 (1.008, 2.183)	0.04522	0.906 (0.653, 1.258)	0.55721	1.009 (0.712, 1.430)	0.95979
BMI: obese	2.390 (1.485, 3.846)	0.00033	1.043 (0.692, 1.572)	0.84043	0.995 (0.645, 1.536)	0.98268

255

256 (E) IL-10 Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-10	0.919 (0.690, 1.224)	0.56257	1.080 (0.851, 1.371)	0.52773	1.129 (0.876, 1.454)	0.34912

257

258 (F) IL-10 Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-10	0.859 (0.643, 1.148)	0.30490	1.057 (0.829, 1.348)	0.65443	1.150 (0.891, 1.483)	0.28272
Age (scaled)	1.387 (1.135, 1.696)	0.00138	1.226 (1.035, 1.452)	0.01813	0.818 (0.691, 0.968)	0.01950
BMI: overweight	1.513 (1.027, 2.229)	0.03624	0.905 (0.652, 1.255)	0.54878	1.004 (0.710, 1.418)	0.98342
BMI: obese	2.474 (1.534, 3.989)	2E-04	1.031 (0.684, 1.554)	0.88457	0.987 (0.641, 1.519)	0.95122

259

260 (G) MCP-1 Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
MCP-1	1.053 (0.757, 1.466)	0.75861	1.151 (0.861, 1.538)	0.34180	1.002 (0.742, 1.353)	0.98814

261

262 (H) MCP-1 Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
MCP-1	0.886 (0.630, 1.247)	0.48810	1.061 (0.784, 1.435)	0.70067	1.106 (0.812, 1.506)	0.52246
Age (scaled)	1.406 (1.149, 1.720)	0.00093	1.225 (1.031, 1.455)	0.02116	0.800 (0.673, 0.951)	0.01119
BMI: overweight	1.483 (1.010, 2.177)	0.04435	0.906 (0.652, 1.258)	0.55447	1.014 (0.718, 1.432)	0.93783
BMI: obese	2.417 (1.506, 3.881)	0.00026	1.037 (0.687, 1.564)	0.86293	0.999 (0.649, 1.538)	0.99777

263



264 (I) TNF Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
TNF	1.108 (0.817, 1.503)	0.50938	1.213 (0.935, 1.573)	0.14628	1.071 (0.809, 1.419)	0.63173

265

266 (I) TNF Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
TNF	0.938 (0.684, 1.286)	0.69235	1.121 (0.853, 1.474)	0.41092	1.183 (0.889, 1.576)	0.24941
Age (scaled)	1.400 (1.141, 1.717)	0.00126	1.212 (1.020, 1.440)	0.02926	0.792 (0.667, 0.940)	0.00768
BMI: overweight	1.483 (1.009, 2.179)	0.04499	0.907 (0.654, 1.259)	0.56052	1.017 (0.721, 1.436)	0.92279
BMI: obese	2.416 (1.501, 3.891)	0.00028	1.026 (0.681, 1.547)	0.90162	0.989 (0.643, 1.521)	0.96151

267 CWP, chronic widespread pain; DED, dry eye disease; IBS, irritable bowel syndrome; NPX,

268 Normalized Protein eXpression; OR, odds ratio; CI, confidence interval; IL-6, interleukin-6; IL-8,

269 interleukin-8; IL-10, interleukin-10; MCP-1, monocyte chemoattractant protein-1; TNF, tumor

270 necrosis factor.

271 Univariate model: adjusted for Family ID (random effect); no fixed effects

272 Multivariate model: adjusted for Family ID (random effect), age (scaled) and BMI category

273 (nominal) (fixed effects)

274 **S11 Table. Discordant twin conditional logistic regression analyses**

275 (A) IL-6 Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-6	1.103 (0.752, 1.619)	0.61468	0.813 (0.542, 1.218)	0.31453	0.765 (0.483, 1.210)	0.25234

276

277 (B) IL-6 Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-6	0.996 (0.667, 1.488)	0.98522	0.804 (0.530, 1.221)	0.30623	0.836 (0.514, 1.361)	0.47225
BMI: overweight	1.107 (0.589, 2.079)	0.75320	0.900 (0.456, 1.774)	0.76033	0.806 (0.403, 1.615)	0.54370
BMI: obese	2.163 (0.776, 6.028)	0.13996	1.188 (0.468, 3.011)	0.71729	0.626 (0.253, 1.549)	0.31056

278

279 (C) IL-8 Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-8	1.735 (0.969, 3.108)	0.06384	0.949 (0.583, 1.545)	0.83317	1.261 (0.767, 2.072)	0.36107

280

281 (D) IL-8 Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-8	1.665 (0.932, 2.973)	0.08479	0.936 (0.573, 1.529)	0.79108	1.289 (0.777, 2.138)	0.32600
BMI: overweight	1.208 (0.647, 2.255)	0.55370	0.861 (0.443, 1.673)	0.65831	0.706 (0.360, 1.383)	0.30975
BMI: obese	1.856 (0.712, 4.836)	0.20591	1.249 (0.519, 3.005)	0.61926	0.614 (0.266, 1.419)	0.25391

282

283 (E) IL-10 Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-10	0.864 (0.495, 1.507)	0.60657	0.744 (0.436, 1.272)	0.28003	1.087 (0.635, 1.861)	0.76111

284

285 (F) IL-10 Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
IL-10	0.903 (0.512, 1.590)	0.72276	0.727 (0.421, 1.255)	0.25239	1.133 (0.658, 1.954)	0.65222
BMI: overweight	1.226 (0.656, 2.294)	0.52288	0.889 (0.456, 1.733)	0.73024	0.712 (0.363, 1.398)	0.32363
BMI: obese	2.059 (0.794, 5.341)	0.13747	1.341 (0.550, 3.271)	0.51924	0.615 (0.267, 1.419)	0.25471

286

287 (G) MCP-1 Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
MCP-1	1.316 (0.719, 2.411)	0.37311	1.076 (0.596, 1.941)	0.80770	1.167 (0.596, 2.285)	0.65154

288

289 (H) MCP-1 Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
MCP-1	1.276 (0.692, 2.352)	0.43534	1.055 (0.580, 1.918)	0.86166	1.206 (0.612, 2.376)	0.58862
BMI: overweight	1.208 (0.650, 2.243)	0.55013	0.872 (0.446, 1.705)	0.68888	0.712 (0.363, 1.395)	0.32209
BMI: obese	1.972 (0.765, 5.086)	0.16001	1.253 (0.520, 3.017)	0.61538	0.620 (0.269, 1.429)	0.26198

290

291 (I) TNF Univariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
TNF	0.850 (0.458, 1.578)	0.60719	0.843 (0.537, 1.324)	0.45883	1.061 (0.614, 1.834)	0.83225

292

293 (I) TNF Multivariate Analyses

Term	CWP		DED		IBS	
	OR (95% CI)	P-value	OR (95% CI)	P-value	OR (95% CI)	P-value
TNF	0.766 (0.402, 1.459)	0.41676	0.833 (0.530, 1.308)	0.42638	1.100 (0.633, 1.912)	0.73517
BMI: overweight	1.167 (0.631, 2.158)	0.62318	0.841 (0.431, 1.643)	0.61316	0.716 (0.365, 1.404)	0.33092
BMI: obese	2.155 (0.823, 5.644)	0.11787	1.233 (0.511, 2.973)	0.64137	0.619 (0.268, 1.429)	0.26138

294

295 CWP, chronic widespread pain; DED, dry eye disease; IBS, irritable bowel syndrome; NPX,

296 Normalized Protein eXpression; OR, odds ratio; CI, confidence interval; IL-6, interleukin-6; IL-8,

297 interleukin-8; IL-10, interleukin-10; MCP-1, monocyte chemoattractant protein-1; TNF, tumor

298 necrosis factor.

299 Univariate model: adjusted for Family ID (strata); no covariates

300 Multivariate model: adjusted for Family ID (strata) and BMI category (nominal)