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Intestinal Permeability in Irritable Bowel Syndrome With and Without Comorbid Conditions: Results From the European DISCOvERIE Cohort

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ABSTRACT

Background: Irritable bowel syndrome (IBS) is a disorder of gut-brain interaction often accompanied by psychiatric and/or somatic comorbidities. Intestinal-barrier dysfunction has been implicated in IBS pathophysiology. However, the contribution of psychiatric and somatic comorbidities to intestinal permeability remains unclear. We aimed to assess intestinal permeability in a

The last two authors contributed equally to this article.

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large multicentric cohort of IBS patients with and without psychiatric and/or somatic comorbidities, compared with disease controls, and healthy subjects.

Methods: IBS patients and disease controls were recruited across nine European centres. Small intestinal and colonic permeabilities were assessed using the lactulose-mannitol-sucralose urinary excretion test. Urinary sugar concentrations were quantified by gas chromatography-mass spectrometry. Analyses were adjusted for relevant clinical and lifestyle confounders.

Results: The lactulose/mannitol (L/M) ratio, but not the percentage of sucralose (%S), was significantly increased in IBS patients ($N = 418$) compared with disease controls ($N = 200$), regardless of IBS subtype or sex. Notably, the increased L/M in IBS patients was primarily driven by a significant decrease in mannitol excretion. IBS patients with psychiatric or combined psychiatric and somatic comorbidities showed a higher L/M than their corresponding disease control groups. However, L/M values did not differ between IBS patients with and without comorbidities. Substantial between-centre variability in L/M ratios awas observed.

Discussion: Small intestinal, but not colonic, permeability, as reflected by an elevated L/M ratio, is increased in IBS patients. These alterations seem to be related to the presence of IBS per se rather than to comorbid conditions. Interestingly, substantial geographic variability was observed, which may reflect environmental or population-related factors influencing barrier function. Further investigations are needed to better understand the clinical relevance of these findings.

1 | Introduction

Irritable bowel syndrome (IBS) is a female predominant disorder of gut-brain interaction (DGBI) characterised by chronic abdominal pain and altered bowel habits [1]. Although classified into four subtypes according to their predominant stool pattern (diarrhoea (IBS-D), constipation (IBS-C), mixed (IBS-M), or unclassified (IBS-U)) [2], IBS is increasingly recognised as a heterogeneous condition in which different pathophysiological mechanisms may coexist. Although IBS is not associated with increased mortality, it severely impairs quality of life due to its chronicity, lack of sensitive and specific biomarkers guiding prevention, and unsatisfactory management [3], highlighting the need for a better understanding of its pathophysiological basis.

In addition to GI symptoms, IBS frequently coexists with psychiatric and somatic comorbidities. IBS patients have a threefold higher risk of anxiety and depressive disorders, and a two-fold higher risk of somatic comorbidities, such as fibromyalgia and chronic fatigue syndrome, compared with healthy subjects [4]. However, the specific mechanisms linking them to IBS remain unclear [4].

Among the proposed pathophysiological mechanisms for IBS, intestinal barrier dysfunction has received considerable attention. Previous research has reported an increased intestinal permeability in subsets of IBS patients compared with healthy subjects, and has been associated with abdominal pain, stool frequency and consistency, and visceral hypersensitivity [5]. An interplay between gut microbiota dysbiosis and increased intestinal permeability has also been widely hypothesised, exposing neural and immune intestinal cells to luminal noxious molecules, thereby leading to mucosal immune activation and disruption of the gut-brain axis [6].

Sugar excretion tests are widely used to assess paracellular permeability but are technically demanding and not readily available in most laboratories. Therefore, circulating biomarkers, such as lipopolysaccharide-binding protein (LBP) and soluble CD14 (sCD14), have been used as surrogate indicators of increased intestinal permeability despite their limited specificity for intestinal barrier dysfunction [7]. Lipopolysaccharide (LPS), a component of the outer membrane of Gram-negative bacteria, may enter the systemic circulation under physiological conditions, primarily via transcellular pathways, either as free LPS or associated with whole bacteria. However, impaired intestinal permeability could also permit paracellular translocation of free LPS. Once LPS reaches the liver, it stimulates the production of LBP and sCD14, which participate in LPS binding and neutralisation. Moreover, sCD14 is also released by activated monocyte/macrophage following LPS exposure. Therefore, circulating levels of LBP and sCD14 are considered indirect markers of increased bacterial translocation rather than direct measures of intestinal permeability.

Importantly, as recently summarised in a systematic review by Hanning et al. [5], previous studies assessing intestinal permeability using multi-sugar tests have been conducted mainly in unicentric cohorts, limited by small sample sizes and methodological heterogeneity (e.g., variation in sugar composition, dosing, etc.), and have yielded inconsistent results. Furthermore, the differences between small intestinal and colonic barrier dysfunction, as well as the potential link of IBS-related comorbidities to intestinal barrier dysfunction, have been poorly explored. In fact, increased intestinal permeability has also been described in a few studies in patients with anxiety [8], depression [9], chronic fatigue syndrome [10], and fibromyalgia [11]. However, it remains unclear whether these conditions further modulate barrier function in IBS.

Importantly, gut microbiota composition and dietary habits differ substantially across geographical regions [12], suggesting that intestinal permeability may vary between countries. However, whether intestinal permeability differs across the European population has also not been completely elucidated.

Therefore, our study aimed to assess in vivo small intestinal and colonic permeability in a large, well-characterised multicentric European cohort of IBS patients with and without psychiatric and/or somatic comorbidities, to compare with disease controls,

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Key Summary

- Summarise the established knowledge on this subject
  - Increased intestinal permeability, assessed using sugar excretion tests, has been reported as a contributing factor in the pathophysiology of irritable bowel syndrome (IBS), particularly in the diarrhoea-predominant subtype.
  - However, studies, often conducted in small cohorts and failing to account for relevant psychiatric and somatic comorbidities, have yielded conflicting results.
- What are the significant and/or new findings of this study?
  - In our multinational cohort comprising more than 600 participants, the lactulose/mannitol (L/M) ratio was significantly increased in IBS patients, irrespective of stool pattern or sex, compared with disease controls. This finding supports the presence of increased paracellular intestinal permeability in IBS patients.
  - Among individuals with psychiatric and somatic comorbidities, those diagnosed with IBS exhibited a higher L/M ratio compared with comorbid patients without IBS, indicating that impaired intestinal barrier function appears to be more strongly associated with IBS than with psychiatric or somatic comorbidities alone.
  - Notably, substantial variability among IBS patients from different countries was observed, which may reflect differences in dietary patterns, lifestyle, environmental exposures, and genetic factors that could modulate intestinal barrier function.

and to evaluate sex- and geographical-related differences in intestinal barrier function.

2 | Materials and Methods

2.1 | Study Participants

Participants were recruited as part of the multinational European DISCOvERIE project, as recently described [13]. The cohort included adults recruited at nine centres across eight European countries that were classified into three groups: (1) IBS patients fulfiling Rome IV criteria for IBS [1] stratified according to the presence or absence of psychiatric (anxiety and/or depressive disorders) and/or somatic comorbidities (fibromyalgia and/or chronic fatigue syndrome), (2) disease controls with psychiatric and/or somatic conditions but without IBS, and (3) healthy controls. More details on the cohort, inclusion and exclusion criteria are provided in the Supplementary Material.

2.2 | In Vivo Intestinal Permeability: Lactulose-Mannitol-Sucralose (LMS) Test

Intestinal permeability was assessed as described in supplementary material using a multi-sugar test measuring the urine excretion of orally administered sugar probes absorbed in different intestinal regions as an indication of small intestinal and colonic permeability.

2.3 | LMS Sugars Quantification

The concentration of the excreted LMS was determined using gas chromatography-mass spectrometry (GC-MS) as previously described [14] and further described in supplementary material.

2.4 | Systemic LPS Translocation

Plasma levels of lipopolysaccharide-binding protein (LBP) and soluble CD14 (sCD14) were measured using a colorimetric Enzyme-Linked Immunosorbent Assay (ELISA) (Hycult Biotech, The Netherlands) according to the manufacturer's instructions.

2.5 | Statistical Analysis

Data are presented as mean ± SEM or as median and interquartile range (IQR), depending on their distribution. Normality of all data sets was determined using the Shapiro-Wilk test. In the demographic table, Kruskal-Wallis with post hoc Dunn test with Bonferroni correction for multiple testing was used. Male/female ratio comparisons between the groups were performed using a Z-test. Given that the data were non-normally distributed, permeability values were log-10 transformed, and a 2-way analysis of variance (ANOVA) using the following factors: presence/absence of IBS, type of comorbidity (non-comorbid, psychiatric, somatic, multi-comorbid), and a Tukey's post hoc test for multiple comparisons was performed. Additionally, clinical variables were screened for their association with permeability using Welch's t-tests or ANOVA with Tukey post hoc tests. Variables showing significant differences in permeability were then included as covariates in two-way analysis of covariance (ANCOVA) models to control for potential confounding factors.

Statistical significance of the Welch's *t*-test, ANOVA, ANCOVA and Tukey's test was determined at $p < 0.05$. Statistical analyses were performed using GraphPad Prism 10.1 and R 4.2.2.

3 | Results

3.1 | Demographic Characteristics

We collected urine samples from 658 patients. Samples from 40 subjects were excluded due to disruption of the freeze-thaw cycle during transport. Therefore, a total of 618 participants were included in the final analysis and stratified into predefined groups: IBS without comorbidities (IBSnc, $n = 180$), IBS with psychiatric comorbidities (IBSpsych, $n = 108$), IBS with somatic comorbidities (IBSsom, $n = 40$), and IBS with both psychiatric and somatic comorbidities (IBSmulti (multicomorbid), $n = 90$). Additionally, healthy controls (HC, $n = 60$), and disease controls without IBS (DC) were included, comprising patients with psychiatric disorders (DCpsych, $n = 48$), somatic disorders (DCsom, $n = 44$), or both psychiatric and somatic disorders (DCmulti, $n = 48$). The demographic characteristics are shown in Table 1 and Supporting Information S1: Table 1.

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TABLE 1 | Baseline clinical and demographic information in IBS_{TOTAL}, namely the sum of IBSnc, IBSpsych, IBSsom, and IBSmulti, also shown separately; and in $Control_{TOTAL}$, namely the sum of HC, DCpsych, DCsom DCmulti, also shown separately.

	$Control_{TOTAL}$ (n = 200)	**HC (n = 60)**	**DCpsych (n = 48)**	**DCsom (n = 44)**	**DCmulti (n = 48)**	**IBS_{TOTAL} (n = 418)**	**IBSnc (n = 180)**	**IBSpsych (n = 108)**	**IBSsom (n = 40)**	**IBSmulti (n = 90)**
Age (median years (IQR))	**45 (27)**	29.5 (21.5)	44.5 (26)	53 (21.5)	51 (26.25)	**44 (24)**	41.5$^{\#\#\#}$ (25.25)	37.5 (23)	53 (12)	47 (14.75)
BMI (median kg/m^2 (IQR))	**25.2*** (7.14)**	23.3 (4.35)	26 (8.4)	28.1 (8.1)	27 (6.7)	**24.1 (5.74)**	23.4 (5.02)	24.2 (5.95)	24.5 (6.5)	24.9 (7.2)
Male:Female (%)	**33:67******	38:62^{++++}	33:67	23:77	35:65	**17:83**	22:78$^{\#}$	15:85$^{\$\$}$	15:85	11:89$^{£££}$
IBS subtype										
IBS-C (%)	—	—	—	—	—	**24**	24	24	24	26
IBS-D (%)	—	—	—	—	—	**37**	41	40	26	32
IBS-M (%)	—	—	—	—	—	**33**	26	30	45	41
IBS-U (%)	—	—	—	—	—	**6**	9	6	5	1
Symptoms severity score (SSS) (median (IQR))	**90**** (138)**	4^{++++} (31)	100 (90)	130 (120)	114 (126.3)	**250 (136)**	220$^{\#\#\#\#}$ (129)	261$^{\$\$\$\$}$ (132)	259$^{°°°°}$ (160)	277$^{£££}$ (133)
Smokers (%)	**26******	23^{+}	33	27	23	**13**	12$^{\#}$	10$^{\$\$\$}$	17.5	19
Alcohol intake units per week (median (IQR))	**1**	2.5^{++++}	0	0	0.5	**0**	1$^{\#}$	1	0	0
Comorbidity diagnosis										
Anxiety (%)	**37**	-$^{+++}$	56	7.9	70	**30**	2.4	50	2.6	71
Chronic fatigue (%)	**31**	—	2.8	42	57	**22**	—	—	63	73
Fibromyalgia (%)	**39*****	—	—	79	62	**21**	0.9	—	77	60
Dietary patterns										
Gluten-free (%)	**6****	3.3^{+}	6.3	4.5	10	**13**	14$^{\#}$	9.3	15	16
Lactose-free (%)	**8.5******	3.3^{++++}	15	11	6.3	**28**	24$^{\#\#\#}$	26	30	34$^{£££}$
Vegan (%)	**2**	3.3	2.1	—	2.1	**1.9**	1.7	3.7	—	1.1
High-rich fibre (%)	**7******	1.7	10	6.8	10	**1**	1.1	0.9$^{\$}$	2.5	–$^{££}$
Medications										
Antidepressants (%)	**42******	3^{+++}	66	43	64	**25**	10	36$^{\$\$\$}$	17.5°	46$^{£}$

(Continues)

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TABLE 1 | (Continued)

	$Control_{TOTAL}$ (n = 200)	HC (n = 60)	DCpsych (n = 48)	DCsom (n = 44)	DCmulti (n = 48)	IBS_{TOTAL} (n = 418)	IBSnc (n = 180)	IBSpsych (n = 108)	IBSsom (n = 40)	IBSmulti (n = 90)
Benzodiazepines (%)	**13.5**	0^{++}	15	14	29	**11**	4	12	18	21
NSAIDs (%)	**24.5******	8	17	41	38	**10**	17	$4^{\$\$}$	$7.5^{\circ\circ\circ}$	$5^{£££}$
Probiotics (%)	**5**	$0^{p\,=\,0.051}$	6	4	10	**6**	$8^{\#}$	4.5	10	$1^{£}$
Laxatives (%)	**1.5******	0^{++}	2	0	4	**12**	$16^{\#\#\#}$	$12^{\$}$	10	4
Anti-acids (%)	**13**	1.5^{++}	14	16	22	**17**	$24^{\#\#\#}$	7	12.5	16.5
Antidiarrheal (%)	**2.5**	0	4	0	6	**3**	4	4	5	0

Note: Differences between IBS_{TOTAL} versus HC and versus $Control_{TOTAL}$, and between each IBS group and the corresponding DC group were assessed. * *IBS_{TOTAL} vs $Control_{TOTAL}$, + IBS_{TOTAL} vs HC, # IBSnc vs HC, $ IBSpsych vs DCpsych, ° IBSsom vs DCsom, £ IBSmulti vs DCmulti.* Bold values correspond to the $Control_{TOTAL}$ and IBS_{TOTAL} groups.

Abbreviations: DCmulti: disease control with both psychiatric and somatic disorder, DCpsych: disease control with psychiatric disorder, DCsom: disease control with somatic disorder, HC: healthy controls, IBSmulti: IBS with both psychiatric and somatic comorbidities, IBSnc: IBS non-comorbid, IBSpsych: IBS with psychiatric comorbidity, IBSsom: IBS with somatic comorbidity.

$^{*}p < 0.05$.
$^{**}p < 0.01$.
$^{***}p < 0.001$.
$^{****}p < 0.0001$.

3.2 | Small Intestinal Paracellular Permeability

3.2.1 | The L/M Ratio was Higher in IBS Patients Compared to DC, Independently of Stool Pattern and sex

More than 50% of IBS patients presented with psychiatric or somatic comorbidities. To provide a more representative control group, we defined a total control group where we included healthy subjects and disease controls without IBS (psychiatric, somatic, or both: $Control_{TOTAL}$). The L/M in the 0–2h urine collection was significantly higher in IBS_{TOTAL} compared to $Control_{TOTAL}$ (Figure 1A). The individual excretion of lactulose (%L) and mannitol (%M) is shown in supplementary material (Supporting Information S1: Figures 1A, B and 2B, C). Neither IBS subtype (Supporting Information S1: Figure 2A) nor sex (data not shown) significantly influenced the L/M. Interestingly, although the DC group has a higher percentage of smokers (26 vs. 13%, $p < 0.0001$) and NSAIDs consumers (24,5 vs. 10%, $p < 0.0001$), smoking did not affect L/M values. NSAID use was associated with a lower L/M ratio ($p < 0.05$, Supporting Information S1: Figure 3C).

3.2.2 | The L/M Ratio was Higher in IBS Patients with Psychiatric and Somatic Comorbidities Compared to Respective DC Controls

When stratifying IBS patients by comorbid conditions, the L/M ratio remained significantly higher in IBSpsych compared with DCpsych, and in IBSmulti compared with DCmulti. However, L/M ratios did not significantly differ among IBS with and without comorbidities (IBSpsych, IBSsom, and IBSmulti and IBSnc) or between these IBS subgroups and HC (Figure 2A). %L and %M in the 0–2h urine collection are included in supplementary material (Supporting Information S1: Figure 7A, B). Interestingly, a significant increase in the L/M ratio was observed in non-comorbid IBS patients with severe abdominal pain when compared with HC (see Supporting Information S1: Figure 8A).

L/M was influenced by several clinical variables, including chronic fatigue, anxiety, and NSAIDs and benzodiazepines treatment (see Supporting Information S1: Figure 3A–D). These

FIGURE 1 | Small intestinal and colonic permeability in vivo assessed with the LMS sugar test in IBS patients (IBS_{TOTAL} n = 418) and disease controls ($Control_{TOTAL}$ n = 200). (A) L/M ratio is increased in IBS patients (1.21, CI: 1.09–1.33). (B) %S is decreased in IBS patients (0.86, CI: 0.77–0.96). Boxes represent the median, the 25th (bottom) and the 75th percentile (top), and whiskers the 5th/95th percentiles. Adjusted mean ratios and their 95% confidence intervals (CI) are given. $^{**}p < 0.01$, $^{***}p < 0.001$. (CI).

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covariates were then included in the ANCOVA model. After adjustment, differences in L/M between HC, DCpsych, and DCmulti were no longer significant when anxiety and benzodiazepine use were accounted for, suggesting that these factors contributed to the reduced L/M in the disease control groups (Supporting Information S1: Table 2).

Overall, these findings indicate that the presence of IBS, rather than the presence of psychiatric and somatic comorbidities, is associated with an increased L/M ratio.

3.2.3 | *The L/M Ratio Differs Across Centres*

To explore potential geographical variability, small intestinal permeability was analysed in all IBS patients according to the recruiting centre. The L/M was higher in patients recruited at UGOT and UM compared with those from KUL, UNIBO, and VHIR (Figure 3A). The %L and %M in the 0–2h urine collection are included in supplementary material (Supporting Information S1: Figure 9A, B). Although geographical variability was significant in the ANCOVA models, IBS and

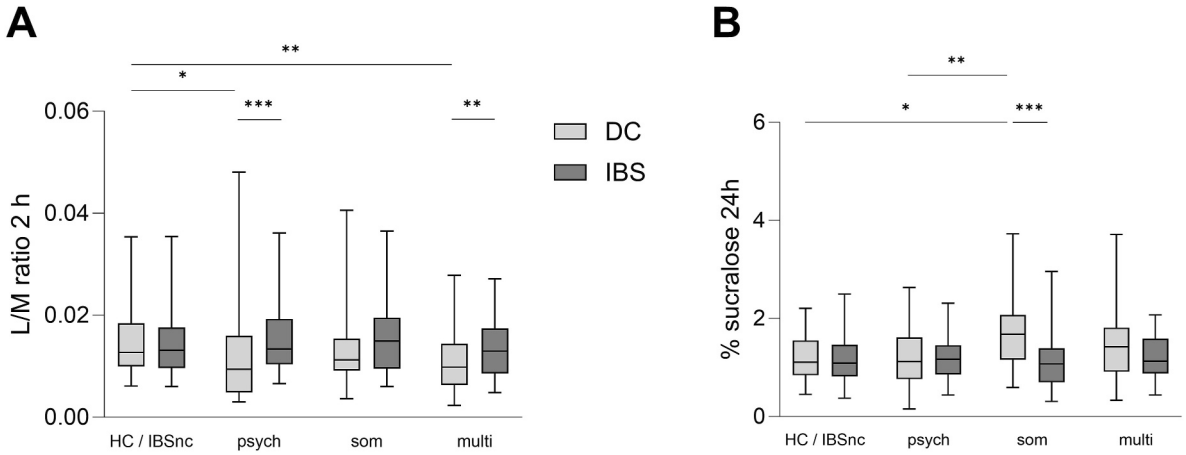

FIGURE 2 | Small intestinal and colonic permeability in vivo assessed with LMS sugar test in IBS patients with and without psychiatric and/or somatic comorbidities versus respective disease controls. (A) L/M ratio is increased in HC versus DCpsych (0.72, CI: 0.54–0.96) and DCmulti (0.70, CI: 0.53–0.93), in IBSpsych versus DCpsych (1.47, CI: 1.21–1.78), and in IBSmulti) versus DCmulti (1.32, CI: 1.08–1.61). (B) %S was increased in DCsom versus IBSsom (0.60, CI: 0.46–0.79), DCpsych (1.73, CI: 1.22–2.47) and HC (1.55, CI: 1.12–2.16)). HC ($n = 60$), IBSnc ($n = 180$), DCpsych ($n = 48$), IBSpsych ($n = 108$), DCsom ($n = 44$), IBSsom ($n = 40$), DCmulti ($n = 48$), IBSmulti ($n = 90$). DCmulti: disease control with both psychiatric and somatic disorder, DCpsych: disease control with psychiatric disorder, DCsom: disease control with somatic disorder, HC: healthy controls, IBSmulti: IBS with both psychiatric and somatic comorbidities, IBSnc: IBS non-comorbid, IBSpsych: IBS with psychiatric comorbidity, IBSsom: IBS with somatic comorbidity. Boxes represent the median, the 25th (bottom) and the 75th percentile (top), and whiskers the 5th/95th percentiles. Data expressed as adjusted mean ratios with their corresponding 95% confidence intervals (CI) *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

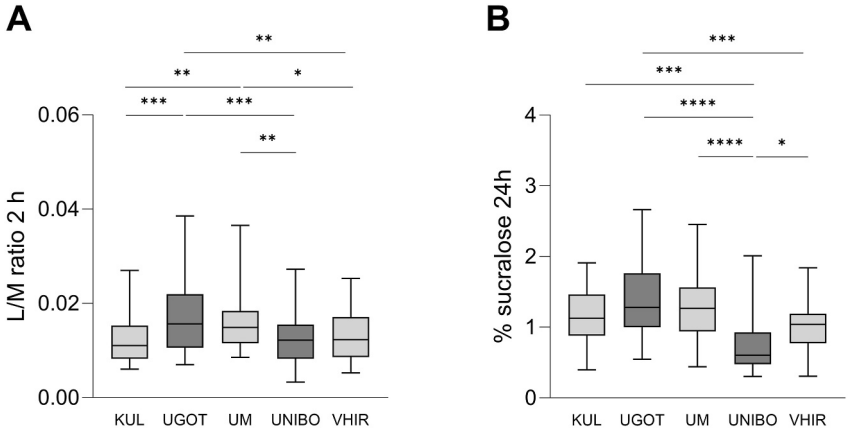

FIGURE 3 | Geographical differences in small intestinal and colonic permeability in vivo assessed with the LMS sugar test only in IBS patients. (A) L/M was increased in IBS patients from UGOT versus KUL (1.35, CI: 1.1–1.68), UNIBO (1.44, CI: 1.11–1.87), and VHIR (1.28, CI: 1.04–1.56). UM patients had increased L/M versus KUL (1.30: 1.05–1.60), UNIBO (1.39: 1.07–1.80)), and VHIR (1.22, CI: 1.00–1.50). (B) %S was decreased in UNIBO patients) versus KUL (0.64, CI: 0.47–0.89), UGOT (0.53, CI: 0.39–0.72), UM (0.59: 0.43–0.80), and VHIR (0.74, CI: 0.54–1.00). %S was decreased in VHIR compared to UGOT patients (0.72, CI: 0.57–0.91). KUL (Belgium, $n = 81$), UGOT (Sweden, $n = 101$), UM (The Netherlands, $n = 97$), UNIBO (Italy, $n = 42$), VHIR (Spain, $n = 94$). Boxes represent the median, the 25th (bottom) and the 75th percentile (top), and whiskers the 5th/95th percentiles. Data expressed as adjusted mean ratios with their corresponding 95% confidence intervals (CI). *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$, ****$p < 0.0001$.

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comorbidity remained significant predictors, showing that their effects were independent of geographical differences (Supporting Information S1: Tables 2, 3 and 4).

3.3 | Colonic Paracellular Permeability

3.3.1 | The % of Sucralose Excretion was Lower in IBS Patients Compared to DC, Independently of Stool Pattern and sex

The urinary %S in the 2–24 hours collection was lower in IBS_{TOTAL} patients than in $Control_{TOTAL}$ patients (Figure 1B). However, no significant differences were observed between IBS subtypes (IBS-C, IBS-D, IBS-M, and IBS-M) (Supporting Information S1: Figure 2D). The lower %S in IBS was independent of the sex (data not shown).

3.3.2 | The % of Sucralose Excretion was not Altered in IBS Patients With Psychiatric and Somatic Comorbidities Compared to Non-Comorbid IBS

No significant differences in %S were observed among IBS groups with and without comorbidities (Figure 2B). Colonic permeability assessed did not differ between the HC and the different IBS groups. These results were confirmed ex vivo in Ussing chambers when colonic biopsies were studied: no significant differences in TEER and passage of FD4 were observed between HC and IBS; see supplementary material (Supporting Information S1: Table 6 and Figure 10A, B).

%S was higher in DCsom than in HC, DCpsych, and IBSsom, suggesting an increased colonic permeability in patients with somatic comorbidities without IBS. Several clinical variables were identified as covariates based on their effects on %S (see Supporting Information S1: Figure 6A–E). When adding these variables to the ANCOVA model, none of them affected the increase in %S in the DCsom patients (Supporting Information S1: Table 5).

Interestingly, lower %S values were seen in IBSmulti patients compared with DCmulti patients (see Supporting Information S1: Figure 8B).

Material describes additional effects of diagnosis, medications, and diet on intestinal permeability, as well as correlations between L/M, %S and symptom-severity score (see Supporting Information S1: Figure 4A–C, Figure 5A–C, and 13A, B).

3.3.3 | Colonic Paracellular Permeability Differs Across Centres

When stratifying IBS patients by recruiting centre, significant differences were observed. IBS patients from UNIBO had lower %S than those from KUL, UGOT, UM, and VHIR. Moreover, %S was lower in IBS patients from VHIR than in those from UGOT (Figure 3B). Geographic origin did not modify the %S difference between the DCsom and HC and DCpsych and IBS groups (Supporting Information S1: Table 5).

3.4 | LPS Translocation was Increased in IBS Patients Compared to Control Groups

Plasma sCD14 concentrations were significantly higher in IBS_{TOTAL} compared with $Control_{TOTAL}$ group ($p = 0.023$), whereas LBP levels were not different between groups ($p = 0.096$) (Figure 4A,B and supplementary material). Supplementary material also shows additional comparisons of LBP and sCD14 levels between the IBS and DC patient groups (Supporting Information S1: Figures 11 A, B and 12 A, B).

4 | Discussion

In this large multicentric European cohort, we observed an increased small intestinal, but not colonic, permeability in IBS patients as reflected by a higher L/M ratio in the 0–2h urine collection, regardless of stool pattern or sex. Importantly, the presence of IBS itself, rather than the psychiatric or somatic comorbidities, was associated with this increase in the L/M ratio. Of interest, we identified geographical differences in intestinal permeability, which may be influenced by dietary, lifestyle, environmental, or genetic factors [15]. Moreover, IBS patients exhibited a small increase in plasma sCD14 levels, suggesting bacterial translocation.

Previous studies using sugar probes highlighted mostly a higher intestinal permeability in IBS patients [16–19], especially in IBS-D [20–24], and less pronounced in IBS-C [19, 25], while other studies showed no differences compared to healthy subjects or between subtypes (e.g., IBS-D, IBS-C, IBS-M) [26–31]. These discrepancies may stem from methodological variations, such as differences in sugar composition, dosing, urine collection timing, and restrictions before testing, as well as other factors (e.g., use of probiotics, laxatives, etc.) [32]. Although the studies mentioned [16–31] used the same sugars as in the present work, differences in dosing and sampling protocols limit direct comparability and complicate interpretation. In the present study, small intestinal permeability was assessed using %L, %M and their ratio in 0–2h urine fraction, based on evidence that

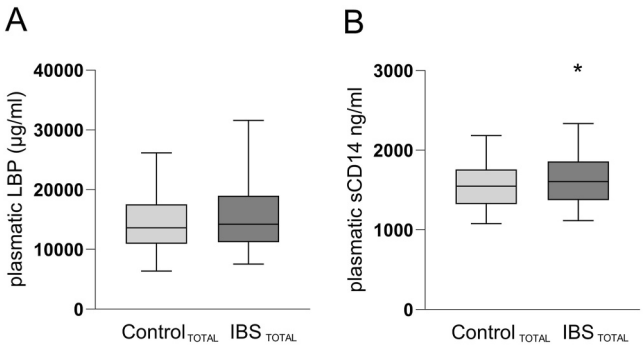

FIGURE 4 | LPS translocation assessed with LBP and sCD14: (A) Plasmatic LBP was not significantly higher in IBS_{TOTAL} patients compared with $Control_{TOTAL}$ (1705, CI: 685.5–2724). (B) Plasmatic sCD14 was significantly higher in IBS_{TOTAL} patients compared with $Control_{TOTAL}$ (73.95, CI: 17.56–130.3). Boxes represent the median, the 25th (bottom) and the 75th percentile (top), and whiskers the 5th/95th percentiles. Data expressed as adjusted mean differences with their corresponding 95% confidence intervals (CI). *$p < 0.05$.

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sugars reach the colon after approximately 2 hours [30]. Moreover, we assess the L/M ratio only as a marker for the small intestinal permeability because both sugars are fermented by gut microbiota in the colon, making them unreliable as markers for colonic permeability [14, 33]. For this reason, we quantified sucralose in the 2–24h urine collection to assess colonic permeability, since sucralose is not degraded by the gut microbiota [5, 33], and provides a more specific assessment of colonic permeability.

Given the controversy of previous studies [5], we first compared intestinal permeability between IBS patients and all Control subjects with and without psychiatric and somatic comorbidities, and we found an increased L/M ratio in the IBS group. When stratifying the cohort according to their comorbidities and comparing IBS comorbid patients and their corresponding disease control groups, we found an increased L/M in the IBS comorbid groups. Interestingly, this difference was not driven by higher %L but rather by lower %M excretion. Essentially, mannitol reflects the intestine's overall absorption capacity. The L/M ratio is commonly used in intestinal permeability tests, as it provides a more comprehensive assessment of barrier integrity [5, 14]. An elevated ratio suggests increased permeability to larger molecules, potentially indicating a compromised intestinal barrier. However, if an increased L/M ratio is driven by reduced mannitol excretion, it may reflect that factors affecting absorptive capacity, for instance, a reduced villus length or intestinal physiology such as increased intestinal motility, may contribute to the observed changes [34] and highlight the need for a cautious interpretation of the L/M ratio when individual sugar excretion differs. In this sense, some more recent research also considers %L and %M for a more nuanced analysis [20, 34].

Most studies report higher intestinal permeability in IBS-D compared to other subgroups (although most compare only IBS-D with healthy subjects, not with other subtypes) [5]. Overall, our data do not support a clear association between stool pattern and small intestinal permeability. This discrepancy could be driven by the inclusion of a higher proportion of postinfectious IBS patients in previous studies.

Given the high prevalence of psychiatric and somatic comorbidities in IBS [4], we compared intestinal permeability between IBS patients with and without psychiatric (i.e., anxiety and depression) and/or somatic (i.e., fibromyalgia and chronic fatigue) comorbidities, and their corresponding disease controls. The L/M was higher in IBS patients with psychiatric comorbidities and IBS multi-comorbid patients compared to the respective disease control groups. Interestingly, a covariate analysis showed that both anxiety diagnosis, and the use of benzodiazepines were associated with L/M values. Benzodiazepines have been shown to affect intestinal motility and water absorption [35, 36], which could indirectly affect intestinal permeability [37]. Therefore, it remains uncertain whether the observed differences reflect the psychiatric comorbidity itself, medication effects or an interaction between both.

With regard to colonic permeability, only a few studies used the non-fermentable sucralose to assess colonic and total intestinal permeability in IBS patients compared with HC. Two studies reported higher sucralose excretion in IBS patients in urine collected over 0–5h [16] and 0–24h [22] intervals, timeframes in which the sugar solution is still in the small intestine. In contrast, two other studies [24] that measured sucralose excretion at 5–24h, more representative of colonic passage, did not find significant alterations in IBS patients. In our study, %S in 2–24h urine did not differ between IBS non-comorbid patients and HC. Interestingly, higher %S values were observed in disease controls with somatic comorbidities, particularly fibromyalgia, compared with their corresponding IBS subgroups (Figure 2B). Moreover, no differences in %S were detected among IBS stool pattern subtypes or among subjects taking NSAIDs, suggesting that altered stool patterns and NSAIDs do not influence colonic paracellular permeability.

The very weak or absent association between sCD14 and LBP with small intestinal and colonic permeability suggests that translocation likely occurs via the transcellular route, and that LBP and sCD14 should be interpreted as indicators of immune activation in response to microbial products rather than as direct measures of paracellular intestinal permeability. Whether LPS translocation occurs in the small intestine or the colon of IBS patients, and the relative contribution to IBS pathogenesis, given the small increase in plasmatic sCD14, requires further investigation.

Dietary patterns, environmental exposures and lifestyle factors have been shown to influence intestinal barrier function [38], and regional variations in intestinal permeability have been reported [39]. Here, we identified geographical differences in both small intestinal and colonic permeability. Interestingly, IBS patients recruited in southern European centres (Italy and Spain) exhibited lower lactulose and sucralose excretion than those recruited from Belgium, the Netherlands, and Sweden. Although unified study protocols and centralised biochemical analyses minimise methodological variability, we cannot rule out differences in environmental or population-related factors between centres. In our cohort, smoking was not associated with intestinal permeability, and alcohol consumption was associated with L excretion but not with the L/M ratio, suggesting that these lifestyle factors alone do not explain the observed geographical variability. Other environmental factors, such as dietary patterns or previous viral and/or bacterial infections, may also contribute, but the absence of detailed data on these factors limits further interpretation. These results may partially explain the discrepancy across previous single-centre studies.

In the present study, several limitations are worth consideration. Firstly, detailed dietary/lifestyle data were not systematically collected, limiting the interpretation of observed geographic variability. Secondly, the absence of additional intermediate timepoints during urine collection. As aforementioned, after 2 hours, most of the sugar solution has reached the colon [30]. However, for example, an additional fraction at 5 hours would have facilitated comparisons with other studies that assess GI permeability (combined small intestine and colon) in 0–5h urine collections. Finally, due to the cross-sectional design, a causal relationship between altered permeability and symptom severity could not be established. Another limitation relates to the study design, as not all patient groups were recruited across all participating centres. Recruitment was performed according to the specific expertise of each centre,

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potentially leading to an unbalanced cohort. Nevertheless, after adjustment for recruiting centre, the results remained essentially unchanged, indicating that this factor did not substantially affect the study findings.

Despite these limitations, we believe our study has notable strengths, as it represents the largest comprehensively characterised multinational cohort to assess intestinal permeability in IBS. In addition, the use of a standardised methodology, including unified collection protocols and centralised quantification of urinary sugars, reduces variability and enhances results reliability.

In conclusion, this large, multicentric European study shows that small intestinal, but not colonic, permeability is increased in IBS compared with disease controls, independent of stool pattern or sex. In patients with comorbid IBS, these changes appeared to be more strongly associated with IBS than with psychiatric or somatic comorbidities alone. We also observed substantial variability across countries, suggesting that diet, lifestyle, environmental, and genetic factors may influence barrier function. Additionally, IBS patients had elevated sCD14 levels, indicating immune activation in response to microbial LPS. Our results highlight that relying solely on the L/M ratio may be insufficient to assess small intestinal permeability. More standardised methods and complementary physiological and immunological measures are needed to better clarify the pathophysiological and clinical relevance of intestinal permeability in IBS.

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Ethics Statement

The study protocol was elaborated under DISCOvERIE and implemented by 9 recruiting centres by the principles of the ethical guidelines of the revised Declaration of Helsinki after approval by the Medical Ethics Committees at: Hospital Vall d'Hebron (PR(AG)26–2020 A), Goeteborgs Universitet (2020–04072), Universiteit Maastricht (NL75159.068.20/METC 20–076), Semmelweis Egyetem (19461–5/2021/EUIG), Stichting Radboud Universitair Medisch Centrum (2021–1049), Johann Wolfgang Goethe—Universität Frankfurt am Main (20–939), Alma Mater Studiorum—Università di Bologna (632/2020/Sper/AOUBo), KU Leuven (B3222021000445, S64753), Universitatea de Medicina Si Farmacie Iuliu (294/15/09/2020). Ethics approval was obtained in all participating centres, including Material and Data Transfer agreements for joint analyses. All study participants received written and oral study information and provided oral and written consent to participate in the study before inclusion.

Conflicts of Interest

Miriam Almirall received honoraria for scientific advice and travel funds from Janssen and speaker honoraria from UCB Pharma and Eli Lilly. Miriam Almirall served on one advisory board from Janssen-Cilag and received travel funds from Janssen-Cilag. Andreas Reif has received honoraria for lectures and/or advisory boards from Janssen/Johnson & Johnson, Boehringer Ingelheim, Compass, GH Research, SAGE/Biogen, LivaNova, Medice, Shire/Takeda, Newron, MSD, AbbVie, Cyclerion and Polpharma. Istvan Bitter received outside of this work in the past 5 years speaker or consultation fees from Gedeon Richter, Janssen/Janssen Cilag/Johnson@&Johnson, Lundbeck, Medichem Pharmaceuti-cals, Mitsubishi Tanabe Pharma Singapore Inc. by Unilab, Newron; research grant rom Gedeon Richter and royalties from Oxford University Press. Javier Santos serves/has served as a medical consultant for Noventure, Devintecpharma, Reckitt, Hipra, Ipsen, Viatris, Ordesa and Sequentia and discloses present or past recent scientific collaborations with Ordesa, Salvat, Norgine, Alfa-Sigma, Cosmo, Adare, Pileje, Danone, Farmasierra, Italfarmaco, Venter, Takeda, and Menarini. Beatriz Lobo has past/present scientific collaborations with Alfa-Sigma, Venter, Devintecpharma, GE HealthCare, Schwabe Farma Ibérica and is a consultant/Advisory Board member in Schwabe Farma Ibérica and VenterPharm. Amanda Rodriguez-Urrutia has received travel awards for taking part in annual psychiatric meetings from Lundbeck and has acted as a speaker at various training courses financed by Organon, Janssen-Cilag, Lundbeck and Farmasierra. She has eventually been a consultant at Danone. Carmen Alonso-Cotoner has past/present scientific collaborations with Alfa-Sigma, Venter, Norgine, Menarini, Noventure, Bioproject, Devintecpharma and is a consultant/Advisory Board member in Alfasigma and VenterPharma. Jose Alegre-Martin has received financial support from VITAE Health Innovation SL, Inmunotec Europa, Phama Nord Sapin and Laboratorios Viñas SA. Miriam Almirall has received consultancy/speaker's/advisory boards honoraria and/or travel support for attending meetings and/or participated in clinical trials sponsored by AbbVie, Johnson&Johnson, Lilly, Novartis and UCB. Jose Alegre-Martin has received financial support for attending meetings from VITAE Health Innovation, S.L., Laboratorios Viñas, S.A., Immunotec Europe, and Pharma Nord Spain. Ramon Sanmartin-Sentañes has participated in clinical trials sponsored by Vitae, Immunotec and Viñas, received consultancy/speaker's honoraria payments from Esteve, and travel support from Ferrer. Giovanni Barbara has consultancies, business interests or sources of honoraria payments from Aboca, AB Biotics, Agave, Alfa Sigma, AGPharma, Bayer, Biocodex, Boeringer, Bromatech, Cadigroup, Danone, Diadema, Falk Pharma, GE Healthcare, Giuliani, Mayoly, Malesci, Sanofy, Sofar, Yakult. Daisy Jonkers has received funding outside the submitted work from public-private partnership grants of top Knowledge institute (TKI) Agri&Food and Health Holland, by Danone Nutriticia Nederland B.V. and by Organic A2BV/Mothersfinest BV. All the remaining authors of this work declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

References

1. M. Camilleri, "Sex as a Biological Variable in Irritable Bowel Syndrome," *Neuro-Gastroenterology and Motility* 32, no. 7 (2020): e13802, https://doi.org/10.1111/nmo.13802.

2. F. Mearin, B. E. Lacy, L. Chang, et al., "Bowel Disorders," *Gastroenterology* 150, no. 6 (February 2016): 1393–1407.e5, https://doi.org/10.1053/j.gastro.2016.02.031.

3. R. Gendi and N. Jahan, "Pharmacological and Non-Pharmacological Treatments of Irritable Bowel Syndrome and Their Impact on the Quality of Life: A Literature Review," *Cureus* 12, no. 7 (2020), https://doi.org/10.7759/cureus.9324.

4. M. G. Shiha and I. Aziz, "Review Article: Physical and Psychological Comorbidities Associated With Irritable Bowel Syndrome," supplement,

26506414, 2026, 9, Downloaded from https://onlinelibrary.wiley.com/doi/10.1002/ueg2.70301 by Jesus Castro-Marrero - Spanish Cochrane National Provision (Ministerio de Sanidad) , Wiley Online Library on [03/11/2026]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

Alimentary Pharmacology & Therapeutics 54, no. S1 (2021): S12–S23, https://doi.org/10.1111/apt.16589.

5. N. Hanning, A. L. Edwinson, H. Ceuleers, et al., "Intestinal Barrier Dysfunction in Irritable Bowel Syndrome," *A systematic review* (2021): 1–31, https://doi.org/10.1177/1756284821993586.

6. E. Quigley, "The Gut-Brain Axis and the Microbiome: Clues to Pathophysiology and Opportunities for Novel Management Strategies in Irritable Bowel Syndrome (IBS)," *Journal of Clinical Medicine* 7, no. 1 (2018): 6, https://doi.org/10.3390/jcm7010006.

7. T. Vanuytsel, J. Tack, and R. Farre, "The Role of Intestinal Permeability in Gastrointestinal Disorders and Current Methods of Evaluation," *Frontiers in Nutrition* 8 (2021): 717925, https://doi.org/10.3389/fnut.2021.717925.

8. X. Li, E. M. Kan, J. Lu, et al., "Combat-Training Increases Intestinal Permeability, Immune Activation and Gastrointestinal Symptoms in Soldiers," *Aliment Pharmacol Ther* 37, no. 8 (2013): 799–809, https://doi.org/10.1111/apt.12269.

9. B. R. Stevens, R. Goel, K. Seungbum, et al., "Increased Human Intestinal Barrier Permeability Plasma Biomarkers Zonulin and FABP2 Correlated With Plasma LPS and Altered Gut Microbiome in Anxiety or Depression," *Gut* 67, no. 8 (2018): 1555–1557, https://doi.org/10.1136/gutjnl-2017-314759.

10. G. Morris, M. Berk, A. F. Carvalho, J. R. Caso, Y. Sanz, and M. Maes, "The Role of Microbiota and Intestinal Permeability in the Pathophysiology of Autoimmune and Neuroimmune Processes With an Emphasis on Inflammatory Bowel Disease Type 1 Diabetes and Chronic Fatigue Syndrome," *Current Pharmaceutical Design* 22, no. 40 (2016): 6058–6075, https://doi.org/10.2174/1381612822666160914182822.

11. A. Goebel, S. Buhner, R. Schedel, H. Lochs, and G. Sprotte, "Altered Intestinal Permeability in Patients With Primary Fibromyalgia and in Patients With Complex Regional Pain Syndrome," *Rheumatology* 47, no. 8 (2008): 1223–1227, https://doi.org/10.1093/rheumatology/ken140.

12. V. K. Gupta, S. Paul, and C. Dutta, "Geography, Ethnicity or Subsistence-Specific Variations in Human Microbiome Composition and Diversity," *Front Microbiol* 8 (2017): 1162, https://doi.org/10.3389/fmicb.2017.01162.

13. I. Midenfjord, M. Khadija, E. Sundelin, et al., "Increased Disease Burden in Irritable Bowel Syndrome With Comorbid Conditions and Psychiatric Diagnoses in a Multinational European Cohort: Results From the Discoverie Project," *United European Gastroenterology Journal* 14, no. 1 (2026): e70157, https://doi.org/10.1002/ueg2.70157.

14. E. Houben, T. Vanuytsel, J. Tack, and K. Verbeke, "Validation of a GC-MS and HPLC-ELSD Method to Study Intestinal Permeability," *Trends in Chromatography* 8 (2013): 83–96, https://www.researchgate.net/publication/265858622.

15. T. R. Dmytriv, K. B. Storey, and V. I. Lushchak, "Intestinal Barrier Permeability: The Influence of Gut Microbiota, Nutrition, and Exercise," *Front Physiol* 15 (2024): 1380713, https://doi.org/10.3389/fphys.2024.1380713.

16. A. Y. Del Valle-Pinero, H. E. Van Deventer, N. H. Fourie, et al., "Gastrointestinal Permeability in Patients With Irritable Bowel Syndrome Assessed Using a Four Probe Permeability Solution," *Clinica Chimica Acta* 418 (2013): 97–101, https://doi.org/10.1016/j.cca.2012.12.032.

17. R. C. Spiller, D. Jenkins, J. P. Thornley, et al., "Increased Rectal Mucosal Enteroendocrine Cells, T Lymphocytes, and Increased Gut Permeability Following Acute Campylobacter Enteritis and in Post-Dysenteric Irritable Bowel Syndrome," *Gut* 47, no. 6 (2000): 804–811, https://doi.org/10.1136/gut.47.6.804.

18. J. K. Marshall, M. Thabane, A. X. Garg, W. Clark, J. Meddings, and S. M. Collins, "Intestinal Permeability in Patients With Irritable Bowel Syndrome After a Waterborne Outbreak of Acute Gastroenteritis in Walkerton, Ontario," *Alimentary Pharmacology & Therapeutics* 20, no. 11–12 (2004): 1317–1322, https://doi.org/10.1111/j.1365-2036.2004.02284.x.

19. J. H. Park, D. Il Park, H. J. Kim, et al., "The Relationship Between Small-Intestinal Bacterial Overgrowth and Intestinal Permeability in Patients With Irritable Bowel Syndrome," *Gut Liver* 3, no. 3 (2009): 174–179, https://doi.org/10.5009/gnl.2009.3.3.174.

20. M. R. Barbaro, C. Cremon, G. Marasco, et al., "Molecular Mechanisms Underlying Loss of Vascular and Epithelial Integrity in Irritable Bowel Syndrome," *Gastroenterology* 167, no. 6 (2024): 1152–1166, https://doi.org/10.1053/j.gastro.2024.07.004.

21. F. Russo, G. Chimienti, G. Riezzo, et al., "Adipose Tissue-Derived Biomarkers of Intestinal Barrier Functions for the Characterization of Diarrhoea-Predominant IBS," *Dis Markers* 2018 (2018): 1827937, https://doi.org/10.1155/2018/1827937.

22. J. Zeng, Y.-Q. Li, X.-L. Zuo, Y.-B. Zhen, J. Yang, and C.-H. Liu, "Clinical Trial: Effect of Active Lactic Acid Bacteria on Mucosal Barrier Function in Patients With Diarrhoea-Predominant Irritable Bowel Syndrome," *Alimentary Pharmacology & Therapeutics* 28, no. 8 (2008): 994–1002, https://doi.org/10.1111/j.1365-2036.2008.03818.x.

23. M. I. Vazquez-Roque, M. Camilleri, T. Smyrk, et al., "Association of HLA-DQ Gene With Bowel Transit, Barrier Function, and Inflammation in Irritable Bowel Syndrome With Diarrhea," *American Journal of Physiology-Gastrointestinal and Liver Physiology* 303, no. 11 (2012): G1262–G1269, https://doi.org/10.1152/ajpgi.00294.2012.

24. L. Li, L. Xiong, J. Yao, et al., "Increased Small Intestinal Permeability and RNA Expression Profiles of Mucosa From Terminal Ileum in Patients With Diarrhoea-Predominant Irritable Bowel Syndrome," *Italian Society of Gastroenterology and the Italian Association for the Study of the Liver* 48, no. 8 (2016): 880–887, https://doi.org/10.1016/j.dld.2016.05.002.

25. F. Mattioli, C. Fucile, V. Marini, et al., "Assessment of Intestinal Permeability Using Sugar Probes: Influence of Urinary Volume," *Clinical Laboratory* 57, no. 11–12 (2011): 909–918, https://pubmed.ncbi.nlm.nih.gov/22239021/.

26. R. W. Lobley, P. C. Burrows, R. Warwick, D. J. Dawson, and R. Holmes, "Simultaneous Assessment of Intestinal Permeability and Lactose Tolerance With Orally Administered Raffinose, Lactose and L-arabinose," *Clinical Science* 79, no. 2 (1990): 175–183, https://doi.org/10.1042/cs0790175.

27. M. Linsalata, G. Riezzo, B. D'Attoma, C. Clemente, A. Orlando, and F. Russo, "Noninvasive Biomarkers of Gut Barrier Function Identify Two Subtypes of Patients Suffering From Diarrhoea predominant-IBS: A Case-Control Study," *BMC Gastroenterol* 18, no. 1 (2018): 167, https://doi.org/10.1186/s12876-018-0888-6.

28. A. P. M. Kerckhoffs, L. M. A. Akkermans, M. B. M. de Smet, et al., "Intestinal Permeability in Irritable Bowel Syndrome Patients: Effects of NSAIDs," *Digestive Diseases and Sciences* 55, no. 3 (2010): 716–723, https://doi.org/10.1007/s10620-009-0765-9.

29. N. Valentin, M. Camilleri, P. Carlson, et al., "Potential Mechanisms of Effects of Serum-Derived Bovine Immunoglobulin/Protein Isolate Therapy in Patients With Diarrhea-Predominant Irritable Bowel Syndrome," *Physiological Reports* 5, no. 5 (2017): e13170, https://doi.org/10.14814/phy2.13170.

30. A. S. Rao, M. Camilleri, D. J. Eckert, et al., "Urine Sugars for in Vivo Gut Permeability: Validation and Comparisons in Irritable Bowel syndrome-Diarrhea and Controls," *American Journal of Physiology-Gastrointestinal and Liver Physiology* 301, no. 5 (2011): G919–G928, https://doi.org/10.1152/ajpgi.00168.2011.

31. S. A. Peters, S. Edogawa, W. J. Sundt, et al., "Constipation-Predominant Irritable Bowel Syndrome Females Have Normal Colonic

26506414, 2026, 9, Downloaded from https://onlinelibrary.wiley.com/doi/10.1002/ueg2.70301 by Jesus Castro-Marrero - Spanish Cochrane National Provision (Ministerio de Sanidad) , Wiley Online Library on [03/11/2026]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

Barrier and Secretory Function," *American Journal of Gastroenterology* 112, no. 6 (2017): 913–923, https://doi.org/10.1038/ajg.2017.48.

32. Z. Mujagic, S. Ludidi, D. Keszthelyi, et al., "Small Intestinal Permeability is Increased in Diarrhoea Predominant IBS, While Alterations in Gastroduodenal Permeability in all IBS Subtypes are Largely Attributable to Confounders," *Alimentary Pharmacology & Therapeutics* 40, no. 3 (2014): 288–297, https://doi.org/10.1111/apt.12829.

33. M. Shaikh, K. Rajan, C. B. Forsyth, R. M. Voigt, and A. Keshavarzian, "Simultaneous Gas-Chromatographic Urinary Measurement of Sugar Probes to Assess Intestinal Permeability: Use of Time Course Analysis to Optimize its Use to Assess Regional Gut Permeability," *Clinica Chimica Acta* 442 (2015): 24–32, https://doi.org/10.1016/j.cca.2014.12.040.

34. Y. Magnus, J. BouSaba, W. Sannaa, S. McKinzie, I. Busciglio, and M. Camilleri, "Bile Acid Diarrhea Is Associated With Increased Intestinal Permeability Compared With Irritable Bowel Syndrome-Diarrhea," *Gastroenterology* 162, no. 4 (2022): 1343–1345.e1, https://doi.org/10.1053/j.gastro.2021.12.243.

35. C. Bonnafous, B. Scatton, and L. Buéno, "Benzodiazepine-Induced Intestinal Motor Disturbances in Rats: Mediation by Omega 2 (BZ2) Sites on Capsaicin-Sensitive Afferent Neurones," *British Journal of Pharmacology* 113, no. 1 (1994): 268–274, https://doi.org/10.1111/j.1476-5381.1994.tb16204.x.

36. R. Fogel, G. Michelson, T. Senler, D. Marshall, T. Brown, and T. Gaginella, "Central Administration of Benzodiazepines Alters Water Absorption by the Rat Ileum in Vivo," *Gastroenterology* 93, no. 2 (1987): 330–334, https://doi.org/10.1016/0016-5085(87)91023-7.

37. C. Graziani, C. Talocco, R. De Sire, et al., "Intestinal Permeability in Physiological and Pathological Conditions: Major Determinants and Assessment Modalities," *European Review for Medical and Pharmacological Sciences* 23, no. 2 (2019): 795–810, https://doi.org/10.26355/eurrev_201901_16894.

38. D. D. Nascimento, A. C. Mota, M. C. Carvalho, et al., "Can Diet Alter the Intestinal Barrier Permeability in Healthy People? A Systematic Review," *Nutrients* 16, no. 12 (2024): 1871, https://doi.org/10.3390/nu16121871.

39. I. S. Menzies, M. J. Zuckerman, W. S. Nukajam, et al., "Geography of Intestinal Permeability and Absorption," *Gut* 44, no. 4 (1999): 483–489, https://doi.org/10.1136/gut.44.4.483.

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