Non-Celiac Gluten Sensitivity and Irritable Bowel Disease: Looking for the Culprits

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ABSTRACT

During the last 30 years, a gluten-free diet has been classified among the most popular diets mainly due to the ambiguous notion that gluten avoidance promotes health. Gluten intolerance has been implicated in non-celiac gluten sensitivity (NCGS) and irritable bowel syndrome (IBS), 2 disorders with overlapping symptoms and increasing trend. Together with gluten, other wheat components; fermentable oligo-, di-, monosaccharides, and polyols (FODMAPs); and amylase trypsin inhibitors (ATIs), are implicated in the pathogenesis of both disorders. Gut microbiota alterations in IBS and NCGS have been described, while microbiota manipulations have been shown to be promising in some IBS cases. This literature review summarizes our current knowledge on the impact of wheat ingredients (gluten, FODMAPs, and ATIs) in IBS and NCGS. In both disorders, FODMAPs and ATIs trigger gut dysbiosis, suggesting that gluten may not be the culprit, and microbiota manipulations can be applied in diagnostic and intervention approaches.

Keywords: non-celiac gluten sensitivity, irritable bowel syndrome, gluten-free diet, microbiota, gluten, FODMAP, ATIs

Introduction

During the last 30 years, the consumption of gluten-free products (GFPs) has become increasingly common, resulting in often being included in the list offad diets with a great impact on the sales market of the Western world (1). The therapeutic role of a gluten-free diet (GFD) is undisputed in wheat allergy (WA) and celiac disease (CD), in which the reactions to gluten are mediated by the adaptive immune system (2). Nevertheless, the prevalence of these diseases is low, accounting for only 1% for CD and 0.1% for WA, and cannot justify the exponential rise in popularity of GFPs (3).

To date, 2 syndromes with unclear pathogenesis, diagnostic criteria, and epidemiology have been included under the umbrella of gluten-related disorders: non-celiac gluten sensitivity (NCGS) and irritable bowel syndrome (IBS), both having gluten among the causative agents of induction of their symptoms (4–9). Self-reporting of the implicated foods into symptom development is an integral part of diagnosis of both disorders, after the exclusion of CD and WA (4, 10), as a genetic predisposition has not yet been identified, nor damage to small-intestine villi or an antigen-triggering allergic reaction (11, 12). Nevertheless, some groups have reported a greater prevalence of the HLA-DQ2/DQ8 genes known to predispose to CD in NCGS patients than in the general population (12). The epidemiology of IBS and NCGS varies greatly in the Western world (10–15% and 0.6–6%, respectively), with estimations being questionable, as many patients start a GFD without any relevant clinical examination and diagnosis (6–9). The similar presentation patterns recognized so far for IBS and NCGS have led to proposing the term “IBS-like disorders,” which also comprises NCGS (13). In general, the onset of symptoms starts after wheat consumption in both NCGS and IBS patients, leading to the assumption that gluten is the culprit. However, 2 other groups of wheat components have been implicated, as follows:

1. The fermentable oligo-, di-, monosaccharides, and polyols (FODMAPs); short-chain fructose oligosaccharides (fructans); galacto-oligosaccharides (GOS; stachyose, raffinose); disaccharides (lactose); monosaccharides (fructose); and polyols (sugar alcohols), which are poorly absorbed in the human small intestine and are partially fermented in the large intestine by gut bacteria (14).

2. The wheat amylase trypsin inhibitors (ATIs), a family of up to 17 proteins with molecular weights of ∼15 kDa and a variable
primary but conserved secondary structure characterized by 5 intrachain disulfide bonds and α-helices and mostly form di- and tetramers.

Both these wheat ingredients induce inflammatory processes and alterations to gut microbiota. Intestinal dysbiosis in NCGS and IBS (15, 16) is known to affect various metabolic and inflammatory processes. The gut microbiota plays a crucial role in intestinal motility regulation and neuroimmune signaling (5). The so-called hidden organ of our body presents contiguos among people with similar genetic background, ethnicity, age, and sex, but remains malleable to noninvasice nutritional interventions (17–19). As such, microbiota manipulations could be of enormous therapeutic potential in inflammatory gastrointestinal diseases, like NCGS and IBS.

This literature review aims to investigate the current knowledge related to whether gluten is the actual culprit for NCGS and IBS disorders or the scapegoat for the benefit of the sales market of GFPs, with other wheat ingredients being responsible, in addition to the putative pathways related to microbiota dysbiosis in those who suffer from these disorders.

**Methods**

A literature search was performed of relevant published original research and reviews that were pertinent to the aim of this review. This involved searching databases of peer-reviewed published literature (Cochrane Library, EMBASE and CINAHL, MEDLINE, and Google Scholar) of both human and animal studies published from 2000 to 2020 on the involvement of gluten, ATIs, FODMAPs, and gut microbiota in IBS and NCGS.

A comparative outline of research findings used is presented in Table 1. These in vivo and in vitro studies have published data regarding the distinct and overlapping pathophysiology of NCGS and IBS, which is summarized in Table 2. Although the entities remain obscure, the knowledge gained and further discussed can initiate future research for more accurate diagnosis and future therapeutic potentials and stop any unnecessary GFP consumption.

**IBS presentation and pathogenesis**

In IBS, obvious abnormalities or intestinal mucosal damage are usually absent. According to the Rome IV classification (20), IBS patients suffer from abdominal pain on average at least once a week for >6 mo before the diagnosis (5, 21). Clinical signs vary widely and include alterations in bowel habits, abdominal pain or distension, bloating or flatulence, absence of constitutional symptoms, and absence of alarming features such as weight loss, anorexia, gastrointestinal bleeding, and fever (5). Along with abdominal pain, changes in stool consistency and frequency or pain at defecation, flatulence, and bloating might occur (22). Based on the predominant bowel habit, patients are classified into 4 types: IBS with predominant constipation (IBS-C), IBS with predominant diarrhea (IBS-D), mixed IBS (IBS-M), and unsubtype IBS.

The outcome of IBS can be influenced by psychosocial stressors, whereas socio-relational status, work ability, and productivity, as well as everyday life activities, can be hindered (23). Emotional and personality patterns can contribute to IBS clinical features, symptomatology, and immune response (21) and can consequently affect treatment (Table 2).

The complexity of IBS pathogenesis is related to the multifactorial impact of symptom exacerbation: diet, sex, antibiotics, regulation of the gut–brain axis, stressful life changes, genetic factors, gut barrier permeability, defective immune responses, gut microflora alterations, and psychosocial factors are all implicated in symptom worsening (24). Notably, IBS is a disease of a gut–brain axis dysregulation, involving altered signaling between immune cells and neurotransmitters. Within the intestinal mucosa, the signaling between immune cells and nerve fibers of the enteric nervous system, such as mast cells and nerves, plays a key role in IBS. The symptom intensity is associated with the activation of immune and neuroendocrine cascades that correlate with changes in the gut microflora, intestinal permeability, and in dysfunctional sensorimotor outputs in the intestine (6).

**Possible triggers in IBS**

Diet along with stress and menstruation are the most common precipitating or exacerbating factors in IBS (25). Most IBS patients attribute their symptoms to food, with a long list of putative culprits (26). IBS patients’ complaints often increase after the consumption of high-carbohydrate meals, resulting in the decision to remove wheat from their diet. Despite the various putative triggers among wheat components, gluten was considered by patients to be the culprit for recurrent gastrointestinal symptoms (23).

A similar presentation of increased colonic motility in IBS-D patients was observed in gliadin-sensitized HLA-DQ8 mice, where gluten stimulated a significant increase in the production of acetylcholine in the myenteric plexus and in high-amplitude propagating contractions, causing an increased colonic motility and a mild inflammation. Gluten removal from the diet eliminated these motor changes, indicating that gluten triggered the gut motor dysfunction (27). Similarly, in a study by Vazquez-Roque et al. (28), the intake of gluten-containing food (mean: 3.10 ± 0.46) in IBS-D patients who were carriers of the HLA-DQ2 and/or -DQ8 haplotypes increased the permeability of the small intestine, which was then accompanied by mild inflammation (6).

Therefore, gluten can be a trigger for IBS patients with CD genetic predisposition. In another double-blind placebo-controlled study [double-blind placebo-controlled food challenge (DBPCFC)] in IBS-D patients without genetic predisposition, a GFD led to a significant improvement in their symptoms such as pain, bloating, stool consistency, and tiredness (29). In addition, Fritscher-Ravens and his group (30) used confocal laser endomicroscopy and reported intestinal leakage and epithelial breaks in half of IBS patients challenged with wheat, while all patients benefited from the GFD in the long term.
### TABLE 1 Outline of research findings from the main studies on the role of gluten, ATIs, FODMAPs, and gut microbiota in IBS and NCGS

| Study (reference)            | Subjects                        | Number of participants | Methods                                      | Research findings                                                                                                                                 |
|------------------------------|---------------------------------|------------------------|----------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------|
| Human studies                |                                 |                        |                                              |                                                                                                                                                  |
| Biesiekierski et al., 2011   | CD genetically predisposed       | 34                     | Double-blind, randomized, controlled study  | A GFD in IBS-D patients significantly improved their IBS-like symptoms                                                                            |
| Fritscher-Ravens et al., 2014| IBS patients                    | 36                     | Confocal laser endomicroscopy                | Half of patients presented intestinal leakage and epithelial breaks after wheat challenge                                                         |
| Pedersen et al., 2017        | IBS patients                    | 89                     | Randomized controlled trial                 | FODMAP removal from the diet significantly decreased abdominal pain and bloating                                                                  |
| Bennet et al., 2018          | IBS patients                    | 67                     | Randomized controlled trial                 | Low-FODMAP diet improved IBS symptoms and correlated with reduced *Bifidobacterium* and *Actinobacteria* fecal bacteria and with lactose consumption |
| Böhn et al., 2015            | IBS patients                    | 75                     | Multicenter, parallel, single-blind study    | Low-FODMAP diet improved IBS symptoms                                                                                                             |
| Frieling et al., 2019        | IBS patients                    | 93                     | Prospective study                           | Low-FODMAP diet improved IBS symptoms but patients lost weight and received insufficient nutrients                                               |
| Staudacher et al., 2017      | IBS patients                    | 104                    | Randomized, controlled study                | Low-FODMAP diet improved IBS symptoms and co-administration with multistrain probiotic increased *Bifidobacterium* fecal bacteria                  |
| Hustoft et al., 2017         | IBS patients                    | 20                     | Double-blind, randomized, controlled study  | Low-FODMAP diet improved IBS symptoms and decreased serum IL-6, IL-8, fecal *Actinobacteria*, *Bifidobacterium*, and *Faecalibacterium*, SCFAs, and n-butryic acid |
| O’Keeffe et al., 2018        | IBS patients                    | 103                    | Long-term prospective study                 | Low-FODMAP education can be nutritionally adequate for 18 mo                                                                                     |
| Klem et al., 2017            | IBS patients                    | 45 studies             | Meta-analysis from 1994                     | IBS onset is due to bacterial, viral, or parasitic infections in the microbiota                                                                    |
| Kerckhoffs et al., 2009      | IBS patients                    | 41                     | FISH and PCR analysis of fecal and duodenal brush samples for microbiota composition | Decreased *Bifidobacteria* levels in IBS                                                                                                          |
| Rajilić-Stojanović et al., 2011| IBS patients                 | 62                     | Phylogenic microarray and PCR analysis of microbiota composition | Decreased *Bifidobacterium*, *Faecalibacterium*, and *Bacteroidetes* and increased ratio of *Firmicutes* to *Bacteriodes*                      |
| Parkes et al., 2012          | IBS patients                    | 47                     | Hybridization of rectal biopsies for microbial quantification | Increased Bacteroides and Clostridia and reduced *Bifidobacteria* in mucosa-microbiota in IBS                                                   |
| Tana et al., 2010            | IBS patients                    | 26                     | Liquid chromatography and PCR analysis on fecal samples and abdominal X-ray films for gas quantification | Increased levels of *Veillonella* and *Lactobacillus*, acetic acid, propionic acid, and total organic acids |

(Continued)
| Study (reference)               | Subjects         | Number of participants | Methods                                                                 | Research findings                                                                                                                                 |
|-------------------------------|------------------|------------------------|--------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------|
| Rigsbee et al., 2012 (42)     | IBS-D children   | 20                     | Phylogenetic microbiota array, FISH, PCR analysis on fecal samples       | Different microbiota taxonomy in IBS with increased Clostridia levels                                                                                 |
| Labus et al., 2017 (43)       | IBS patients     | 29                     | 16S rRNA sequencing on fecal samples and structural brain images         | Microbial composition correlated with structural measures of brain regions                                                                           |
| Vandeputte et al., 2016 (44)  | IBS patients     | 9                      | 16S rRNA sequencing on fecal samples and lactulose breath testing       | Increased levels of M. smithii methanogen in IBS-C and correlated with breath methane                                                              |
| Tap et al., 2017 (45)         | IBS patients     | 110                    | Assessment of 16S rRNA sequencing on fecal samples and mucosal samples for microbiota, exhaled H₂ and CH₄, psychological and gastrointestinal symptoms, and fecal methanogens | IBS symptom severity associated with decreased microbial richness, exhaled CH₄, methanogens, and enterotypes with Clostridiales or Prevotella species |
| Silk et al., 2009 (46)        | IBS patients     | 44                     | Randomized, parallel, crossover, controlled clinical trial              | Prebiotics increased fecal bifidobacteria                                                                                                             |
| Hunter et al., 1999 (47)      | IBS patients     | 21                     | Double-blind crossover study                                           | Oligofructose prebiotics did not improve IBS symptoms                                                                                                 |
| Olesen et al., 2000 (48)      | IBS patients     | 98                     | Multicenter, prospective, randomized, double-blind, placebo-controlled parallel study | Oligofructose prebiotics did not affect IBS symptoms                                                                                                  |
| Paineau et al., 2008 (49)     | IBS patients     | 105                    | Comparative, randomized, double-blind study                            | Oligofructose prebiotics improved significantly the IBS symptoms                                                                                     |
| Didari et al., 2015 (50)      | IBS patients     | 24 studies             | Meta-analysis on the efficacy of probiotics in IBS                      | Probiotics improve IBS symptoms                                                                                                                     |
| Min et al., 2012 (51)         | IBS patients     | 130                    | Randomized controlled study                                            | Yogurt with acacia fiber and B. lactis has significant therapeutic effects in IBS                                                                |
| Tsuchiya et al., 2004 (52)    | IBS patients     | 68                     | Randomized, blind control study                                        | Administration of symbiotic novel symbiotic Microflora F (SCM-III) increased Lactobacillus, Eubacteria, and Bifidobacteria and improved IBS symptoms |
| Chey et al., 2015 (53)        | IBS patients     | 1074                   | Phase 3, randomized, double-blind, controlled study                    | Repeated rifaximin treatment was efficacious in IBS-D patients with relapsing symptoms                                                              |
| Dieterich et al., 2019 (22)   | NCGS patients    | 19                     | Clinical trial                                                         | Low-FODMAP diet improved clinical and psychological NCGS symptoms. NCGS patients present a microbiota dysbalance                                        |
| Zanini et al., 2015 (54)      | NCGS patients    | 35                     | Randomized, double-blind, clinical study                               | Symptom recurrence occurred in one-third of the patients after gluten challenge                                                                    |
| Dale et al., 2018 (55)        | NCGS patients    | 20                     | A randomized, double-blind controlled study                           | NCGS symptoms did not re-appear after gluten challenge in most patients                                                                             |

(Continued)
| Study (reference) | Subjects | Number of participants | Methods | Research findings |
|------------------|----------|------------------------|---------|-------------------|
| Skodje et al., 2018 (56) | NCGS patients | 59 | Randomized, double-blind crossover study | Fructans rather than gluten-induced NCGS symptoms in 24 patients |
| Molina-Infante et al., 2017 (57) | NCGS patients | 231 | Data analysis from 10 double-blind, controlled study | Heterogeneity and methodology flaws among studies of gluten challenge; the role of gluten in NCGS is questionable |
| Tovoli et al., 2017 (58) | NCGS patients | 44 | Questionnaire-based study | About 70% of patients continued to have NCGS symptoms after 1 y of a GFD |
| Garcia-Mazcorro et al., 2018 (59) | NCGS patients | 12 | 16S rRNA sequencing on fecal and duodenal samples | Significant changes in duodenal Pseudomonas levels after 4 wk of a GFD |
| Animal studies | | | | |
| Verdu et al., 2007 (27) | CD genetically predisposed subjects | 15 | Gliadin-sensitized HLA-DQ8 mouse model | Gluten induced IBS-D like symptoms (increased acetylcholine production and colonic motility) that improved upon gluten removal from the diet |
| Junker et al., 2012 (60) | TLR-4–deficient subjects | 12 | Mouse model challenged with gliadin and ATIs | Mice with defective TLR-4 or TLR-4 pathways are protected from the intestinal and immune responses when they are challenged with ATIs |
| Zevallos et al., 2017 (61) | TLR-4–responsive mice | 38 | TLR-4–sensitized mouse and human cell line model | Gluten-containing cereals have the highest concentrations of ATIs that activate TLR-4 |
| Bellinghausen et al., 2018 (62) | Humanized mice | 10 | Mice were engrafted with the PBMCs from allergic donors and were challenged | ATIs are strong allergen activators |

1 ATI, α-amylase/trypsin inhibitor; CD, celiac disease; FISH, fluorescent in situ hybridization; FODMAPs, fermentable oligo-, di-, mono-saccharide, and polyols; GFD, gluten-free diet; IBS, irritable bowel syndrome; IBS-C, IBS with predominant constipation; IBS-D, IBS with predominant diarrhea; NCGS, non-celiac gluten sensitivity; PBMC, peripheral blood mononuclear cell; PCR, polymerase chain reaction; rRNA, ribosomal RNA; TLR, Toll-like receptor.
nevertheless, in ~70% of IBS patients bloating and pain are induced by FODMAPs (13, 63, 64). As FODMAPs are not absorbed properly in the small intestine, they retain water and are rapidly fermented by the bacteria in the colon, leading to gas and SCFA production, accompanied by luminal distension and abnormal motility (6). In turn, this aggregation of fluids and gases results in visceral hypersensitivity, gut microflora alterations, and changes in intestinal hormones and neurotransmitters that characterize IBS (65). There are ~15 different types of gastrointestinal endocrine cells releasing different types of hormones depending on the types of sensed nutrients (66). The interactions between FODMAPs and gastrointestinal endocrine cells induce changes in cell densities. Restoring the densities of the gastrointestinal endocrine cells results in IBS symptom improvement. Therefore, the removal of FODMAPs from the diet of the IBS patients is anticipated to result in a remarkable improvement in gastrointestinal and extra-intestinal symptoms in 68–86% of individuals (67, 68, 31, 32). However, it should be stressed that a low-FODMAP diet is very restrictive, resulting occasionally in significant weight loss, but in the majority of the cases in significant nutrient deficiencies (33, 34, 36) and gut dysbiosis due to limited intake of dietary prebiotics; a respectable number of studies reported that a low-FODMAP diet significantly decreased the levels of fecal Bifidobacterium, Faecalibacterium, and Actinobacteria populations in the microflora, leading to reduced SCFAs, n-butyric acid, and proinflammatory cytokines IL-6 and IL-8 (15, 32, 35, 43).

Among other triggering agents, wheat ATIs have been shown to elicit inflammatory and immune responses as they have been identified to be strong inducers of innate immune responses (36). ATIs, which represent 2–4% of wheat proteins, are resistant to digestion by the gastric proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases. In a standard Western diet, the daily mean consumption is 10–15 g of gluten/d, containing the co-fractionated with gliadin tritic proteases.
ral, or parasitic infections in the microbiota (37). Furthermore, data reveal a predominance of bacterial phyla connected with dysbiosis, such as Enterobacteriaceae, with an increase in proinflammatory cytokines, decreased concentrations of the tolerogenic dendritic cells, and reduced levels of Lactobacillus and Bifidobacterium (6, 70, 38–40). The reduced levels of Bifidobacterium, Clostridiales, Ruminococaceae, and Erysipelotrichaceae, which produce SCFAs (6); the reduced levels of Faecalibacterium (33); and the impaired Firmicutes-to-Bacteroidetes ratios, together with an abundance of Lactobacillus species (43, 41, 42) and an increase in Veillonella and Ruminococcus, indicate a perturbation of the bacterial colonization in the gastrointestinal tract (6).

Patients with IBS-D have decreased methane production, whereas it is increased in IBS-C (8, 71). Methane in the gut microbiota is produced exclusively from methanobacteria. It slows down the intestinal transit and has anti-inflammatory effects (45). Increased methane production in IBS-C patients, however, correlates with microbial overgrowth of the methanobacteria Clostridiales or Prevotella species, which further reduces food transition, by an average of 59% in animal models (45, 44, 72).

Potential therapeutic pathways of IBS

Elimination diet.

Due to the lack of reliable biomarkers, the IBS therapeutic milieu is mainly based on subjective estimations of each unique patient of their symptoms' exacerbating agents, among which there is also a long list of implicated foods. Food avoidance of all irritating agents is unavoidable, at least at the initiation of the diet therapy or during symptom eruption. The "IBS Food Pyramid" (73) is an encouraging educational tool for IBS patients to aid in following a healthy diet pattern over the long term. Regular physical activity, adequate fluid intake, and regular eating habits are encouraged. Foods from all food groups can be consumed with appropriate personalized recommendations regarding consumption of gluten-free cereals, low FODMAPs, and lactose-free products. In the case of fat, the anti-inflammatory PUFAs are thought to be beneficial, but further research is needed to confirm this. The avoidance of spicy foods, alcohol, and highly processed foods is recommended. As patients following a low-FODMAP diet are at high risk of developing deficiencies in vitamins B and D, zinc, calcium, iron, folate, and natural antioxidants (56), the strict food avoidance should be reduced and, together with this, the nutrient deficiencies resulting from low fiber intake and extreme dietary choices, like a GFD, could be limited.

Microbiota manipulations.

As IBS symptoms are strongly correlated with microbiota synthesis and methane production, manipulations of the gut microbiota have been investigated for more than a decade, but their beneficial potential is still not confirmed. Probiotic supplements are recommended via the IBS Food Pyramid, to reduce exacerbating symptoms, for a 4-wk period in a dose as recommended by the manufacturer, for the individual patient to evaluate the beneficial effect (73).

Intervention trials in IBS aiming to alter microflora and to improve IBS symptoms are presented in detail in Table 3. In short, they were based on the following:

1. Probiotics supplementation, which consist of live bacteria aiming to shift gut microbiota towards abundance of the beneficial bacteria. It is noteworthy that Lactobacillus and Bifidobacterium species lead to a decrease in pathogens by hampering the intestinal mucosal adhesion and they can reduce mild inflammation by regulating the TLRs, the intestinal permeability, visceral hypersensitivity, intestinal motility, and even neurotransmitter release (50). Data meta-analysis reports suggest that probiotics largely improve IBS symptoms, especially in low doses and short-term administration, and restore the intestinal mucosal barrier, particularly in women (50, 74, 75).

2. Prebiotic supplements confer a health benefit by stimulating the growth of probiotic bacteria, mainly Bifidobacteria and Lactobacilli (76). Even during early life they are added to infant formulas aiming to resemble breast milk, as they have been found to benefit the gut microbiota of breastfed infants. Prebiotics are predominantly carbohydrate-based, but other substrates, such as polyphenols and PUFAs, might also exert prebiotic effects. In the case of IBS, a prebiotics mixture of fructo-oligosaccharides (FOS) and trans-GOS (TGOS) has been suggested as capable of improving symptoms, whereas TGOS are correlated with abundance of Bifidobacterium in feces (76, 77, 46, 78, 79, 49). However, the lower prebiotic dose provided the optimum outcome (6), whereas 2 other controlled trials showed no improvement after a similar prebiotic administration (47, 48).

3. Synbiotics contain selected bacteria species in combination with prebiotic components that favor beneficial bacterial growth. Prebiotics that are mainly used include disaccharides, such as lactulose; oligosaccharides, such as FOS, Tgos, and GOS; and polysaccharides, such as fructan, inulin, and cellulose (77). A symbiotic preparation of yogurt supplemented with acacia fiber and Bifidobacterium lactis, a combination of L. acidophilus, L. helveticus, and Bifidobacterium in a vitamin-supplemented medium, Bacillus coagulans, and FOS improved IBS symptoms (6, 51, 52).

Drug therapy.

Nonabsorbable antibiotics are known to improve symptoms in IBS, probably due to their ability to lower the concentrations and compositions of intestinal bacteria and alter the intestinal permeability and fecal microbiome (5). Neomycin has been found to induce up to 50% improvement in all IBS symptoms, but also led to bacterial resistance (5). Rifaximin, a broad-spectrum nonsystemic antibiotic, has proven to be effective by managing the bacterial overgrowth in the small intestine in IBS patients and to increase bacterial diversity and the Firmicutes-to-Bacteroidetes ratio (80, 53, 81). Short courses of therapy (2–4 wk) with rifaximin in IBS treatment is recommended; however, bacterial antibiotic resistance should be monitored, especially in patients who require repeat courses of rifaximin and other antibiotics including clarithromycin and metronidazole (80).

NCGS presentation and pathogenesis.

NCGS is characterized by both intestinal, such as alterations in bowel habits, abdominal pain, bloating, and flatulence, and extra-intestinal symptoms, such as body aches, depression, severe fatigue, anxiety, skin manifestations, and oral ulceration (23). In addition, the contribution...
| Study (reference)          | Study design                                                                 | Subjects                                                                 | Probiotic's synthesis             | Substrate                                                                 | Effectiveness                                                                                               | Side effects                              |
|---------------------------|------------------------------------------------------------------------------|--------------------------------------------------------------------------|-----------------------------------|---------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------|-------------------------------------------|
| Koebnick et al., 2003 (82) | Double-blind placebo-controlled randomized trial                             | 35 active; 35 placebo; 18–65 y; males and females                         | Lactobacillus casei Shirota       | Beverage (65 mL/d)                                                        | Significant improvement in constipation and stool consistency                                               | No side effects                           |
| Niv et al., 2005 (83)     | Double-blind placebo-controlled randomized trial                             | 54 included/39 completed; 19–70 y; males and females                      | Lactobacillus reuteri ATCC 55,730 | Capsule (1 × 10^8 CFU)                                                   | No symptom improvement                                                                                       | Several side effects                      |
| Whorwell et al., 2006 (84)| Double-blind placebo-controlled randomized trial; multicenter dose ranging   | 362 females, 270 active group/92 placebo group; 18–55 y; males and females | Bifidobacterium infantis 35,624   | Lyophilized capsule (1 × 10^6, 1 × 10^8, 1 × 10^10 CFU/mL)              | B. infantis (1 × 10^8 CFU) improved the symptoms of IBS, such as abdominal pain, bloating, bowel dysfunction, and incomplete emptying | 17 (~5%) of the 362 patients reported adverse effects and withdrew from the study |
| Sinn et al., 2008 (85)    | Double-blind placebo-controlled randomized trial                             | 20 active group/20 placebo group; 18–65 y                                 | Lactobacillus acidophilus-SDC 2012, 2013 | Lyophilized capsules (2 × 10^7 CFU/mL)                                    | Improvement in abdominal pain and discomfort                                                              | No side effects                           |
| Guglielmetti et al., 2011 (86) | Double-blind placebo-controlled randomized trial                          | 60 active group/62 placebo group; 18–68 y; males and females             | Bifidobacterium bifidum MIMBb75   | Uncovered capsule (1 × 10^7 CFU)                                          | GI symptom relief: pain, discomfort, dilation, bloating, digestive disorders, QoL improvements             | 23/60 patients reported abdominal distension, abdominal pain, diarrhea, nausea, constipation                  |
| Ducrotté et al., 2012 (87) | Double-blind placebo-controlled randomized trial                             | 214; 18–70 y; males and females                                          | Lactobacillus plantarum 299v (DSM 9843) | Lyophilized capsule (1 × 10^10 CFU)                                       | Pain reduction, daily frequency, and bloating in patients                                                 | No side effects; only 1 patient mentioned transient vertigo                                           |
| Niedzielin et al., 2001 (88) | Multicenter double-blind, placebo-controlled study with parallel groups     | 10^8 active group/10^6 placebo group; males and females                  | Lactobacillus plantarum 299V (LP299V) | Fermented fruit juice with 5% oats                                       | Symptom improvement in 95% of the IBS patients                                                             | No side effects                           |
| Murakami et al., 2012 (89) | Double-blind cross-matched trial                                             | 35 males and females >6 y                                                 | Lactobacillus brevis KB290        | Lyophilized capsule (1 × 10^10 CFU)                                       | Improvement in IBS symptoms; increased abundance of Bifidobacterium and Clostridium in the intestinal microflora | Side effects: abdominal pain and diarrhea                                                        |

(Continued)
TABLE 3 (Continued)

| Study (reference)       | Study design                     | Subjects                                      | Probiotic’s synthesis                      | Substrate                                      | Effectiveness                                                                 | Side effects                                                                                     |
|-------------------------|----------------------------------|-----------------------------------------------|---------------------------------------------|-----------------------------------------------|--------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------|
| Stevenson et al., 2014  | Double-blind placebo-controlled randomized trial | 54 active group/27 placebo group; 96% females | *Lactobacillus plantarum* 299V (LP299V)   | Capsule (5 × 10^9 CFU)                         | No relief in patients from abdominal pain                                      | Several side effects/not described in detail                                                   |
| Pineton de Chambrun et al., 2015 | Double-blind placebo-controlled randomized trial | 86 active group/93 placebo group              | *Saccharomyces cerevisiae*                   | Capsule (8 × 10^9 CFU/g)                      | Improvement only in abdominal pain and discomfort                                  | Side effects included diarrhea, constipation, headache, abdominal pain, bloating, back pain, gastroesophageal reflux disease, bladder infection, influenza, and hemorrhoidal crisis |
| Thijssen et al., 2016   | Double-blind placebo-controlled randomized trial | 39 active group/41 placebo group; males and females | *Lactobacillus casei* Shirota              | Fermented milk with L. casei Shirota (6.5 × 10^9 CFU) | No improvement during intervention; positive effect after completion              | No side effects                                                                                 |
| Spiller et al., 2016    | Double-blind placebo-controlled randomized trial | 192 active group/187 placebo group; 18–75 y; males and females | *Saccharomyces cerevisiae* CNCM I-3856 | Capsule (500 mg)                              | No overall benefit in IBS; improvement in abdominal pain, discomfort, bloating    | Several side effects/not described in detail                                                   |
| Sadrin et al., 2017     | Double-blind placebo-controlled randomized trial | 40 active/40 placebo; 18–65 y                 | *Lactobacillus acidophilus* NCFM και LAFTI L10 | Capsule (2.5 × 10^9 CFU)                      | Reduced abdominal pain; overall improvement of IBS symptoms                      | Several side effects/not described in detail                                                   |

1 ATCC, American Type Culture Collection; GI, gastrointestinal; IBS, irritable bowel syndrome; QoL, quality of life.
of gender has been reported in NCGS, with a female-to-male predominance of 3:1 (12, 95, 96). The basic difference between IBS and NCGS is that patients with the latter assert that their symptoms occur after wheat consumption and blame gluten as the culprit. The main clinical manifestations of NCGS are presented in Table 2.

Even a 3-d challenge with 16 g/d gluten has been related to feelings of depression in NCGS patients (4). This, however, was not associated with cortisol secretion, suggesting the involvement of gluten exorphins, which are opioid peptides that derive from partially digested food proteins (97). Exorphins have been shown to pass through the blood–brain barrier and can therefore directly interfere with pain, emotional pathways, and other hormonal or neurotransmitter systems through the endogenous and exogenous opioid receptors. Orally administered gliadinexorphin A5 was shown to modify learning and anxiety behavior during several laboratory stressors in mice, thus indicating that orally delivered exorphins can influence both the peripheral and central nervous system and suggesting that gluten exorphins possess opioid activity (4).

The onset of symptoms after gluten ingestion appears from hours up to few days and their resolution time also varies and can last up to weeks. Due to the lack of specific serological markers, the diagnosis of NCGS is made after the exclusion of gastrointestinal malignancies and allergic (WA) or autoimmune (CD, dermatitis herpetiformis, and gluten ataxia) reactions (9). Although, for the accurate diagnosis of NCGS, a DBPCFC with 8 g of gluten and at least 0.3 g of the proinflammatory ATIs per day for at least 7 d has been proposed by the Salerno experts in 2015, to date, a proper vehicle to carry out the challenge has not been developed. As trials conducted so far have high heterogeneity, it is difficult to define a diagnostic protocol (4, 69, 98).

Although, and in contrast to CD, there is no evident genetic predisposition identified so far, the activation of innate immunity without any implication of the adaptive immune response has been described (22, 23, 95, 99). Some studies suggest that ATIs or a combination of ATIs and gluten can induce this immune response (61, 62). In NCGS patients, the intestinal permeability and the expression of tight junction proteins claudin-1 and zonulin-1 (ZO-1) appear to be normal, but there is an increased expression of claudin-4 accompanied by the high expression of TLRs (TLR-1, TLR-2, and TLR-4) and a lower number of regulatory T cells (9). TLRs strongly maintain the intestinal epithelial homeostasis by mediating the dynamic host–microbe interactions. The intestinal microbes have been shown to decrease intestinal permeability by upregulating the expression of tight junction proteins. Since the gut microbiota has an essential role in regulating the antigen milieu of enterocytes, it has been suggested that it can activate the immune processes in certain individuals towards CD or NCGS. Moreover, human gastric, pancreatic, and brush-border enzymes cannot completely degrade dietary gluten because of the unusually high proportion of proline residues and T-cell stimulatory gluten peptides up to 33 amino acids in length. Numerous gut micro-organisms, including many sourdough bacteria belonging to Streptococcaceae, Lactobacillaceae, and Bifidobacteriaceae, as well as fungi and yeasts, are able to completely degrade gluten proteins (100). By producing gluten-degrading enzymes, the gut microbiota can convert an immunogenic peptide to a nonimmunogenic peptide, and consequently build up driving function towards an NCGS profile (100).

Possible triggers in NCGS.

Gluten has been considered to be the stimulator of NCGS, forcing patients, similarly those with CD, to a life-long adherence to a GFD. Nevertheless, today we know, via several blinded placebo-controlled studies (4, 22, 54, 55), that ATIs and wheat FODMAPs, apart from gliadin, trigger the innate immune responses (Table 2) (56, 61, 54, 101).

ATIs contribute to symptoms in NCGS by the activation of gut myeloid cells (56, 101). In murine models, dietary ATIs worsened the allergic inflammation in the airways, allergen IgE-dependent colitis, and gut inflammation (62). Since ATIs can trigger and sustain gut inflammation, they should undoubtedly be considered an additional trigger in NCGS pathogenesis.

A meta-analysis reported that only 16% of NCGS patients developed symptoms after challenge with <8 g/d pure gluten, whereas another Double Blind Placebo Challenge (DBPC) demonstrated a significant improvement in gastrointestinal symptoms after a low-FODMAP diet (4, 57). Wheat FODMAPs (fructans) have been blamed for the abdominal pain in NCGS, as they resist digestion in the proximal small bowel and are processed by bacteria in the distal small bowel and colon (102, 103). Many NCGS patients benefit from a low-FODMAP diet, in both their gastrointestinal and psychological symptoms, despite being on a gluten-containing diet (101, 103, 104). In some cases, NCGS patients develop more severe intestinal symptoms after consuming only 2.1 g fructans than when they introduce gluten in their diet (56). Additional evidence comes from a recent study showing that NCGS patients adhering to a GFD have only partial symptom alleviation (2) and FODMAPs were suggested as also being responsible for symptom development in NCGS (2, 22).

Microbiota in NCGS.

As described in Table 2, an abundance of Firmicutes with decreased Bacteroidetes populations, similar to IBS, has been reported in NCGS patients (5, 6, 98). In a study by Dieterich and Zopf (98), a low-FODMAP diet improved NCGS symptoms and further reduced the levels of Bifidobacterium compared with corresponding controls. However, a low-FODMAP diet also eliminates prebiotics and can result in dysbiosis. The gut microbiome in NCGS patients presents high metabolic activity, suggesting that their microflora is more susceptible to dietary changes than in their healthy controls.

In a recent study in Mexico a GFD improved NCGS patients’ symptoms and induced a significant increase of an average of 14.8% of Pseudomonas in gut microbiota and in duodenal biopsies (59) 4 wk after implementation. Since Pseudomonas comprises strains with gluten-degrading capabilities, the authors suggested that some Pseudomonas strains could be tested as a probiotic supplement for alleviating symptoms in gluten-related disorders. Nevertheless, research to date is limited and the role of microbiota in NCGS onset and/or development should be further investigated via prospective clinical trials.

Potential therapeutic pathways in NCGS.

The NCGS profile is still obscure in terms of clinical characteristics, diagnostic criteria, and therapy. Although a GFD is generally recommended, there does not need to be lifelong adherence at the level of CD. Usually, patients after a period of 1–2 y of a strict GFD can attempt to reintroduce small and gradually increasing amounts of gluten in their
The literature thus far refers to gluten, FODMAPs, and ATIs as 3 ambiguous villains for symptom exacerbation of IBS and NCGS. These overlapping disorders, in terms of clinical features and some inducing agents, have increasing prevalence. The insufficient knowledge to date, however, and the lack of proper accurate diagnostic criteria make them dependent on subjective report, making self-diagnosis difficult. When wheat products provoke symptoms, a GFD is the diet of choice, which is strict and hard to adhere to in the long term with parallel sufficient macro- and micronutrient intake, unless sufficient personalized dietary guidance is provided by a specialized dietitian. A GFD is low in fiber and high in fat and sugar (105), so patients need to be informed regarding the long-term complications, such as hyperlipidemia, cardiovascular disease, and obesity. Evidence shows that gluten is not the actual cause of NCGS symptoms as patients challenged with pure gluten in a DBPCFC did not present more symptoms than the placebo group (54, 55, 57). In addition, the activation of the innate immune responses and the resulting intestinal inflammation and dysbiosis make the microbiome an open area for intervention.

Opinions on the topic
Taking into consideration that 1) IBS and NCGS respond in similar ways when wheat is the triggering food; 2) NCGS symptom deterioration is probably dependent on the total dose of wheat consumed, similar to lactose intolerance (58), therefore a strict elimination might not be obligatory for all patients; 3) exacerbation of both intestinal and extra-intestinal symptoms after wheat consumption occurs; and 4) clinical trials with prebiotics in IBS patients induced shifts in healthier microbiota populations, whereas prebiotics did not induce changes in the microflora composition but mainly resulted in metabolic changes in the local bacteria populations, we suggest the following for future research:

1. Standardize the challenge method for NCGS according to the Salerno Experts criteria with a proper challenge vehicle
2. Investigate the microbiome’s shifts from a cereal-free diet to a cereal-containing diet in NCGS patients and parallel metabolic changes
3. Aim for personalized diet therapy in clinical trials with parallel cognitive behavioral therapeutic interventions to minimize anxiety and stress upon interventions and investigate individualized response to diet therapy and psychotherapy (106)
4. Perform clinical trials with symbiotics in NCGS patients, according to results so far from IBS trials, to investigate their actual anti-inflammatory potential in NCGS
5. Enlarge clinical trials including IBS patients sensitive to wheat and NCGS patients and compare the 2 disorders under identical investigation conditions

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References
1. Food formulation trends: ingredients consumers avoid: market research report. 2016. [cited 2016 Apr 29] [Internet]. Packaged Facts, 208.
2. Sapone A, Bai JC, Ciacci C, Dolinsek J, Green PHR, Hadjivassiliou M, Kaukinen K, Rostami K, Sanders SD, Schumann M, et al. Spectrum of gluten-related disorders: consensus on new nomenclature and classification. BMC Med 2012. [cited 2020 May 19];10(1):13. [Internet]. Available from: http://bmcmedicine.biomedcentral.com/articles/10.1186/1741-7015-10-13.
3. Niland B, Cash BD. Health benefits and adverse effects of a gluten-free diet in non-celiac disease patients [Internet]. Gastroenterol Hepatol 2018;14 [cited 2020 Jul 3]. Available from: http://pmc.biomedcentral.com/articles/PMC5866307/report#abstract.
4. Peters SL, Biesiekierski JR, Yolland GW, Muir JG, Gibson PR. Randomised clinical trial: gluten may cause depression in subjects with non-coeliac gluten sensitivity—an exploratory clinical study. Aliment Pharmacol Ther 2014;39(10):1104–12.
5. Distrutti E, Monaldi L, Ricci P, Fiorucci S. Gut microbiota role in irritable bowel syndrome: new therapeutic strategies. World J Gastroenterol 2016;22:2219–41.
6. Rodiño-Janeiro BK, Vicario M, Alonso-Cotoner C, Pascua-García R, Santos J. A review of microbiota and irritable bowel syndrome: future in therapies. Adv Ther 2018;35:289–310.
7. Singh P, Arora A, Strand TA, Leffler DA, Catassi C, Green PH, Kelly P C, Ajuja V, Makharia GK. Global prevalence of celiac disease: systematic review and meta-analysis. Clin Gastroenterol Hepatol 2018;16(6):823–36, e2.
8. Choung RS, Ditah IC, Nadeau AM, Rubio-Tapia A, Marietta E V, Brantner TL, Camilleri MF, Rajkumar SV, Landgren O, Erhart JE, et al. Trends and racial/ethnic disparities in gluten-sensitive problems in the United States: findings from the National Health and Nutrition Examination Surveys from 1988 to 2012. Am J Gastroenterol [Internet] 2015;110(10):455–61. Available from: http://www.ncbi.nlm.nih.gov/pubmed/25665935.
9. Sapone A, Bai JC, Ciacci C, Dolinsek J, Green PHR, Hadjivassiliou M, Kaukinen K, Rostami K, Sanders DS, Schumann M, et al. Spectrum of gluten-related disorders: consensus on new nomenclature and classification. BMC Med 2012;10.
10. Harris LA, Park JY, Voltaggio L, Lam-Himlin D. Celiac disease: clinical, endoscopic, and histopathologic review. Gastroint Endosc 2012;76(3):625–40.
11. Sapone A, Lammers KM, Casolaro V, Cammarota M, Giuliano MT, De Rosa M, Stefanié R, Mazarella G, Tolone C, Russo ML, et al. Divergence of gut permeability and mucosal immune [BMC Med. 2011]. PubMed result. BMC Med [Internet] 2011;9(1) [cited 2020 Jan 10]:455–61. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21392369.
12. Volta U, Bardella MT, Calabrò A, Troncone R, Corazza GR, Bagnato C, Study Group for Non-Celiac Gluten Sensitivity. An Italian prospective multicenter survey on patients suspected of having non-celiac gluten sensitivity. BMC Med 2014;12(1).
13. Borghini R, Donato G, Alvaro D, Picarelli A. New insights in IBS-like disorders: Pandora’s box has been opened; a review. Gastroenterol Hepatol Bed Bench 2017;10:79–89.
14. Yan YL, Hu Y, Gänzle MG. Prebiotics, FODMAPs and dietary fiber—conflicting concepts in development of functional food products? Curr Opin Food Sci 2018;20:30.
15. Staudacher HM, Lomer MCE, Farquharson FM, Louis P, Fava F, Franciosi E, Scholz M, Tuohy KM, Lindsay JO, Irving PM, et al. A diet low in

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Culprits for wheat sensitivity 11
FODMAPs reduces symptoms in patients with irritable bowel syndrome and a probiotic restores Bifidobacterium species: a randomized controlled trial. Gastroenterology 2017;153(4):936–47.

16. Makaria A, Catassi C, Makaria GK. The overlap between irritable bowel syndrome and non-celiac gluten sensitivity: a clinical dilemma. Nutrients [Internet] 2015;7(12) [cited 2020 Jan 9];10417–26. Available from: http://www.ncbi.nlm.nih.gov/pubmed/26690475.

17. Koumbi L. Gut microbiota alterations in liver diseases. EC Gastroenterol Dig Syst 2018;5(4):191–8.

18. Brooks AW, Priya S, Blekhman R, Bordenstein SR. Gut microbiota diversity across ethnicities in the United States. PLOS Biol [Internet] 2018;16(12) [cited 2020 Oct 31];e2006842. Available from: https://dx.plos.org/10.1371/journal.pbio.2006842.

19. Fontana A, Panebianco C, Picchianti-Diamanti A, Laganà B, Cavalieri D, Potenza A, Pracella R, Binda E, Copetti M, Piazzena V. Gut microbiota profiles differ among individuals depending on their region of origin: an Italian pilot study. Int J Environ Res Public Health [Internet] 2019;16(21) [cited 2020 Oct 31]. Available from: http://pmc/articles/PMC6862301/?report=abstract.

20. Lacy B, Patel N. Rome criteria and a diagnostic approach to irritable bowel syndrome. J Clin Med [Internet] 2017;6(11) [cited 2020 Jun 20];99. Available from: http://pmc/articles/PMC5704116/?report=abstract.

21. Farnam A, Somi MH, Sarami F, Farhang S. Five personality dimensions in patients with irritable bowel syndrome. Neuropsychiatr Dis Treat 2008;4(5):959–62.

22. Dieterich W, Schuppan D, Schink M, Schwappacher R, Wirtz S, Agaimy A, Agaimy A, Neurath MF, Zopf Y. Influence of low FODMAP and gluten-free diets on disease activity and intestinal microbiota in patients with non-celiac gluten sensitivity. Clin Nutr [Internet] 2019;38(2):697–707. Available from: https://doi.org/10.1016/j.clnu.2018.03.017.

23. Makaria A, Catassi C, Makaria GK. The overlap between irritable bowel syndrome and non-celiac gluten sensitivity: a clinical dilemma. Nutrients 2015;7(12):10417–26.

24. Muscatello MRA, Bruno A, Mento C, Pandolfi G, Zoccali RA. Personality traits and emotional patterns in irritable bowel syndrome. World J Gastroenterol 2016;22(28):6402–15.

25. Bharadwaj S, Barber MD, Graff LA, Shen B. Symptomatology of irritable bowel syndrome and inflammatory bowel disease during the menstrual cycle. Gastroenterol Rep 2015;3(3):185.

26. Wellang ME, Palmer WC, Lacy BE. Irritable bowel syndrome and dietary interventions. Gastroenterol Hepatol 2019;15(1):16–26.

27. Verdu EF, Huang X, Natividad J, Lu J, Brennerhassett PA, David CS, McKay S, Böhn L, Störsrud S, Liljebo T, Collin L, Lindfors P, Törnbloom H, Öhman L SM. Multivariate modelling of faecal bacterial profiles of patients with IBS predicts responsiveness to a diet low in FODMAPs. Gut 2018;67(5):872–81.

28. Vazquez-Roque MI, Camilleri M, Smyrk T, Murray JA, O’Neill J, Carlson JD, Shepherd SJ, Muir JG, Gibson PR. Gluten causes DQ genotype with bowel transit, barrier function, and inflammation in irritable bowel syndrome. Am J Gastroenterol [Internet] 2010;105(1) [cited 2020 Jan 9];512–9, e114-5. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20066003.

29. Vandeputte D, Falony G, Vieira-Silva S, Tito RY, Joossens M, Raes J. Potential metabolic and physiological effects of prebiotics and probiotics on the gut microbiota. Gut Microbes 2017;8(2):50–62.

30. Tana C, Umesaki Y, Imaoka A, Handa T, Kanazawa M, Fukudo S. Altered prevalence of mucosa-associated and fecal microbiota in irritable bowel syndrome patients. World J Gastroenterol 2009;15(23):2887–92.

31. Rajilić-Stojanović M, Biagi E, Heilig H, Kajander K, Kekkonen RA, Tana C, Umesaki Y, Imaoka A, Handa T, Kanazawa M, Fukudo S. Altered profiles of intestinal microbiota and organic acids may be the origin of symptoms in irritable bowel syndrome. Neurogastroenterol Motil [Internet] 2017;9(1):e12619. Available from: http://www.ncbi.nlm.nih.gov/pubmed/28470862.

32. Pedersen N, Andersen DV, Felding M, Wachmann H, Végh Z, Molzen L, Burisch J, Andersen JR, Munkholm P. Low-FODMAP diet reduces irritable bowel symptoms in patients with inflammatory bowel disease. World J Gastroenterol 2017;23(18):3356–66.

33. Fritscher-Ravens A, Schuppan D, Ellrichmann M, Schoch S, Röcken C, Brashc J, Bethge J, Bottner M, Klose J, Milla PJ. Confocal endomicroscopy shows food-associated changes in the intestinal mucosa of patients with irritable bowel syndrome. Gastroenterology 2014;147(5):1012–20, e4.

34. Pedersen N, Andersen DV, Felding M, Wachmann H, Végh Z, Molzen L, Burisch J, Andersen JR, Munkholm P. Low-FODMAP diet reduces irritable bowel symptoms in patients with inflammatory bowel disease. World J Gastroenterol 2017;23(18):3356–66.

35. Fritscher-Ravens A, Schuppan D, Ellrichmann M, Schoch S, Röcken C, Brashc J, Bethge J, Bottner M, Klose J, Milla PJ. Confocal endomicroscopy shows food-associated changes in the intestinal mucosa of patients with irritable bowel syndrome. Gastroenterology 2014;147(5):1012–20, e4.

36. Pedersen N, Andersen DV, Felding M, Wachmann H, Végh Z, Molzen L, Burisch J, Andersen JR, Munkholm P. Low-FODMAP diet reduces irritable bowel symptoms in patients with inflammatory bowel disease. World J Gastroenterol 2017;23(18):3356–66.

37. Fritscher-Ravens A, Schuppan D, Ellrichmann M, Schoch S, Röcken C, Brashc J, Bethge J, Bottner M, Klose J, Milla PJ. Confocal endomicroscopy shows food-associated changes in the intestinal mucosa of patients with irritable bowel syndrome. Gastroenterology 2014;147(5):1012–20, e4.

38. Pedersen N, Andersen DV, Felding M, Wachmann H, Végh Z, Molzen L, Burisch J, Andersen JR, Munkholm P. Low-FODMAP diet reduces irritable bowel symptoms in patients with inflammatory bowel disease. World J Gastroenterol 2017;23(18):3356–66.

39. Pedersen N, Andersen DV, Felding M, Wachmann H, Végh Z, Molzen L, Burisch J, Andersen JR, Munkholm P. Low-FODMAP diet reduces irritable bowel symptoms in patients with inflammatory bowel disease. World J Gastroenterol 2017;23(18):3356–66.

40. Pedersen N, Andersen DV, Felding M, Wachmann H, Végh Z, Molzen L, Burisch J, Andersen JR, Munkholm P. Low-FODMAP diet reduces irritable bowel symptoms in patients with inflammatory bowel disease. World J Gastroenterol 2017;23(18):3356–66.
47. Hunter JO, Tuftnell Q, Lee AJ. Controlled trial of oligofructose in the management of irritable bowel syndrome. J Nutr 1999;129(7):1451S–3S.

48. Olesen M, Gudmand-Hoyer E. Efficacy, safety, and tolerability of fructooligosaccharides in the treatment of irritable bowel syndrome. Am J Clin Nutr [Internet] 2000;72(6) [cited 2020 Jan 10]:1570–5. Available from: http://www.ncbi.nlm.nih.gov/pubmed/11101487.

49. Paineau D, Payen F, Panserrie S, Coubliomer G, Sobaszek A, Lartigau I, Lartigau I, Brabet M, Galmiche JP, Tripodi D, et al. The effects of regular consumption of short-chain fructo-oligosaccharides on digestive comfort of subjects with minor functional bowel disorders. Br J Nutr 2008;99(2):311–8.

50. Didari T, Mozaffari S, Nikfar S, Abdollahi M. Effectiveness of probiotics in irritable bowel syndrome: updated systematic review with meta-analysis. World J Gastroenterol [Internet] 2015;21(10) [cited 2020 Jan 10]:3072–84. Available from: http://www.wjgnet.com/wjgnet25780308.

51. Min YW, Park SU, Jang YS, Kim Y-H, Rhee P-L, Ko SH, Joo N, Kim SI, Kim CH, Chang DK. Effect of composite yogurt enriched with acacia fiber and Bifidobacterium lactis. World J Gastroenterol [Internet] 2012;18(33) [cited 2020 Jan 10]:4563–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/22969230.

52. Tsuchiya J, Barreto R, Okura R, Kawakita S, Fesce E, Marotta F. Single-blind follow-up study on the effectiveness of a symbiotic preparation in irritable bowel syndrome. Chin Dig Dis 2004;5(4):169–74.

53. Chey WD, Chang L, Lembo A, Aggarwal K, Bortey E, Paterson C, Forbes WP. 313 Effects of rifaximin on urgency, bursting, and abdominal pain in patients with IBS-D: a randomized, controlled, repeat treatment study. Gastroenterology 2015;148(4):S–69.

54. Zanini B, Baschi R, Ferraresi A, Ricci C, Lanzarotto F, Marullo M, Villanacci V, Hidalgo A, Lanzini A. Randomised clinical study: gluten challenge induces symptom recurrence only in a minority of patients who meet clinical criteria for non-coeliac gluten sensitivity. Aliment Pharmacol Ther 2015;42(8):968–76.

55. Dale HF, Hatlebakaj BK, Hvodonak N, Ystad SO, Lied GA. The effect of a controlled gluten challenge in a group of patients with suspected non-coeliac gluten sensitivity: a randomized, double-blind placebo-controlled challenge. Neurogastroenterol Motil 2018;30(8):e13032.

56. Skodje GI, Sarna VK, Minelle IH, Rolfsen KL, Muir JG, Gibson PR, Veierød MB, Henriksen C, Lundin KEA. Fructan, rather than gluten, induces symptoms in patients with self-reported non-celiac gluten sensitivity. Gastroenterology 2018;154(3):529–39, e2.

57. Molina-Fantje J, Carroccio A. Suspected non-celiac gluten sensitivity confirmed in few patients after gluten challenge in double-blind, placebo-controlled trials. Clin Gastroenterol Hepatol 2017;15(3):339–48.

58. Tovoli F, Granito A, Negri G, Guidetti E, Faggiano C, Bolondi L. Long term effects of gluten-free diet in non-coeliac wheat sensitivity. Clin Nutr 2017;38(1):357–63.

59. Garcia-Macorso JR, Rivero-Gutierrez X, Cobos-Quevedo OJ, Grube-Pagola P, Meixueiro-Daza A, Hernandez-Flores K, Cabrera-Jorge FJ, Vivanco-Cid H, Dowd SE, Remes-Troche JM. First insights into the gut microbiota of Mexican patients with celiac disease and non-celiac gluten sensitivity. Nutrients 2018;10(11):1641.

60. Junker Y, Zeissig S, Kim SJ, Barisani D, Wieser H, Leffler DA, Zevallos V, Scholz, Tuohy KM, Lindsay JO, Irving PM, et al. A diet low in FODMAPs reduces symptoms in patients with irritable bowel syndrome and a probiotic restores Bifidobacterium species: a randomized controlled trial. Gastroenterology 2017;153(4):936–47.

61. Mazzawi T, El-Sally M. Effect of diet and individual dietary guidance on gastrointestinal endocrine cells in patients with irritable bowel syndrome [review]. Int J Mol Med [Internet] 2017;40 [cited 2020 Oct 31]:943–52. Available from: http://pmc/articles/PMC5593462/?report=abstract.

62. Nanayakkara WS, Skidmore PM, O’Brien L, Wilkinson TJ, Garey RB. Efficacy of the low FODMAP diet for treating irritable bowel syndrome: the evidence to date. Clin Exp Gastroenterol 2016;9:131–42.

63. Tuck CJ, Muir JR, Barrett JS, Gibson PR. Fermentable oligosaccharides, disaccharides, monosaccharides and polyols: role in irritable bowel syndrome. Exp Rev Gastroenterol Hepatol 2014;8:819–34.

64. Bardella MT, Elli L, Ferretti F. Non celiac gluten sensitivity. Curr Gastroenterol Rep [Internet] 2016;18 [cited 2020 Oct 31]:1–7. Available from: https://link.springer.com/article/10.1007/s11894-016-0536-7.

65. Staudacher HM, Lomer MCE, Farquharson FM, Louis P, Fava F, Franciosi E, Scholz, Tuohy KM, Lindsay JO, Irving PM, et al. A controlled gluten challenge in a group of patients with suspected non-celiac gluten sensitivity (NCGS): the Salerno experts’ criteria. Nutrients [Internet] 2015;7(6) [cited 2020 Apr 9]:4966–77. Available from: http://www.mdpi.c om/2072-6643/7/6/4966.

66. Dupont FM, Vensel WH, Tanaka CK, Hurkman WJ, Altenbach SB. Deciphering the complexities of the wheat flour proteome using quantitative two-dimensional electrophoresis, three proteases and tandem mass spectrometry. Proteome Sci 2011;9:10.

67. Kim G, Deepinder F, Morales L, Huang W, Weitsman S, Chang C, Gunsalus R, Pimentel M. Methanobrevibacter smithii is the predominant methanogen in patients with constipation-predominant IBS and methane on breath. Dig Dis Sci [Internet] 2012;57(12) [cited 2020 Jan 30]:3213–8. Available from: http://www.ncbi.nlm.nih.gov/pubmed/22573345.

68. Pimentel M, Lin HC, Enayati P, Van Den Burg B, Lee HR, Chen JH, Park S, Kong Y, Conklin J. Methane, a gas produced by enteric bacteria, slows intestinal transit and augments small intestinal contractile activity. Am J Physiol Gastrointest Liver Physiol [Internet] 2006;290(6) [cited 2020 Nov 1]:1089–95. Available from: http://www.ajpgi.org:1089.

69. Cosma-Petrut A, Loghin F, Miere D, Dumitrascu DL. Diet in irritable bowel syndrome: what to recommend, not what to forbid to patients! [Internet]. World J Gastroenterol 2017;23 [cited 2020 Apr 9]:3771–83. Available from: http://www.ncbi.nlm.nih.gov/pubmed/28638217.

70. Ford AC, Quigley EM, Lacy CJ, Aalto SA, Schiller LR, Soffer EE, Spiegel BM, Moayyedi. Efficacy of prebiotics, probiotics, and synbiotics in irritable bowel syndrome and chronic idiopathic constipation: systematic review and meta-analysis. Am J Gastroenterol 2014;109(10):1547–62.

71. Zhang Y, Li L, Guo C, Mu D, Feng B, Zuo X, Li Y. Effects of probiotic type, dose and treatment duration on irritable bowel syndrome diagnosed by Rome III criteria: a meta-analysis. BMC Gastroenterol 2016;16(1):62.

72. Hill C, Guaner F, Reid G, Gibson GR, Merenstein DJ, Pot B, Morelli L, Canani RB, Flint HJ, Salminen S, et al. Expert consensus document. The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. Nat Rev Gastroenterol Hepatol [Internet] 2014;11(8) [cited 2020 Jan 10]:506–14. Available from: http://www.ncbi.nlm.nih.gov/pubmed/24912386.

73. Hosseini A, Nikfar S, Abdollahi M. Are probiotics effective in management of irritable bowel syndrome? Arch Med Sci 2012;8:403–5.

74. Vogt I, Meyer D, Pullens G, Faas M, Smelt M, Venema K, Ramasamy U, Schols HA, De Vos P. Immunological properties of inulin-type fructans. Immunological properties of inulin-type fructans. Immunologically relevant properties of inulin-type fructans. Curr Rev Food Sci Nutr [Internet] 2015;5(3) [cited 2020 Jan 10]:414–36. Available from: http://www.ncbi.nlm.nih.gov/pubmed/24915372.

75. Mego M, Manichanh C, Accarino A, Campos D, Pozuelo M, Varela E, Vulevic J, Tzortzis G, Gibson G, Guaner F, et al. Metabolic adaptation of colon microbiota to galactooligosaccharides: a proof-of-concept study.
Aliment Pharmacol Ther [Internet] 2017;45(5) [cited 2020 Jan 10]:670–80. Available from: http://www.ncbi.nlm.nih.gov/pubmed/28078750.

Meneses SB, Maneerattanaporn M, Kim HM, Chey WD. The efficacy and safety of rifaximin for the irritable bowel syndrome: a systematic review and meta-analysis. Am J Gastroenterol 2012;107:28–35.

Halfvarson J, Brislawn CJ, Lamendella R, Vázquez-Baeza Y, Walters WA, Bramer LM, D’Amato M, Bonfiglio F, McDonald D, Gonzalez A, et al. Dynamics of the human gut microbiome in inflammatory bowel disease. Nat Microbiol 2017;2.

Koebnick C, Wagner I, Leitzmann P, Stern U, Zunft HF. Probiotic beverage containing Lactobacillus casei Shiroti improves gastrointestinal symptoms in patients with chronic constipation. Can J Gastroenterol 2003;17(11):655–9.

Niv E, Naftali T, Hallak R, Vaisman N. The efficacy of Lactobacillus reuteri ATCC 55730 in the treatment of patients with irritable bowel syndrome—a double blind, placebo-controlled, randomized study. Clin Nutr [Internet] 2005;24(6) [cited 2020 Apr 10]:925–31. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16051399.

Worhorne PJ, Altringer L, Morel J, Bond Y, Charbonneau D, O’Mahony L, Kiely B, Shanahan F, Quigley EMM. Efficacy of an encapsulated probiotic Bifidobacterium infantis 35624 in women with irritable bowel syndrome. Am J Gastroenterol [Internet] 2006;101(7) [cited 2020 Apr 10]:1581–90. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16863564.

Sinn DH, Song JH, Kim HJ, Lee JH, Son HJ, Chang DK, Kim YH, Kim JJ, Rhee JC, Rhee PL. Therapeutic effect of Lactobacillus acidophilus–SDFC 2012, 2013 in patients with irritable bowel syndrome. Dig Dis Sci 2008;53(10):2714–8.

Guglielmetti S, Mora D, Gschwender M, Popp K. Randomised clinical trial: Bifidobacterium bifidum MIMBb75 significantly alleviates irritable bowel syndrome and improves quality of life—a double-blind, placebo-controlled study. Aliment Pharmacol Ther [Internet] 2011;33(10) [cited 2020 Apr 9]:1123–32. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21418261.

Ducrotté P, Sawant P, Jayanthi V. Clinical trial: Lactobacillus plantarum 299v (DSM 9843) improves symptoms of irritable bowel syndrome. World J Gastroenterol 2012;18(30):4012–8.

Niedzielin K, Kordecki H, Birkenfeld B. A controlled, double-blind, randomized study on the efficacy of lactobacillus plantarum 299V in patients with irritable bowel syndrome. Eur J Gastroenterol Hepatol 2001;13(10):1143–7.

Murakami K, Habukawa C, Nobuta Y, Moriguchi N, Takemura T. The effect of Lactobacillus brevis KB290 against irritable bowel syndrome: a placebo-controlled double-blind crossover trial. Biopsychosoc Med 2012;6:16.

Stevenson C, Blaauw R, Fredericks E, Visser J, Roux S. Randomized clinical trial: effect of lactobacillus plantarum 299v on symptoms of irritable bowel syndrome. Nutrition [Internet] 2014;30(10) [cited 2020 Apr 10]:1151–7. Available from: http://www.ncbi.nlm.nih.gov/pubmed/25194614.

Pineton de Chambrun G, Neut C, Chau A, Cazaubiel M, Pelerin F, Justen P, Desreumaux P. A randomized clinical trial of Saccharomyces cerevisiae versus placebo in the irritable bowel syndrome. Dig Liver Dis [Internet] 2015;47(2) [cited 2020 Apr 10]:119–24. Available from: http://www.ncbi.nlm.nih.gov/pubmed/25488056.

Thijssen AJ, Clemens CHM, Vankerckhovken V, Goossens H, Jonkers D, Mascele AAM. Efficacy of Lactobacillus casei Shirori for patients with irritable bowel syndrome. Eur J Gastroenterol Hepatol 2016;28(1):8–14.

Spiller R, Pelerin F, Cayzeele Decherf A, Maudet C, Houssez B, Cazaubiel M, Jüsten P. Randomized double blind placebo-controlled trial of Saccharomyces cerevisiae CNCM I-3856 in irritable bowel syndrome: improvement in abdominal pain and bloating in those with predominant constipation. United European Gastroenterol J 2016;4(3):353–62.

Sadrin S, Sennoune SR, Gout B, Marque S, Moreau J, Grillasca J, Pons O, Maixent JM. Lactobacillus acidophilus versus placebo in the symptomatic treatment of irritable bowel syndrome: the LAPIBSS randomized trial. Cell Mol Biol (Noisy-le-grand) 2017;63(9):122–131, doi: 10.14715/cmb.2017.63.9.21, PMID: 28980935.

Caio G, Riegler G, Patturelli M, Facchiano A, De Magistris L, Sapone A. Pathophysiology of non-celiac gluten sensitivity: where are we now? Minerva Gastroenterol Dietol [Internet] 2017;63(1) [cited 2020 Jan 9]:16–21. Available from: http://www.ncbi.nlm.nih.gov/pubmed/27808487.

Half JO, Wyatt HR, Peters JC. The importance of energy balance. Eur Endocrinol [Internet] 2016;9(2) [cited 2019 Nov 21]:111. Available from: http://www.eurendumocrinology.com/articles/importance-energy-balance-e-9.

Peters SL, Biesiekierski JR, Yelland GW, Muir JG, Gibson PR. Randomised clinical trial: gluten may cause depression in subjects with non-coeliac gluten sensitivity—an exploratory clinical study. Aliment Pharmacol Ther 2014;39(10):1104–12.

Dieterich W, Zopf Y. Gluten and FODMAPS—sense of a restriction/when is restriction necessary? Nutrients 2019;11(8):1957.

Brottveit M, Beitnes A-CR, Tollefsen S, Bratlie JE, Jahnsen FL, Johansen F-E, Sollid LM, Lundin KEA. Mucosal cytokine response after short-term gluten challenge in celiac disease and non-celiac gluten sensitivity. Am J Gastroenterol [Internet] 2013;108(5) [cited 2020 Jan 8]:842–50. Available from: http://www.ncbi.nlm.nih.gov/pubmed/23588237.

Makharia A, Catassi C, Makharia GK. The overlap between irritable bowel syndrome and non-celiac gluten sensitivity: a clinical dilemma. Nutrients 2015;7(12):10417–26.

van Gils T, Nijensooer P, Ljissenagger CE, Sanders DS, Mulder CJJ, Bouma G. Prevalence and characterization of self-reported gluten sensitivity in The Netherlands. Nutrients 2016;8(11):714.

Catassi C, Bui JC, Bonaz B, Bouma G, Calabró A, Carroccio A, Castillejo G, Ciacci C, Christofori F, Dolinske J, et al. Non-celiac gluten sensitivity: the new frontier of gluten related disorders. Nutrients 2013;5(10):3839–53.

Fasano A, Sapone A, Zevallos V, Schuppan D. Non-celiac gluten sensitivity. Gastroenterology 2015;148(6):1195–204.

Ulevicius J, Juric A, Walton GE, Claus SP, Tzortzis G, Toward RE, Gibson GL. Influence of galacto-oligosaccharide mixture (B-GOS) on gut microbiota, immune parameters and metabolomics in elderly persons. Br J Nutr 2015;114(4):586–95.

Jones AL. The gluten-free diet: fad or necessity? Diabetes Spectr 2010;9(2) [cited 2019 Nov 21]:111. Available from: http://www.touchendocrinology.com/articles/importance-energy-balance-e-9.