CME

Food Choice as a Key Management Strategy for Functional Gastrointestinal Symptoms

Peter R. Gibson, MD[1] and Susan J. Shepherd, PhD[1]

Recognition of food components that induce functional gut symptoms in patient's functional bowel disorders (FBD) has been challenging. Food directly or indirectly provides considerable afferent input into the enteric nervous system. There is an altered relationship between the afferent input and perception/efferent response in FBD. Defining the nature of food-related stimuli may provide a means of minimizing such an input and gut symptoms. Using this premise, reducing the intake of FODMAPs (fermentable oligo-, di-, and mono-saccharides and polyols)—poorly absorbed short-chain carbohydrates that, by virtue of their small molecular size and rapid fermentability, will distend the intestinal lumen with liquid and gas—improves symptoms in the majority of patients. Well-developed methodologies to deliver the diet via dietician-led education are available. Another abundant source of afferent input is natural and added food chemicals (such as salicylates, amines, and glutamates). Studies are needed to assess the efficacy of the low food chemical dietary approach. A recent placebo-controlled trial of FODMAP-poor gluten provided the first valid evidence that non-celiac gluten intolerance might actually exist, but its prevalence and underlying mechanisms require elucidation. Food choice via the low FODMAP and potentially other dietary strategies is now a realistic and efficacious therapeutic approach for functional gut symptoms.

INTRODUCTION

The association of ingestion of food with the induction of gastrointestinal (GI) and, less commonly, non-GI symptoms is widely accepted by the community. In patients with irritable bowel syndrome (IBS), the frequency of perceptions of food intolerance is at least twofold more common than in the population in general (1). Furthermore, many take actions in accord with their perceptions. For example, in a Norwegian population study, 70% of 84 patients with IBS had symptoms related to food intake and most of those limited or excluded the perceived foods from their diet (2). In fact, food choice is the major method that women use to influence their symptoms (3). The belief that food is causing or at least triggering gut symptoms has led to the application of a profusion of investigations purporting to guide dietary design (such as skin prick tests, food-specific immunoglobulins, and many non-validated tests) (4), and of dietary therapies (such as gluten-free, wheat-free, anti-candidal, carbohydrate-free, and other complex exclusion diets).

The most profound evidence that ingestion of food itself really does have an important role in triggering symptoms was provided by observations of the effects of prolonged fasting, where marked improvement was noted in symptoms (5). However, recognition of what foods or food components are triggering the symptoms has been difficult. Broad differences in diet between those with and without functional gut symptoms, as assessed by the Harvard Food Frequency Questionnaire, have not been identified (1). Specific intolerances can be sought from patients' observations, but these correlate poorly with specific tests such as skin prick test and assessment of food-specific circulating antibodies (6). Even when intestinal provocation tests combined with exclusion diet-rechallenge methodology are performed, only a very small proportion of patients proven to have reactions to specific foods (7). The application of a bland exclusion diet (to minimize symptoms) followed by blinded rechallenge of a range of food types has been reported to be highly efficacious, but such enthusiasm has been dampened by the failure of other groups to reproduce the benefits, as well as the intensity and prolonged duration of such methodology (8). Furthermore, the intensity of dietary restrictions may lead to nutritional inadequacy (2).

Such a scenario leaves gastroenterologists generally to have a defensive role when patients request dietary intervention; while food is an undoubted trigger, recognizing what the specific food triggers is difficult at best, tests designed to do this have unproven or poor predictive value in their clinical utility, and the resulting diets are often overrestrictive with the potential to leave the patient nutritionally compromised (2). There have, however, been some notable exceptions, which include manipulation of dietary fiber intake (9,10), restricting lactose in those with hypolactasia (11), to a lesser extent, reduction of fructose intake in association with fructose malabsorption (12), and more recently, application of the very low carbohydrate diet in those with diarrhea-predominant IBS (13).

[1]Department of Gastroenterology, Alfred Hospital, Monash University, Melbourne, Victoria, Australia. **Correspondence:** Peter R. Gibson, MD, Department of Gastroenterology, Alfred Hospital, Monash University, Melbourne, 55 Commercial Road, Victoria 3004, Australia. E-mail: peter.gibson@monash.edu

REVIEW

A large degree of the confusion and skepticism over dietary therapies resides in the overriding obsession with "allergy" or with hypotheses that currently carry few physiological correlates. This review will approach concepts about the way specific foods potentially trigger bowel symptoms and discuss three approaches, each with a specific target, which have or may potentially have a significant impact on clinical practice for gastroenterologists.

CONCEPTS OF HOW FOOD TRIGGERS GI SYMPTOMS

To understand how food might trigger functional gut symptoms, it must be accepted that the symptoms primarily generate from the gut and specifically from the enteric nervous system (ENS). The brain can be considered as a modifying player only. The ENS is a major controller of multiple gut functions. These include secretion, motility, blood flow, and mucosal growth. In the normal situation, afferents respond to low intensity stimuli from lumen principally via mechanoreceptors and chemoreceptors. Little of such activity, as might be experimentally observed in association with minimal inflation of a rectal balloon during barostat studies, will reach the conscious brain and have few discernible effects on motility. In contrast, intense stimuli, such as those associated with large volume distension of a rectal balloon, will trigger sensations that are uncomfortable or painful and will alter motility. In most patients with functional bowel disorders (FBD), there is a change in relationship between stimulus intensity and perception (the hallmark of visceral hypersensitivity) and efferent motility response. Thus, low intensity stimuli may be felt as painful and noxious, and abnormal motility responses may be observed.

Luminal events are monitored via two main stimuli—mechanical/physical forces, principally associated with distension of the gut wall, and chemical stimuli. Enteroendocrine cells of the gut are believed to be the main receptors for chemical stimuli (14). In response, these cells release serotonin, which in turn stimulates primary afferents of the ENS. There is also evidence that some enteric neurons might directly respond to mechanical stimuli (15). Evidence is mounting that some members of the superfamily of transient receptor potential (TRP) cation channels are intimately involved in most levels of control of GI function including visceral hypersensitivity (16,17). TRPV1 (vanilloid) channels appear to be central to the initiation and persistence of visceral hypersensitivity in an animal model (18). Furthermore, increased expression of TRPV1 channels in neurons of the gut has been observed in patients with IBS and such expression appeared to correlate with visceral hypersensitivity (19) and with abdominal pain (20,21). Other potentially relevant receptors, such as T1R and T2R G-protein-coupled taste receptor families (22), glutamate-sensing receptors (23), and acid sensors (24), have also been identified (see recent reviews (16,17,22–24)). Dietary chemicals and products of bacterial metabolism of dietary components can also directly influence epithelial ion transport as recently reviewed (25).

Low-grade inflammation in the intestinal wall has been demonstrated in many patients with IBS, particularly but not exclusively in patients with post-infectious IBS, as reviewed in detail elsewhere (26). There is a body of evidence that suggests this inflammation

may be a mechanism through which many luminal molecules may lead to afferent and efferent responses in the ENS as well as altered ion transport. For example, inflammation in patients with IBD can lead to increased expression of TRPV1 channels in intestinal wall, which may lead to visceral hypersensitivity (21,27). Similarly, inflammatory cytokines such as interleukin-6 and prokineticin 2 induce visceral hypersensitivity and changes in epithelial ion flux via neuronal activation (28,29). Why the inflammation is present in patients with IBS is not clear. It is possible that this represents neuroinflammation, that is, inflammation being driven by the ENS. For example, in the microenvironment of intestinal neurons, there may be an increased density of mast cells (30), which can degranulate as a direct response to neuronal activation with subsequent release of chemoattractants for polymorphs (31). Likewise, neuropeptides can change transport protein expression in colonic epithelium with subsequent alterations in ion transport (32) and can influence immune function via binding to specific receptors on the surface of lymphocytes.

Food can potentially interact with the mechanisms involved in the genesis of symptoms and the complexity of such potential interactions is demonstrated in **Figure 1**. Food components can influence the composition of the microbiota, but this will not be discussed further. Food is a major physiological source of stimuli for the ENS. It will induce luminal distension by its physical presence and by secondary events such as gas production from bacterial fermentation. Food also contains a large soup of potentially stimulating chemicals. If the food constituents that stimulate the ENS can be identified, then these would be obvious targets for dietary manipulation. In the presence of visceral hypersensitivity and/or abnormal motility responses, reducing exposure to food-induced intestinal distension and/or specific food chemicals may then minimize triggering of symptoms.

Alternatively, if food components induce changes in the gut wall that directly (such as stimulation of receptors and/or induction of neuroinflammation) or indirectly (such as inducing a specific immune response) lead to altered afferent and/or efferent function of the ENS, then those components should also be the targets for dietary manipulation. Moreover, in this case, the avoidance of such food components may lead to the resolution of the underlying neural abnormalities. There is some evidence supporting food as a cause of visceral hypersensitivity. At a mechanistic level, plant chemicals are well documented to stimulate TRP channels (33). At the patient level, prolonged fasting, which improves symptoms of IBS (5), was associated with resolution of the visceral hypersensitivity in a single case report (34). In a study of 25 patients with IBS, food-specific IgG4 antibody-guided exclusion diets not only improved symptoms but also normalized rectal compliance (35). One interpretation of these observations is that visceral hypersensitivity was caused by food components and that this resolves with removal of the offending component.

On the basis of these concepts, three specific areas of proven or suspected food-induced gut symptoms in patients with FBD will be addressed: dietary FODMAPs (fermentable oligo-, di-, and mono-saccharides and polyols) that induce luminal distension, food chemicals that potentially stimulate the ENS, and gluten that
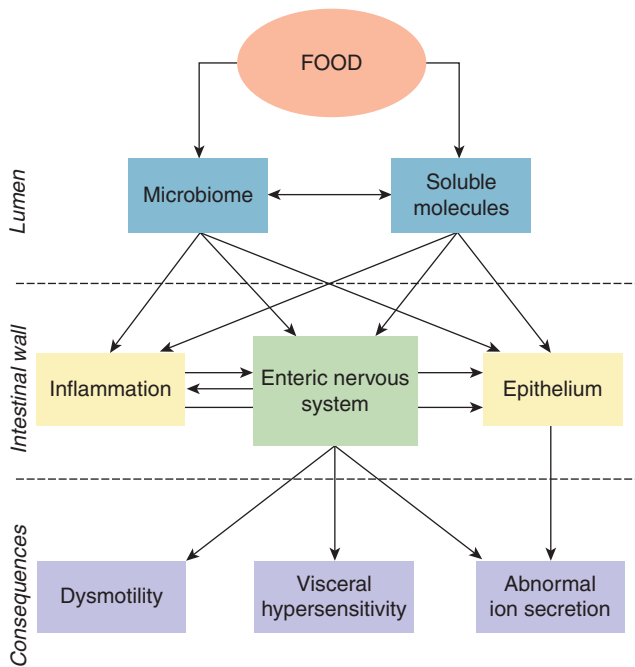
Figure 1. The complexity of potential interactions of food components with the intestinal wall and potential mechanisms for triggering of symptoms.

may trigger symptoms in non-celiacs by as yet unknown mechanisms. Specific immune-mediated reactions to food antigens and effects on the microbiota will not be addressed.

TARGETING LUMINAL DISTENSION—THE FODMAP APPROACH

Distension of the intestinal lumen results from changing volume of its contents. The luminal liquid content is dictated by the number of osmotically active particles present, since osmolality of the intestinal lumen maintained within the physiological range of the internal milieu. Gas in the distal small intestine and colon derived from swallowed air and that generated by fermentation of principal carbohydrates by bacteria. Food components that are small enough to be osmotically active yet able to be fermented readily by bacteria are short-chain carbohydrates that are poorly and/or slowly absorbed in the small intestine. Such molecules are plentiful in the diet and have been collectively termed as FODMAPs (36,37).

FODMAPS

There are several types of FODMAPs in the diet and they, together with common dietary sources, are listed in **Table 1**. Not all of the carbohydrates listed in the table behave as FODMAPs in specific individuals due to different small intestinal absorptive patterns. These are outlined in **Table 2**. However, FODMAPs share similar physiological effects, though the extent to which each exerts some those effects, such as osmotic effects and rapidity of fermentation, varies according to the chain length of the FODMAPs (**Table 3**).

In fasting patients with FBD, ingestion of pure FODMAPs, such as lactose, fructose, or sorbitol, alone or in combination, induce gut symptoms that mimic what they experience in association of their FBD (12). Very large doses will also cause symptoms in healthy persons, as evident after a large dose of lactulose or fructo-oligosaccharide (38). The same has been observed when diets high in FODMAPs are provided for patients with IBS, where significantly elevated symptoms are observed even on the first day of such a diet, as opposed to minimal or no symptoms in healthy volunteers (39). Whether actual fermentation of the ingested FODMAP is needed to induce the symptoms has recently been questioned. Mannitol in a dose of 10 g was well absorbed in a cohort of 20 patients with IBS possibly related to slow transit in this groups compared with the matched controls, but, despite this, symptoms were still induced (40). In the same study, sorbitol also induced symptoms but this bore no relationship to whether breath hydrogen responses (and, therefore, sorbitol malabsorption) were seen. These observations did not reflect "placebo" effects since blinded glucose induced minimal symptoms, but more likely reflected the small intestinal distension that has been demonstrated following the ingestion of mannitol due to its osmotic effect and slow absorption (41).

The definitive demonstration of the role of dietary FODMAPs in the induction of functional gut symptoms was reported in a randomized, placebo-controlled, double-blinded, quadruple arm crossover, rechallenge study of 25 patients with IBS and documented fructose malabsorption who previously had durable symptomatic response to reduction of FODMAPs in their diets (42). Variables associated with background diet were controlled by providing all food ingested in the study. The participants had a low FODMAP diet and were asked to take identically tasting drinks containing fructose, fructans, fructose, and fructans or glucose (as placebo) with food three times a day in a graded introduction. The doses were judged according to average intake in the community. Symptoms similar to those previously experienced were specifically induced in the FODMAP arms of the study in a dose-dependent way. Furthermore, there was evidence for an additive effect of fructose and fructans.

The low FODMAP diet—structure, efficacy, and implementation
The principles of the design of the low FODMAP diet are shown in **Figure 2** and have largely remained constant since its first design (43). However, the knowledge of food composition has been greatly expanded by the measurement of FODMAP content in a wide variety of Australian foods using a combination of high-performance liquid chromatography and enzymatic assays (44–46). This has led to modification of safe and unsafe food lists (**Table 1**). While most of the information is applicable worldwide, there is likely to be some variation of content of common foods and country-specific food types that have not been evaluated.

The evidence base for efficacy of the low FODMAP diet covers aspects that include challenge and rechallenge studies, observational studies, comparative studies, and randomized controlled trials. These are outlined in **Table 4**. Adherence to the diet has been assessed in two retrospective studies and these indicated durable adherence at least most of the time can be expected in up to 75% of patients (43,47).

Table 1. Some common food sources of FODMAPs (42,43,72)

Food type	Free fructose	Lactose	Fructans	Galacto-oligosaccharides	Polyols
Fruits	Apple, cherry, mango, pear, watermelon		Peach, persimmon, watermelon		Apple, apricot, pear, avocado, blackberries, cherry, nectarine, plum, prune
Vegetables	Asparagus, arti-chokes, sugar snap peas		Artichokes, beetroot, Brussels sprout, chicory, fennel, garlic, leek, onion, peas		Cauliflower, Mushroom, Snow peas
Grains and cereals			Wheat, rye, barley		
Nuts and seeds			Pistachios		
Milk and milk products		Milk, yoghurt, ice-cream, custard, soft cheeses			
Legumes			Legumes, lentils, chickpeas	Legumes, chickpeas, lentils	
Other	Honey, high-fructose corn syrup		Chicory drinks		
Food additives			Inulin, FOS		Sorbitol, mannitol, maltitol, xylitol, isomalt

FODMAPs, fermentable oligo-, di-, and mono-saccharides and polyols; FOS, fructo-oligosaccharides.

Additional applications of the low FODMAP diet

There are several clinical situations, in addition to adults with IBS and/or IBD, where reducing FODMAP intake might improve symptoms. The effectiveness in children needs formal evaluation, although anecdotal experience suggests efficacy. Patients with a high output ileostomy or an ileal pouch should benefit from a low FODMAP diet, as suggested by observations in patients without a colon (48,49). An important role of FODMAPs in inducing diarrhea during enteral nutrition has been postulated (50) and supported by a retrospective review of predictors of diarrhea (51). Unfortunately, nearly all enteral nutrition products on the market appear to have a high FODMAP content in a preliminary evaluation (52). This concept provides new challenges for the nutrition industry. The induction of fatigue with a high FODMAP diet in patients with IBS (39) raises the issue of the potential role of FOD-MAPs in chronic fatigue states (see **Table 3**).

Unanswered questions

There are many recommendations in the low FODMAP diet that are based upon clinical observation and have not been formally tested. These include the cutoff values that define "safe" from "unsafe" for the content of FODMAP classes (37,43). It is recommended that large doses of free fructose be avoided in all patients (37,43). The concept is that fructose in excess of glucose is slowly absorbed along the entire small intestine and will, therefore, exert an osmotic effect in the small intestine, increasing the volume of fluid in its lumen even when fructose malabsorption is not evident on breath hydrogen testing. In support of this, mannitol, also a slowly absorbed small molecule, increases the volume of fluid in the small bowel lumen and has recently been shown to induce symptoms even without evidence of malabsorption (see above) (40). However, this concept has not been

formally tested for fructose. At present, there is limited understanding of how to predict who will be more likely to respond to the diet before its initiation, so that there might be more efficient targeting of the diet. In the IBD populations, higher educational status, working part-time, and the use of recommended cookbooks were associated with better adherence and response (47). Correlations of symptom patterns and pathophysiology (such as the presence of visceral hypersensitivity) with response have yet to be evaluated.

The efficacy of different methods of patient education has not been formally evaluated. At the present time, the only method used has been the delivery of the dietary education by a trained dietician, together with the use of written educational material, recipe books, and group education sessions (37,43). Whether the diet can be successfully initiated via information sheets only or via detailed publications of the dietary approach (53) has not been tested.

The safety of long-term FODMAP restriction has not been formally reported. Restrictive diets are at risk of being nutritionally inadequate. As the diet discourages excluding whole food groups, it is anticipated that the low FODMAP diet would not compromise nutritional adequacy. Foods are substituted with a suitable alternative form within the same food group family. The difficult food group is legumes (including the baked beans, red kidney beans, lentils, and chickpeas) as these all contain fructans and galacto-oligosaccharides. Fortunately, people on a low FODMAP diet can still enjoy tofu, and foods such as seeds, nuts, and quinoa are encouraged, as well as eating legumes in small amounts. Dietary fiber intake may potentially reduce because of the restriction of wheat products. There is no information regarding the safety of long-term restriction of oligosaccharides with regard to the potential loss of the putative prebiotic benefits to health, such as prevention of colorectal cancer (12,54).

Table 2. Absorptive patterns of different FODMAPs

Absorption pattern	FODMAP	Mechanisms	Assessment of individual's capacity
Poorly absorbed in some only	Lactose (sucrose)	Deficiency of disaccharidases • Lactase deficiency common depending on ethnicity • Sucrose-isomaltase deficiency rare	Breath hydrogen test Lactose tolerance test Hydrolase activity in duodenal biopsy
Slowly absorbed in all	Free fructose (fructose in excess of glucose), Polyols (e.g., sorbitol, mannitol), Xylose	Low capacity absorptive mechanisms: GLUT5 facultative transporter for fructose (12); passive diffusion for polyols and xylose. Degree of absorption dependent upon • individual capacity of transporter or passive diffusion • small intestinal transit time • small intestinal microbiota (bacterial overgrowth) (73,74)	Breath hydrogen test • Large dose fructose (35 g) used to determine those who have more efficient absorption (no breath hydrogen rise)—about 60%) (73) • 10 g sorbitol, 10 g mannitol – clinical value determined as slowly absorbed in all and symptom induction independent of positive hydrogen production (48)
Poorly absorbed in all	Fructans, Galacto-oligosaccharides	No small intestinal hydrolases to split fructose-fructose and galactose-galactose bonds	Not applicable

FODMAPs, fermentable oligo-, di-, and mono-saccharides and polyols.

Table 3. Physiological effects of FODMAPs

Effect	Evidence
Osmotic effect	• Ileostomy output increases in proportion to recovered FODMAPs (48) • Increased liquid content of small intestine after mannitol by magnetic resonance imaging (41) • Severity of diarrhea directly proportional to the chain length of FODMAPs (38)
Bacterial fermentation	• Marked increase in breath hydrogen over entire day with high vs. low FODMAP diet; also reduced methane production (39) • Rapidity of hydrogen production inversely proportional to chain length of FODMAPs (75)
Motility effects: acceleration of small intestinal and colonic transit; increase in gastroesophageal reflux	• Osmotic effect of FODMAPs (38,76) • Neural feedback pathways and/or hormonal changes from short-chain fatty acid production secondary to bacterial fermentation (12,77,78)
Prebiotic effects	• Oligosaccharides in small doses preferentially metabolized by bifidobacteria and lactobacilli (79)
Systemic effects	• Mild depression in young women associated with lower serum tryptophan concentrations (80) • Tiredness induced in patients with irritable bowel syndrome by high but not by low FODMAP diet (39) • Toxic metabolites of malabsorbed FODMAPs postulated (72)

FODMAPs, fermentable oligo-, di-, and mono-saccharides and polyols.

Additional questions regarding the intake of dietary FODMAPs in patients with FBD or other intestinal diseases have yet to be addressed. For example, it is unclear as yet whether excessive intake of FODMAPs is associated with a higher risk of IBS or Crohn's disease, as previously hypothesized (36). To facilitate such studies, a comprehensive Food Frequency Questionnaire detailing the FODMAP content of an extensive range of fruits, vegetables, grains, and cereal foods has been developed and validated as a potentially useful tool for population studies (55).

TARGETING FOOD CHEMICALS

Plants are the major chemical factories. They use chemicals, for example, for protection (bad taste), reproduction (colors and odors attracting pollinating bees), and antibacterial or preservative functions. Potentially bioactive chemicals are found in most food types. Some common sources rated "very high" in chemicals are shown in **Table 5** and can be sourced in detail from http://www.allergy.net.au. They include salicylates (monohydroxybenzoates) that are found widely in plants and have a protective role, their concentrations reducing with ripening; amines

and glutamates that are products of protein breakdown, their concentrations increasing with aging; and food additives such as glutamates to enhance flavor, benzoates, sulfites, and nitrates as preservatives, and various colors. In general, the stronger the flavor of the food, the higher the chemical content will be. In clinical practice, food chemicals have received some attention in the pathogenesis and management of urticaria, headaches, asthma, and anaphylactoid reactions. They are believed to induce problems by non-immune mechanisms (see below). Patients with these non-GI conditions often have associated gut symptoms and observations that these improve in concert with the other manifestations have led to extending the food chemical dietary approach to patients with functional gut symptoms without non-GI manifestations. Benefits of applying elimination diet-rechallenge methodologies are claimed (56) and a detailed guide to this approach has been published (57), but the evaluation of response rates has not been reported.

As outlined above, food chemicals are major afferent stimuli to the ENS. In the presence of visceral hypersensitivity, normal physiological stimulation by such chemicals might result in exaggerated effector responses, as hypothesized for luminal distension. Perhaps
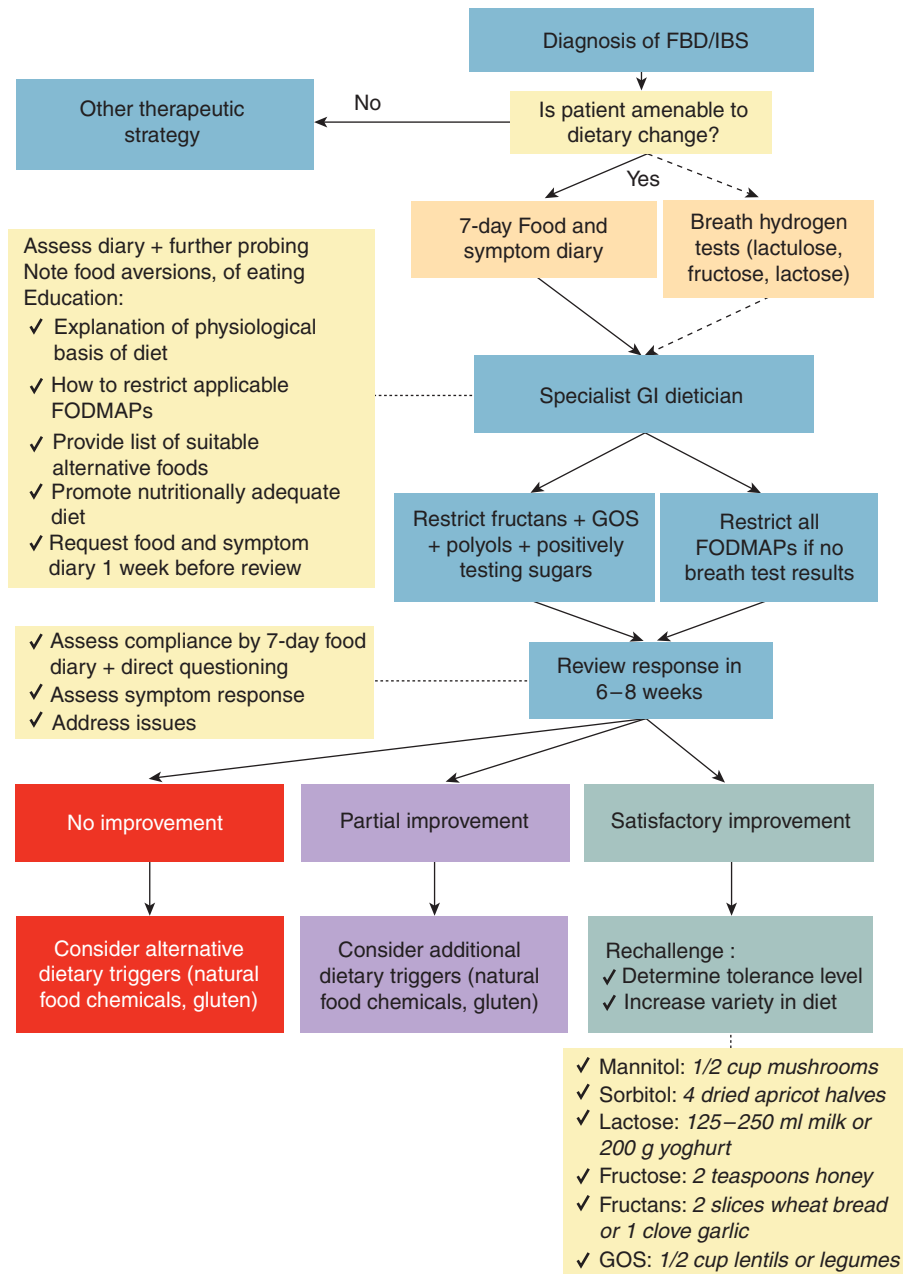
Figure 2. A dietary strategy for the management of patients with functional bowel disease (FBD)/irritable bowel syndrome (IBS). FODMAPs, fermentable oligo-, di-, and mono-saccharides and polyols; GI, gastrointestinal; GOS, galacto-oligosaccharides.

of more interest and importance is that food chemicals may themselves induce visceral hypersensitivity. Plant chemicals are able to activate TRP channels (33), which appear to be central to visceral hypersensitivity, at least in an animal model (18). It is possible that chronic exposure to certain chemicals will lead to increased expression of TRP channels and that this contributes via heightening of the sensitivity of the ENS to the development of functional gut symptoms. Likewise, withdrawal of the offending chemicals from the diet may lead to reversal of the TRP channel overexpression and subsequent resolution of the gut symptoms. Unfortunately, this mechanistic concept has not been directly evaluated.

There are other potential mechanisms by which food chemicals might be causally related to gut symptoms. Salicylates are found in most plant foods and are well documented to be a trigger of non-GI illness such as anaphylactoid reactions, urticaria, and asthma in susceptible individuals (56,58). Such reactions are not immune mediated and may involve direct effect on mast cells to produce cysteinyl leukotrienes (59), which are pro-inflammatory, promote smooth muscle contraction, and increase vascular permeability. Whether salicylates in food have such an effect on mast cells in the gut has not been studied. What underlies the individual susceptibility to salicylates is also unknown.

Table 4. Evidence base for the efficacy of the low FODMAP diet in relieving functional gastrointestinal symptoms

Type of study	Methodology	Results	References
Challenge	Challenge with individual or combination of FOD-MAPs; some randomized, some placebo controlled	Fructose, fructans, sorbitol, mannitol, lactose, fructose and sorbitol, fructans and lactose induce symptoms in acute challenge in patients with IBS >> healthy control	Reviewed in (12,40)
Randomized placebo-controlled rechallenge	Randomized, placebo-controlled, double-blinded, quadruple arm cross-over, rechallenge with fructose, fructans, fructose, and fructans or glucose (as placebo) in 25 patients with IBS and documented fructose malabsorption who previously had durable symptomatic response to reduction of dietary FODMAPs	Fructose and fructans specifically induced symptoms that mimicked IBS symptoms; fructose and fructans had additive effect. Supported the concept of restriction of FODMAPs in general rather than restriction of one component alone	(42)
Observational cohort studies	Retrospective studies of efficacy of instruction in low FODMAP diet in patients with IBS and fructose malabsorption, IBS and no breath hydrogen testing, inflammatory bowel disease, or patients without a colon (ileal pouch, ileostomy)	Consistent response of 75% (IBS), 50–75% (IBD) of patients across all symptoms. Adherence to a predictor of efficacy. In IBD, higher educational status, working part-time and use of cookbooks predicted efficacy	(43,47,49,81)
Comparative studies	Comparison of low FODMAP diet with standard dietary approach. Non-randomized	Significantly better response with low FODMAP across all functional gut symptoms	(82)
Randomized controlled trials	Two-day intervention of low versus high FODMAP diet (food all supplied) in 15 patients with IBS and 15 healthy controls	Significantly greater symptoms (functional gut and tiredness) with high FODMAP diet in IBS patients (not controls)	(38)

FODMAPs, fermentable oligo-, di-, and mono-saccharides and polyols; IBS, irritable bowel syndrome.

The only food chemical that has been systematically studied with respect to gut symptoms is salicylates and related molecules such as non-steroidal anti-inflammatory drugs where, for example, 2–4% of patients with inflammatory bowel disease or food allergies were identified to be salicylate/non-steroidal anti-inflammatory drug intolerant by historical and/or rechallenge methodologies (60). Identifying individuals susceptible to chemicals in general or specifically at present can only be achieved with exclusion diet placebo-controlled-rechallenge methodology. There have been suggestions that chemical sensitivity can be assessed *in vitro* with high specificity and positive predictive value, and fair sensitivity by the basophil activation test. This has been shown in relation to salicylates (61,62), although not universally accepted (63).

SUSPECTED MOLECULES WITHOUT A KNOWN MECHANISM—GLUTEN

There is little doubt that gluten is causally associated with celiac disease. There has been a strong belief that gluten causes other illnesses, including FBD, in some patients with no features of celiac disease. However, this so-called "non-celiac gluten intolerance" (NCGI) has only recently been formally studied. The wheat-induced gut symptoms in non-celiacs have been used as evidence of NCGI in the past, but the elucidation of one of the carbohydrate moieties in wheat, fructans, as a potent trigger for functional gut symptoms has made any such conclusions invalid. Likewise, demonstration of efficacy of gluten-free diet in patients with HLA-DQ2/DQ8 who did not have villous atrophy on duodenal biopsy, but may have had celiac-associated antibodies also does not convincingly define patients with NCGI, especially as many had intraepithelial lymphocytosis in the duodenum and

typical lesions may have been missed because of the patchy nature of the intestinal involvement in celiac disease (64,65). Study of mice expressing HLA-DQ8 has demonstrated that gluten can lead to changes in neuromotor function and microbiota independently of inducing intestinal inflammation or injury (66).

The strongest evidence that NCGI does indeed exist in humans derived from a double-blind randomized rechallenge trial of FOD-MAP-depleted gluten in 34 patients who had celiac disease excluded on best available criteria (one half were HLA-DQ2/DQ8 negative) and who had IBS that had markedly improved on a gluten-free diet to which the patients were adherent on dietary assessment (67). Muffins and bread containing the gluten or not (placebo) were fed to the patients over 6 weeks in addition to the patient remaining on their usual gluten-free diet. The muffins and bread from each arm were indistinguishable, as the gluten used had lost baking functionality. Within the first week of the intervention period and maintained through the 6 weeks, the gluten-treated subjects had significantly greater gut symptoms and tiredness than did the placebo group. A mechanism for the difference was not identified; inflammation was not evident on fecal lactoferrin, intestinal permeability did not change, systemic inflammation (as judged by highly sensitive C-reactive protein) was not observed, and celiac-associated antibodies did not emerge. The HLA-DQ status did not influence the results. One can speculate on likely mechanism(s) (68,69), but further study is indicated.

While this was only a small study, it did provide the first evidence of the existence of NCGI. Many have seen this as legitimizing claims made for many years by alternative practitioners and the public. However, before this can be applied in clinical practice, a few issues need to be addressed. First, like all initial reports, the findings need to be reproduced. Second, the prevalence of NCGI

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Table 5. Examples of food sources containing very high amounts of salicylates, amines, or glutamates (source: http://www.allergy.net.au)

	Salicylates	Amines	Glutamates
Fruits	Avocado, berries, blackcurrant, cherry, citrus, date, grape, kiwifruit, passionfruit, pineapple, plum, tomato		Dried prunes, raisins, sultanas, grapes, plum, tomato
	Strawberry	Redcurrant	
Vegetables	Pickled vegetables, mushrooms, sauerkraut, spinach, tomato		
	Capsicum, chicory, chilli, eggplant, ginger, herbs, spices, onion	Eggplant, olives	
Grains and cereals	Breakfast cereals, mueslis and bread containing cocoa, coconut, dried fruit, honey or nuts, pasta, noodles, rice crackers, biscuits, cakes, muesli bars, pastries containing chocolate, coconut, fruit, nuts, ham or spices, flavored snack foods, hot potato chips, potato chips		
Nuts	Almond meal, hazelnut meal, marzipan, nut paste (all), peanut butter, roasted nuts (all), coconut (desiccated, shredded)		—
Seeds	Mustard seeds, sesame seed paste (tahini)		—
Milk and milk products	Milk with chocolate, strawberry or banana flavor, flavored cheeses, yoghurt		
		Brie, camembert, parmesan, tasty cheeses	Brie, camembert, parmesan
Legumes	Bean mixes, broad beans, felafel, textured vegetable protein		Canned baked beans in sauce, textured vegetable protein
	Canned baked beans in sauce	Surimi, miso, soy sauce, tamari, tempeh	
Meat, fish, chicken	Beef: aged, corned, dried, smoked; chicken: nuggets, smoked; devon, sausages; meat pastes, meat pies, offal, salami; fish pastes		
	Gravy, chicken: pressed, seasoned	Ham, bacon; fish: pickled, salted, smoked; anchovies, canned fish roe, prawns tuna	Beef: biltong, jerky; chicken: pressed, seasoned, gravy
Fats and oils	Almond oil, avocado oil, extra virgin olive oil, sesame oil, walnut oil		
Other	Jam, lemon butter, marmalade, maple-flavored syrup, fruit flavored sweets, yeast extract, fermented products, chicken salt, vinegar (cider, red, white wine), stock (cubes, powders)		Jam, fruit flavored sweets, stock (cubes, powders) yeast extract, fermented products, chicken salt, sauces (including tomato, soy, fish, and oyster)
	Honey, peppermints, liquorice, yeast extract, tomato sauce, soy sauce	Chocolate, sauces (including tomato, soy, fish, and oyster)	
Beverages	Cordials, flavored mineral waters, spirits (except gin, tonic, whisky, vodka), wine		
	Fruit juices, ginger beer, vegetable juice, beer, champagne, cider		
	Apple cider, herbal teas, tea	Chocolate drinks, cocoa powder	

in an IBS population needs to be defined. The patient group from that report was highly selected. From the use of gluten-free diet in the population at large, it might be anticipated that NCGI is common. However, in a survey of 132 patients with self-perceived NCGI who responded to advertising, two in three did not have celiac disease adequately excluded, one in three did not follow a strict gluten-free diet in any case, and one in four had uncontrolled symptoms despite a gluten-free diet (70). This survey indicated that there is likely to be a major difference in perceived vs. actual NCGI. Third, a mechanism needs to be defined (as outlined above) so that biomarkers of NCGI can be developed. At the present time, the only diagnostic process for NCGI is to exclude celiac disease by HLA-DQ genotype or serology and duodenal biopsy on a gluten-rich diet, followed by symptomatic marked improvement on a gluten-free diet, followed by blinded placebo-controlled rechallenge.

This is not a methodology that works well with population studies. Fourth, practical issues such as whether the gluten-mediated effect is all-or-none (like in celiac disease) or a dose-related phenomenon. This is of relevance since a strict gluten-free diet may not meet all nutritional requirements (71). Thus, there is no clinical evidence yet to recommend a gluten-free diet as first-line dietary therapy in patients with IBS.

CONCLUSION

Simple concepts of how food might trigger functional GI symptoms have led to at least one efficacious dietary approach that is effective in the majority of patients with FBD. The evidence base for the low FODMAP diet is strong, provided dieticians with the skills to implement it are available. Other approaches, most of

which are, at least theoretically, complementary to the low FOD-MAP diet, are being actively pursued in clinical practice, if not always in scientific enquiry. There is a real need for biomarkers or other clinical predictors to enable individualization of the dietary approach, particularly as many diets require the use of elimination diet methodologies. Gastroenterologists can no longer ignore specific dietary intervention for patients with functional gut symptoms.

CONFLICT OF INTEREST

Guarantor of the article: Peter R. Gibson, MD.

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REFERENCES

1. Saito YA, Locke III GR, Weaver AL et al. Diet and functional gastrointestinal disorders: a population-based case-control study. Am J Gastroenterol 2005;100:2743–8.
2. Monsbakken KW, Vandvik PO, Farup PG. Perceived food intolerance in subjects with irritable bowel syndrome—etiology, prevalence and consequences. Eur J Clin Nutr 2006;60:667–72.
3. Jamieson AE, Fletcher PC, Schneider MA. Seeking control through the determination of diet: a qualitative investigation of women with irritable bowel syndrome and inflammatory bowel disease. Clin Nurse Spec 2007;21:152–60.
4. Mullin GE, Swift KM, Lipski L et al. Testing for food reactions: the good, the bad, and the ugly. Nutr Clin Pract 2010;25:192–8.
5. Kanazawa M, Fukudo S. Effects of fasting therapy on irritable bowel syndrome. Int J Behav Med 2006;13:214–20.
6. Dainese R, Galliani EA, De Lazzari F et al. Discrepancies between reported food intolerance and sensitization test findings in irritable bowel syndrome patients. Am J Gastroenterol 1999;94:1892–7.
7. Bischoff SC, Herrmann A, Manns MP. Prevalence of adverse reactions to food in patients with gastrointestinal disease. Allergy 1996;51:811–8.
8. Niec AM, Frankum B, Talley NJ. Are adverse food reactions linked to irritable bowel syndrome? Am J Gastroenterol 1998;93:2184–90.
9. James SL, Muir JG, Curtis SL et al. Dietary fibre: a roughage guide. Intern Med J 2003;33:291–6.
10. Ford AC, Talley NJ, Spiegel BM et al. Effect of fibre, antispasmodics, and peppermint oil in the treatment of irritable bowel syndrome: systematic review and meta-analysis. Br Med J 2008;337:a2313.
11. Brandt LJ, Chey WD, Foxx-Orenstein AE et al. An evidence-based position statement on the management of irritable bowel syndrome. Am J Gastroenterol 2009;104 (Suppl 1): S1–S35.
12. Gibson PR, Newnham E, Barrett JS et al. Review article: fructose malabsorption and the bigger picture. Aliment Pharmacol Ther 2007;25:349–63.
13. Austin GL, Dalton CB, Hu Y et al. A very low-carbohydrate diet improves symptoms and quality of life in diarrhea-predominant irritable bowel syndrome. Clin Gastroenterol Hepatol 2009;7:706–8.
14. Sternini C, Anselmi L, Rozengurt E. Enteroendocrine cells: a site of "taste" in gastrointestinal chemosensing. Curr Opin Endocrinol Diabetes Obes 2008;15:73–8.
15. Schemann M, Mazzuoli G. Multifunctional mechanosensitive neurons in the enteric nervous system. Auton Neurosci 2010;153:21–5.
16. Boesmans W, Owsianik G, Tack J et al. TRP channels in neurogastroenterology: opportunities for therapeutic intervention. Br J Pharmacol 2011;162:18–37.
17. Holzer P. TRP channels in the digestive system. Curr Pharm Biotechnol 2011;12:24–34.
18. Winston J, Shenoy M, Medley D et al. The vanilloid receptor initiates and maintains colonic hypersensitivity induced by neonatal colon irritation in rats. Gastroenterology 2007;132:615–27.
19. Chan CL, Facer P, Davis JB et al. Sensory fibres expressing capsaicin receptor TRPV1 in patients with rectal hypersensitivity and faecal urgency. Lancet 2003;361:385–91.
20. Akbar A, Yiangou Y, Facer P et al. Increased capsaicin receptor TRPV1-expressing sensory fibres in irritable bowel syndrome and their correlation with abdominal pain. Gut 2008;57:923–9.
21. Akbar A, Yiangou Y, Facer P et al. Expression of the TRPV1 receptor differs in quiescent inflammatory bowel disease with or without abdominal pain. Gut 2010;59:767–74.
22. Rozengurt E, Sternini C. Taste receptor signaling in the mammalian gut. Curr Opin Pharmacol 2007;7:557–62.
23. Kusano M, Zai H, Hosaka H et al. New frontiers in gut nutrient sensor research: monosodium L-glutamate added to a high-energy, high-protein liquid diet promotes gastric emptying: a possible therapy for patients with functional dyspepsia. J Pharmacol Sci 2010;112:33–6.
24. Holzer P. Acid sensing by visceral afferent neurones. Acta Physiol (Oxf) 2011;201:63–75.
25. Kaji I, Karaki S, Kuwahara A. Chemosense for luminal environment in the large intestine. Yakugaku Zasshi 2011;131:1691–8.
26. Akiho H, Ihara E, Nakamura K. Low-grade inflammation plays a pivotal role in gastrointestinal dysfunction in irritable bowel syndrome. World J Gastrointest Pathophysiol 2010;1:97–105.
27. Yiangou Y, Facer P, Dyer NH et al. Vanilloid receptor 1 immunoreactivity in inflamed human bowel. Lancet 2001;357:1338–9.
28. O'Malley D, Liston M, Hyland NP et al. Colonic soluble mediators from the maternal separation model of irritable bowel syndrome activate submucosal neurons via an interleukin-6-dependent mechanism. Am J Physiol Gastrointest Liver Physiol 201;300:G241–52.
29. Watson RP, Lilley E, Panesar M et al. Increased prokineticin 2 expression in gut inflammation: role in visceral pain and intestinal ion transport. Neurogastroenterol Motil 2012;24:65–e12.
30. Barbara G, Stanghellini V, De Giorgio R et al. Activated mast cells in proximity to colonic nerves correlate with abdominal pain in irritable bowel syndrome. Gastroenterology 2004;126:693–702.
31. De Winter BY, van den Wijngaard RM, de Jonge WJ. Intestinal mast cells in gut inflammation and motility disturbances. Biochim Biophys Acta 2012;1822:66–73.
32. Surawicz CM. Mechanisms of diarrhea. Curr Gastroenterol Rep 2010;12:236–41.
33. Vriens J, Nilius B, Vennekens R. Herbal compounds and toxins modulating TRP channels. Curr Neuropharmacol 2008;6:79–96.
34. Kano M, Fukudo S, Kanazawa M et al. Changes in intestinal motility, visceral sensitivity and minor mucosal inflammation after fasting therapy in a patient with irritable bowel syndrome. J Gastroenterol Hepatol 2006;21:1078–9.
35. Zar S, Mincher L, Benson MJ et al. Food-specific IgG4 antibody-guided exclusion diet improves symptoms and rectal compliance in irritable bowel syndrome. Scand J Gastroenterol 2005;40:800–7.
36. Gibson PR, Shepherd SJ. Personal view: food for thought-western lifestyle and susceptibility to Crohn's disease. The FODMAP hypothesis. Aliment Pharmacol Ther 2005;21:1399–409.
37. Gibson PR, Shepherd SJ. Evidence-based dietary management of functional gastrointestinal symptoms: the FODMAP approach. J Gastroenterol Hepatol 2010;25:252–8.
38. Clausen MR, Jørgensen J, Mortensen PB. Comparison of diarrhea induced by ingestion of fructooligosaccharide Idolax and disaccharide lactulose: role of osmolarity versus fermentation of malabsorbed carbohydrate. Dig Dis Sci 1998;43:2696–707.
39. Ong DK, Mitchell SB, Barrett JS et al. Manipulation of dietary short chain carbohydrates alters the pattern of gas production and genesis of symptoms in irritable bowel syndrome. J Gastroenterol Hepatol 2010;25:1366–73.
40. Yao CK, Tan HL, Langenberg DR et al. Abnormal intestinal handling of sorbitol and mannitol in patients with IBS: mechanistic insights and potential clinical implications. J Gastroenterol Hepatol 2011;26 (Suppl 4): 70.
41. Marciani L, Cox EF, Hoad CL et al. Postprandial changes in small bowel water content in healthy subjects and patients with irritable bowel syndrome. Gastroenterology 2010;138:469–77.
42. Shepherd SJ, Parker FC, Muir JG et al. Dietary triggers of abdominal symptoms in patients with irritable bowel syndrome: randomized placebo-controlled evidence. Clin Gastroenterol Hepatol 2008;6:765–71.

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43. Shepherd SJ, Gibson PR. Fructose malabsorption and symptoms of irritable bowel syndrome: guidelines for effective dietary management. J Am Diet Assoc 2006;106:631–9.

44. Muir JG, Shepherd SJ, Rosella O et al. Fructan and free fructose content of common Australian vegetables and fruit. J Agric Food Chem 2007;55:6619–27.

45. Muir JG, Rose R, Rosella O et al. Measurement of short-chain carbohydrates in common Australian vegetables and fruits by high-performance liquid chromatography (HPLC). J Agric Food Chem 2009;57:554–65.

46. Biesiekierski JR, Rosella O, Rose R et al. Quantification of fructans, galacto-oligosaccharides and other short-chain carbohydrates in processed grains and cereals. J Hum Nutr Diet 2011;24:154–76.

47. Gearry RB, Irving PM, Barrett JS et al. Reduction of dietary poorly absorbed short-chain carbohydrates (FODMAPs) improves abdominal symptoms in patients with inflammatory bowel disease-a pilot study. J Crohns Colitis 2009;3:8–14.

48. Barrett JS, Gearry RB, Muir JG et al. Dietary poorly absorbed, short-chain carbohydrates increase delivery of water and fermentable substrates to the proximal colon. Aliment Pharmacol Ther 2010;31:874–82.

49. Croagh C, Shepherd SJ, Berryman M et al. Pilot study on the effect of reducing dietary FODMAP intake on bowel function in patients without a colon. Inflamm Bowel Dis 2007;13:1522–8.

50. Barrett JS, Shepherd SJ, Gibson PR. Strategies to manage gastrointestinal symptoms complicating enteral feeding. JPEN J Parenter Enteral Nutr 2009;33:21–6.

51. Halmos EP, Muir JG, Barrett JS et al. Diarrhoea during enteral nutrition is predicted by the poorly absorbed short-chain carbohydrate (FODMAP) content of the formula. Aliment Pharmacol Ther 2010;32:925–33.

52. Halmos EP, Liels KL, Rosella O et al. Enteral nutritional formulas deliver laxative doses of FODMAPs which cannot be predicted by ingredients lists. J Gastroenterol Hepatol 2011;26 (Suppl 4): 73.

53. Shepherd SJ, Gibson PR. Food intolerance management plan. Penguin: Melbourne, 2011.

54. Sengupta S, Muir JG, Gibson PR. Does butyrate protect from colorectal cancer? J Gastroenterol Hepatol 2006;21:201–18.

55. Barrett JS, Gibson PR. Development and validation of a comprehensive semi-quantitative food frequency questionnaire that includes FODMAP intake and glycemic index. J Am Diet Assoc 2010;110:1469–76.

56. Allen DH, Van Nunen S, Loblay R et al. Adverse reactions to foods. Med J Aust 1984;141 (5 Suppl): S37–42.

57. Swain A, Scoutter V, Loblay R. RPAH Elimination Diet Handbook. Allergy Unit Royal Prince Alfred Hospital: Sydney, 2011. www.allergy.net.au.

58. Grattan CE. Aspirin sensitivity and urticaria. Clin Exp Dermatol 2003;28:123–7.

59. Suzuki Y, Ra C. Analysis of the mechanism for the development of allergic skin inflammation and the application for its treatment: aspirin modulation of IgE-dependent mast cell activation: role of aspirin-induced exacerbation of immediate allergy. Pharmacol Sci 2009;110:237–44.

60. Raithel M, Baenkler HW, Naegel A et al. Significance of salicylate intolerance in diseases of the lower gastrointestinal tract. J Physiol Pharmacol 2005;56 (Suppl 5): 89–102.

61. Gamboa P, Sanz ML, Caballero MR et al. The flow-cytometric determination of basophil activation induced by aspirin and other non-steroidal anti-inflammatory drugs (NSAIDs) is useful for in vitro diagnosis of the NSAID hypersensitivity syndrome. Clin Exp Allergy 2004;34:1448–57.

62. Rodríguez-Trabado A, Cámara-Hijón C, Ramos-Cantariño A et al. Basophil activation test for the in vitro diagnosis of nonsteroidal anti-inflammatory drug hypersensitivity. Allergy Asthma Proc 2008;29:241–9.

63. Celik GE, Schroeder JT, Hamilton RG et al. Effect of in vitro aspirin stimulation on basophils in patients with aspirin-exacerbated respiratory disease. Clin Exp Allergy 2009;39:1522–31.

64. Wahnschaffe U, Ullrich R, Riecken EO et al. Celiac disease-like abnormalities in a subgroup of patients with irritable bowel syndrome. Gastroenterology 2001;121:1329–38.

65. Wahnschaffe U, Schulzke JD, Zeitz M et al. Predictors of clinical response to gluten-free diet in patients diagnosed with diarrhea-predominant irritable bowel syndrome. Clin Gastroenterol Hepatol 2007;5:844–50.

66. Verdu EF, Huang X, Natividad J et al. Gliadin-dependent neuromuscular and epithelial secretory responses in gluten-sensitive HLA-DQ8 transgenic mice. Am J Physiol Gastrointest Liver Physiol 2008;294:G217–25.

67. Biesiekierski JR, Newnham ED, Irving PM et al. Gluten causes gastrointestinal symptoms in subjects without celiac disease: a double-blind randomized placebo-controlled trial. Am J Gastroenterol 2011;106:508–14.

68. Newnham ED. Does gluten cause gastrointestinal symptoms in subjects without coeliac disease? J Gastroenterol Hepatol 2011;26 (Suppl 3): 132–4.

69. Verdu EF. Editorial: can gluten contribute to irritable bowel syndrome? Am J Gastroenterol 2011;106:516–8.

70. Biesiekierski JR, Newnham ED, Shepherd SJ et al. Self-diagnosis of non-coeliac gluten intolerance by Australian adults: failure to exclude coeliac disease or benefit from a gluten-free diet. J Gastroenterol Hepatol 2011;26 (Suppl 4): 70.

71. Stuckey C, Lowdon J, Howdle P. Joint BAPEN and British Society of Gastroenterology Symposium on 'Coeliac disease: basics and controversies'. Dietitians are better than clinicians in following up coeliac disease. Proc Nutr Soc 2009;68:249–51.

72. Campbell AK, Matthews SB, Vassel N et al. Bacterial metabolic "toxins": a new mechanism for lactose and food intolerance, and irritable bowel syndrome. Toxicology 2010;278:268–76.

73. Barrett JS, Irving PM, Shepherd SJ et al. Comparison of the prevalence of fructose and lactose malabsorption across chronic intestinal disorders. Aliment Pharmacol Ther. 2009;30:165–74.

74. Nucera G, Gabrielli M, Lupascu A et al. Abnormal breath tests to lactose, fructose and sorbitol in irritable bowel syndrome may be explained by small intestinal bacterial overgrowth. Aliment Pharmacol Ther 2005;21:1391–5.

75. Brighenti F, Casiraghi MC, Pellegrini N et al. Comparison of lactulose and inulin as reference standard for the study of resistant starch fermentation using hydrogen breath test. Ital J Gastroenterol 1995;27:122–8.

76. Madsen JL, Linnet J, Rumessen JJ. Effect of nonabsorbed amounts of a fructose-sorbitol mixture on small intestinal transit in healthy volunteers. Dig Dis Sci 2006;51:147–53.

77. Piche T, Zerbib F, Varannes SB et al. Modulation by colonic fermentation of LES function in humans. Am J Physiol Gastrointest Liver Physiol 2000;278:G578–84.

78. Piche T, des Varannes SB, Sacher-Huvelin S et al. Colonic fermentation influences lower esophageal sphincter function in gastroesophageal reflux disease. Gastroenterology 2003;124:894–902.

79. Kolida S, Gibson GR. Prebiotic capacity of inulin-type fructans. J Nutr 2007;137 (11 Suppl): 2503S–6S.

80. Ledochowski M, Widner B, Murr C et al. Fructose malabsorption is associated with decreased plasma tryptophan. Scand J Gastroenterol 2001;36:367–71.

81. Shepherd SJ, Willet I, Fone D et al. The value of "fructose malabsorption" (FM) diet and fructose breath hydrogen test in patients with irritable bowel syndrome (IBS). J Gastroenterol Hepatol 2003;18 (Suppl): B26.

82. Staudacher HM, Whelan K, Irving PM et al. Comparison of symptom response following advice for a diet low in fermentable carbohydrates (FODMAPs) versus standard dietary advice in patients with irritable bowel syndrome. J Hum Nutr Diet 2011;24:487–95.