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Glucose breath test for the detection of small intestine bacterial overgrowth: Impact of diet prior to the test

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Abstract

Background: Glucose breath test (GBT) is used for the diagnosis of small intestine bacterial overgrowth. A restrictive diet without fibers and/or fermentable food is recommended on the day before the test. The aim of our retrospective study was to evaluate the impact of two different restrictive diets on the results of GBT.

Methods: A change of the pretest restrictive diet was applied in our lab on September 1, 2020. The recommended diet was a fiber-free diet before this date, and a fiber-free diet plus restriction of all fermentable food afterward. We thus compared the results of GBT performed before (group A) and after (group B) this pretest diet modification. Demographics, reasons to perform GBT, digestive symptoms, and hydrogen and methane baseline values and variations after glucose ingestion were compared between the two groups.

Key Results: 269 patients underwent GBT in group A, and 316 patients in group B. The two groups were comparable in terms of demographics. Methane and hydrogen baseline values were significantly higher in group A (respectively 14 [18] vs. 8 [14] ppm, $p < 0.01$ and 11 [14] vs. 6 [8] ppm, $p < 0.01$). The percentage of positive tests was higher in group A for methane (43% vs. 28%, $p < 0.05$), and for hydrogen (18% vs. 12%, $p = 0.03$).

Conclusion & Inferences: This retrospective study suggests the importance of the restrictive diet prior to GBT. A strict limitation of fibers and fermentable food decreased hydrogen and methane baseline values, and the prevalence of positive GBT. Thus a strict restrictive diet should be recommended on the day before the test, in order to limit the impact of food on hydrogen and methane breath levels, and possibly improve the diagnosis quality of GBT.

KEYWORDS

diet, glucose breath test, irritable bowel syndrome, procedure, SIBO

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1 | INTRODUCTION

Small intestinal bacterial overgrowth (SIBO) and intestinal methanogen overgrowth (IMO) are defined as abnormalities of the intestinal microbial flora associated with excessive production of respectively hydrogen or methane by certain categories of bacteria and archaea, either at baseline or after ingestion of specific carbohydrates.¹ Glucose or lactulose breath tests, with measurement of hydrogen and methane levels in exhaled gases, represent the most frequently used tests for the diagnosis of intestinal microbial overgrowth, despite their known relatively poor sensibility and specificity.²⁻⁴ Jejunal aspiration and culture is currently considered as the reference method for the diagnosis of SIBO, but remains relatively invasive, and cumbersome due to the difficulties of anaerobic cultures.⁵ New techniques allowing the molecular analysis of duodeno-jejunal aspirates are promising, but do not appear correlated to the results of jejunal aspirate cultures.⁶

SIBO or IMO may be expressed clinically by multiple symptoms, similar in many ways to irritable bowel syndrome (IBS). Because there are no specific therapy for IBS, SIBO or IMO diagnosis is attractive to patients because of the possibility of adapted therapies such as a low FODMAP diet, or antibiotic therapy, with encouraging results in several studies.⁷⁻¹¹

Nevertheless, several studies have shown the low diagnostic value (sensitivity and specificity) of these breath tests.^{12,13} In order to improve the diagnostic value of these tests, and to avoid unnecessary restrictive diets or expensive antibiotic treatments, it is probably important to limit digestive fermentation in relation to ingested food prior to the test. Consensus papers indicated the need of restrictive diet before the test.^{1,14} The North American consensus on breath testing suggested to avoid “fermentable foods such as complex carbohydrates” 1 day before the breath test, and a 8–12 h fasting period.¹⁵ According to the 2022 Europeans Guidelines, “foods containing poorly absorbed, fermentable carbohydrates and dietary fibers should be avoided for a minimum of one day before breath testing”, with at least 8 h fasting.¹⁶ The European guidelines thus appear more specific in terms of diet prior to the test than North American ones. However, the evidence for such recommendations is mainly based on articles describing the effect of the ingestion of fibers and complex carbohydrates, while the effect of food containing fructose or lactose has been poorly studied.^{17,18}

In order to evaluate the importance of diet restrictions before breath test, we retrospectively analyzed our GBT results, around a protocol change in the restrictive diet proposed on the day before the test.

2 | SUBJECTS AND METHODS

Our retrospective study focused on a longitudinal cohort of 585 adult patients who performed a GBT between July 2017 and June 2021 in our lab. The initial cohort was composed of 616 GBT. In the

Key Points

- A strict diet without all kinds of fermentable food on the day before the test decreases significantly the baseline values of breath hydrogen and methane, compared to a diet without dietary fibers
- Glucose breath tests results are less frequently positive with a strict diet on the day before the test
- The impact of the pre-glucose breath test diet must be emphasized

event of repeated breath tests in the same subjects, only the first test was included, which brought the total number of subjects to 585. There was no exclusion criterion. During the test, all patients filled in an electronic questionnaire, containing **Irritable bowel syndrome-severity scoring system (IBS-SSS)**,¹⁹ and **Hospital Anxiety and Depression scale (HAD)**,²⁰ main symptoms, and previous medical history. Patients received an information notice from which they could oppose the use of their data. Data collection and analysis was carried out retrospectively. The study was approved by the HCL scientific and ethics committee. Data were compared between the two groups, before and after the implementation of the change in pretest restrictive diet instructions. Additionally, GBT results were compared based on the past history of patients (digestive surgery, diabetes), and between patients who did acknowledge smoking on the day of the test and those who did not.

The following instructions were given before the GBT: fasting at least 8 h before the test, no smoking and no intense physical activity on the day of the test, as described in North American Consensus. Laxative use was prohibited for 8 days before the test, and antibiotics for 4 weeks. For diabetic patients, rapid insulin injections were suspended on the day of the test.

A change in the pre-GBT diet protocol was introduced for tests performed after September 1, 2020, and the constitution of the groups A and B are determined by the date of the performance of the GBT. Before that date, patients were instructed to follow a fiber-free diet on the day before the test: potatoes, animal protein, dairy products, jam, honey, bread, butter, and dried biscuits were allowed (group A).

After that date, more drastic instructions with the exclusion of all fermentable foods (including dairy products, dried biscuits, potatoes) the day before the test were put in place. Patients were instructed to consume only rice, animal protein, hard cheese, vegetable oil on the day before the test. Drinks were limited to plain or sparkling water (group B).

On the day of the test, a mouth rinse with chlorhexidine was performed in order to limit the action of oral bacteria, in accordance with European guidelines.¹⁶ Then, two baseline breath samples were obtained, in glass tubes (EasySampler™, Quintron, Milwaukee, WI, USA). Patients then ingested 75 g of glucose dissolved in 250 mL of plain water, and breath samples were obtained every 15 min after

glucose ingestion during 2 h. Samples were then analyzed for hydrogen and methane levels (and CO₂ to ensure sample quality) with a BreathTracker SC Analyser (Quintron, Milwaukee, WI, USA).

We used the North American consensus cut-offs to define a positive breath test. GBT was considered positive in the event of an increase in the level of hydrogen ≥ 20 ppm compared to baseline values (positive test for SIBO), or in the event of methane value ≥ 10 ppm at any time during the test, including baseline values (positive test for IMO), within 120 min following ingestion.¹⁵

Two groups of GBT results were thus compared, before (group A) and after diet restrictions changes (group B).

Categorical variables are described in terms of numbers and percentages and continuous variables in terms of medians and interquartile ranges.

For categorical variables, comparisons between groups were performed using the chi-squared test or the Fisher exact test as appropriate. For continuous variables, comparisons between groups were performed by a *t*-test in case of normal distribution and sample size >30 in each group or by the non-parametric Mann-Whitney *U*-test otherwise. Normality was tested using the Shapiro-Wilk test.

A value of $p < 0.05$ was considered to be statistically significant. All analyses were performed using R software version 3.5.3 (R Foundation for Statistical Computing, Vienna, Austria).

3 | RESULTS

269 patients underwent GBT after a fiber-free diet only (group A), and 316 patients with the more restrictive fermentable food-free diet (group B). The 2 groups were similar in terms of sex, age, BMI, severity of symptoms, diabetes and digestive tract surgery. (Table 1). Baseline values of hydrogen and methane were significantly higher in group A than in group B (Table 2). Maximal increase of hydrogen levels after glucose ingestion was higher in group A than in group B (17 [31] ppm versus 12 [26] ppm, $p < 0.01$). Maximal increase of methane levels was significantly higher in group A (6 [8] ppm vs. 4 [7] ppm, $p < 0.01$). The % of positive GBT for methane and hydrogen was significantly higher in group A.

With regards to methane positivity, most of patients had elevated baseline values of exhaled methane (190 out of 246 cases, 77%).

GBT tests positive for both hydrogen and methane were found in 61 cases (10.4%): the majority were in group A (45/61: 74%). 72% of hydrogen positive GBT were also methane positive (92% in group A, versus only 43% (16/37) in group B, $p < 0.01$). On the other hand, 32% of methane positive GBT were also hydrogen positive, mostly in group A (45/106, 42% vs. 16/84, 19% in group B, $p < 0.01$).

Past history of digestive tract surgery was significantly associated with a higher prevalence of positive GBT for hydrogen (48% vs. 9%; $p < 0.01$), but not for methane (45% vs. 35%, $p > 0.05$). Small bowel resections ($n = 18$, including resection of the ileocecal valve) were associated with positive GBT for hydrogen in 56%, while left colon resections ($n = 12$) were associated with negative GBT for hydrogen (100%). 100% of GBT were positive

for hydrogen after gastric bypass surgery ($n = 18$), and 80% after sleeve gastrectomy ($n = 5$).

Diabetes ($n = 23$) was strongly associated with hydrogen positive GBT (55.6% vs. 12.1% in non-diabetics, $p < 0.01$), but not with methane (16.7% vs. 12.1%, $p = 0.06$).

Fifty-nine patients recognized smoking on the day of the test. Baseline hydrogen values were slightly but significantly higher compared to the nonsmoking group (11 [11] ppm vs. 7 [10] ppm, $p < 0.05$), unlike baseline methane values which were similar (10 [16] ppm vs. 11 [16] ppm, $p > 0.05$). The % of positive GBT for hydrogen and methane was similar in the smoking group compared to the nonsmoking group (19% vs. 11% and 32% vs. 32%, respectively, $p > 0.05$).

4 | DISCUSSION

The results of our retrospective study suggest the importance of the restrictive diet prior to GBT, in order to limit elevated hydrogen and methane baseline values in relation with food fermentation, and possibly false positive results. International recommendations do insist on the need to restrict the ingestion of fermentable complex carbohydrates.^{15,16} Despite the low level of evidence in the literature to increase the restriction to all FODMAPS, this attitude should be recommended, based on the results of our study.^{17,18} The importance of these diet restrictions should be emphasized for all carbohydrates breath test protocols, especially at a time when home tests are being commercialized. For methane evaluation, fasting exhaled values are important since they determine the positivity of GBT in the majority of cases (77% in our series). Takakura et al. found similar results: 86% of positive GBT or lactulose BT positive for methane had increased baseline values of methane.^{21,22} These authors proposed that a single fasting exhaled methane measurement could be used for the diagnosis and follow-up of IMO. In this case, the implementation of a strict diet without any fermentable foods on the day before the test seems even more important. For hydrogen evaluation (SIBO diagnosis), our results show the impact of a more restrictive diet on baseline exhaled hydrogen values, as well as on maximal difference to baseline values after glucose ingestion. However, the difference of GBT positivity between the two groups was not as dramatic as for methane. This was probably due to the fact that the GBT positivity for hydrogen is by definition independent of fasting exhaled hydrogen values.¹⁵

Patients with GBT tests positive for both hydrogen and methane were found predominantly in group A (74%), and concerned the majority (92%) of GBT positive for hydrogen in this group. Due to the fact that methanogeni archaea use hydrogen as substrate, this unequal repartition of this population in the two groups may explain a lesser effect of the diet modifications on the GBT positivity for hydrogen.²³

In our study, the impact of smoking on the day before GBT was mild at best, with a slight increase of hydrogen, but not methane, baseline values. This observation is in line with previous results showing that 15 min was the time needed for hydrogen baseline values to return to normal after smoking a cigarette.²⁴

	Group A (n = 269)	Group B (n = 316)	p-Value
Age (years), Mean (SD)	46 (16)	47 (16)	0.9
Gender			
Female	205 (76%)	226 (72%)	0.21
Male	64 (24%)	90 (28%)	
BMI (kg/m²) (mean [SD])			
<18	32 (12%)	29 (9%)	0.9
18–25	170 (63%)	200 (63%)	
25–30	39 (15%)	56 (18%)	
>30	28 (10%)	31 (10%)	
Past surgery of the digestive tract (N [%])			
Gastric bypass	10	9	0.9
Sleeve gastrectomy	3	4	
Ileocecal valve resection	8	13	
Left colon resection	8	10	
Diabetes			
Diabetes	14 (5%)	9 (3%)	0.8
IBS (n = 247)			
IBS-D	29 (30%)	30 (20%)	0.15
IBS-C	15 (15%)	31 (21%)	
IBS-M	29 (30%)	52 (34%)	
IBS-U	24 (25%)	37 (25%)	
IBS-SSS			
(Mean score [SD])	317 (85)	327 (88)	0.6
(IBS-SSS ≤175)	5 (5%)	8 (5%)	
(175 < IBS-SSS < 300)	37 (38%)	39 (26%)	
(IBS-SSS ≥300)	55 (57%)	103 (69%)	
HAD score			
	(n = 47)	(n = 300)	
HAD-A (mean (SD))	9.6 (4.4)	9.1 (3.9)	0.12
HAD-D (mean (SD))	5.8 (3.8)	5.4 (3.6)	0.45

Abbreviations: BMI: body mass index; HAD: Hospital Anxiety and Depression Scale; IBS: irritable bowel syndrome (IBS-D: diarrhea; IBS-C: constipation; IBS-M: mixed transit; IBS-U: undetermined).

TABLE 2 Glucose breath tests results before and after the change of instructions for the restrictive diet on the day before the test.

	Group A, before diet change (n = 269)	Group B, after diet change (n = 316)	p-Value
Mean baseline methane values (SD) (ppm)	14 (18)	8 (14)	<0.01
Mean baseline hydrogen values (SD) (ppm)	11 (14)	6 (8)	<0.01
Number of GBT tests positive for methane	144 (53%)	102 (33%)	<0.01
Number of GBT tests positive for hydrogen	49 (18%)	37 (12%)	0.03

Past digestive surgery is clearly a factor associated with positive GBT for hydrogen, as well as diabetes. It must be stressed that these positive tests may be in fact false positive, especially in the context of bariatric surgery, or after resection of the ileocecal valve, since glucose may enter more readily the colon before complete absorption in these conditions.^{25–28}

Our results point out to several implications: because carbohydrate breath tests have a relatively poor sensitivity and specificity, their interpretation should be used with caution. In order to improve

their clinical significance, and especially to limit the number of false positive tests, these tests should be performed as rigorously as possible, especially with regards to the pretest restrictive diet. Recent publications proposed the development of portable breath analyzer to assess lactose malabsorption: the correlation was good compared to a calibrated clinic-based breath analyzer.²⁹ Although this approach may be appealing to patients, the need of pretest restrictive diet should be emphasized. Similarly, the therapeutic options for SIBO (and possibly IMO) include rather complex diet restrictions

TABLE 1 Demographic characteristics of the patients undergoing glucose breath test before (group A) and after (group B) the change in the pretest restrictive diet protocol.

such as low FODMAPS diet, antibiotic therapies that may increase the bacterial antibiotic resistance pressure, and the frequent need to treat patients more than once when the diagnosis is made.^{1,30-33} Limiting the number of false positive SIBO or IMO diagnoses should be on the priority list of all researchers involved in this field.

There are specific limitations in our study. It remains retrospective and we did not compare the impact of pre-GBT diets in the same patients. The two groups were not performed at the same time, thus a time bias cannot be excluded. The use of proton pump inhibitors (PPI) was not recorded in our study, although PPI may be associated with GBT positivity, both for methane and hydrogen, due to the inhibition of gastric acid secretion.³⁴⁻³⁶ Similarly, the possibility of having patients on long-term restrictive diets because of disorders of brain gut interactions (DBGI) was not recorded. However, the % of DBGI patients was similar in both groups, as well as the severity of symptoms: from this, one would expect a similar proportion of long-term restrictive diet followers in both groups. Finally, in order to limit variations, we analyzed only the results of GBT: our findings may not be applicable to other breath tests.

In conclusion, this retrospective study suggests that a restrictive diet removing all kinds of fermentable foods the day before the test limits food fermentation and thus elevated baseline values of hydrogen and methane, more efficiently than a simple fiber-free diet. This diet should thus be clearly recommended to improve the diagnosis quality of GBT, and probably also for all other carbohydrate breath tests.

AUTHOR CONTRIBUTIONS

FM initiated the study, NM, PP, CM, AM, SR, CC and FM planned the study, NM collated the data, PP did the statistical analyses, NM and FM drafted the manuscript. Each author has approved the final draft submitted.

CONFLICT OF INTEREST STATEMENT

The authors have no conflict of interest to declare.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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