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Methane Production During Lactulose Breath Test Is Associated with Gastrointestinal Disease Presentation

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It has recently been determined that there is an increased prevalence of bacterial overgrowth in IBS. Since there are two gases (hydrogen and methane) measured on lactulose breath testing, we evaluated whether the different gas patterns on lactulose breath testing coincide with diarrhea and constipation symptoms in IBS and IBD. Consecutive patients referred to the gastrointestinal motility program at Cedars-Sinai Medical Center for lactulose breath testing were given a questionnaire to evaluate their gastrointestinal symptoms. Symptoms were graded on a scale of 0–5. Upon completion of the breath test, the results were divided into normal, hydrogen only, hydrogen and methane, and methane only positive breath tests. A comparison of all subjects and IBS subjects was undertaken to evaluate diarrhea and constipation with regards to the presence or absence of methane. This was further contrasted to Crohn's and ulcerative colitis (UC) patients in the database. After exclusion criteria, 551 subjects from the database were available for comparison. Of the 551 subjects ($P < 0.05$, one-way ANOVA) and in a subgroup of 296 IBS subjects ($P < 0.05$, one-way ANOVA), there was a significant association between the severity of reported constipation and the presence of methane. The opposite was true for diarrhea ($P < 0.001$). If a breath test was methane positive, this was 100% associated with constipation predominant IBS. Furthermore, IBS had a greater prevalence of methane production than Crohn's or UC. In fact, methane was almost nonexistent in the predominantly diarrheal conditions of Crohn's and UC. In conclusion, a methane positive breath test is associated with constipation as a symptom.

KEY WORDS: bacterial overgrowth; irritable bowel syndrome; inflammatory bowel disease; methane.

Small intestinal bacterial overgrowth (SIBO) is a condition in which the small bowel is colonized by excessive amounts of upper or lower gastrointestinal tract flora. Although there are many conditions associated with SIBO, recent studies have demonstrated an increased prevalence of SIBO in irritable bowel syndrome (IBS) (1), and it is a recognized cause of diarrhea in inflammatory bowel disease (IBD) (2–4).

One method of diagnosing SIBO is the lactulose breath test (LBT), where overgrowth is considered to be present if a rise greater than 20 ppm in breath hydrogen or methane concentration is observed within 90 min of oral administration of lactulose (5). Hydrogen and methane are common gases excreted during breath testing (6). Although hydrogen production appears more ubiquitous, methane production is seen in 36–50% of healthy subjects (7–9).

Although methane excretion is not present in all individuals, data suggest there may be clinical implications of these different gas profiles. For example, in diarrheal

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conditions such as Crohn's disease (CD) and ulcerative colitis (UC), methane excretion is uncommon (7, 8), whereas it is more prevalent in constipating conditions such as diverticulosis (10) and encoparesis (11). Recently, we reported a double-blind study where the relationship of abnormal lactulose breath test was compared in IBS by treating with an antibiotic (12). In this study, all the subjects with an abnormal breath test consisting of only methane gas production had constipation predominant IBS.

The goal of this study was to confirm and further investigate the relationship between gastrointestinal complaints (specifically, diarrhea and constipation) in IBS subjects with SIBO and gas excretion on LBT in a large prospectively collected database. The prevalence of gas excretion patterns in IBS and the predominantly diarrheal conditions of Crohn's disease and ulcerative colitis will also be compared.

MATERIALS AND METHODS

Patient Population. Consecutive patients referred for a lactulose breath test (LBT) to the Cedars-Sinai Medical Center, GI Motility Program from 1998 to 2000 completed a questionnaire designed to assess bowel symptoms as previously described (1) after approval from the institutional review board. Subjects were requested to rate the severity of nine symptoms (diarrhea, constipation, abdominal pain, bloating, sense of incomplete evacuation, straining, urgency, mucus, and gas) on a scale of 0–5, with 0 signifying the absence of the symptom. The questionnaire also inquired whether subjects had CD or UC. Of subjects reporting a history of IBD, only those whose diagnosis had been confirmed by the Cedars-Sinai Inflammatory Bowel Disease Center were included in the analysis. The diagnosis of IBS was identified if subjects fulfilled Rome I criteria (13). Subjects found to have both IBD and IBS were assigned to the IBD subgroup.

Subjects with conditions predisposing to rapid transit (short bowel syndrome, gastrectomy, etc), those taking narcotic medications, and those without evidence of overgrowth on LBT were excluded.

Lactulose Breath Test (LBT). After an overnight fast, subjects completed the questionnaire. A baseline breath sample was then obtained, after which subjects ingested 10 g of lactulose syrup (Inalco Spa, Milano, Italy, packaged by Xactdose, Inc., South Beloit, Illinois, USA). This was followed by 1 oz of sterile water. Breath samples were then collected every 15 min for 180 min. Each sample was analyzed for hydrogen, methane, and carbon dioxide gas concentration within 15 min of collection using a model SC Quintron gas chromatograph (Quintron Instrument Company, Milwaukee, Wisconsin, USA). CO_2 was analyzed to correct for the quality of the alveolar sampling.

Three abnormal gas patterns were described upon completion of the test: (1) Hydrogen-positive breath test: a rise in breath hydrogen concentration of >20 ppm within 90 min of lactulose ingestion (14–17). (2) hydrogen- and methane-positive breath test: a rise in both breath hydrogen and methane concentrations of >20 ppm within 90 min of lactulose ingestion, and (3) methane-positive breath test: a rise in breath methane concentration of >20 ppm within 90 min of lactulose ingestion.

Data Analysis. For all subjects with SIBO, mean diarrhea and constipation severity scores among the three abnormal gas patterns were compared. Based on symptom severity scores, the entire IBS group was further subdivided into diarrhea-predominant and constipation-predominant subgroups. Constipation-predominant IBS was identified if a subject's constipation severity score exceeded their diarrhea severity score, whereas the reverse applied for diarrhea-predominant IBS. Subjects who had a constipation severity score equal to the diarrhea severity score (indeterminate pattern) were excluded from the IBS subgroup analysis. The percentage of IBS subjects within each abnormal gas pattern who reported constipation-predominant or diarrhea-predominant symptoms was tabulated. The prevalence of methane production between the IBS subgroups was also compared.

Subsequently, a mean C − D score was obtained by calculating the difference between the constipation (C) and diarrhea (D) severity scores. This was used to examine the relative weight of constipation to diarrhea in individual subjects. The C − D score was compared among the three abnormal breath gas patterns in the group as a whole and among IBS subjects.

Finally, the prevalence of each of the three abnormal gas patterns was evaluated in subjects with CD and UC. The prevalence of methane production was contrasted between subjects with IBS and IBD.

Statistical Analysis. A one-way ANOVA was conducted to compare symptom severity scores among the three gas patterns on LBT. Prevalance data was analyzed with a chi-square test.

RESULTS

Subjects. At the time of analysis, 772 patients were referred for a LBT and entered into the database. One hundred eighty-three subjects with negative breath tests and 38 subjects either taking narcotic medications or with conditions predisposing to rapid transit were excluded. A total of 551 subjects remained for analysis. Of these, 78 carried the diagnosis of IBD (49 with CD and 29 with UC) and 296 without IBD fulfilled Rome I criteria for IBS. Of the subjects with IBS, 120 reported constipation-predominant symptoms, 111 had diarrhea-predominant symptoms, and 65 had a constipation severity score equal to the diarrhea severity score.

Bacterial Overgrowth Analysis. When the entire group of subjects with SIBO was evaluated ($N = 551$), the diarrhea severity scores differed significantly among the three abnormal breath test patterns (one-way ANOVA, $P < 0.00001$; Figure 1). Subjects who excreted methane reported significantly lower diarrhea severity scores than those who produced hydrogen only. Constipation severity also differed significantly among the breath test patterns ($P < 0.05$), with higher severity scores reported by subjects who produced methane.

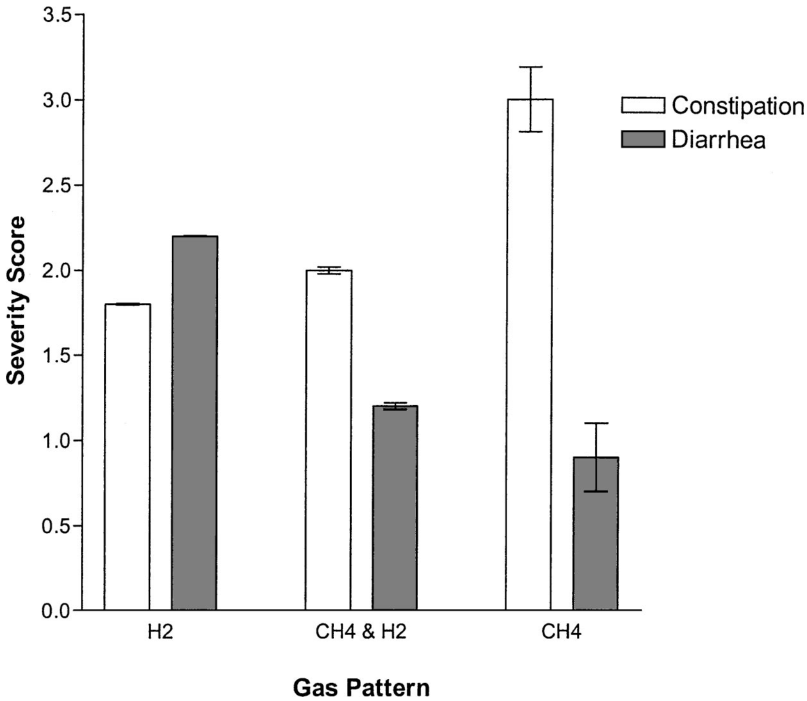

Fig 1. Mean diarrhea and constipation severity scores of all subjects ($N = 551$) with SIBO as a function of the of type of gas pattern produced on LBT. $P < 0.00001$ for trend in reduction of diarrhea with the presence of methane (one-way ANOVA). $P < 0.05$ for the trend towards increasing constipation with the presence of methane (one-way ANOVA).

Among all IBS subjects ($n = 296$), diarrhea severity scores also differed similarly (one-way ANOVA, $P < 0.001$) with a lower severity reported by those who produced methane than those who produced hydrogen gas alone (Figure 2).

When the C-D score was evaluated as a reflection of the degree of constipation with respect to diarrhea, the effect of methane was even more obvious (Figure 3). In both the total group and the IBS subjects, constipation was by far the prevailing symptom in individuals, whereas diarrhea was the prevailing symptom in subjects with only hydrogen.

When IBS subgroups were compared, constipation-predominant IBS was reported by 91 (37%) of the hydrogen-excreting subjects, 23 (52.3%) of the hydrogen- and methane-excreting subjects, and 6 (100%) of the methane-excreting subjects. By contrast, diarrhea-predominant IBS was observed in 105 (42.7%) of the hydrogen excretors, 6 (13.6%) of the hydrogen and methane excretors, and none of the methane excretors (Figure 4).

Inflammatory Bowel Disease and Methane. The predominant gas excreted by patients with IBD was hydrogen alone, detected in 47 of 49 subjects (95.9%) with Crohn's disease and 29 of 29 (100%) of subjects with ulcerative colitis (Figure 5).

Methane Production in Subjects with IBS and IBD. The percentage of subjects with IBS who produced each of the three gas patterns was tabulated. Of 296 IBS subjects, 246 (83.1%) produced hydrogen gas alone, 44 (14.9%) produced hydrogen and methane gas, and 6 (2.0%) produced methane gas alone. Methane production depended significantly upon whether or not subjects had IBS or IBD. IBS subjects were more likely to produce methane gases than subjects with ulcerative colitis or Crohn's disease (OR 7.7, CI 1.8–47.0, $P < 0.01$ Table 1).

DISCUSSION

In this study, we found a significantly higher proportion of breath methane excretion during LBT among subjects with constipation than those with diarrhea. Methane

TABLE 1. COMPARISON OF PREVALENCE OF METHANE TO NONMETHANE GAS PRODUCTION BETWEEN SUBJECTS WITH IBS AND IBD*

Disease type	CH_4	Non-CH_4
IBS ($N = 296$)	50	246
UC or CD ($N = 82$)	2	76

*$\chi^2 = 9.4$, OR = 7.7, CI: 1.8–47.0, $P < 0.01$.

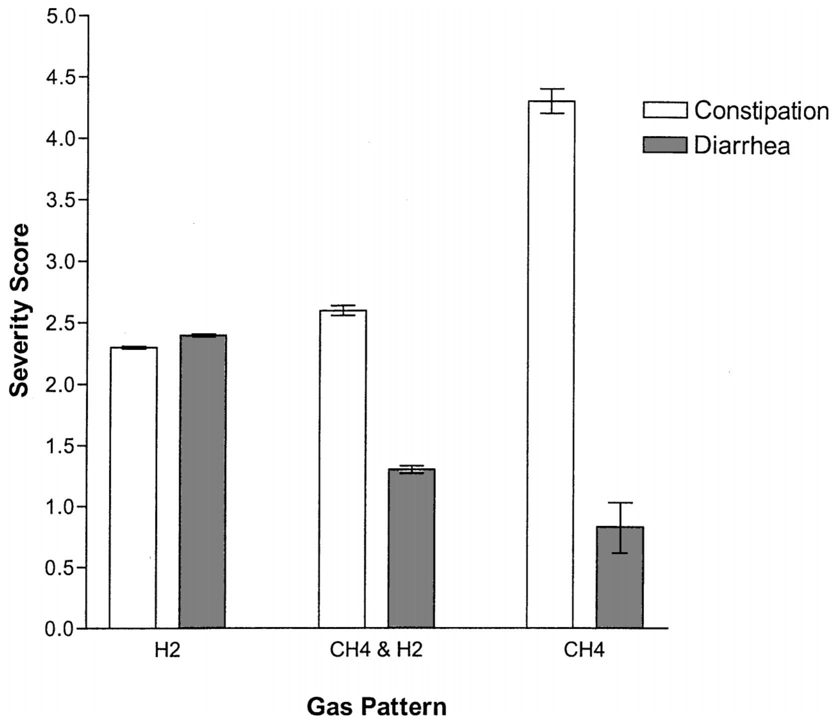

Fig 2. Mean diarrhea and constipation severity scores of IBS subjects ($N = 296$) with SIBO as a function of the of type of gas pattern produced on LBT. $P < 0.001$ for trend in reduction of diarrhea with the presence of methane (one-way ANOVA). $P < 0.05$ for the trend towards increasing constipation with the presence of methane (one-way ANOVA).

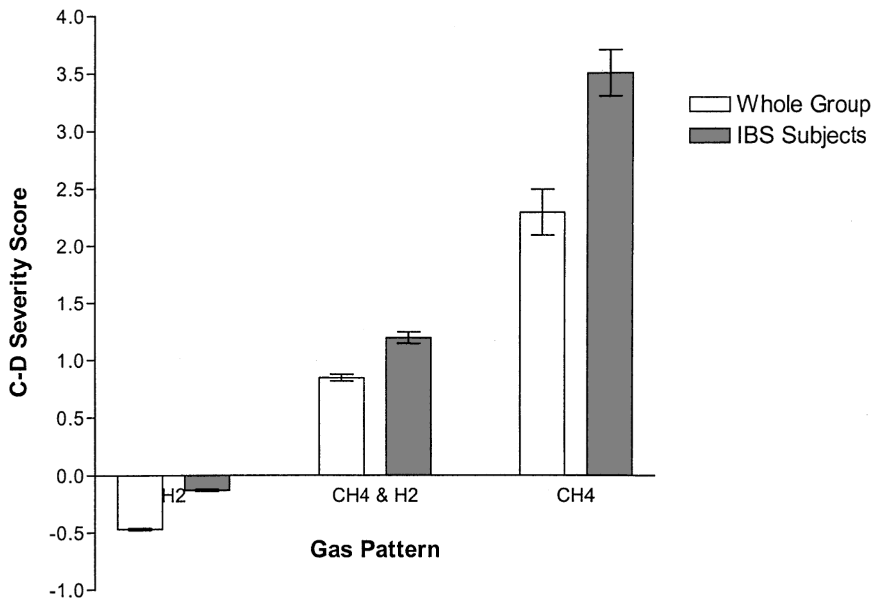

Fig 3. Mean constipation − diarrhea (C − D) severity score for the whole group ($N = 551$) and IBS subjects ($N = 296$) as a function of the type of gas pattern produced on LBT. $P < 0.00001$ for trend in C−D for whole group (one-way ANOVA). $P < 0.0001$ for trend in C−D for IBS subjects (one-way ANOVA).

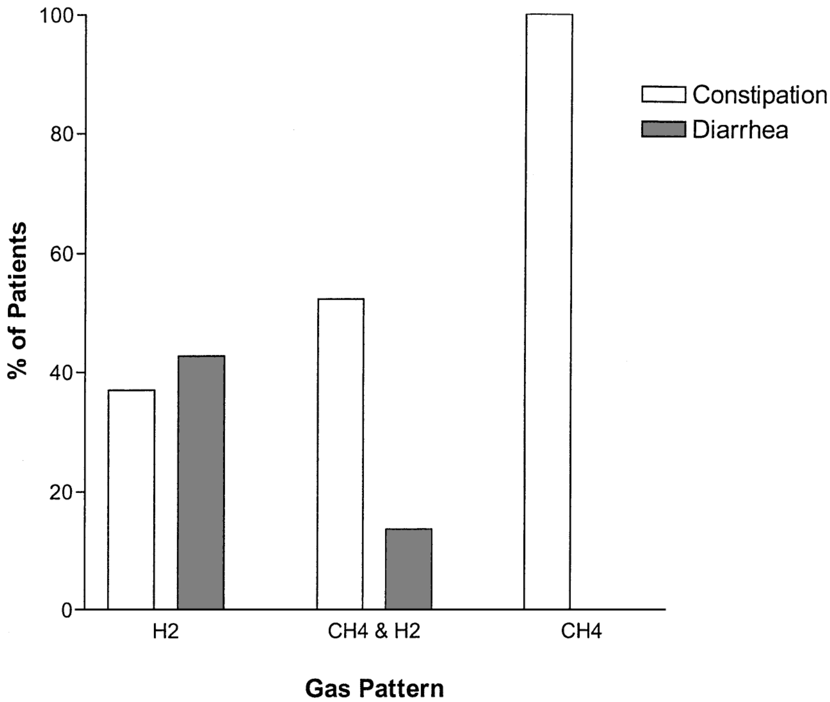

Fig 4. Percentage of IBS subjects within each gas pattern who reported constipation versus diarrhea predominant symptoms. (Chi-square for difference between constipation and diarrhea predominant IBS = 16.6, $P < 0.001$.)

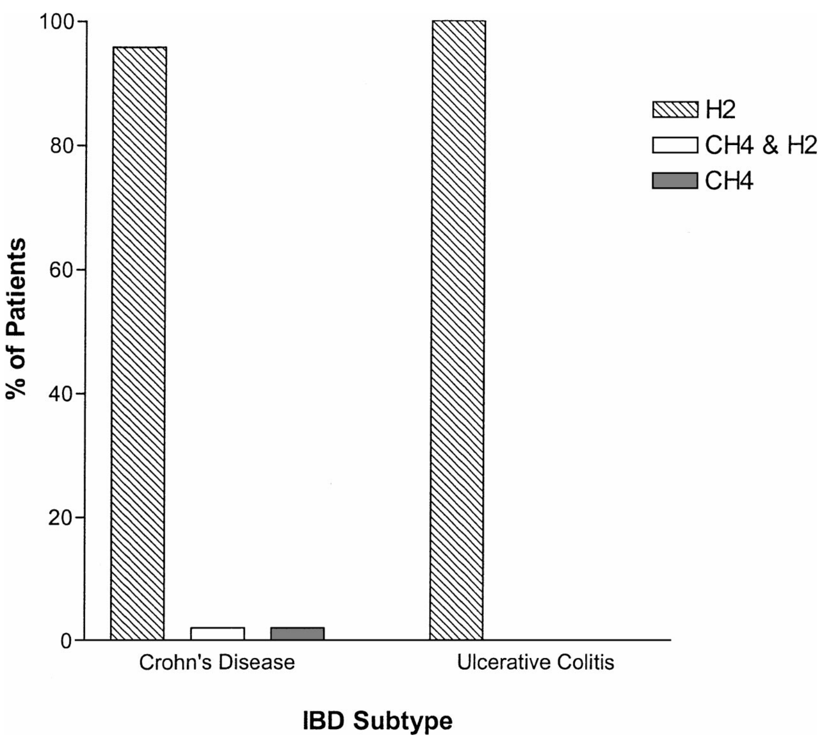

Fig 5. Percentage of subjects with IBD who produced each of the three abnormal gas patterns on LBT.

excretion among subjects with SIBO and IBS was associated with higher constipation severity scores and lower diarrhea severity scores, as well as with the constipation-predominant subgroup of IBS. The findings from this large prospectively collected database continue to support findings similar to a recent study (12). By contrast, methane excretion was infrequent in diarrhea-predominant IBS and virtually absent in IBD.

Relationships between certain gastrointestinal diseases and methane excretion have been described. Previous investigators have observed the prevalence of breath methane excretion to be significantly lower in subjects with diarrheal conditions such as Crohn's disease and ulcerative colitis (7, 8) compared to healthy controls. By contrast, an increased proportion of breath methane excretion has been observed in constipating conditions such as encopresis (11) and high stool concentrations of methanogens have also been found in subjects with diverticulosis (10). The IBS literature is less clear, but one paper suggests an increased prevalence of methane excretion among IBS patients complaining of constipation compared to those who complained of diarrhea (8).

Methane is produced extensively by strict anaerobic bacterial fermentation in the gut and generally has not been found to have a physiologic role in humans (18). Since approximately 20% of colonic methane is excreted via the breath, breath methane analysis has been used as an indirect assessment of intracolonic bacterial metabolism (18). The predominant methanogenic bacteria found in humans is *Methanobrevibacter smithii* (10, 13, 19, 20), which preferentially colonizes the left colon (20–22). It is possible that the lower prevalence of methane excretion in IBD and diarrhea-variant IBS may be an artifact of colonic purging. Diarrhea theoretically may inhibit proliferation of methanogenic bacteria. In support of this hypothesis, colonic lavage can reduce and even eliminate methane excretion for extended periods of time (11). Therefore, one could argue that breath methane may be a marker of constipation.

Another possibility is that the methane production is more proximal in origin. It has been suggested that methane, unlike hydrogen production, does not usually vary with the ingestion of nonabsorbable carbohydrates such as lactulose (18). However, the rise in breath methane excretion we observed upon lactulose ingestion suggests otherwise. Since lactulose would not be expected to reach the left colon (the location of the *Methanobrevibacter smithii*) within 90 mins, other bacteria may be involved in methane excretion in our subjects with small intestinal bacterial overgrowth. For instance, *Clostridia* and *Bacterioides* species are known to liberate methane by using hydrogen produced by regional organisms to reduce carbon dioxide (23). This warrants further investigation, however.

There is further evidence for the role of intestinal gases in symptoms. It has been postulated that the absence of methanogenic flora may be associated with an increase in gastrointestinal complaints (24). Evidence for this relationship derives from studies where patients with lower rates of methanogenesis tend to have higher concentrations of sulfate-reducing bacteria (25–27). The product of bacterial sulfate reduction, hydrogen sulfide, may damage the colonic epithelium. In fact, hydrogen sulfide has been suggested to have a role in the pathogenesis of ulcerative colitis (28, 29). It is known that sulfate-reducing bacteria and methanogenic bacteria compete for hydrogen in the colon via mutually exclusive pathways (26, 27). Thus, if the balance of flora is skewed toward sulfate-reducing bacteria, or if the sulfate-reducing bacteria outcompete the methanogenic bacteria for the available hydrogen substrate needed to support their metabolism, higher concentrations of hydrogen sulfide may be produced, resulting in inflammation and epithelial cell damage. Hydrogen sulfide, however, is not measured by the LBT using current technology.

The observation of methane being associated with constipation in IBS and in general patients with gastrointestinal disease suggests potential therapeutic options. Perhaps altering the balance of organisms to either favor or reduce methanogen populations may benefit subjects with constipation or diarrhea, respectively. Based on this paper, an obvious potential application is in inflammatory bowel disease, whereby observing the effects of administering methanogens may produce benefial results. These potential applications need to be further researched.

In summary, this study demonstrates that the presence of methane on LBT is associated with constipation as a symptom. Likewise, diarrhea and conditions that produce this symptom, such as IBD, are associated with hydrogen production on LBT. Whether the respective gas excretion is simply a marker of symptoms or whether the type of flora causally determines symptoms is as yet unknown.

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