Fructose Malabsorption Is Associated with Lower Plasma Folic Acid Concentrations in Middle-Aged Subjects, *Maximilian Ledochowski,[1*] Florian Überall,[2] Theresia Propst,[3] and Dietmar Fuchs[2]* (Departments of [1] Clinical Nutrition and [2] Gastroenterology, and [3] Institute of Medical Chemistry and Biochemistry, University of Innsbruck, A-6020 Innsbruck, Austria; * address correspondence to this author at: Institute of Medical Chemistry and Biochemistry, University of Innsbruck, Anichstrasse 35, A-6020 Innsbruck, Austria; fax 43-512-504-2017, e-mail Maximilian.Ledochowski@uibk.ac.at)

Fructose malabsorption *(1, 2)*, which is frequently seen in the general population, is characterized by the inability to absorb fructose efficiently. As a consequence, fructose reaches the colon where it is broken down by bacteria to short-chain fatty acids, CO_2, and H_2. Bloating, cramps, osmotic diarrhea, and other symptoms of irritable bowel syndrome are the consequence. It is believed that up to 36% of the European population has fructose malabsorption in a more or less severe form, and approximately one-half of affected individuals are symptomatic *(3)*. We recently found that fructose malabsorption is associated with early signs of mental depression *(4)*. Because folic acid deficiency may contribute to the development of mental depression *(5)*, we examined folic acid concentrations in subjects with fructose malabsorption.

We studied 73 otherwise healthy adults (47 women, 26 men), ages 40–81 years (mean 52.0 years; SD, 9.1 years), who visited physicians' offices for a medical health check-up. None of the patients showed signs of inflammatory bowel disease or any other chronic or infectious diseases, and none was receiving medication. No vitamin supplements were taken because this was an exclusion criterion. Blood samples were collected after an overnight fast. Breath H_2 content was measured with a Bedfont gastrolizer (Bedfont) *(6, 7)*. A baseline H_2 breath test was performed after a 12-h overnight fast. An oral dose of 50 g of fructose was given in 250 mL of tap water. All tests were performed between 0800 and 0830, and body weight and height were measured. After the fructose load, H_2 exhalation was monitored in 30-min intervals for at least 2 h. Maximum H_2 exhalation after fructose load was registered, and the differences from baseline (ΔH_2) were calculated. Fasting plasma samples were drawn into a 5-mL EDTA syringe, and folic acid was measured by an immunoassay (Elecsys System 2010; Boehringer Mannheim) according to the manufacturer's instructions.

The cutoff point for the diagnosis of fructose malabsorption was an increase in breath H_2 >20 μL/L above baseline *(1, 8)*. Subjects with increases in breath H_2 ≤20 μL/L above baseline were considered normal fructose absorbers.

In 46 patients (17 men and 29 women; ages 51.7 ± 9.3 years, mean ± SD), breath H_2 concentrations increased >20 μL/L above baseline fasting values; they were therefore classified as fructose malabsorbers. The remaining 27 subjects (9 men and 18 women, ages 52.4 ± 9.1 years) were normal fructose absorbers (ΔH_2 <20 μL/L). Plasma folic acid was significantly lower in fructose malabsorbers (7.14 ± 2.2 μg/L, mean ± SD) than in normal fructose absorbers (9.1 ± 3.7 μg/L; P <0.01, Student t-test; P <0.02, nonparametric Mann–Whitney U-test; Fig. 1.). The lower plasma folic acid concentration in fructose malabsorption was independent of sex or age.

Low plasma folic acid concentrations may be attributable to dietary deficiency or malabsorption of folic acid. Another cause may be an unfavorable bacterial composition in the gut because folic acid derived from colonic bacterial metabolism is a major source of resorbed folic acid. Because malnutrition is highly unlikely in our study population, the lower folic acid concentrations in fructose malabsorbers compared with normals are probably attributable to malabsorption of alimentary folic acid or changes in intestinal bacterial colonization. Fructose malabsorption is known to accelerate gastrointestinal transit when patients are exposed to fructose, thus reducing the contact time that is necessary for the absorption of (micro)nutrients. On the other hand, fructose malabsorption leads to a profound change in bacterial colonization, especially in the colon. Because a substantial amount of folic acid derives from intestinal bacteria, it seems reasonable that a change in the population of gastrointestinal bacteria could lead to a change in the plasma concentrations of folic acid. Fructose malabsorption can be seen in approximately one-third to one-half of the Western European population; therefore, fructose malabsorption could be a major cause of lower folic acid status.

Folic acid deficiency, like vitamin B_6 and B_{12} deficiencies, may increase concentrations of homocysteine *(9)*, which is known to be an additional risk factor for cardiovascular disease *(10)*. Further studies are needed to determine whether fructose malabsorption is indeed associated with increased plasma homocysteine concentrations. In addition, folic acid deficiency has also been shown to increase the risk for development of neural tube defects in newborns *(11, 12)*, and folic acid supplementation was found to reduce the relative risk for the development of

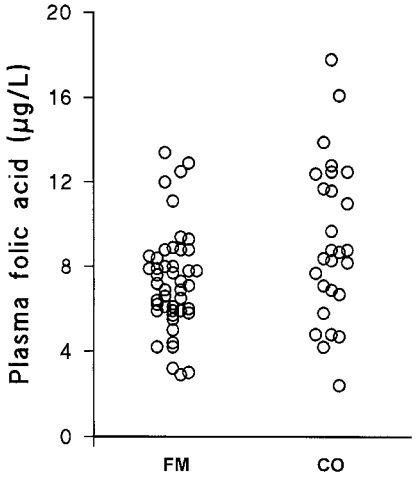

Fig. 1. Plasma folic acid concentrations in individuals with (*FM*) and without (*CO*) fructose malabsorption.

colon carcinoma *(13)*. These findings suggest that fructose malabsorption could be a risk factor in the development of these diseases.

Because bacterial metabolism alters folic acid status and intestinal bacterial colonization is altered by nutritional factors and carbohydrate malabsorption syndromes, dietary measurements should not rely solely on folic acid supplementation but should also consider carbohydrate malabsorption syndromes, especially fructose malabsorption. It is suggested that fructose malabsorption be considered in the elderly with folic acid deficiency. Further studies will be necessary to compare not only folic acid concentrations but also homocysteine, vitamin B_{12}, and hematological indices in patients with and without fructose malabsorption.

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Comparison of the Diagnostic Accuracy of Three Commercially Available Enzyme Immunoassays for Anti-p53 Antibodies, *Jacques Rohayem,[1*] Karsten Conrad,[1] Thomas Zimmermann,[2] and Karl-Heinz Frank[1]* ([1] Institute for Immunology and [2] Department of Surgery, Medical Faculty "Carl Gustav Carus", Technical University Dresden, Dresden 01101, Germany; * author for correspondence: fax 49-351-8832778, e-mail rohayem@rcs.urz.tu-dresden.de)

The *p53* tumor suppressor gene encodes a 53-kDa nuclear phosphoprotein that is thought to protect cells against the accumulation of genetic alterations *(1)*. The p53 phosphoprotein is involved in cycle arrest, apoptosis, inhibition of tumor growth, and preservation of genetic stability *(2)*. Abnormalities of the *p53* gene are reported to be the most common genetic alterations in human cancer *(3, 4)*. Mutated *p53* gene encodes for mutant p53 proteins that may serve as targets of the host immune system as tumor-specific antigens *(5)*. However, accumulation of the p53 protein in the cell is considered the main cause of anti-p53 antibody production *(6)*. The presence of anti-p53 antibodies has been demonstrated in sera of patients with various cancers *(7–9)*, including lung *(10)*, breast *(11, 12)*, liver *(13)*, colorectal *(14)*, and prostate cancer, and blood cell malignancies *(15)*.

Anti-p53 antibodies initially were detected by immunoblot and immunoprecipitation with extracts of transformed cells as a source of antigen. In recent years, various ELISAs have been described that use mutant *(8)* or wild-type *(16)* p53 as antigen, solid-phase or sandwich methods, and prokaryotically or eukaryotically expressed p53 proteins.

Our purpose was to evaluate the potential of the tests to provide correct diagnostic classification *(17)*. We studied three commercial ELISAs for anti-p53 antibodies by use of ROC curve analysis. We selected 72 patients presenting with suspicion of malignancy (unexplained body weight loss and chronic fatigue associated with chronic rectal hemorrhage and/or intermittent obstipation, hemoptysis, and/or pathognomonic aspect of tumoral development on chest radiographs). Ethics approval by the Ethics Commission of the University of Dresden (Germany) according to the Helsinki Declaration of 1975 as revised in 1996 as well as informed consent from the study subjects was obtained. Blood samples from patients with suspicion of lung cancer and/or other malignancies were collected during the control examination for occupational lung diseases at the Medical Opinion Community Niederdorf (Germany). Blood samples from patients with suspicion of colorectal malignancy were collected in the Department of Surgery of the University Hospital of Dresden (Germany). Forty-nine percent of the patients recruited presented symptoms of colorectal malignancy (men, n = 15; women, n = 20), 33.3% presented symptoms of lung malignancy, and 18.1% showed suspicion of melanoma (n = 1), blood cell malignancies (n = 2), or oral (n = 3), esophageal (n = 2), urogenital (n = 3), or hepatic (n = 2) cancer. The mean age (± SE) was 68 years (± 3 years) for the men and 71 years (± 4 years) for the women. Before therapy was started, blood samples were collected by venipuncture. After centrifugation, serum was stored at −28 °C. Later in the course of the disease, diagnosis of cancer was confirmed by histopathological examination.

Young, healthy blood donors, thus with a low probability to harbor "silent" cancer, were selected as a control population (n = 72). The mean age (± SE) was 19 years (± 1 year). Blood samples from healthy donors and patients with suspicion of cancer underwent the same procedure.

The anti-p53 antibody value was assessed for each serum sample with three different ELISAs: test A, a solid-phase ELISA using eukaryotically expressed wild-type p53 (PharmaCell; distributed by Coulter-Immuno-